About the book

Part one (*Health Care : Between Innovation And Cost-Effectiveness*) is an update of a report authored by A. Sasson and published in 2008 by the United Nations University Institute of Advanced Studies (Yokohama, Japan), with particular emphasis on how medical biotechnology can improve the whole health-care system – a major concern of all governments. Better and accurate diagnostics, used in the earliest detection of diseases, effective new vaccines (including candidate vaccines against HIV/AIDS, tuberculosis and malaria), innovative and targeted drugs, therapies of the future based on stem cells and new approaches to combat cancers are presented.

Sequencing the full (diploid) human genome at a cost which is falling steadily, is opening new vistas for the association of gene mutations with diseases or predictability of illnesses. It is not however that simple. Personal medicine may remain utopian, while precision medicine – the result of the combination of advanced imagery, bioinformatics and human genomics – is becoming a reality. The Human Genome Project has also shown the complexity of gene structure, expression and interaction, and opened the way to even more ambitious endeavours such as the Genographic Project, the Personal Genome Project, the International Cancer Genome Consortium. It also highlighted the importance of epigenetics and the need to devote much attention to the study of the human epigenome.

Progress in all these areas has led the pharmaceutical industry to review its strategies in order to cope with new developments and to contribute to health-care effectiveness. Mergers and acquisitions, the purchase of biotechnology companies, restructuration, and the review of business models aim to give the highest priority to innovation in the search of really effective and targeted drugs (when the innovation pipeline tends to dry out and the "blockbuster drug" model is becoming obsolete). These measures also aim to face the harsher competition from generics and biosimilars manufacturers, as well as the loss of patents, and the pressure from governments regarding the decrease of drug prices. Increasing their presence in emerging markets is a short-term solution for the big pharmas, but the medium- and long-term solution remains research and development and innovation in the life sciences and medical biotechnology.

Part two (*Food And Nutrition*) highlights the perceptions of the complex relationships between, food, nutrition and health, which are rooted in the traditions and cultures of peoples and civilizations. Food and eating habits are borrowed from one culture and adapted to another. Family values, and flavours and meals of the past are submitted to globalization and uniformization, even though a number of associations believe that finding inspiration in the past could help applying good ideas to better produce and consume food. With the help of food research and biotechnology, globalization is turning foodstuffs into global products that can also suit local tastes and eating habits. Another global trend is to have a healthier diet : food companies commit themselves to produce healthier foodstuffs, without losing too much on the pleasure and flavour side, fast-food firms adapt their strategies and an industry is being developed at the interface of food and pharmaceuticals.

While it should not be forgotten that the major challenge for humankind is the eradication of hunger, under- and malnutrition across the world, obesity is now a serious concern of all nations. Henceforth the need for an increasing number of people to eat less and better. Dietary recommendations exist at national and international level and concern the wide range of foodstuffs and beverages, produced by the agrifood industry as well as through traditional know-how. In many cases, the food and beverage processes can be improved by biotechnology, e.g. the fermentation and preservation processes. Many examples of current foodstuffs and beverages, their history, production and nutritional value are presented.

Consumers are very concerned by the improvement of the quality of their foodstuffs and particularly their potential benefit for health and in preventing diseases of all sorts. They are exposed to nutritional and health claims by the food producers, but they must be able to make a difference between allegations and deeds. This is where food science and nutrition, biomedical research and biotechnology can establish proven health benefits of the so-called functional foods or beverages, food additives and/or supplements. In other words, the current evolution of food supply and dietary habits requires a nutrivigilance system aimed at better protecting the consumer and that could be compared with pharmacovigilance dealing with the consumption of medicines. In a way, we are following the very old precept of Hippocrates : "Your food should be your first medicine."

Eating natural products, with no artificial inputs for their production, is giving a strong impetus to organic agriculture worldwide : the acreages devoted to biofarming are increasing and, despite their high prices, bioproducts are popular. There are nevertheless problems of supply, distribution, cost and access, and the superiority of bioproducts in terms of health benefits is not always scientifically proven.

Fair trade, which represents a tiny part of global food and beverage trade, is also becoming significant across the planet. It offers a broad range of products, including those derived from the large biological diversity of several tropical countries. They are sold at a price that brings higher income and other social benefits to poor and small farmers from the developing world participating in the fair-trade networks. Analysts consider that fair trade should bring more income to poor farmers. It is also expected that the use of biological diversity should lead to more mutual benefit sharing between those who own this heritage and those who prospect it for novel foods, pharmaceuticals, cosmetics, fragrances and flavours.

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Albert SASSON . **HEALTH CARE, FOOD AND NUTRITION**
Opportunities And Challenges For The Life Sciences And Biotechnology

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Publication supported by :

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The opinions expressed in this publication are those of the author and do not necessarily reflect those of the Hassan II Academy of Science and Technology of Morocco and of the Centre for Global Sustainability Studies (CGSS)-Universiti Sains Malaysia (USM), Penang, Malaysia.

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“La victoire avant tout sera
de bien voir au loin
et tout voir de près
et que tout ait un nom nouveau.”

Guillaume Apollinaire
French poet (1880-1918)

“Victory will result, above all,
from foreseeing correctly,
from having a close look at everything,
and that everything has a new name.”

PREFACE

The Hassan II Academy of Science and Technology has been created by a Royal Decree on October 6, 1993 and enjoys the protection and tutorship of His Majesty the King of Morocco. The Academy's missions are to promote scientific and technological research particularly through assessing, supporting and funding research programmes and projects; to issue recommendations on the national science and technology policy and priorities; and to contribute to the dissemination of scientific culture and progress.

In addition to the *Proceedings* of its sessions, and in particular of its annual plenary session devoted to a specific theme of worldwide relevance, the Academy publishes a *Bulletin* and a *Newsletter*. It also promotes and supports publications and reports dealing with subjects of relevance to its scientific Sections.

The Centre for Global Sustainability Studies (CGSS), inaugurated on December 14, 2009 by the Minister of Higher Education on the campus of Universiti Sains Malaysia (USM) in Penang, has been assigned the mission to :

- contribute, through education for sustainable development, scientific assessment, policy research and capacity building, to efforts to resolve pressing problems confronting Malaysian society and the global community today and into the future;
- address the impact of climate change and ecosystems degradation on the economies of developing countries;
- explore how developing countries can become or remain low-carbon societies while addressing development challenges.

The CGSS-Universiti Sains Malaysia is an international centre bringing together global expertise as well as knowledge from within USM – a university of global standing – and Malaysia. It adopts a multidisciplinary approach to research issues through bringing together experts from a wide range of social and natural sciences in order to understand and resolve some of the key challenges of sustainable development. It also seeks to collaborate with institutions working in and outside Malaysia to leverage on complementarities and to share information and know-how.

Both the Academy and CGSS-USM are very glad to support the publication titled *Health Care, Food And Nutrition. Opportunities And Challenges For The Life sciences And Biotechnology*, authored by Professor Albert Sasson, a founding member of the Hassan II Academy of Science and Technology and director of the Academy's Section of Life Sciences and Technologies. Professor Albert Sasson enjoys a long-standing reputation for disseminating scientific and technological knowledge, especially with respect to biotechnology in developing countries over the last 38 years.

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HEALTH CARE :
BETWEEN INNOVATION AND COST-EFFECTIVENESS

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HEALTH-CARE SPENDING AND REFORM : A HEADACHE FOR GOVERNMENTS

Health-care spending in developed countries

All-developed countries have seen their populations age significantly, and the combination of ageing populations, rising expectations for better health care and technological innovation have pushed up costs over the last 50 years. Kenneth Clarke, one of the United Kingdom's most highly regarded former health secretaries, stated : "in my experience, at international meetings of health ministers, all they and the other people there ever talk about is how to control costs – and they never really seem to find a way to it" (Timmins, 2008).

Indeed the health systems of Western Europe, which already offer universal health coverage, are struggling to cope with soaring costs. Nicolaus Henke, head of the London health practice for the management consultants McKinsey, has underlined that over 50 years health spending has outstripped growth in the economy by 2% a year on average in every country in the Organisation for Economic Cooperation and Development (OECD). The trend, he stated, is "startling." If it continues, "by 2050 most countries will spend more than 20% of gross domestic product (GDP) on health care. The United States will be spending well over 30%." By 2100, the American health-care spend would take 97% of national income, the United Kingdom's two-thirds. "That is difficult to conceive," stated N. Henke. "But in 1960, most observers would have said that 40 years on it would have been pretty inconceivable that Western Europe on average would be spending 9% of GDP on health. But that, of course, has happened" (Timmins, 2008).

The marked increase in life expectancy at birth explains the pressure on the health-care systems. It soared, for males, from 63.5 years in 1950 to 71.0 years in 2000 and would reach 79.4 years in 2050; for females, from 68.6 years in 1950 to 78.5 years in 2000, and would reach 85.4 years in 2050, in developed countries, according to United Nations and OECD sources. There are also social reasons to higher spending on health care.

In the case of the United States, for instance, President Barack Obama wanted to bring 46 million or so uninsured Americans into his country's health-care system. This is also a major challenge of his presidency which has of course a very high cost, particularly amidst a major economic downturn; but social equity must prevail.

The American health-care system, which consumes about 16% of national economic output (about US$7,000 per person per year in 2007, half of this amount from public funding) is by far the most expensive in the world. The Congressional Budget Office (CBO) estimated that on current trends spending on Medicare and Medicaid, the government schemes for the old and the poor, would rise from 4% of GDP in 2007 to 12% in 2050. This might lead to a fiscal disaster and therefore reform of the health-care system was a priority of President B. Obama's government (*The Economist*, 2009 f).

For the defenders of the system, countries should expect to spend more on health care as people age. Americans are wealthy enough to choose extra health care over other things. Their spending approach calls forth the discovery and speedy adoption of valuable new drugs, devices and procedures. The doubters consider that granted, medical inventions are readily embraced by American doctors and patients. But if the system in general were providing value for money, the vast expenditure would at least be reflected in a healthier population than in more frugal countries. Comparisons with other rich countries and within the United States show that the American health-care system is not only growing at an unsustainable pace, but also provides questionable value for money (*The Economist*, 2009 f).

Passage of the US health-care bill : a historic accomplishment

"It is abnormal for any industry to throw back upon the community the human wreckage due to its wear and tear, and the hazards of sickness … should be provided for through insurance," stated Theodore Roosevelt in 1912, at the beginning of the progressive era. The work of building a social safety net for the industrial age proceeded for the following 50 years. The excesses of that endeavour brought the pendulum in the opposite direction during the Ronald Reagan era, and the need for a new safety net became necessary. President Barack Obama's signature on health-reform legislation in the East Room of the White House on 23 March 2010 was a historical achievement. The bill represented a national commitment to the worst elements of the current system. It aimed to provide coverage to tens of millions of insured Americans, prevent the worst insurance

company abuses, and begin to come to grips with relentlessly rising costs, while slightly reducing future deficits. The bill will have to be modified in the future, but, however flawed and imperfect, the health-care reform bill was a sign that major, concerted public reforms were once again possible, and that the difficult work of transforming America to compete successfully in a new world with many challenges could begin (Klein, 2010).

The new law will be judged on whether it actually fulfils its promise of a better and fairer health-care system or instead sends costs skyrocketing, and opens up a world of unintended medical implications. Economists and health-care experts have long agreed on the ails of the health-insurance system in the United States. It leaves too many people out. On the other hand, by some estimates, as much as 30% of the more than US$2 trillion Americans spend on health care each year goes towards treatments that are unnecessary and even harmful. This staggering investment does not result in a better health or life expectancy that lag behind those of most other industrial democratic countries (Tumulty et al., 2010).

The most ambitious element of the new health-care bill – the expansion of coverage to an additional 32 million Americans – will not even take effect until 2014. "It'll take four years to implement fully many of these reforms because we need to implement them responsibly," President B. Obama said as he prepared to sign the bill. Governors in all 50 States were looking at the prospect of having to set up new health-insurance market places for small businesses and individuals who had no coverage at work. In short, the bill is far from a finished product, it is only a start according to health experts (Tumulty et al., 2010).

The implementation schedule of the reform is the following :

- 2010

The uninsured : receive immediate access to coverage through high-risk pools if they are uninsured because of pre-existing conditions; children can remain on parents' plans until they are 26 years old.
Insurers : barred from removing coverage when a person becomes sick, denying coverage to children with pre-existing conditions and imposing lifetime coverage caps.
Employers : small businesses can receive tax credits to purchase insurance for employees.
Medicare prescription-drug beneficiaries : receive a US$250 rebate when they hit the "doughnut hole" gap in drug coverage (currently, when enrollees pass US$2,700 in costs, they lose coverage until they reach US$6,154).

- 2011
Insurers: required to spend at least 80% of premiums on medical services.
Medicare prescription-drug beneficiaries : receive a 50% discount on brand-name drugs while in the doughnut hole.

- 2013
Tax payers : Medicare payroll taxes increase and expand to include unearned income for individuals making more than US$200,000 and families earning more than US$250,000.

- 2014
The uninsured : most Americans are required to be covered or pay a penalty; families can receive subsidies to buy insurance if they earn up to four times the federal poverty level (currently about US$88,000 a year); individuals and small businesses can buy packages through state exchanges.
Insurers : prohibited from refusing to sell policies and limited in their ability to set prices on the basis of health status.
Employers : businesses with 50 or more employees must provide coverage or pay a penalty.

- 2018
Tax payers : high-cost employer-provided policies (US$27,500 for family or US$10,200 for single coverage) are subject to a 40% excise tax.

- 2020
Medicare prescription-drug beneficiaries : the prescription-drug coverage gap is eliminated.

The following figures illustrate the situation prevailing before the signature of the health-care reform bill :

- 30% of Americans between 19 and 29 years of age were uninsured;
- 10 million seniors were enrolled in Medicare Advantage plans;
- 50 employees was the maximum number a company can have without providing benefits and paying a penalty;
- 98% of companies with 200 or more employees offered health benefits;
- 16 million low-income Americans will be added to Medicaid;
- 36% of Americans were turned down or charged higher premiums because of pre-existing conditions in 2007 (Tumulty et al., 2010).

Most Americans – those who already had employer-based insurance – will not see much change for a while. They will nevertheless have an important

benefit quickly : for an additional fee, parents will be able to keep adult dependent children on their policy through age 26. That was good news when so many young people were struggling to find jobs during the recession. The biggest difference for Americans who had employer-based insurance was the security of knowing that, starting in 2014, if they lost their job and had to buy their own policy, they could be denied coverage or charged high rates because of pre-existing conditions. Before then, the chronically ill could gain temporary coverage from enhanced high-risk pools and chronically ill children were guaranteed coverage. The focus of the reform was on improving the dysfunctional and hugely expensive insurance markets for individuals and small businesses, and on expanding Medicaid coverage for the poor. The large expansion of coverage was to start in 2014, but some reforms will begin quickly, such as tax credits to help small businesses to provide coverage.

Cost of the reform

While the health-care reform bill was expected to reduce the federal deficit by US$124 billion in the next ten years, according to the Congressional Budget Office, its impact on family budgets is much more difficult to evaluate. Unlike plans to extend coverage and end discrimination against the sick, there seems to be no proven strategy in the bill guaranteed to slow down the rise of health-care costs in the United States at twice the rate of inflation. Some experts believed the reform would lay the structural framework to mount the most serious effort ever made to control medical inflation. Many economists stated it would be impossible to control health-care spending unless everyone was covered. That was because the uninsured tended to wait until they were sick to seek treatment. Then they attended the emergency services where costs were astronomical (Tumulty et al., 2010).

It should be recalled the United States was the only advanced industrial country that did not provide or guarantee health-care coverage for virtually all of its citizens. The bill did not quite reach full universality, but by 2019, fully 94% to 95% of American citizens and legal residents below Medicare age will have coverage. The bill achieved that by requiring most Americans to obtain health insurance, providing subsidies to help the middle classes buy policies on new competitive exchanges, and extending Medicaid coverage of the poor to include childless adults and others who were not eligible previously.

In the next ten years, the bill foresaw the spending of about US$350 billion on subsidies for 24 million low- and middle-income Americans who buy

insurance independently. These people and small businesses will have access to a new coverage market place in which insurers will compete against one another to offer the most attractive package of benefits at the lowest price. With this streamlining, administrative costs should go down, and with more transparency and competition, insurance premiums should become far more stable (Tumulty et al., 2010).

But those premiums will continue to rise, because the health-insurance industry operates on a simple principle : collect enough premium dollars to cover overhead and claims plus, in the case of commercial insurers, earn a profit margin of 3% to 6%. Insurance rates in most cases are rising steadily not because of price gouging but rather because underlying health-care costs are increasing at an unsustainable rate. This growth is due to a wasteful and inefficient payment system and to an innovative, and therefore expensive, approach to medicine. Slowing the rate of increase was the only solution to a health-care crisis still looming. The reform's ultimate success will hinge on whether it could transform an industry that currently rewarded volume and accounts for one-sixth of the American economy to one that would pay for the results (Tumulty et al., 2010).

The reform most likely to be effective in controlling costs is a tax on the country's most expensive insurance benefits, known as Cadillac plans. These insurance policies, which require tiny or non-existent co-payments and out-of-pocket spending, provide no incentive for patients to seek cost-effective care and in turn promote overuse of the health-care system, all of which increases costs. Faced with this new tax, employers and individuals will undoubtedly turn to cheaper policies, which economists consider will save money in the whole system. In general, economists loathe any tax exemptions for health benefits, which currently cost the government some US$200billion in lost tax revenue per year (Tumulty et al., 2010).

The second way reformers hope to control rising costs is to start rewarding physicians and hospitals on the basis of health outcomes – such as chronic-disease management and effective treatments for injuries – and not the volume of services they provide. In a system where providers are reimbursed separately for every procedure and service rendered, it is not surprising that costs are skyrocketing. No legislation could overturn this long-standing system, but the reform will enable the federal government to experiment with ways to compensate providers for quality. A demonstration project, for instance, would pay hospitals a set fee for a single episode of care – e.g. bypass surgery. Then everyone involved would have to divide it up. Reformers were optimistic about the pilot project's chances of success, but really slowing the rate of spending

growth would not happen until a successful experiment becomes widespread policy. That is where pilot projects have fallen short in the past (Tumulty et al., 2010).

The hard data about which treatments work more efficiently than others will enable the adoption of standard protocols. But lawmakers were careful to avoid giving any support for those who said health-care reform would substitute government decision-making for that of the physician and the patient. The law explicitly prevented comparative-effectiveness research from being used to decide which services Medicare would pay for and how much it would reimburse. It was a victory of politics over science (Tumulty et al., 2010).

Implications for Medicare

Medicare, introduced into law in 1965 and covering 38 million in 2010, is one of the most popular social programmes in United States history; that is why the health-reform bill, in its early stages, will not fundamentally change the way the programme operates. Americans 65 years old and above will continue to receive comprehensive health insurance; doctors and hospitals will continue to be paid by procedure. The lawmakers insisted that the US$500 billion in cuts to the programme over the next ten years – out of the total US$6.1 trillion expected to be spent – were to come from the elimination of waste in the system and from small but widespread reductions in reimbursements to hospitals and physicians. Seniors were, however, sceptical (Tumulty et al., 2010).

Regarding purchasing health-care services, the new law phased out the wasteful subsidies to private insurers that contracted with the federal government to provide Medicare-type benefits to seniors. Some 10 million elderly Americans received coverage from these Medicare Advantage plans, which often required lower co-payments than traditional Medicare and provide extra benefits like eye care, hearing aids and even gym memberships. These extra benefits, however, came to an extra cost, and experts stated the government paid about 14% more for each Medicare Advantage beneficiary than for a traditional Medicare patient. These overpayments would be gradually ended beginning in 2011 (Tumulty et al., 2010).

Further cuts – more than US$150 billion – would be made through what the legislation called "productivity adjustment": eliminating small amounts off the annual growth in reimbursements to hospitals and other facilities in the hope that they would squeeze out unnecessary expenses of their operations. At the same time, the government would

start spending more money, not less, on Medicare Part D, which provided prescription-drug coverage. Currently, millions of seniors every year found themselves in the Part D "doughnut hole" : a gap in coverage that existed once beneficiaries' costs exceeded US$2,830. Coverage did not kick back in until these seniors paid US$6,440 out of pocket. The law would begin closing this gap immediately. Seniors on Medicare would also receive free preventive services under the reform (Tumulty et al., 2010).

The new bill would set up an independent board to study clinical outcomes and evidence, and come up with ways Medicare could reduce spending without sacrificing quality or access. Hospitals with the highest rates of avoidable infections and unnecessary readmissions would be penalized – although not as much as many health-care experts would have liked (Tumulty et al., 2010).

Treating 32 million Americans gaining health-care coverage

The next challenge is addressing the needs of 32 million Americans gaining health-care coverage. Family physicians, who are on the front line of this surge in demand, are already in short supply, as are nurses, whom the new bill identified as critical players in meeting some of the expected new demand for services. By 2020, when most of the currently uninsured would have been fully brought into the health-care system, the American Academy of Family Physicians predicted a shortfall of 40,000 in the ranks of primary-care providers to treat them. One way to address that gap would be to make primary-care medicine a more attractive field – not just for physicians but also for nurse practitioners who receive an additional one to two years of training to extend their range of care into areas like anaesthesia. The new bill called for appropriations over five years to fund further training programmes, scholarships and loan repayments for those entering primary care. But even if new students took up those offers in 2010, they would not be ready to treat patients for three to seven years. The law would also temporarily boost what primary-care providers received for treating patients insured by Medicaid, the plan that would pick up nearly half of the newly covered (Tumulty et al., 2010).

Adding 32 million Americans to the health-care system implies a change in the way care is delivered. The new bill, for instance, recognized that doctors could not be the only ones to provide care and that hospitals and physicians'offices could not be the only places where people received health services. The reform would ultimately spend US$11 billion to create more health-care centres based in communities and schools as well as nurse-managed clinics. It would also enhance the government-

salaried National Health Service Corps of primary-care physicians, nurse practitioners and physician's assistants who target undeserved regions and receive loan repayments or scholarships to subsidize their medical education (Tumulty et al., 2010).

The funding would also support programmes like the "medical home," a team-based approach to delivering health care that breaks down the traditional hierarchy in which all health decisions are made by the physician. Instead, a medical home spreads responsibility across a range of providers and facilities, which allows existing hospitals and clinics to accommodate the increased demand for services without costly investments in capacity building. In New Jersey, for instance, AtlantiCare's pilot Special Care Center, a medical-home programme collaborating with Atlantic City's largest hotel and restauration union, was successfully reducing ER visits and hospital admissions. Two family physicians worked with a nurse practitioner, and together the team discussed the best ways to treat those with chronic conditions such as diabetes and heart disease. Patients received no claim forms or bills for services at the Center, and salaried providers were not reimbursed on the basis of the volume of services they generated, so they could focus on providing appropriate, quality care (Tumulty et al., 2010).

But innovative health-system administrators acknowledged that as long as the fee-for-service reimbursement structure remained in place with private insurers, physicians would be forced to practise two kinds of medicine : one in which they were reimbursed on the basis of the volume of services they provided and another in which the health outcomes and efficiency of their care were prioritized. When Massachusetts subsidized health-care coverage in 2006 and mandated universal coverage by 2007, visits to the State's emergency rooms swelled 7%, adding to a US$146 million jump in health-care costs from 2005 to 2006. This experience highlighted how much of the Americans' health behaviour was embedded in the current fee-for-service system – which, for the time being, was not disappearing. The key to achieving real reform was to persuade new – and existing – patients to seek care to maintain their heath rather than to treat a disease after it has taken hold. The bill's greatest achievement may be to make it possible for more people to access the health system in many different ways. Health care will increasingly take the form of preventive services such as regular diabetes check-ups and weight-loss programmes, instead of patients waiting to see their doctors until they need coronary-bypass operations or kidney dialysis. Those who were currently uninsured may turn out to be at the forefront of this trend, since 55% of them were under the age of 35 years, according to the Rand Corp. This group tends to be

the healthiest segment of the population and uses health care the least. That makes it the ideal cohort to begin thinking of health-care providers as wellness coaches (Tumulty et al., 2010).

Variation and experimentation of the health-care reform in the States

After 2014, the health-care system would indeed look far more uniform across the nation than it did in 2010. But the new bill left a lot of room for variation and experimentation by individual States. Some States have been far ahead of the federal government in expanding coverage and cracking down on dubious insurance-industry practices. Massachusetts, for instance, moved to cover nearly all its citizens in 2006; its system is in many ways a model for much of what is anticipated for the rest of the country. But other States – like Texas, where an estimated 28% of the population was uninsured in 2010 – will have a lot of catching up to do. Governors have been lukewarm to the Obama health-care bill, in large part because it called for adding 16 million people to the Medicaid rolls, which are jointly administered by the States and the federal government. Governors indeed stated expanding Medicaid would add billions to their health-care costs at a time when their States were facing record deficits (Tumulty et al., 2010).

The most important challenge for the States would be setting up health-insurance exchanges – market places where small businesses and individuals would be able to choose from a selection of insurance policies, much as federal government employees (including members of Congress) did before the bill. President B. Obama stated in a speech shortly after he had signed the bill : "What we are going to do is create exchanges all across the country where uninsured people – small businesses – are going to be able to purchase affordable, quality insurance." But each exchange would need to have enough enrollees and enough insurance-plan offerings to ensure vigourous competition. That is not a small challenge, given the near monopoly power insurers have in many States. While some States would be able to operate these exchanges on their own, others were likely to join with their neighbours in regional operations. And some States may well return to a government-run public option for the uninsured, similar to Medicare. This idea generated a lot of controversy during the yearlong battle over the health reform. Oregon was studying the feasibility of including a public option as part of its State exchange (Tumulty et al., 2010).

Conservative legislators in 39 States have introduced bills – or planned to do so – that would exempt their citizens from the new requirement in the law,

effective in 2014, to have health insurance or else face a fine. Such measures have already been enacted by the Virginia and Idaho legislatures, were on the November 2010 ballot in Arizona and have passed one chamber in Georgia, Missouri, Oklahoma and Tennessee (Tumulty et al., 2010).

Fulfillment of the new legislation

Administration officials wanted to deliver the parts of the new legislation that go into effect almost right away. Among them were tax credits to help an estimated 4 million small businesses to provide coverage for their workers, assistance for people having pre-existing conditions, a provision allowing young adults up to age 26 to stay with their parents' policies and a US$250 rebate for seniors whose initial Medicare drug benefits had run out (Tumulty et al., 2010).

Another challenge is filling key posts in the Executive Branch that remained empty for more than 14 months after B. Obama was inaugurated. By April 2010, no one had yet been named to head the Centers for Medicare and Medicaid Services, the huge agency that ran those two programmes. And Senate Republicans had held up the nomination of Columbia University professor Sherry Glied for the important post of Assistant Secretary for Planning and Evaluation at the Department of Health and Human Services (Tumulty et al., 2010).

Among the questions one would be asking about the fulfilment of the law are the following ones : are the exchanges working? Are the billions that are going into health information technology – like electronic health records – paying off as promised? Is it a good thing or a bad one if businesses decide to start sending their employees into the government-subsidized exchanges rather than providing coverage themselves? And are the subsidies the government provides adequate for the middle-income people who are required to buy insurance? "The worst thing would be to end up mandating coverage people cannot afford," stated Andy Stern, president of the Service Employees International Union, which represented many of the country's health-care workers (Tumulty et al., 2010).

The reform process has begun despite all the obstructionism. It is true that it will cost close to a staggering trillion dollars over the next ten years, a vast amount of money at any time when America's budget deficit was gobbling up nearly 11% of GDP and unemployment seemed stuck at close to 10%. The reform aims to control the relentless above-inflation rise in health-care costs that has gone on for decades, squeezing corporate and personal budgets alike and threatening, if unchecked, to

overwhelm the federal budget entirely. The reform also aims to shift away from a payment model that encourages physicians to prescribe too many overpriced tests (*The Economist*, 2010 e).

The bill is valuable for two reasons. The first has to do with coverage. It is unacceptable for a country as rich as America to have tens of millions of people without health insurance. Beyond them is the much higher number of people who fear falling into that position through losing their jobs; and the larger number again who cannot buy affordable insurance because they have an existing medical condition, or because they are too old, or because they have exhausted the "lifetime caps" imposed by insurance companies. The health-reform plan represents the last chance, perhaps for decades, of eliminating one of the least creditable differences between America and the rest of the industrialized world (*The Economist*, 2010 e).

The second reason, somewhat paradoxical, is that this bill will have to be improved, especially when it comes to costs. America's health-care system includes incentives that are in many cases unjustified. Because employer-provided health insurance is not considered to be a taxable benefit, people feel insulated from the real cost of their coverage and consequently overconsume. Because hospitals and medical practices in many areas face too little competition, they charge absurdly too much even for simple procedures. Because of the rapacity of American lawyers, the fear of lawsuits encourages physicians to practise "defensive" medicine, again driving up costs. The new health-care bill does not fully tackle all these problems. Gold-plated insurance policies will in effect lose their tax-exempt status, though not for a while, and not in full. An independent presidential commission will have some power to force down the rates paid to medical-service providers, although surprisingly hospitals were exempted. The bill is a historical and moral achievement, but it will still need future improvements with regard to the drastic reduction of costs of the whole US health-care system (*The Economist*, 2010 e). See Cypel (2010).

OECD countries

Among OECD countries, spending on long-term care is already equivalent to 15% of total health spending and is rising fast. In 2007, health-care spending as per cent of GDP was about 11% in France (average life expectancy at birth : 80.9 years), 10% in Germany (79.8 years), 10% in Canada (80.4 years), 9% in Sweden (80.8 years), 8% in Italy (80.9 years), 7.5% in the United Kingdom, (79.1 years), 7.5% in Spain (81.1 years), 7.5% in Japan (82.4 years), the average across OECD being 9% of GDP and 78.9 years for life expectancy at birth (*The Economist*, 2009 e).

In France, the deficit of the state's illness insurance amounted to €11.4 billion in 2009; it was expected to reach €11.6 billion in 2010. It is considered a major problem for the French government which was striving to reduce its overall budget deficit (as a percentage of the gross domestic product) through the decrease of expenses, including those of the social security system. In addition the deficit of the Fund of solidarity with old people amounted to €4.3 billion and was considered in a "critical situation." Within the 2010 deficit (€26.88 billion), health-care insurance represented half of the deficit (€13.1 billion), while the pension branch deficit was estimated at €9.3 billion and the family branch at €3.8 billion. Such situation was due to the fall in income (two-thirds) caused by the economic crisis.

In 2009, French people had spent €223 billion for their health, a 4% increase compared with the amount spent in 2008. Three-quarters of these expenses corresponded to health care and consumption of medical goods, but other expenses had been rising up strongly, e.g. health care for aged people in retirement homes (+15%) and expenses associated with preventive measures (+12.9).

The great bulk of the care for old people – an estimated 80% - is still provided by family and friends, the traditional support for the elderly. But more women are going out to work, so fewer of them have time to look after old people and formal help is becoming increasingly important. The latter is even considered a major source of employment in the ageing societies. As keeping old people in nursing homes or hospitals is expensive, many countries are providing grants to adapt homes, paying families for the care they provide and supplying helpers to give a hand in such tasks as dressing and bathing (*The Economist*, 2009 e).

In the United Kingdom, in 2010, the new coalition government pushed ahead with Conservative election commitments to create a £200 million cancer fund to pay for expensive new medicines, while the National Institute for Health and Clinical Excellence (NICE) has been facing fresh criticism for a series of recent rulings calling on the National Health System (NHS) not to use a number of expensive new medicines including for cancer, such as Avastin (Jack, 2010 b).

NICE has been preparing for budget reductions during the round of austerity measures compared with past plans for ambitious expansion, but it has also been recasting the way it presents its findings in a more positive light. Where its public communications on decisions in the past often talked about rejecting a drug it had considered too expensive for the benefits it provides, NICE is now using more nuanced language (Jack, 2010 b).

NICE's recent statement on cancer drug Avastin [known generically as bevacizumab and the world's best-selling cancer drug with sales in 2009 of about US$6 billion (Pollack, 2010 b)] for metastatic colorectal cancer meant that it would consult again on the treatment, stressing that it had not yet reached a final decision. It also explained that it was not currently recommending Avastin use, given that the drug cost £20,800 per patient and the limited evidence of its "modest benefit" was "highly uncertain." Data suggested the drug might extend the life of patients with bowel cancer by an average of six weeks, although none have survived for longer. Critics suggested NHS should pay only for drugs that do provide significant added benefits, and that otherwise limited resources are used less efficiently than they could be elsewhere in the health service (Jack, 2010 b).

Of the 102 specific uses of cancer drugs scrutinized by the National Institute of Health and Clinical Excellence since 2002 until the end of August 2010, the agency approved 73%. An analysis of 372 conclusions reached between March 2000 and July 2010 for its full range of appraisals on all drugs and treatments showed 309 had been approved, or 83%. By early October 2010, NICE reversed opposition to the use of drugs in mild Alzheimer's disease in draft guidance four years after a previous rejection (Jack, 2010 b).

In the United States, Avastin was given accelerated approval as a treatment for breast cancer in 2008 under a programme that allows for speedy approval of drugs for serious diseases. But accelerated approval is subject to further studies to confirm a drug effectiveness. And in two new trials, in which it was combined with different chemotherpy agents, Avastin did not perform as well. It delayed cancer progression by just one to three months, did not prolong lives and for many, had severe side-effects (Pollack, 2010 b).

The US Food and Drug Administration has moved to revoke the approval of Avastin as a treatment for breast cancer by mid-December 2010. Some patient advocacy groups welcomed the decision, stating that Avastin had never been shown to prolong lives and that women with breast cancer needed more than false hope. The US FDA decision was issued on the same day that the European Medicine Agency (EMEA), operating under different rules but with the same data, left Avastin available to breast cancer patients, but in a narrower way than it had before (Pollack, 2010 b).

In the United States, Avastin was likely to remain approved for use against breast cancer at least for a few more months while the drug manufacturer, Genentech (Roche), appealed the decision. Disappointed by the US FDA's

decision, the company had 30 days to submit its arguments, after which the US FDA will decide whether to grant it a hearing. Other approved uses of Avastin – to treat colorectal, lung, kidney and brain cancers – were not to be affected. So even if the revocation for use against breast cancer is final, physicians could still use the drug off-label. But insurers would be less likely to pay for off-label use of a drug that is very expensive. If approval is withdrawn, Genetech would remove breast cancer treatment from its price-cap programme, which limited yearly expenditures on Avastin at about US$57,000 for those with income under US$100,000. Without the price cap, Avastin would cost about US$88,000 a year. Genentech stated the drug was used by about half of the 28,000 Americans who were diagnosed a metastatic breast cancer each year (Pollack, 2010 b).

Health care in India

India faces perhaps the world's heaviest disease burden, ranging from infectious diseases to illnesses of affluence such as diabetes, hypertension and obesity. The public sector is overwhelmed, as the Indian government spends only about 1% out of 5% of the country's GDP devoted to health care (2005); nearly four-fifths of all health services are being supplied by private firms and charities – a higher proportion than in any other big country (*The Economist*, 2009 c).

However, Technopak Healthcare, a consulting firm, expected spending on health care in India to increase from US$40 billion in 2008 to US$323 billion in 2023. In part that is the result of the growing affluence of India's emerging middle classes. Another cause is the boom in health insurance, now offered by private firms, but also, in some cases, by the state. In addition, the government has eased restrictions on lending and foreign investment in health care, thereby encouraging public-private partnerships and offering tax breaks for health-care investments in smaller cities and rural areas. This has attracted a wave of investment from some of India's biggest corporations, including Ranbaxy (the generic-drugs pioneer behind the New Delhi-based hospital chain, Fortis) and Reliance (one of India's biggest conglomerates). New business models have been devised, some of them doing things cheaply, but others have helped India to move towards the rich world (*The Economist*, 2009 c).

India's private health-care providers, such as Apollo Hospitals, have been focusing for a long time on the affluent upper social classes, but they are now increasingly interested in less privileged groups. For instance, Vishal Bali, head of Bangalore's Wockhardt hospital, intended to take advantage of tax breaks to build hospitals in small and medium-sized cities, i.e.

with up to 3 million inhabitants. Prathap Reddy, Apollo Hospitals' founder, planned to do the same. Cutting costs in half for patients was his objective: a quarter saved through lower overheads, and another quarter by eliminating travel to bigger cities (*The Economist*, 2009 c).

Columbia Asia, a privately owned US firm with over a dozen hospitals across Asia in 2009, is also interested in investing in India. It advocates the building of hospitals using standardized designs, connected to a hub that can handle more complex illnesses. The firm offered modestly priced services to those earning US$10,000-20,000 a year within wealthy cities (in 2009), thereby reaching customers overlooked by more expensive chains. Its small hospital on the outskirts of Bangalore lacks glittering facilities and expensive imagery tools, but it has fully integrated health information technology (HIT) systems, including electronic health records (EHRs). Around Hyderabad, a chain of small maternity hospitals, LifeSpring Hospitals, offers normal deliveries by private doctors for just US$40 in its general ward, and Caesarean deliveries for about US$140 – one-fifth of the price at the large private hospitals. The chain has cut costs thanks to the elimination of canteens and the outsourcing of laboratory tests and pharmacy services. It also achieves economies of scale by attracting large numbers of patients using marketing. Monitor, a consultancy, estimates that LifeSpring Hospitals' doctors perform four times as many operations a month as their counterparts do in other private clinics, and, crucially, obtain better results because of high volumes and specialization (*The Economist*, 2009 c).

India's approach to cutting costs of the health-care system is supplemented by the better use of health information technology (HIT). According to a study carried out by the Journal of the *American Medical Association*, fewer than 20% of doctors' surgeries in the United States use HIT. In contrast, according to Technopak Healthcare, nearly 60% of Indian hospitals do so. Electronic health records circulate between hospitals, clinics and pharmacies, and they also handle patient registration and billing. Apollo Hospitals is selling its expertise in HIT and EHRs to American hospitals (*The Economist*, 2009 c).

Another example of cost- and clinical-effectiveness is that of Aravind, the world's biggest eye-hospital chain based in Madurai, in southern India. There are about 12 million blind people in India, with most cases arising from treatable or preventable causes such as trachoma or cataracts. Rather relying on government subsidies or charity, Aravind's founders use a tiered pricing structure that charges wealthier patients more (for instance, for meals and air-conditioned rooms), letting the firm cross-

subsidize free care for the poorest. Aravind also takes advantage from its scale. Its staff screen over 2.7 million patients a year via clinics in remote areas, referring 285,000 of them for surgery at its hospitals. International experts have estimated that the care was good, not only because physicians performed so many operations that they acquired very good experience, but also because the staff were rotated to deal with both paying and non-paying patients so there was no difference in quality. Aravind's successful model combines scale, pricing, technology and process (*The Economist*, 2009 c).

India's health-care entrepreneurs are therefore trying to channel the country's technological and medical skills towards frugal approaches that may inspire the rich world's overwhelmed health-care systems. According to Vivek Jawali, a heart surgeon at Bangalore's Wockhardt hospital and a pioneer of a new technique of heart bypass, "in our country's patient-centric health system you must innovate." This does not mean adopting every new piece of equipment. Over the years V. Jawali has rejected surgical robots and "keyhole surgery" kit because the costs did not justify the benefits. Instead, he has looked for tools and techniques that spare resources and improve results. Shivinder Singh, head of Fortis, a hospital chain based in New Delhi, stated that most of the new, expensive imagery machines were only a little better than older models. Meanwhile, vast markets for poorer patients are not taken care of. Fortis now promises only that its scanners are "world class," not the newest (*The Economist*, 2009 c).

Paul Yock, head of the bio-design laboratory at Stanford University, which develops medical devices, thinks that amidst growing concern about soaring health-care spending, the medical-technology industry could find inspiration in India. He has extended its bio-design programme to this country, in part to make his students understand the benefits of frugality. He believes that India's combination of poverty and medical and engineering talents could produce a world-class medical devices industry. P. Yock considers that medical-technology giants have "looked at need, but been blind to cost." Tim Brown, the head of Ideo, a design consultancy, agrees, and pointing to another recent example of India's low-cost engineering, stated : "In health care, as in life, there is need for both Ferraris and Tata Nanos" (*The Economist*, 2009 c).

Health care in China

According to the World Health Organization (WHO), "urbanization, environment degradation, increasing consumption of tobacco and the widespread use of motor-cars have worsened the risks of chronic diseases

in China." Some 23% of total population are overweight, 160 million Chinese suffer from hypertension, the number of diabetics would double in 2030 and reach 42 million. About 66% of men and 3% of women smoke, and lung cancer kills 300,000 persons per year; in 2020, lung cancer would be responsible of one death out of three. At least 400 million Chinese people carry the tuberculosis bacillus, 38 million the hepatitis C virus and 120 million that of hepatitis B. Enterovirus EV71 or "feet-hands-mouth" syndrome, that affected in 2008 several thousands of children and killed several dozens of them, was the last epidemic that stressed the deficiencies of China's health-care system (Pedroletti, 2009 a).

Official recognition of these deficiencies became apparent in 2003 during an outbreak of SARS (Severe Acute Respiratory Syndrome), an often fatal respiratory ailment. Its spread highlighted the difficulty of handling such emergencies when many Chinese were afraid of going to hospital because of the potential cost. New leaders who took over in 2002 and 2003 tried to consolidate their power by emphasizing the party's concern for the plight of the poor. But it was only in 2006 that work began on drawing a comprehensive plan for health-care reform (*The Economist*, 2009 b).

Two documents issued on 6 and 7 April 2009 set out reform targets through to 2020 as well as more specific objectives for the period 2009-2011. The broad goals remained unchanged from draft proposals released in October 2008 after a delay of several months. Officials said 200 million Chinese had no insurance in 2009, but by 2020 China was to have a "relatively robust" government-financed health-insurance system, with more than 90% of citizens covered by 2011. Also unchanged was the figure of 850 billion yuan (US$125 billion or €92.6 billion), which the government stated in January 2009 it intended to spend on these reforms during 2009 and the two subsequent years (*The Economist*, 2009 b).

In a World Bank's report published in March 2009 on poverty alleviation in China, it was underlined that the proportion of Chinese population living under the threshold of extreme poverty (i.e. whose consumption was under 888 yuans per annum) had decreased from 15.56% in 2001 to 10.38% in 2004 and to 4% in 2007. These achievements hide an extreme vulnerability, as twice more people can move under the threshold of poverty, at least once every three years. And in 2004, 41% of total population, i.e. 538 million Chinese, were living under twice the threshold of poverty. However, according to the World Bank's report, even the poorest households spare money for precautionary reasons (Pedroletti, 2009 a).

Over the 30 years of market economy and opening-up policy, government's commitment to health care has been decreasing, and the share of public expenses fell down from 90% during the communist era to 17% in 2007. This percentage is about 50% in the United States and 80% in Western Europe and Japan. In 2007, China has devoted only 1% of its GDP to public health care, a little less than India or Indonesia, three times less than Brazil and twice less than Thailand. In addition, the share of the Chinese central government in health expenses is only 10.5%, compared with 89.5% contributed by local governments. And in the least developed regions of the country, the lack of resources lead local governments to invest in poor-quality health infrastructures (Pedroletti, 2009 a). *The Economist* (2009 b) has summarized the situation : "Health-care provision, once rudimentary but accessible and widely admired by other developing countries, has been turned into a profit-driven system notorious for its corruption, indifference and expense."

"The major challenge is therefore to rebuild a public health service that functions with public funds and to find out alternative funding for hospitals that meet 30% to 50% of their budget from the sale of drugs," stated the social affairs adviser of the French embassy in China. Government subsidies account for a small amount of hospitals' revenue. Reports in the state-run press indicate that 90% or even more of their income comes from charges (poorly regulated and often excessive) for providing services and medicines. Weaning hospitals and physicians off these sources of funds will be a huge task. Even for those with government insurance, a substantial amount still has to be paid out of the patient's own pocket. In the countryside, despite the government's rapid deployment of a new insurance scheme in the past few years, many peasants still shy away from hospitals (*The Economist*, 2009 b).

The reform forecast the building of some 2,000 district hospitals as well as 5,000 basic health-care centres in the rural areas, where medical staff and equipments are rudimentary. Since 2005, rural populations have benefited from a cooperative system of rural voluntary insurance, which cost 10 yuans per year and has been extended to about 86% of rural districts in 2007. But peasants could only receive a maximum of 60% of their health costs in rural clinics and basic health-care centres (Pedroletti, 2009 a).

For district and city hospitals, where costs could be very high, reimbursement to the patient reached 30%, and only 10% for hospitalization. The reform forecast the creation of 3,700 health centres

in the cities, and some 11,000 basic health-care centres should be built or refurbished (Pedroletti, 2009 a).

Another key element of the reform is to regulate the almost private pharmacy services within the hospitals, through the setting up of the prices of about 300 basic medicines. Over the next three years (2009-2011), government-run medical facilities will be required to give preference to drugs on the list of essential medicines and profits made on them by health-care providers will be phased out. They will receive extra-subsidies to make up for their losses (*The Economist*, 2009 b).

According to Hubert Stüker, a German expert on medical insurance systems based in Beijing to participate in a cooperation project between China and the European Union, "the Chinese reform is a gradual one; the indicators are geared towards more social justice, but, in contrast to Europe, it will not be a real universal health insurance; the system will have several velocities. And Chinese people will have to continue to save money, in so far as they will have to pay an important part of the medical bill." Some key issues remain, such as the financial reform of hospitals' budget, as well as the overhaul of the public management of the health system. At least seven ministries are involved in carrying out the reform. The fragmentation of insurance systems among the regions of China and the subsequent lack of protection of migrant workers are also a headache for the central government. Another big obstacle to reform could be a lack of enthusiasm among local governments. Of the planned 850 billion yuans in spending, only 40% would come from the central government. Provincial and lower-level authorities may be reluctant to divert resources to areas that do not produce immediate benefits in terms of boosting employment and GDP growth, especially in a period when the economic crisis has markedly reduced their resources (*The Economist*, 2009 b; Pedroletti, 2009 a).

On the other hand, amidst the global economic crisis, many economists argue that the Chinese government should spend much more in the social arena (rather than boosting exports), so as to mitigate the crisis by stimulating domestic consumption. Chinese people are advised to save less and to spend more, but with a strong stimulus from the government.

The following figures from the World Health Organization summarize the health situation in China :

total population (2006)	: 1,328,474,000
life expectancy at birth (average)	: 72 years (men), 75 years (women)
life expectancy in good health at birth (2003)	: 63 and 65 years
infant mortality	: 24 per 1,000 live births
total expenses (%GDP) devoted to health	: 4.67% (compared with 11.2% in France)
total expenses for health per inhabitant per year	: €93.94 (compared with €3,819 in France)
share of private and public sources of funding in health	: 59.35% and 40.65% (compared with 20.1% and 79.9% in France).

Rationale and need for reform of the health-care system

For the same reasons as Bismarck carried out the reform of social insurance (including health) in Germany at the end of the 19th century, the other European countries followed suit, and nowadays President B. Obama wants to rebuild the American health-care system. It is indeed not acceptable to maintain in a growing economy inequalities that keep families entirely without defence in front of disease, ageing, casualties, without a substitution revenue. It is not just a humanitarian issue, it is based on the need to preserve the working force : one must care about the population in order to maintain its productivity (Reverchon, 2009).

In developing countries, it is also about a fair and balanced distribution of the benefits of economic growth. While it was believed that economic wealth should be created first and thereafter social protection would follow, it is nowadays agreed that social protection is a prerequisite and a tool for development and for wealth redistribution between the North and the South (Reverchon, 2009).

Traditional economies cannot do much against the globalization of diseases, such as HIV/AIDS, or influenza; the struggle against these diseases and others (cancers) is very costly and the average individual cannot cope alone with them. That is why a universal system that guarantees a fixed and permanent funding through mandatory participation is necessary. But if only the patients pay, it would not work. Disease cannot be fought successfully if there is not a permanent health structure or system that distributes medical staff across the national territory. And this can be funded only by a compulsory and fair tax (Reverchon, 2009).

It is true that due to the heavy weight of informal economy in developing countries and to the reluctance of people who tend to reject the idea that

risks should be covered even though they are not certain, the principle of universal contribution of everyone to a health-insurance system is not easy to apply. That is why a partial solution is often to rely on mutual agreements within a profession or a regional community, in which the participation of everyone is voluntary and not mandatory. But in order to reach the full objectives of public health, the universalization, even gradual, of the system is necessary (Reverchon, 2009).

Jean-Marie Spaeth leads a French organism on Health-International Social Protection, that promotes international cooperation and technical assistance in the areas of health and social protection. The organism has several cooperation agreements with China, including one within a French, German and British consortium which aims to train staff for the Chinese social security and to help the government to conceive and implement its policy. Other developing countries are following the same approach : a long national debate followed by the promulgation of a law. Thereafter, many years are needed to build up the system and make it operational. In France, social insurance became universal after a century (Reverchon, 2009).

For instance, in Central and Eastern European countries, important changes have been made in the early 1990s to improve the efficiency of the health-care system. A compulsory insurance against disease was set up in Poland, the Czech Republic, Slovakia and Hungary. Regional offices of the insurance system were created. Hospital management has been decentralized in Poland, Hungary and the Czech Republic, which resulted in the displacement of the funding issue towards local communities, without sufficiently consolidating the available funds. In order to reduce the debt of hospitals, the number of beds has been decreased : less than 75,000 beds in Poland between 1990 and 2005, according to the European Observatory on Health Policies and Systems. The average number of beds for intensive care dropped from 6.3 to 4.7 per 1,000 inhabitants, coming close to the OECD average in 1990 (Rodier, 2009).

When these countries evolved through the democratic transition, social protection was not the top priority among the state reforms. But in 2004, when some of them became members of the European Union, the reform of their health-care systems became urgent in order to comply with the European standards. Remedies had to be brought to a number of ailments : funding of the health-care system, low quality of health care, underpayment of medical staff, difficult access to health care and persistence of some kind of bribery. In 2009, despite a series of measures, health reform was not yet fully implemented, even though it was considered a priority (Rodier, 2009).

"In Poland, for instance, the salary of a physician is lower than the average national salary, despite two increases of more than 20% in 2004 and 2006. Their earnings had not changed between 1998 and 2004," stated Marek Naczyk, research assistant at the University of Oxford. That explains why bribery was still prevailing in public health services (Rodier, 2009).

The margin of salary increases of medical staff has been seriously reduced by the investments made by hospitals to comply with European equipment standards as well as by the high increase in the prices of medicines. The share of the budget allocated to drugs is one of the highest in Europe : in 2005, it amounted to 28% of total health expenses in Poland, 31% in Hungary, and 32% in Slovakia, compared with 16.4% in France. Drug prices had to be adjusted to the single European market (Rodier, 2009).

In addition to the underpayment of medical staff, another urgent aspect of the unaccomplished health reform concerns the disbalance between general practitioners and specialists. The study *Eco-Santé OCDE 2009*, published at the end of June 2009, indicates that in 2007, there were 2.9 specialists and only 0.7 general practitioners per 1,000 inhabitants in the Czech Republic (compared with 1.7 and 1.6, respectively, in France). A similar disbalance existed in Slovakia and Hungary (Rodier, 2009).

A new paradigm for health and development ?

On 28 August 2008 a report was released by a group of experts who had been requested by the World Health Organization (WHO) to take a broad look at the issue of inequality and health. The experts included Amartya Sen, an Indian-born economist and Nobel laureate in economics. After more than two years' work, the panel issued the report entitled *Closing the gap in a generation* (*The Economist*, 2008 f).

The gap can be illustrated by the following question. Why do America's Asian females live, on average, to 87, while the life expectancy of black males is only 69? The explanation, according to the WHO's Commission on Social Determinants of Health, is not merely a matter of income. Nor can it be reduced to the varying capacities of health systems. In addition to those factors, stated the report, there are social, political and economic forces that ostensibly have little to do with health but can still end up determining "whether a child can grow up and develop to its full potential and live a flourishing life, or whether its life will be blighted." To reduce the risk of the latter, the experts have called on governments to improve the quality of everyday life, particularly for women and girls in poor countries, through investment in child care and education, and by insisting on better working

conditions. They stressed the need to "tackle the inequitable distribution of power, money and resources" – through better governance, support for civil society, and more equitable economic policies. The report is now a paradigm for development, claimed Sir Michael Marmot, a professor at University College London, who chaired the panel (*The Economist*, 2008 f).

Ruth Levine, of the Centre for Global Development, an American think tank, described the report as imperfect but still useful. On the other hand, she noted, the report fell to provide any ranking for its list of laudable aims. But it made a worthwhile point, in her view, by urging a rediscovery of an earlier view of global health that was more prevalent before 2000. That was the year when a different WHO-inspired panel – convened by Jeffrey Sachs of Columbia University – put a controversial emphasis on the way in which poor health leads to bad economic performances by individuals and nations (*The Economist*, 2008 f).

In other words, there is renewed stress on the way that poverty and inequality lead to worse health. Julio Frenk, a former Mexican health minister now dean of Harvard University School of Public Health, stated the new report offered a way out of a "sterile debate" about whether poor health causes poverty, or vice-versa. Regarding the other possible flaw of the report, that it downplayed the link between income (as opposed to inequality) and health, Adam Wagstaff, a World Bank economist, stated he still believed income "is causal" when it comes to health; so that faster economic growth is likely to benefit the health of society as a whole, even if income inequality is constant. He cited the example of South Africa, where the health of older people improved after they started receiving pensions at the age of 65. In fact the authors of the report did not dismiss the role of growth – which they described as "without question important" – although they did state it could lead to greater inequity unless there were policies specifically designed to improve public health (*The Economist*, 2008 f).

Sir Michael Marmot argued that even in rich societies people became healthier as they climbed the social gradient in ways that could not be explained by wealth alone. Hence his interest, and the report's focus, on "social determinants" of health that are non-monetary. For instance, job insecurity and the resulting stress, have a proven link with mental health. So does the immunization of children, even in countries with free and universal access to vaccination. The report listed many reforms – ranging from the extension of social safety-nets to the education of girls and better public information about nutrition – that might increase the chances of better health (*The Economist*, 2008 f).

The structure of a country's health services plainly mattered too. The panel pointed out that societies with universal medical coverage enjoyed better health than places of comparable wealth that chose a different approach. That, for instance, gave the citizens of Costa Rica an advantage which many uninsured Americans lacked. The report, on the other hand, did not highlight people's responsibility with regard to their health condition. J. Frenk recalled that as Mexico's health minister he successfully made the argument that raising taxes on the sort of cigarettes smoked by the poor would in the long run help the worst off. As he saw it, such a tax needed to imply a rejection of choice : diehard smokers can still put away, but they must pay a price that reflects the cost to society of their habit (*The Economist*, 2008 f).

Clinical effectiveness versus cost-effectiveness

Does quality improve with more spending?

Spending more on health does not necessarily mean a better and a more effective system. For instance, OECD economists have found that the United States does indeed do well on some measures, such as breast-cancer survival rates and cervical-cancer screening, compared with other rich countries. William Tauzin, head of America's pharmaceutical lobby, warned that cutting costs too much could stiffle life-saving innovation. Diabetes monitors and pacemakers have improved dramatically in the past 20 years and have fallen in price – but costs have gone up because they are now being used by more patients (*The Economist*, 2009 a, e).

However, many studies show that America's spending on health care is soaring, yet its medical outcomes remain mediocre. American infant mortality was 6.7 per 1,000 births in 2007, compared with an OECD average (excluding Mexico and Turkey) of 4.0. The death rate after haemorrhagic strokes was 25.5% in American hospitals but only 19.8% in OECD countries as a group (*The Economist*, 2009 e).

Mark McClellan of the Brookings Institution, an American think tank, stated that a big problem is the overuse of technology. Whether a scan is needed, the system usually pays if a doctor orders it – and the scan might help defend the doctor against a malpractice claim. M. McClellan, a former head of the American Food and Drug Administration, pointed out that other innovative industries often sold new products at a loss, and recouped their investments later. In genuinely competitive industries, innovators are rarely rewarded with the "cost plus" reimbursements demanded by medical-device makers (*The Economist*, 2009 a).

OECD countries have an average of 11 magnetic-resonance imaging machines per 1 million people. The United States has 25.9. America uses them more often, too : 91.2 times per 1,000 people per year, compared with the OECD average of 39.1. Similar situations exist for other expensive medical tools (*The Economist*, 2009 e).

David Cutler of Harvard University stated that "technological change is the predominant reason for medical cost increases in the past half-century"…"studies of aggregate medical spending, and of particular medical conditions, show that at least half of all cost growth is a result of increased use of technology" (Cookson and Jack, 2008).

"Evidence-based medicine"

The search about the cost-effectiveness of the health-care system has led to what is known as "evidence-based medicine," attempting to remove treatments and procedures that are at best unnecessary and at worst harmful, and concentrate resources on what works. This approach, if it is systematic, could help avoid dire predictions for the future of the health-care system, particularly in developed countries. Thus Jonathan Anscombe, head of health of A.T. Kearney's European health practice, predicted that countries would have to restrict tax and social insurance systems to a "core" offering. This would consist mainly of preventive and primary care services that help restrain costs, along with emergency services and support for the poor, he suggested. Everything else – most non-emergency care, let alone costly end-of-life cancer drugs – would have to be covered by private individual insurance or by out-of-pocket payments. This, he stated, is "inevitable" and "it is hard to see how this can be achieved without making care more unequal" (Timmins, 2008).

But there are less pessimistic viewpoints. For instance, dangerous and expensive surgery for gastric ulcers has disappeared, thanks to antibiotherapy against *Helicobacter pylori*, the infectious pathogen that causes a very high proportion of gastric ulcers. In June 2008, researchers announced the discovery of a genetic test for breast cancer that could allow screening programmes to be focused on those at most risk, lowering costs (Timmins, 2008).

Evidence-based medicine is, nevertheless, a sensible approach, a useful way to promote transparency and value of the health-care system. For instance, the United Kingdom's National Institute of Clinical Excellence (NICE) which assesses new drugs and technologies, and recommends which ones the National Health Service (NHS) should fund, is widely

seen internationally as a success, being copied, or about to be copied, by almost a dozen countries. The United States is discussing a similar initiative, although it might concentrate only on clinical effectiveness, not cost-effectiveness. Sir Michael Rawlins, chairperson of NICE, commented : "Even if the United States does not include cost-effectiveness now, it will have to do so at some stage in the future" (Timmins, 2008).

For Mark Sculpher, professor of health economics at New York University, what counts is cost-effectiveness : "It may be appropriate to devote considerable additional resources to new technology if it is good value," he stated. Joe Hogan, head of General Electric Healthcare, one of the world's leading diagnostics and medical imaging companies, highlighted the surge in the power of diagnostic devices and the extraordinary drop in costs of scanners in recent years. David Cutler of Harvard University published an extensive study of "revascularization" (heart bypass surgery) to restore blood flow after a heart attack. Analyzing 17 years of data, he concluded that the procedure was associated with more than a year of extra life expectancy at a cost of about US$40,000 – making it "highly cost-effective" (Cookson and Jack, 2008).

According to Paul Ginsburg, president of the Centre for Studying Health System Change in Washington, D.C., the majority of technology improvements are cost-effective and valuable, "but the benefits are diminished when the technology is applied beyond those patients most likely to benefit from it." For instance, arthroscopic, key-hole, operations on the knee were very beneficial for the original patients, who had a clearcut requirement for each surgery, but it was extended too far, to people who did not really need it. Similarly, the pain-killer Vioxx undoubtedly benefited many patients with arthritis, but Merck withdrew it from the market in 2004 after side-effects emerged (cardiovascular risks) when it was used by a far larger number of patients to whom, critics argued, it should never have been prescribed (Cookson and Jack, 2008).

While the prices of some new drugs – notably against cancers and certain extremely rare diseases – have risen sharply in recent years, Chris Brinsmead, president of the Association of the British Pharmaceutical Industry, stated that the overall proportion of the UK National Health Service budget spent on medicines was not only modest, at less than 10%, but had declined. That is partly explained by generic competition once patents expire for drugs such as proton pump inhibitors, which have contributed to the elimination of expensive ulcer surgery, and statins, that have helped ease cardiovascular problems (Cookson and Jack, 2008).

Booz Allen Hamilton, a consultancy, found "substantial evidence that overutilization and misuse of technology is leading to spending that exceeds its value for patients." Diagnostic imaging, a US$100 billion global business, is a good example of increases in spending being "driven to a large extent by the growth in the number of machines installed in hospitals, as well as in doctors' offices and at imaging centres," the consultants stated. The result is a strong incentive for physicians to prescribe unnecessary scans that provide little help for a closer diagnosis (Cookson and Jack, 2008).

The overall conclusion is that health-care systems throughout the world are facing up to the need for better evaluation of the costs and benefits of new technologies before they are introduced on a large scale.

Other means of tackling health-care costs

But tackling inflation costs in health care cannot find an appropriate solution just through the evaluation of medical technology cost-effectiveness (drugs, diagnostic devices, treatments). In the case of the US health-care system, polls showed that 61% of respondents thought the high cost of care and insurance was a bigger problem than the number of uninsured, against 31% who believed the reverse. Only 21% would be willing to support a reform plan if they had to pay more in insurance and tax; 62% would not (*The Economist*, 2009 e).

Physicians' generous pay is another reason for the high costs of the US health-care system. The OECD estimated that American general practitioners earned 3.7 times the average wage. Their British counterparts earned 4.2 times their national average. American specialists earned 5.6 times the average wage, compared with 7.6 times for their Dutch colleagues. Yet health-care costs in the United Kingdom and the Netherlands remain lower than in the United States. The real problem is not how much American physicians are paid, but how. Most doctors are not paid a fixed salary, but they are paid more if they provide more services, regardless of the results. Predictably, this leads to far higher rates of doctors' visits, specialists referrals, scans, etc. There is strong evidence that American patients consume drugs, and use procedures, scans and other expensive forms of health care than do patients in other rich countries, and not always to good effect. The American insurance system encourages overuse in several ways. One is the tax break that favours health insurance provided by employers, which leads to generous coverage and henceforth to overconsumption. Another is the fact that American health insurers earn a lot of revenue from administering the

health plans provided to employees by big corporations which, in effect, insure themselves. This leaves insurers with no incentive to cut costs, because more spending means higher management fees (*The Economist*, 2009 e).

A second major factor pushing up health costs is a lack of proper competition among operators of American hospitals. Thanks to a wave of consolidation in recent years, argued Regina Herzlinger of Harvard Business School, "most parts of the United States are dominated by oligopolistic hospital systems," and there is almost a total lack of price competition among providers. There are nevertheless challengers of this system in some areas. For instance, specialist heart hospitals, which obtain better results at more reasonable prices than local general hospitals, or retail clinics at Wal-Mart stores. Remote medicine, in the form of technology for tele-care or medical journeys to Thailand and Costa Rica, also challenge the system. Medical-device manufacturers often expect reimbursement for expensive new equipment on a "cost plus" basis, a practice rarely seen in competitive markets, and drug companies enjoy temporary monopolies on new pills that may be no better than cheaper alternatives on the market today. More transparency would help to counter the lack of proper competition, by empowering patients to choose hospitals and physicians providing good value and better results. This can be prompted, in part, by a host of information and communication technologies that should make health care much more portable, precise and personal. The spread of electronic medical records should bring more transparency. Change is also being prompted by the willingness of physicians and politicians, especially ones in poorer countries, to apply at least some economic criteria to medical spending (see the case of India and the efforts made by the United Kingdom's NICE) [*The Economist*, 2009 e].

CAN BIOTECHNOLOGY HELP?

Undeniable contributions

The first biotechnology revolution took place during the second half of the 19th century, thanks to the discoveries and innovations made by Louis Pasteur (1822-1895) and several of his collaborators or contemporary scientists and physicians (e.g. Robert Koch, 1843-1910; Emile Roux, 1853-1903; Shibasaburo Kitasato, 1856-1931) in the areas of microbiology (food and beverage fermentations, bacterial diseases), vaccination and immunology, environmental microbiology (nitrogen cycle). See Sasson (2008).

The second biotechnology revolution started with the discovery and production of antibiotics (Alexander Fleming, 1881-1955, penicillin; Salman Wasksman, streptomycin). The industrial manufacture of a wide range of antibiotics has contributed and still contributes to the control of many human and animal infectious diseases. There is a permanent search for new antibiotics in order to overcome the resistance developed against these molecules by many pathogens. Molecular biology and genetics applied to the better understanding of the resistance mechanism, including the identification and isolation of the resistance genes (thanks to the sequencing of the whole genome of the microbial pathogens), help to meet a major challenge the control of communicable diseases is facing. The appropriate use of antibiotics also raises issues of clinical and cost-effectiveness. Looking retrospectively, there is no doubt that this second wave of medical biotechnology has been very helpful in the global control of animal and human diseases.

The third biotechnology revolution started when, in 1953, James Watson, Francis Crick, Maurice Wilkins and Rosalind Franklin described the structure of DNA. That established molecular and cell biology as the basis of bioindustry. The sequencing of the human genome, in 2001, preceded and followed by the sequencing of many microbial and animal genomes, heralded an era of innovation with respect to diagnosis, prevention, prediction and cure of human diseases. See Sasson (2008).

Convergence of biology and engineering

Nowadays, a fourth revolution is under way : the convergence of biology and engineering. Phillip Sharp, a Nobel laureate at the Massachusetts Institute of Technology (MIT) believes that the tools which have transformed physical sciences, i.e. information technology, advanced materials, imaging, nanotechnology, and sophisticated modelling and simulation, are about to be brought to bear on the life sciences. Robert Langer, a biochemist at MIT who holds over 500 patents in biotechnology and medical technologies and has started or advised more than 100 new companies, considers that innovation in medical technologies is about to take off (*The Economist*, 2009 a).

For instance, researchers at the Massachusetts Institute of Technology (MIT) have created prototypes for cancer monitors the size of a grain of rice, that can fit easily into the bore of a biopsy needle. Tiny coated particles inside the devices can bind with molecules linked to cancer at the site, generating miniscule clumps that can be detected by a non-invasive scan like a magnetic resonance imaging (MRI). Michael J. Cima, professor of materials science and engineering at MIT, was the leader of the team that created such devices (Eisenberg, 2009).

The tiny monitors were tested in mice and they could detect the cancer with the change in the MRI. The method could replace the cancer biopsy which is the gold standard for diagnosis. But the shortcoming is that one has the information only at the time the tissue is sampled. By contrast, the monitors can offer continuous information. J. Cima indicated : "Instead of trying a chemotherapy and waiting a while, you could see early indications of whether the therapy was going to work." These monitors could have many applications, according to W. Mark Saltzman, chairman of the department of biomedical engineering at Yale University, where he is carrying out research on nanoparticles and drug delivery. The devices may hold possibilities not only for sensing the disease, but also for remedying it, stated W.M. Saltzman (Eisenberg, 2009).

Cancer monitors designed by J. Cima are small plastic containers with a reservoir to sequester the magnetic nanoparticles so they cannot leak out. The particles are of the same material as those currently injected to patients intravenously to improve contrast in MRIs. On the top is a membrane through which fluids diffuse into the chamber and make contact with the particles. The magnetic nanoparticle technology used in the device was developed by Ralph Weissleder, professor at Harvard University Medical School and director of the Center for Systems Biology

at Massachusetts General Hospital in Boston. With M.J. Cima, they are two of the co-founders of T2 Biosystems, a company in Cambridge, Mas., that is commercializing the core nanoparticle technology. The particles can detect the biomarkers that are shed by tumour cells as they develop, or respond to therapy (Eisenberg, 2009).

M.J. Cima's implantable devices will detect biomarkers inside the body and could help physicians realize whether a tumour responds to a particular therapy. One may hope that biomarkers detected by the cancer monitors would make cancer treatments a little more rational than they are nowadays, stated R. Weissleder. M.J. Cima is planning a variety of devices, each set up to monitor a different metabolic activity of tissue near a tumour. "If the therapy is having an impact on the survival of that tumour, you will see it in the local metabolites," said M.J. Cima. In the future, the device may not have to be read by a large MRI machine. The American researchers are working on another version of the implantable device, made with a metal coil that acts as a kind of antenna. That version could be read by a hand-held magnetic resonance detector (Eisenberg, 2009).

The next experimental stage was to implant these devices in larger animals with tumours that look like human ones, and much later on tests would be carried out in humans. M.J. Cima hoped that the new devices would lead to a stream of diagnostic information, "to help physicians treat cancer as a chronic disease as opposed to an acute one," he stated (Eisenberg, 2009).

The market for medical innovations of all sorts is poised to grow and cost-effective innovations can help reduce costs and overspending in health care in rich countries, and also improve the current situation in poorer countries through leapfrogging. But would it be a revolution or rather a reformation, as it will take time? We have seen that given soaring health-care costs, governments and insurers may not wish to adopt new technologies unless it is conclusively proved that they produce better results and offer value for money. For instance, genetic tests (derived from advances in human genomics), offered to consumers as a means to predict the occurrence of diseases, are questioned. On the other hand, the benefits of safer and more effective vaccines against viral, bacterial and even parasitic diseases (e.g. malaria) are unquestionable. The concept of a blockbuster drug consumed by large populations is gradually abandoned by drug companies which prefer to develop more effective drugs targeted to smaller populations that will react favourably and with lesser side-effects (this clinical and cost-effectiveness approach is successfully applied to anticancer drugs).

If these obstacles can be overcome and if evidence-based medicine prevails, the health-care sector could embrace information technology and digitization of biomedicine can become a reality. In fact, in 2007, J. Craig Venter stated : "For the past 15 years at ever faster rates we have been digitizing biology. By that I mean going from the analog world of biology through DNA sequencing into the digital world of the computer. The human genome is perhaps the best example of digitizing biology. Our computer databases are growing faster per day than during the first 10 years of DNA sequencing" (see Sasson, 2008).

But digitization also promises to connect physicians not only to everything they need to know about their patients, but also to other physicians who have treated similar disorders. The forthcoming convergence of biology and engineering will be led by information technologies, which in medicine means the digitization of medical records and the setting up of a network for sharing those records. That will enable many other major technological changes to be introduced. It can also make that information available to the patients, so as to play a greater role in managing their own health care. Is it advisable? Many physicians and patients reckon that they lack the knowledge to make informed decisions. But it is a learning process which can progressively engage individuals to take more responsibility in dealing with their health and prevent problems before they require costly treatments, while relying on the effective cooperation of their physicians (see *The Economist*, 2009 a).

Protection of biotechnology-derived drugs and its impact on the American health-care reform

On 31 July 2009, as the Energy and Commerce Committee of the US House of Representatives was putting the final touches on health-reform legislation, a vote took place (47 against 11) and it was considered a great victory for drug companies : the Energy and Commerce Committee bill would extend the protection of biotechnology-derived drugs to 12 years of exclusivity, as would legislation passed a few weeks earlier by the Senate Health, Education, Labor and Pensions (HELP) Committee. Then-chairman Ted Kennedy, whose State of Massachusetts is home to many biotechnology firms, had long supported a 12-year exclusivity period (Tumulty and Scherer, 2009).

The vote was the result of intense lobbying by the Biotechnology Industry Organization (BIO) and by its chairman Jim Greenwood, a former Republican congressman from Pennsylvania, who was a member of the Energy and Commerce Committee. In fact, in the first half of 2009, drug and

biotechnology companies, and their trade associations had spent more than US$110 million – i.e. about US$609,000 a day – to influence lawmakers, according to data compiled by the non-partisan Center for Responsive Politics. The drug industry's registered lobbyists numbered 1,228, or 2.3 for every member of Congress. The campaign contributions to members of the House Energy and Commerce Committee had totalled US$2.6 million over the three-year period 2006-2008 (Tumulty and Scherer, 2009).

Biotechnology-derived drugs or biopharmaceuticals, which differ from the usual chemical-based pharmaceuticals because they are produced from living cells, are generally regarded as the future of the pharmaceutical industry and as having a considerable impact on medicine. While in 2009 only 20% of the drugs on the market were biopharmaceuticals, it is expected that, with 633 biotechnology-derived drugs in development in 2008 for more than 100 diseases, 50% of the new drugs approved in 2015 will be. Biopharmaceuticals average more than 20 times the cost of conventional drugs : for instance, treating breast cancer for a year with Herceptin costs about US$48,000; the treatment of rheumatoid arthritis with Remicade can cost US$20,000 annually; for other rarer diseases, the cost of biotechnology-based treatments can be as high as US$200,000 a year (Tumulty and Scherer, 2009).

It is therefore important to examine ways and means to decrease these costs in order to better control health-care increasing expenses, e.g. bringing in more competition. For instance, in 1984 a law written and presented by Henry Waxman with Republican senator Orrin Watch, lowered the regulatory obstacles that prevented generic drugs from making their way to market. At the time, it was expected that fast-tracking the approval of "bioequivalent" drugs would bring down medical costs by US$1 billion a year. But with generics accounting in 2009 for more than 70% of prescriptions dispensed in the United States, "the actual savings have exceeded our wildest expectations," stated H. Waxman in a 18-September speech before the Generic Pharmaceutical Association. He added "in the last decade alone, generic drugs have saved consumers, businesses and State and federal governments US$734 billion" (Tumulty and Scherer, 2009).

Biotechnology-derived drugs were not dealt with in the 1984 law, and a 2008 analysis by Robert Shapiro, a former Clinton administration official, suggested that generic versions of the top 12 categories of biopharmaceuticals the patents of which have expired or will expire soon could save American citizens up to US$108 billion in the first ten years and as much as US$378 billion over two decades (Tumulty and Scherer, 2009).

To illustrate the reasoning behind this assumption, the following figures are the annual (2008) sales of several biopharmaceuticals :

- Avastin, produced by Genentech and used in the treatment of various cancers – US$9.2 billion;
- Enbrel, produced by Immunex and used in the treatment of rheumatoid arthritis - US$8.0 billion;
- Remicade, produced by Centocor Ortho Biotech and used in the treatment of inflammatory disorders – US$7.9 billion;
- Humira, produced by Abbott Laboratories and used in the treatment of rheumatoid arthritis and psoriatic arthritis;
- Rituxan, produced by Genentech and used in the treatment of non-Hodgkin's lymphoma and rheumatoid arthritis – US$7.3 billion;
- Herceptin, produced by Genentech and used in the treatment of breast cancer – US$5.7 billion;
- Lantus, produced by Sanofi-Aventis and used in the treatment of diabetes – US$5.1 billion;
- Epogen, Procrit, produced by Amgen, Ortho Biotech, and used in the treatment of anaemia – US$5.1 billion;
- Neulasta, produced by Amgen and used in the treatment of neutropenia – US$4.2 billion;
- Novolog, produced by Novo Nordisk and used in the treatment of diabetes – US$3.7 billion.

Cost-saving competition which is a legitimate concern of policy-makers should not eliminate the financial incentives that have fostered the American bioindustry at the forefront of biomedical research and development, nor stiffle the development of even more drugs that could save lives and mitigate suffering. Finding the right equilibrium leads to the question of how long biotechnology firms should be guaranteed exclusivity outside the protection of their patents, before copycats can begin using the data they have developed.

The 12-year exclusivity approved by the House Energy and Commerce Committee at the end of July 2009 had been questioned earlier in June by the Federal Trade Commission (FTC) which argued that giving biopharmaceutical makers any period of exclusivity at all could actually stiffle innovation. Biopharmaceuticals are so much more complex and expensive to produce than conventional drugs that the barriers to would be "biosimilar" competitors are already high, the FTC argued. Giving biologicals further protection – particularly the 12 years of exclusivity that the bioindustry wanted – would merely encourage firms to use what they have rather than drive them toward "new inventions to address unmet medical needs" (Tumulty and Scherer, 2009).

The biopharmaceutical industry has a strong lobbying power, and it is not easy to find a truly independent viewpoint regarding the good balance between cost-saving competition and exclusive protection of biologicals. For instance, in July 2009, the National Health Council wrote letters to members of Congress "on behalf of 133 million Americans" asking for a minimum of 10 years of data exclusivity. The group claimed a membership that included 50 of the American largest patient-advocacy groups, including the American Cancer Society, Easter Seals and the National Kidney Foundation. But its board of directors includes pharmaceutical executives, and its 2007 tax filings showed that almost half of its US$2.3 million budget came from the Pharmaceutical Research and Manufacturers of America (PhRMA) and drug companies. Similarly, on 19 October 2009, PhRMA called for "a fair period of data protection" of 12 years at a "bare minimum." To defend its position, the group cited Duke University economist Henry Grabowski, whose work it had funded, and two patient groups. One is Retire Safe, which receives "general operating support" from Pfizer; the other, the Alliance of Ageing Research, is also run by the drug industry (its vice chairman was with Novartis) [Tumulty and Scherer, 2009].

Most high-profile advocate of the biotechnology-derived pharmaceuticals has been former Democratic National Committee chairman Howard Dean. In July 2009, he wrote an article in the *Hill* newspaper arguing for a "common sense and fair approach" to give biotechnology companies at least 12 years of exclusivity. But the counterarguments also existed. Thus, the world's biggest generic drug company, Teva Pharmaceuticals, had spent a lot of money on lobbying and also sponsored academic work on the issue, aiming to disprove Duke's Grabowski. Generic drug manufacturers are allied with powerful organizations, labour unions, insurance companies, health-maintenance organizations and health-reform advocacy groups. Heated debates were expected in the US Congress (House and Senate). How the issue is resolved – in favour of protecting the bioindustry or opening up the market to generic drugs – may indicate which interest groups will ultimately reap the benefits at stake (Tumulty and Scherer, 2009).

The US health-care reform puts more pressure on the pharmaceutical industry. The decrease in prices for the drugs delivered to the state via Medicaid (health-care insurance provided to people with low resources) and Medicare (health-care insurance provided to old people) would be theoretically compensated by the increase in the market volume, as 32 million Americans will become, as of 2014, newly insured and will obtain drugs and benefit from complete health care. The retail prices demanded by the administration within the framework of Medicaid are significant: they vary between 15% and 23% for patented drugs, and between 11% and 13% for generics (Boder, 2010).

It should be recalled that the American market is among the very few in the world where there is a complete freedom of drug prices, except for the Medicaid and Medicare systems of health-care insurance. This freedom enables pharmaceutical groups to make profits that are unimaginable in Europe or Japan. In the latter, governments negotiate the sale prices of drugs with the companies. For instance, in Switzerland, if no agreement is found, the drug can be sold, but it will not be reimbursed by the social security (Boder, 2010).

The American pharmaceutical market has an annual growth rate of 6%, amounting to US$218.8 billion in 2009 – the world's biggest. The German market, the first in Europe, had a volume of sales of about US$35 billion in 2009. The Pharmaceutical Research and Manufacturers of America (PhRMA) has immediately approved the health-care reform and invested about US$100 million in an advertisement campaign. It estimated the financial impact of the reform on its turnover would vary between -2% and +1%. Other associations, closer to the consumers, were of the opinion that drug expenses would increase by US$3.5 billion per year, because the effect of the substitution of patented drugs by generics would be less important than foreseen in the reform (Boder, 2010).

Contrary to the proposals made by Hillary Clinton in 1994 to reform the health-care system, which were not approved, President Barack Obama did not question the principle of free market and free price devise in the pharmaceutical area. He has also continued to tolerate a practice, that is condemned in Europe and that permits to pharmaceutical companies to negotiate among them the marketing or not of a generic drug – with financial compensation. In addition, the patent system has been consolidated, offering a special 12-year protection to biotechnology-derived drugs (Boder, 2010).

This sector is propelled by the booming sales of drugs manufactured by some companies like Genentech – the American subsidiary of Roche –, and they represent a market of more than US$100 billion in the United States. According to a study by Standard and Poors, the European companies that will most benefit from the American health-care reform are Roche and Novartis. Roche commercializes high added-value drugs against cancer, that will resist to the pressure exerted on prices. Novartis will profit from its diversification in generics, which are supported by the health-care reform, and from its strong presence in the area of controlling hypertension. The latter, in addition to diabetes, will be one of the chronic diseases that will be treated for many years among the newly insured 32 million Americans. Novartis, present in the vaccine area, will also benefit from the financial support of the American administration for research on new vaccines (a US$500 million investment has been foreseen in that area) [Boder, 2010].

DIAGNOSTICS

In vitro diagnostics : a key sector of medical biotechnology and the health-care system

In addition to biochemical tests carried out on samples of blood, urine or tissues to measure the concentration of several metabolites (e.g. glucose, urea, uric acid, lipids, etc.), *in vitro* diagnostics (i.e. performed outside the human body or *in vitro*) comprise immunodiagnostics and molecular biology-based tests. Immunodiagnostics are quantitative analytical techniques that are based on the reaction between an antigen and the corresponding antibody; most of immunodiagnostics also use a third element, a tracer, which originates from the association of the antigen or the antibody with a marker. When the marker is an isotope, the test is a radio-immunodiagnostic; when it is an enzyme, the test is an ELISA test, and when the marker is a fluorescent compound, the test is an FIA test. The enzymes generally used in the ELISA tests are peroxidase or alkaline phosphatase.

The second category of *in vitro* diagnostics consists of using techniques that amplify nucleic acids existing in a sample (e.g. blood, saliva, tissue). In addition to the polymerase chain reaction (PCR), the main amplifying techniques are the nucleic acid sequence based amplification or NASBA, the transcription mediated amplification or TMA and the strand displacement amplification or SDA. Once the quantity of nucleic acids is amplified, partial or whole sequencing of these nucleic acids can be carried out, in order to detect a pathogen (through known sequences of its genome) or to identify gene sequences of the patient that can be associated with a particular disease.

DNA biochips are microscopic slides (generally made of glass or silicium or a polymer) on which are fixed DNA or oligonucleotides in great quantity and with a high density. These chips play the role of a diagnostic tool. In the case of DNA biochips, the substance to be studied or analyzed is a sample from the patient or a foodstuff, that is put in contact with the molecules fixed on the chip. In case of complementarity between the

two, a hybridization takes place (key/lock principle). The complementary hybridization process can be visualized if the sample is pretreated with a fluorescent dye. The use of DNA biochips or microarrays are therefore very useful tools for gathering detailed information on the composition of the sample.

In vitro diagnostics are performed in laboratories of medical analyses, in blood-transfusion centres and hospitals, as well as in physicians' cabinets (not all of them) or even by the patients (e.g. glucose test, pregnancy tests, or rapid detection of haemolytic streptococcus-A group in anginas).

Innovation strategy in the area of *in vitro* diagnostics over the last years has been focused on several complementary approaches :

- automation and robotization of the assays, which lead to process standardization, system miniaturization, reduction in the volume of samples, higher precision and reliability, as well as rapidity of results;
- improvement of the specificity and sensitivity of reagents;
- access to molecular biology techniques, that are increasingly used in a routine way;
- digital management of data that has to be increasingly efficient in order to meet the requirements of patients and physicians.

The fast growth rate of *in vitro* diagnostics and their wide diversification respond to the need for :

- preventing a disease, through the screening of populations at risk of developing the disease (e.g. rubella, cystic fibrosis, hepatitis, cancers); this is generally called primary prevention;
- confirming an early and reliable diagnosis of a disease (detection of the illness or a lesion that precedes it, at a stage when a treatment can be useful); this is called secondary prevention;
- following up the treatment of a disease through the measurement of biological parameters (e.g. in the case of diabetes, cardiovascular diseases and cancers). This tertiary prevention can help improve the clinical effectiveness of treatments as well as the monitoring of disease evolution.

In vitro diagnostics are therefore a key component of a cost-effective health system, and their cost is generally considered moderate with regard to their medical benefit and their impact on costs; they can reduce the length of hospitalization, improve the treatments and the monitoring of the illness. See Sasson (2008) and J. Kadouche (personal communication, 2009).

In 2006, global health-care expenses (consumption by patients) amounted to about €157 billion, and medical diagnostics made up 2.6% of this total expenditure. In 2007, the global annual turnover of the *in vitro* diagnostics industry reached €27 billion : €10.2 billion for the United States, €9.8 billion for Europe, €2.1 billion for Japan and €4.9 billon for the rest of the world. The growth of this key sector of medical biotechnology is strong, due to a high innovation rate; experts even speak of a scientific revolution, fuelled by the discovery of new biomarkers and the applications of DNA sequencing, proteomics, and nanotechnologies (J. Kadouche, personal communication, 2009). According to Kalorama, a market-research firm, medical diagnostics industry was expected to be worth US$56 billion in 2012 (*The Economist*, 2009 a).

In 2007, the value of the global drug market has been estimated at US$712 billion (€508 billion), compared with US$200 billion in 1990 (€143 billion), with an annual growth rate of 6.4%. The North American market (Canada and United States) is the biggest (40% of global market), ahead of Europe (32%) and Japan (10%). In ten years, Europe's share has markedly decreased, even though the European market remained dynamic (annual growth rate of 7% in 2007). Generics market had an annual growth rate of 10%-15% in 2007, Teva Pharmaceuticals (Israel) being the world's leader, and France and Germany being the leading markets in Europe (J. Kadouche, personal communication, 2009 a).

Regarding *in vitro* diagnostics, in 2007, the 16 first companies withheld 86% of the global market. During that year, some 30 major fusions and acquisitions took place. A major event has been the purchase of Bayer Diagnostics by Siemens, at the beginning of 2007, followed by that of the American firm Dode Behring, for €4.5billion and €5 billion, respectively.

Hologic, a leader in imaging technology, acquired Cytyc, specialized in the detection of cervix cancer, for US$6.2 billion. The new group is in a position to offer a comprehensive spectrum of diagnostics for women's health care. Roche Diagnostics had acquired : 454 Life Sciences (sequencing), NimbleGen Systems (biochips), BioVeris (immunochemistry) and the American firm Ventana Medical Systems (world leader in histo-pathological diagnosis).

In the area of molecular diagnosis, the Dutch group Qiagen had made its biggest acquisition when it purchased the molecular biology firm Digene for US$1.6 billion. The record of acquisitions in the diagnostics area was that of Inverness Medical Innovation (a dozen acquisitions in 2007), which acquired the California-based diagnostics company Biosite.

Bio-Rad has become one of the three world leaders in immunohaematology after acquiring the Swiss company Diamed (specialized in the production of reagents and tools for blood typing) in October 2007 (J. Kadouche, personal communication, 2009).

Tissue sampling, storage and property

In *The Immortal Life Of Henriette Lacks*, published in 2010, Rebecca Skloot tells the story of Ms. Lacks and her family, as well as that of one of the most important medical discoveries of the last 100 years (Skloot, 2010). Before Henriette Lacks died from a cervix cancer in 1951, cancer cells were removed through a biopsy and cultured without the permission of the patient. They were named HeLa cells, from the first two letters of her first and last names, and they reproduced in a laboratory at Johns Hopkins University, Baltimore – the first human cells ever to do so. Meanwhile, Ms. Lacks, a 31-year-old African-American who had once been a tobacco farmer, tended her five children and received radiotherapy in the hospital's "coloured" ward (Margonelli, 2010).

Scientists have grown some 50 million tons of HeLa cells, which can be ordered over the telephone. HeLa have been the subject of 60,000 scientific studies, with nearly 10 more being published every day, on numerous areas of biology and medicine (e.g. ageing, cancer). In 1966, it became clear that HeLa cells had contaminated hundreds of cell lines. By 1973, when Ms. Lacks' children were shocked to learn that their mother's cells were still alive, HeLa cells had already been to outer space (Margonelli, 2010).

Some of the family members felt they had been cheated by either Johns Hopkins, although the hospital never sold the cells, or the entire medical establishment, which had made a lot of profit from the cells. R. Skloot traces the family's ordeal, the changing ethics and laws around tissue collections, and the journalists and researchers who violated the family's privacy by publishing everything from Ms. Lacks' medical records to the family's genetic information (Margonelli, 2010).

The book authored by Rebecca Skloot is also a critique of science that insists on ignoring the human provenance of its materials. "Scientists do not like to think of HeLa cells as being little bits of Henriette because it's much easier to do science when you dissociate your materials from the people they come from," Robert Stevenson, a researcher, told R. Skloot in one of the numerous ethical discussions reported in the book (Margonelli, 2010). There are indeed many ethical issues in the story of

HeLa cells. Since 1951, biomedical science has progressed much faster than the ability to figure out what is right and wrong about tissue culture. During the 1980s, a physician who had removed the spleen of a patient suffering from cancer, named John Moore, patented some of the cells to create a cell line then valued at more than US$3 billion, without J. Moore's knowledge. J. Moore sued, and on appeal the court ruled that patients had the right to control their tissues, but soon that was struck down by the California Supreme Court, which stated that tissue removed from the body had been abandoned as medical waste. The cell line created by the physician had been "transformed" via his "inventive effort," and to say otherwise would "destroy the economic incentive to conduct important medical research." The court stated that physicians should disclose their financial interests and called on legislators to increase patient protections and regulation. In 1999, Rand Corp. estimated that American laboratories alone held more than 307 million tissue samples from some 178 million people. Not only is the issue of payment for profitable tissues unresolved, as noted by R. Skloot, but it is also still not necessary to obtain consent to store cells and tissue taken in diagnostic procedures and then use the samples for research (Margonelli, 2010).

Diagnosis of infectious diseases

Tuberculosis is a global public health problem. According to the World Health Organization (WHO), one-third of the world's population runs the risk to catch an active form of the disease. About 1.7 million people die from tuberculosis every year, due to a large extent to the fact that the illness has not been diagnosed or too late to be effectively treated. Of the 9 million people who develop the infection annually, most of them do not receive a diagnosis confirmed by a laboratory. Only 2.2 million cases of tuberculosis are diagnosed and registered officially, using the microscopic examination of a smear of blood or tissue sample spread on a slide and coloured with the Ziehl-Nielsen dye. This is very often the only diagnostic test available in many developing countries; its sensitivity is low, because it is positive only when the concentration of tuberculosis bacteria (*Mycobacterium tuberculosis*) is higher than 10 µ/ml. When the test is positive, it means that acid and alcohol-resistant microorganisms are present – a trait shared by all mycobacteria. Another diagnostic test consists of culturing the sample(s) on Lowenstein-Jensen culture medium, followed by the identification of *M. tuberculosis*; results are available after three weeks at least and specialized laboratories are needed.

The development of improved and accurate diagnostics for tuberculosis responds to a high demand and will contribute to a more effective control

of the disease, which is reemerging in many countries whose health-care system has worsened (e.g. Russia) and which is becoming increasingly difficult to treat due to the resistance developed by the pathogen to the drugs used currently.

The value of the global diagnostic market for tuberculosis has been estimated at twice that of the drugs used to treat the disease. According to the World Health Organization's data published at the end of 2006, about US$1 billion were spent annually throughout the world to carry out diagnostic tests so as to detect the pathogen among some 10 million people, while about US$300 million were devoted to purchase drugs for treating the disease. In low or intermediate income countries where three-quarters of the detection tests are carried out, US$326 million are spent to purchase diagnostics, and there is an important potential market for lower-cost and more accurate tests. Indeed, 70% to 90% of the potential market for new diagnostics applied to tuberculosis exists in the 22 countries where the disease prevails. In these countries, there is an urgent need to acquire and develop new diagnostic tools, because the conventional microscopic examination, bacillus culture and X-ray scan of the chest cannot accurately detect an active form of the disease, particularly among patients suffering from HIV/AIDS (this pandemic is fuelling the spread of tuberculosis and other infectious diseases, such as pneumonia, among immuno-deficient patients), as well as the drug-resistant strains of *M. tuberculosis*.

New diagnostic tools include immunological methods, that consist of stimulating T lymphocytes by specific antigens of the pathogen (ESAT 6 and CFP 10), which induces the production of gamma-interferon. The *in vitro* exposure of the T lymphocytes stimulated by well identified antigens, to a patient's (or a suspected patient) serum will be followed by the significant production of gamma-interferon that can be measured. This response is observed only among persons who have been infected before, and is not blurred by earlier exposures to the tuberculosis vaccine (BCG, Bacillus Calmette Guérin) or to other mycobacteria.

Two tests have been approved and used internationally : QantiFERON TB Gold and T SPOT-TB (see 2005 Guidelines of the Centers for Disease Control – CDC – Division of Tuberculosis Elimination, United States). Their high specificity for detecting the disease is estimated at 85% and 90%, respectively. However, these tests are not quantitative and cannot give indications on when the infection occurred, or predict the risk of transition to the tuberculosis disease. Their specificity is by no means higher than that of the intradermic reaction (IDR). Both tests are generally carried out

for diagnosing latent tuberculosis infection, e.g. among migrants who may have been infected in their country of origin or among groups at risk.

Other new diagnostic tools concern the tests for resistance to antibiotics by *M. tuberculosis*. These tools have been developed because most mutations in the bacterial genome related with drug resistance have been identified. When these mutations are localized on short nucleotide sequences of the genes encoding the targets of the main antibiotics, they can be detected after amplifying them. The mutation can thereafter be precisely detected through sequencing the amplified sequence, which can be carried out in specialized laboratories. The LIPA (for "Line Probe Assay") technique consists of amplifying the nucleotide sequence that may mutate, then of hybridizing it with, on the other hand, probes specific to its normal structure, and on the other with probes specific to the main known point mutations. Probes are fixed on a strip, and the detection of the hybrids is an enzymatic one and shows up as a coloured reaction. This technique leads to results in 24 hours regarding the detection of resistance to rifampicin, which is due to mutations in a well defined region of gene rpo. The LIPA technique gives reliable results, it is costly and should be carried out by well trained staff; it is generally reserved to patients where pathogen multiresistance is suspected (J. Kadouche, personal communication, 2009).

When the mutations relating to drug resistance are located on long nucleotide sequences, or even on several genes, they cannot be detected by simple amplification and hybridization. This is the case for resistance to isoniazide and, to a lesser degree, to pyrazinamide. Recent development of DNA biochips that permit the hybridization of amplified DNA with thousands of probes laid on a very small area, is a very promising diagnostic tool aimed at detecting the resistances of *M. tuberculosis* rapidly (J. Kadouche, personal communication, 2009).

Hepatitis C is a widespread disease, as it is estimated that 3% of the world's population (200 million people approximately) is affected by a chronic disease (mainly liver) caused by the hepatitis C virus – a flavivirus. Viral transmission is mainly through blood; sexual transmission exists, but it is exceptional and it is indeed associated with blood contact. There is a risk of transmission from the mother to the infant during delivery, only if the virus is detectable in the mother's blood. This risk, estimated at 5% or even more, becomes a reality if the mother is infected by both the hepatitis C virus and HIV.

Generally infection by the hepatitis virus starts with a silent incubation period the length of which depends on the quantity of virus transmitted; the average is two months. Thereafter, the disease is diagnosed through an acute hepatitis in 10% to 20% of the cases. The duration of the hepatitis is about three months and the symptoms are digestive ailments, liver pain and jaundice eventually, the patient being very tired. In 70% to 80% of the cases, the disease evolves towards a liver cancer or cirrhosis. Furthermore, hepatitis C can evolve during 15 to 20 years quietly before showing serious symptoms and the prognosis is lethal. According to WHO's estimations, 50% to 70% of the virus carriers are not detected and can become seriously ill (J. Kadouche, personal communication, 2009).

Virus detection is therefore an important public health measure. Diagnosis is simple and consists of searching for the presence of antibodies against the hepatitis C virus (anti-HCV), as well as of analyzing the viral RNA, like in the case of the HIV. The blood test is carried out six to eight weeks after the viral infection in order to detect the anti-HCV antibodies. This presence only means that the person has been in contact with the virus, and not that the person is ill. Other tests have to be performed after the detection of antibodies in order to understand how the disease is evolving.

Qualitative PCR (polymerase chain reaction) is the most accurate technique for bringing the evidence that the virus is multiplying and certainly active against the liver. If this is the case viral RNA will be found in significant amounts in the patient's blood. Virus replication is therefore the actual evidence of its detrimental activity. However, it is not easy to detect it in immunodeficient patients, such as those co-infected by both the HCV and HIV. A new test is being developed to measure the virus antigens and could become a more precise alternative to qualitative PCR. Once the virus activity has been demonstrated, quantitative PCR is performed in order to determine viremia, i.e. the number of virus copies in the blood. Under 100 copies per ml of blood, the interpretation of the test is not relevant; 100 copies of the virus per ml is therefore the limit of sensitivity of the test. A high viremia is about 800,000 copies per ml.

There are six different HCV genotypes and it is important to know the genotype that affects the patient in order to adapt the kind and duration of treatment. Generally, genotypes 1, 4, 5 and 6, with a high viremia, entail a one-year treatment, while genotypes 2 and 3 with a low viremia are treated during half a year. Two tests can be carried out for virus typing (J. Kadouche, personal communication, 2009).

The **Epstein-Barr virus (EBV)**, discovered in 1964, belongs to the herpes virus family and can induce benign or malignant tumours in children and adults. Interaction between environmental factors and genetics is clearly at the origin of cancer development caused by EBV. Three very different diseases are caused by the EBV : infectious mononucleosis ("kiss disease"), widespread in Europe; Burkitt lymphoma in African children; and nasopharyngeal cancer (NPC) that occurs in Southern China, North Africa and in the Arctic regions.

The NPC develops behind the nasal cavities, which play a filter role and a barrier to inhaled dust and microbes; the filter is composed of ciliated respiratory mucosa that covers a sub-mucosa rich in lymphocytes, and it is at the convergence of epithelial cells and lymphocytes that the tumour can develop. The EBV is present in the cancerous epithelial cells. It encodes more than 100 of specific proteins, one of them playing a key role in the interaction with nasopharynx cells.

In collaboration with Guy de Thé of the Institut Pasteur, Paris, a Chinese team has used the EBV for a very early detection of NPC, which can be followed by radiotherapy and the cure of 80% of early detected cancers. An antibody targeted against the EBV can detect not only the tumours, but also individuals that have a high risk to develop NPC within 12 to 24 months (Desgranges and de Thé, 2006).

As the presence and replication of the EBV has been related to high amounts of IgA immunoglobulin, the diagnosis and follow-up treatment of the nasopharyngeal cancer (NPC) associated with EBV, are based on the serum detection (ELISA) of antibodies anti-EA (Early Antigen) and anti-VCA (viral capsid antigen) of the IgA class. The EBV DNA in the bloodstream can also be measured through quantitative PCR in real time and this is also a good marker (J. Kadouche, personal communication, 2009).

The three above-mentioned examples illustrate the clinical and cost-effectiveness of *in vitro* diagnostics, as well as the good prospects they offer for combating infectious diseases, which remain a major cause of mortality and morbidity worldwide. These examples also illustrate the relevance of *in vitro* diagnostics for poor or intermediate income countries, where early detection of diseases can save most of therapy costs and improve the health status of their populations.

Diagnosis of Alzheimer's disease

According to the data published in the report of Alzheimer's Disease International, issued on 21 September 2010 – the 17th global day of the struggle against the disease –, 35 million people were affected by Alzheimer's disease in the world in 2009. This figure may double in 20 years to reach 66 million patients in 2030, and 115 million in 2050. In France, 860,000 patients suffered from the disease in 2009, and 225,000 new cases were diagnosed every year.

In the United States, more than 5 million persons suffered from Alzheimer's disease in 2009, a number that was expected to rise to 13.4 million by 2050. The therapies that exist – drugs and lifestyle behaviour such as keeping the mind sharp with enriching social relationships and stimulating the brain with games and puzzles – can only delay, not stop, the onset of memory loss, confusion and cognitive decline that generally extend over a period of several years or, more often, decades. Health experts estimate that a 65-year-old American has a 10% risk of developing Alzheimer's disease. The illness adds to health-care costs, and there is a considerable psychological price too, for patients and caregivers alike – and a fear factor. In a recent American poll, conducted for *The Shriver Report*: *A Woman's Nation Takes On Alzheimer's*, 84% of adults were concerned that they or someone in their family would be affected by the disease (Park, 2010).

That fear is compounded by the belief that research on Alzheimer's disease is lagging behind that on other diseases. While 81% of those polled in the United States saw great progress being made in curing heart disease and 74% stated the same for cancer, only 48% felt researchers were making strides in combating Alzheimer's disease. "We spend US$5.6 billion a year funding cancer studies, US$1 billion a year for heart disease … and US$500 million to study Alzheimer's," stated Ronald Peterson, director of the Mayo Clinic Alzheimer's Disease Center, jointly based in Florida and Minnesota (Park, 2010).

Hopes of curing Alzheimer's disease have met with great disappointment. In 2002, a promising vaccine caused dangerous inflammation in the brain and spinal column and had to be abandoned after years of research; in August 2010, a highly anticipated drug worsened rather than improved cognitive symptoms. Experts are now convinced that it is crucial to treat patients as early as possible, rather than attempt to improve a brain already scourged by the disease. The second lesson involves the scope of the medical approach : adopting a multipronged approach that

addresses as many of the disease complex abnormalities as possible may improve the chances that new therapies used early on will not only delay symptoms but also reverse them (Park, 2010).

Cognitive decline is a natural consequence of ageing, and confusion and memory loss are often just inconvenient parts of becoming older. It was therefore understandable that physicians were reluctant to introduce more uncertainty by attempting to tease apart Alzheimer's dementia from the so-called senior moments typical of the normal ageing process. Initially it made sense for researchers and drugmakers to focus on finding ways to shrink amyloid protein-plaque buildup and reduce the amyloid burden in the brain. But these efforts have failed and had side effects, because the agents that target amyloid plaques affect other processes, including those regulating how cells communicate as well as the development of heart, pancreas and immune-system cells. In addition, it is not clear that eliminating plaques has any effect on brain function. For instance, when scientists analyzed autopsied brains of patients in the failed vaccine trial, they noted that the subjects had fewer plaques than before they received the vaccine, but yet had shown no improvement on tests of mental function. More confusing was the observation that in tests involving animals with the equivalent of Alzheimer's disease, mice whose brains were loaded with amyloid protein performed as well as those without the plaques. Perhaps amyloid plaques were only one of the many factors of the pathology. It was also possible that amyloid protein was indeed pushing the disease but the vaccine and drugs used to dissolve the plaques were introduced too late and in too small a dose. Many of those drugs were designed to block the breakdown of amyloid into smaller fragments, which have a greater tendency to clump together. "By the time a person is impaired to the point of dementia, there is probably sufficient damage done to the brain that we really can't reverse it," stated Ronald Peterson of the Mayo Clinic Alzheimer's Disease Research Center (Park, 2010).

If that would be the case, then testing the drugs on patients whose brains are just beginning to accumulate amyloid protein might be more successful. But finding such patients, many of whom show no signs of memory loss or decline in mental function, is a real challenge. Consequently, in 2004, the US National Institute of Aging (NIA), part of the National Institutes of Health, partnered with pharmaceutical companies to create the Alzheimer's Disease Neuroimaging Initiative (ADNI), a US$60 million project aiming at identifying easily detectable differences – preferably through blood tests or brain scans – between Alzheimer's patients and unaffected individuals. The project dealt with

600 patients who either already suffered from symptoms of Alzheimer's dementia or had mild cognitive impairment – a preliminary stage of the disease – as well as 200 cognitively normal control-group volunteers (Park, 2010).

In carrying out the project, a few dozen intriguing protein markers in blood and spinal fluid that may indicate the onset of Alzheimer's disease have been isolated, and they could help researchers identify high-risk individuals before symptoms set in. Also, newer, better brain scans are helping detect the amyloid patterns that previously could be verified only by autopsy (Park, 2010).

In July 2010, the NIA and the Alzheimer's Association of the United States decided to update their criteria for helping physicians diagnose Alzheimer's disease by defining three distinct patients : those who are symptom-free but at high risk, those with mild cognitive impairment and those with Alzheimer's dementia. The guidelines fold in the latest understanding of how brain scans and other tests can help distinguish among the three groups and perhaps even specify which treatments among the many being explored might be most effective at each stage of the disease (Park, 2010).

Because Alzheimer's disease cannot be absolutely, definitively diagnosed until death, patients are given a probable diagnosis based on their performance on memory and recall tests, and reports from family members. The blood and spinal-fluid tests, along with the brain scans, could improve the predictive accuracy of these measures. The early data looked promising : the screens may be 80% to 90% accurate in picking up the earliest signs of the disease. In addition to diagnostic value, it allows researchers to start targeting candidate medications and be more confident that the patients who receive them will benefit. Coupling screening tests with treatment could cut a 65-year-old's lifetime risk of developing the disease in half (Park, 2010).

It is true that ever since Alzheimer's disease has been described by a German physician, Alois Alzheimer, in 1906, there was only one way to know for sure that a person had it. A pathologist, examining the brain after death, would see microscopic black freckles, plaque, sticking to brain slices. Without plaque, a person with memory loss did not have the disease. Every major drug company now has new experimental drugs it hopes it will work, particularly if they are taken much earlier. The question though, is who should be given the drugs : who really has Alzheimer's or is developing it? (Kolata, 2010).

On 11 June 2010 several big pharmaceutical companies publicized the profiles of 4,000 patients from 11 trials so that they could learn from each other's failures. The disease is estimated to cost America alone some US$170 billion a year (2009). And the number of people suffering from the disease is expected to triple by 2050 (*The Economist*, 2010 h).

At the turn of the century, research on Alzheimer's disease seemed promising. Many drugs which treated symptoms of the illness had just hit the market and researchers were setting out confidently on a deeper investigation of its causes. Unfortunately a long list of drugs have failed in late-stage clinical trails, at enormous costs to the companies producing them. The latest of these, Dimebon, made by Pfizer, was abandoned in March 2010, after US$725 million had been spent on research and development (*The Economist*, 2010 h).

The sticky plaques of beta-amyloid protein, and nerve-cell-engulfing tangles of a second type, called tau protein, were the subject of research for the past two decades because it was thought the disease was caused by the plaques (and the tangles are mere consequence); and most attention was given to developing drugs that will remove amyloid plaques from an affected brain. Five drugs that do this are on the market, but they only delay the onset of senile dementia. Once their effectiveness has run its course, memory loss and cognitive decline progress unimpeded, and sometimes even accelerate. As a consequence of this, the plaque theory is waning. Most researchers still believe beta-amyloid protein is the culprit, but the idea that free-floating protein molecules, rather than the proteins in the plaques are to blame is gaining ground. This assumption is supported by a study published in April 2010 in the *Annals of Neurology*, which showed that mice without plaques, but with floating beta-amyloid protein, were just as weakened by the disease as mice with both. If that were true in people, too, many more drugs now in clinical trials might prove to be ineffective (Arnold et al., 2010).

Concentrating on amyloid protein alone is not sufficient to reverse Alzheimer's disease, so researchers are trying hard to identify additional targets. Among the potential areas of interest are genes like that coding for apolipoprotein E (Apo E), which in certain forms can promote the formation of amyloid. Also of interest is the neural protein tau, which stabilizes axons – the long extensions that nerve cells send out to communicate with one another and reach tissues like muscles in the fingers and toes. Researchers believe that the disease begins, when for still unknown genetic and other reasons, the brain starts to produce amyloid-protein molecules that clump together, forming a plaque. Once

the plaques form, tau proteins that maintain the structural integrity of the neural highway break down, leaving the equivalent of potholes which interrupt the electrical signals travelling along the nerves. With this communication flow disrupted, nerve cells start to wither and die, leaving behind their tangled remains. That in turn activates the immune system's inflammatory response, which attempts to remove the debris (Park, 2010).

The result is a brain full of dead and dying neurons, and the shutdown of neural connections leads to a drop in cognitive function. Targeting each of these steps – inhibiting amyloid-protein production, controlling the formation of amyloid fragments, and limiting tau breakdown of nerve connections – may be necessary to control the resulting dysfunctioning of the brain. "But the difficult thing is to know what the relative input of each is on the human condition. And we won't know this until we have found drugs that can block each of them or combinations of them, to see how much improvement results," stated Lennart Mucke, director of the Gladstone Institute for Neurological Diseases and a neurologist at the University of California – UCSF (Park, 2010).

But even if therapies are years or decades away, identifying patients earlier in the disease cycle will remain valuable. By knowing they are at risk for Alzheimer's disease, patients can plan better for the future and make changes to their lifestyle, such as exercising and staying mentally and socially engaged – behaviours known to delay the onset of symptoms (Park, 2010).

The Dominantly Inherited Alzheimer Network, based at Washington University in St. Louis, Missouri, is taking another approach. Its researchers are studying families with a genetic mutation that triggers the early onset of Alzheimer's disease. That means it is possible to predict which members of a family are destined to have the disease, and compare their biochemical features with those of relatives who have not the mutation (*The Economist*, 2010 h).

Development of an early accurate diagnosis

Daniel Skovronsky, a physician at the University of Pennsylvania, has been working with the chemist Hank Kung for nine years to find and develop a radioactive dye, that can enter the brain and stick to plaque. They labelled the dye with a commonly used radioactive tracer and use a scanner to directly see the plaque in a living person's brain. D. Skovronsky left academia and formed Avid Radiopharmaceuticals to design a study with

hospice patients to prove that his radioactive dye can stick to plaques and be seen on scans. The research conducted at Avid Radiopharmaceuticals built on earlier work by two scientists at the University of Pittsburgh, who developed an amyloid dye that, while not practical for widespread use, surprised researchers by showing it seemed possible to see amyloid protein in a living brain. The problem was that the compound they relied upon used carbon 11 as its radioactive tracer : its half life is 20 minutes. Consequently, it has to be produced in a cyclotron in the basement of a medical centre and be quickly attached to the dye, injected to a patient lying in a scanner. That was not practical. D. Skovronsky used fluoride 18, whose half life is two hours, so that it could be made in the morning and used in the afternoon. And fluoride 18 is made routinely for 2 million cancer scans each year in the United States (Kolata, 2010).

On 23 October 2008, Avid Radiopharmaceuticals and two other companies, Bayer and General Electric, that were developing fluoride 18-based dyes for amyloid-protein scans, were asked by the US Food and Drug Administration (FDA) to answer the following question : how do you know what you are seeing on scans is the same as the amyloïd protein on autopsy? It seemed impossible to answer. If researchers wait for their patients to die before comparing scans with brain-autopsy examination, they can be waiting for a long time. But this was the condition the FDA put in order to approve the use of the dye (Kolata, 2010).

Avid Radiopharmaceuticals had a plan, and the advisory committee agreed in principle that it would work. Patients in hospices would be study subjects, some with dementia, others without. All would have memory tests and brain scans. And the company suggested that after the first 35 patients died, there should be enough data to know if the scans gave a real picture of the pathological conditions. By the end of 2009, the initial results of the hospice study concerning the first six patients were seen by the company. As more patients were studied, the data were held by the analysis company and Avid Radiopharmaceuticals did not see the results until the study was completed. A man diagnosed with Alzheimer's disease and cancer had a scan showing no plaque. Neither did the autopsy. Alzheimer's disease had been misdiagnosed. Another man with Parkinson's disease and dementia had been diagnosed as having dementia solely due to Parkinson's. His scan showed no amyloid plaque; so did the autopsy. He also had Alzheimer's. A woman with mild memory loss had a scan showing no amyloid plaque; her autopsy also found none. Three others had clinical diagnoses of Alzheimer's disease, confirmed by scans and autopsies. Finally, on 14 May 2010, 35 patients had been scanned and autopsied. The study was completed

and the results were disclosed by the company responsible for analyzing them. D. Skovronsky's assumption was therefore correct, while the other pharmaceutical companies were still carrying out their studies and had not yet data to be examined (Kolata, 2010).

The findings by Avid Radiopharmaceuticals were presented at an international meeting of the Alzheimer's Association in Honolulu on 11 July 2010. They have not yet been approved by the US FDA. But if they are, it will mean that for the first time physicians would have a reliable way to diagnose the presence of Alzheimer's disease in patients with memory loss problems. It will be therefore possible to compare Avid's technique with those developed by the Alzheimer's Disease Neuroimaging Initiative (ADNI) and the Dominantly Inherited Alzheimer Network.

One thing is sure : research on the disease should be enhanced in order to better understand its aetiology and its early accurate diagnosis is an important step towards a possible cure. Yet research on Alzheimer's disease was to receive only US$480 million in 2011, compared with US$643 million spent by NIH in 2006. These cuts are part of a general reduction in the agency's budget. This may not be the wisest thing to do, when the tripling of future sufferers will be very expensive for the nation's health-care system (*The Economist*, 2010 h).

New portable and rapid diagnostic tools

Diagnostic tests are generally carried out on samples taken from, or provided by, the patients, with the help of big, expensive machines in central laboratories. This is often unfeasible in developing countries and therefore the solution would be to have more portable and precise diagnostic tools, so as to favour a decentralized system of health care.

At Becton Dickinson (BD), an American diagnostics giant, a new type of diagnostic toolkit is being developed, thanks to the fusion of genomics, proteomics and information technologies. The impact of this kind of diagnostics would be as big as that of mobile phones, extending the reach of modern health care to places that are unserved or underserved. Diagnostics for All, initiated by students at MIT and Harvard University, has developed a range of diagnostic tests that are printed on ordinary paper and use microfluidics technology to direct the sample (e.g. a drop of blood) through tiny grooved channels to various chambers; chemicals then react with the sample, providing rapid diagnostic results (*The Economist*, 2009 a).

Ustar Biotechnologies, a Chinese start-up, has developed a cheap and portable diagnostic kit that it is marketing with BioHelix, a Boston-based firm. Similar companies from developing countries are researching, designing and manufacturing medical technology tools in order to suit local conditions best (*The Economist*, 2009 a).

The development and gradual spread of portable and rapid diagnostic tools is part of the move to digital medicine, that includes much more targeted drug delivery, telemedicine (e.g. monitoring and offering remote medical care to the elderly in their homes) using digitized data linked by internet video to physicians at a main hospital, ubiquitous and user-friendly medical devices. Medical technologies also have the potential to empower patients and give them the tools and data needed to take charge of their own health (*The Economist*, 2009 a).

Tracking nosocomial infections (*Staphylococcus aureus*)

Staphylococcus aureus is a common bacterium, very often present on the skin, but it can cause serious infections in the body after crossing the skin barrier. Some strains of this bacterium have become resistant to meticillin, one of the antibiotics mostly used against this pathogen. French data collected in 2006 showed that the proportion of patients infected with a meticillin-resistant *Staphylococcus aureus* (MRSA) was 0.39%, i.e. 8% of patients that had been affected by a so-called nosocomial disease during their stay in a hospital or health centre. The frequence of these diseases is higher in intensive-care units. One-fifth (24%) of *S. aureus* isolated during invasive infections were resistant to meticillin (Benkimoun, 2010 b).

Current techniques used to distinguish the various strains of pathogenic bacteria are based on the analysis of certain DNA sequences. They are not sufficiently precise for determining the relationship between these strains. Simon Harris and Stephen Bentley of the Wellcome Trust Sanger Institute at Cambridge, United Kingdom, have used a new method for the identification of the relevant DNA sequences with the help of machines that can sequence the whole bacterial genome. They were able to precisely trace the transmission of MRSA strains between patients of the same hospital, as well as the transmission of strains between one continent and another. Their results were published in *Science* on 22 January 2010 (Harris et al., 2010).

The British team had analyzed two types of samples : on the one hand, 42 samples from patients infected by MRSA strains between 1982 and 2003 worldwide; and on the other, 20 samples from patients that

developed an MRSA infection within seven months and who were in the same hospital located in northeastern Thailand (all of them could belong to a transmission chain from person to person). The researchers found that all the samples, but one, from South America (Brazil, Chile, Argentina and Uruguay), belonged to one MRSA subtype, which may reflect the expansion of the same variant throughout the subcontinent. Despite a wider diversity, the strains isolated in China and Thailand seemed to belong to the same "Asian" subtype. The diversity of strains present in Europe does not seem to exclude a possible European origin for one of the most often found MRSA strains (Harris et al., 2010).

Two strains, one found in Denmark and the other that caused an epidemic for two years in a London hospital, were related to the Thai strain. The enquiry carried out showed that the infection in Denmark was linked to a Thai patient, while that of London had its origin in South-East Asia. The screening of the bacterial samples made in the Thai hospital revealed that a point mutation in the bacterial genome occurred every six weeks. It also showed that out of the 20 samples made in that hospital, five did diverge by only 14 point mutations. These samples had been taken from patients hospitalized in wards that were close to each other. By contrast, these wards were not represented in the most divergent samples (Harris et al., 2010).

Although genomic plasticity was known among bacteria, the British scientists have been able to demonstrate that microevolution with the help of the new high-throughput sequencing tools. The authors consider that their observations reveal a limited number of effective intercontinental transmission events as well as the expression of variants (derived from the same line) which, in some cases, have become dominant in their new geographical area. The British scientists see two potential applications of their work : detection of these new appearances and design of strengthened control measures against these infections, as it was done during the London epidemic; and screening of the strains isolated in the same hospital or health centre, while applying the genetic methodology to epidemiological analysis (Harris et al., 2010).

Bruno Coignard, head of the unit for nosocomial infections and resistance to antibiotics at the French Institute of Health Vigilance (*Institut de veille sanitaire*), has been less optimistic regarding the application of the research carried out by the British scientists to the control of nosocomial infections. According to him, the frequency of "national" strains (MRSA) is such that the likelihood of infection by a strain imported from a foreign country is much lower. In addition, "nothing replaces the washing of hands,"

reiterated François Vandenesch, of the National Reference Centre for Toxaemias due to Staphylococci. In French hospitals, simple prophylactic measures (e.g. washing of hands) have been efficient : between 2005 and 2008, nosocomial infections have decreased by about 4% per year. But the new method developed by the British researchers could apply to emergent and antibiotic-resistant bacteria, for instance to enterobacteria. The latter are more frequent around the Mediterranean basin, particularly in Greece. Another effective means to control nosocomial infections is to limit the use of antibiotics to only the situations where they are really needed. For instance, in Norway, this approach has led to an important decrease in the proportion of MRSA-caused infections : for years this proportion has not exceeded 1% of all infections caused by staphylococci, compared with 24% in France, 38% in Greece, 44% in Israel, 63% in the United States and 80% in Japan. Also in the United Kingdom where this proportion reached 45%, a national prophylactic programme has resulted in a significant decrease of infections caused by MRSAs (Benkimoun, 2010 b). See also Vincent (2010 e).

GENETIC TESTS

Sequencing the human genome : achievements and prospects

Tenth anniversary of the Human Genome Project

June 26th 2010 marked the tenth anniversary of the reading of the human genome – the 3-billion-letter-long message that prompted self-knowledge to humanity (as was recommended by one of the two maxims posted above the chamber of the oracle at Delphi for the edification of those who sought her prophecy : "Know thyself;" the other maxim being "nothing in excess") [*The Economist*, 2010 i].

Six months before the end of his second mandate, on 26 June 2000, President Bill Clinton was standing in the East Room, the large reception room of the White House. In that same room, Thomas Jefferson, the third president of the United States of America, had been handed over by the explorators Meriwether Lewis and William Clark the map of the North-West Territories of the USA, which had been established at the end of an epic expedition carried out from 1804 to 1806. Almost a century after, President Bill Clinton welcomed Francis Collins, then the leader of the International Human Genome Sequencing Consortium, and J. Craig Venter, head of Celera Genomics, a private biotechnology company. They brought him another kind of map, that of the two first sketches of the human genome. Bother runners, from the public and the private sector, won the prize of shaking hand of America's president. The event caught the public imagination at the time. The president stated in the presence of prestigious scientists : "Genome science will have a real impact on our life and even more on that of our children. It will revolutionize the diagnosis, prevention and therapy of most, if not all, human diseases" (*The Economist*, 2010 i, j; Benkimoun, 2010 g).

F. Collins and J. Craig Venter were competing in the race to sequence the human genome. While the United States was playing a leading role, the project of the public sector was based on the power of an international

consortium largely supported by public funding. J. Craig Venter, by contrast, believed in the flexibility of a private structure he created in 1998 and in the development of a newly developed sequencing technology. He also played the role of a stimulus to the whole endeavour. In fact, "the venture started in 1984-1985, with three meetings in which I am the only one who participated each time; the first one took place in Utah on the topic of DNA methods aimed at detecting mutations," reported George Church of Harvard University Faculty of Medicine. He added : "Since 1976, Wally Gilbert has been thinking of the sequencing of DNA; I moved to his laboratory in Harvard the following year." Later on, W. Gilbert founded the biotechnology start-ups Biogen and Myriad Genetics. Further to these meetings, George Church was one of those who received funding for the human genome sequencing from the US Department of Energy (DoE), which launched the *Human Genome Initiative* in 1986. According to G. hurch, the DoE had three reasons for launching this project : it wanted to measure and treat all the health consequences associated with sources of energy, like radiations; it had big capacities in informatics and some of its engineers at Los Alamos had collections of DNA sequences; finally, it possessed powerful lasers that could be used to provoke lesions on the chromosomes (Benkimoun, 2010 g).

In 1987, James Watson, co-discoverer with Francis Crick, Maurice Wilkins and Rosalind Franklin of the double-helical structure of DNA, was responsible for suggesting the human-genome project in the first place. At the time, he was head of America's National Center for Human Genome Research, part of the National Institutes of Health (NIH). He had a strong discrepancy with NIH over the issue of patenting DNA sequences called expressed sequence tags (ESTs). He opposed this, arguing that "you should not patent something a monkey could do." That did not endear him to J. Craig Venter, who created those DNA sequences and wanted to patent them. J. Watson was replaced by Francis Collins, while he continued as head of the Cold Spring Harbour genetics laboratory until 2007, when he made some injudicious remarks about genetics and black people, and found himself suddenly retired (*The Economist*, 2010 j).

The objective was to fully sequence the human genome in 2015. At the same time, initiatives with the same goal were launched in Japan, the United Kingdom and the rest of the European Union. In France the alliance between the researchers of the Centre for the Study of Human Polymorphism, created by Jean Dausset, Daniel Cohen, with Jean Weissenbach, and the French Association against Myopathologies – that organizes the Telethon – was crucial. Private funding coming from the generosity of French citizens allowed the creation of Genethon in 1990

and, between 1990 and 1996, it was possible, under the leadership of Jean Weissenbach, to map the human genome (localization of the genes on the chromosomes) [Benkimoun, 2010 g].

J. Craig Venter, too, left the NIH in the wake of the expressed-sequence-tag incident. At first, he teamed up with William Haseltine, a virus geneticist with a record as an entrepreneur, to create in 1992 an institute and a company, Human Genome Sciences Inc., to exploit expressed sequence tags. The two did not carry on for long, and in 1998 J. Craig Venter went on to help to create with Perkin-Elmer a second firm, Celera Genomics, in the hope of beating the public to the human genome. He used a new DNA-sequencing technique called whole-genome shotgunning that he and a colleague, Hamilton Smith, had invented, and patented a number of human genes on the way. That was anathema to F. Collins and his British counterpart, John Sulston (who was head of the Wellcome Trust's Sanger Centre, now known as the Sanger Institute, which did about a third of the public project), who wanted genes to be public goods and started racing Celera Genomics to stop the firm finding genes first (*The Economist*, 2010 j).

In December 1999, the Human Genome Project (HGP) published in *Nature* the full DNA sequence of chromosome 22, the first to be achieved. In April 2000, J. Craig Venter "retaliated" when he stated that "he had sequenced the genome of an individual and he started to assemble the sequenced fragments of that genome in the right order." Two months later, the HGP consortium declared it had almost completed a map of the human genome; this was the map presented to President Bill Clinton in the White House. On 11 February 2001, the HCP consortium and Celera Genomics, each one presented a "completed" version of the human genome, published in *Nature* and *Science*, respectively, 14 years ahead of the initial schedule (Benkimoun, 2010 g). Eric Lander, of the Whitehead Institute, in Cambridge, Massachusetts, achieved effective leadership of the American arm of the project. It was E. Lander whose name led the list of researchers on the paper published by *Nature* in 2001 (*The Economist*, 2010 j).

Celera Genomics' scientific triumph did not produce much in the way of revenue and J. Craig Venter left the firm in 2002 (W. Haseltine left Human Genome Sciences in 2004). He then went on a round-the-world cruise on his yacht, collecting bacterial samples from the sea for a project he named the Global Ocean Sampling Expedition. He also set up another research institute (which unveiled a microorganism with an artificial genome in May 2010) and another commercial arm, called Synthetic Genomics, in collaboration with Hamilton Smith (*The Economist*, 2010 j).

Eric Lander, with money from two Californian benefactors of that name has set up the Broad Institute, America's largest genome-sequencing laboratory, next door to the Whitehead Institute. John Sulston, meanwhile, had started a scientific-ethics institute at Manchester University and F. Collins had bagged one of the top prizes in American science. In 2010 he was the head of the National Institutes of Health (*The Economist*, 2010 j).

In fact, the race J. Craig Venter and Francis Collins were engaged in had been dogged by difficulties from the beginning. There was a false start (the announcement at the White House that the sequence was complete relied on a generous definition of that word; a truly complete sequence was not published until 2003). The competitors found at first that there were far fewer genes than they had expected, only to discover later that there were far more. These discoveries changed the meaning of the word "gene." They found the way genes were switched on and off as important, both biologically and medically, as the composition of those genes. They found that their methods for linking genetic variation to disease were inadequate. And they found, above all, that they had not enough genomes to work on. Each human genome is different, and that matters. However, the US$3 billion spent on the HGP was not wasted money. As *The Economist* observed at that time, "the race for deciphering the human genome was a race not to the finish but to the starting line" (*The Economist*, 2010 j).

The human genome

The human genome consists of 22 pairs of identical chromosomes, plus a pair of sexual chromosomes – two X chromosomes for the female, and one X and one Y chromosomes for the male. Each chromosome is made of a packed molecule of double-stranded DNA and specific proteins. Each DNA strand is made of four types of nucleotides aligned in a variable sequence, and containing a sugar molecule (desoxyribose), a phosphate molecule and a base : adenine (A), cytosine (C), guanine (G) and thymine (T). Both strands are linked to each other through linkages between the bases [adenine-thymine (AT) and guanine-cytosine (GC)] and the whole is arranged as a double helix.

The whole human genome or DNA contains 3 billion pairs of nucleotides or bases, but only 3% of this DNA codes for the synthesis of proteins, which are the building blocks of tissues and organs; they also catalyze chemical reactions in the body (enzymes) and serve as signals between cells and for the transport of ions and molecules through the cell membranes.

Genes are DNA sequences that contain the information used to synthesize not only one protein, as it was thought formerly, but three as an average. Genes can be activated and therefore expressed, or inactivated. The regulation of their expression involves the non-coding part of DNA. Sequencing the human genome has shown that it consisted of only 23,000 genes encoding proteins, compared with 13,000 genes of *Drosophila melanogaster* (fruit fly).

Progress in genome sequencing

Sequencing the genome has consisted of establishing the order of the nucleotide bases along the whole DNA in each chromosome. In a decade the sequencing equipment has been improved so as to sequence more nucleotides in shorter time lapses and, consequently, the cost of sequencing has plummeted. Thus, in the year 2000, the Sanger technique, developed about ten years earlier, allowed the sequencing of hundreds of thousands of nucleotide pairs in one single operation of the machine, and the cost was US$10,000 per million nucleotide pairs sequenced. In 2005, a new generation of sequencers (automate 454, Roche) could sequence hundreds of millions of nucleotide pairs in one single operation, and the cost plummeted to US$1,000 per million nucleotide pairs sequenced. Other companies like Solexa (which became Illumina) have modified the new generation of sequencers in order to sequence billions of nucleotide pairs in one single operation. By mid-2006, the cost fell to US$100 per million nucleotide pairs sequenced. In 2008, this cost was only US$10, and third-generation sequencers, designed by companies such as Helicos BioSciences, Pacific Biosciences, Oxford Nanopore Technologies or Ion Torrent, were expected to do the same work for just US$1 (Benkimoun, 2010 g).

In 2007, thanks to the new generation of sequencers, the diploid genome, i.e. the DNA contained in all the 23 chromosomes, of J. Craig Venter had been sequenced, while ten years had been necessary to complete the Human Genome Project. In 2008, the genomes of James Watson, of a woman suffering from an acute myeloid leukaemia, of a Yoruba man from Nigeria and of the first Asian individual were sequenced. In 2009, that was the case of two Korean men, a woman suffering from cancer, George Church, two Yorubas (a man and a women), and another four individuals. In 2010, thanks to the last generation of sequencers, the genomes of the four members of a family, a cancer cell line (glioma), Inuk, Gubi, archbishop Desmond Tutu of South Africa and James Lupski were sequenced. The cost of sequencing the whole genome has been divided by 10,000 in a decade, and the technological sophistication

of the machines employed have led to a huge accumulation of DNA data. In 2000, the main genetic databases (the US National Center for Biotechnology Information — NCBI —, the Japanese DNA Data Base and the European Molecular Biology Laboratory's Base of nucleotide Sequences) stored altogether 8 million identified nucleotide pairs. In the following years, these databases doubled in size every 18 months, and DNA sequences corresponding to some 270 billion nucleotide pairs were added. The three structures exchange information within the framework of the International Nucleotide Sequence Data Base Collaboration – INSDC, which includes data from genomes belonging to the human and other species (viruses, bacteria, plants, invertebrates and vertebrates) [Benkimoun, 2010 g].

Two other public structures, both under the aegis of NCBI, store a more considerable number of genetic data : the Trace Archives, which store the chromatogrammes ("traces") of DNA sequences drawn from various large-scale genome-sequencing projects, contain around 2,000 billion; the Sequence Read Archive (SRA) that stores the data derived from the new generation of sequencing platforms (for instance, Roche 454 GS System, Illumina Genome Analyzer, Applied Biosystems SOLID System, Helicos Heliscope) contained 25,000 billion nucleotide pairs in 2010. Both American institutions collaborate with their European and Japanese counterparts (Benkimoun, 2010 g).

In other words, technological breakthroughs have been as important to the development of genomics as intellectual insights have been. The telescope revolutionized astronomy; the microscope, biology; and the spectroscope, chemistry. Genomics development depends on two technological changes. One, in computing power, is generic, although computer-makers are slavering at the amount of data that genomics will need to process, and the kind of evermore sophisticated machines that will be necessary to do the processing. This huge amount of data, however, is the result of the second technological change that is driving genomics, in the power of DNA sequencing (*The Economist*, 2010 j).

Genome-sequencing implications

Computing has increased in potency according to Moore's law : computers double in power roughly every two years – an increase of more than 30 times over the course of a decade, with concomitant reductions in cost. Thus, Eric Lander, the head of the Broad Institute, in Cambridge, Massachusetts, which is America's biggest DNA-sequencing centre, calculated that the cost of DNA sequencing at his institute had

fallen to a hundred-thousandth of what it was a decade ago. The genome sequenced by the International Human Genome Sequencing Consortium took 13 years and cost US$3 billion. Now using the latest sequencers for Illumina, of San Diego, California, a human genome can be read in eight days at a cost of about US$10,000. Another Californian firm, Pacific Biosciences, of Menlo Park, has developed a technology that can read genomes from single DNA molecules; it thinks that in 2012-2013 a human genome could be mapped in 15 minutes for less than US$1,000. And a rival technology being developed in the United Kingdom by Oxford Nanopore Technologies was expected to be competitive in terms of speed and cost. This increase in speed and reduction in cost is an upheaval for the biology business. Until now, firms that claimed to read individual genomes have been using a shortcut. They have employed arrays of DNA probes, known as genetic chips, to look for pre-identified variations in their clients' DNA. Those variations had been discovered thanks to scientific collaborations such as the International HapMap Project, which search for mutations of the genetic code, called single-nucleotide polymorphisms, or SNPs, in blocks of DNA called haplotypes. A SNP is a place where a lone nucleotide base varies from person to person. Some 10 million SNPs were known in 2010, but among the 3 billion nucleotide bases there is reason to believe they are but a smattering of the total variation. Proper sequencing will reveal the lot (*The Economist*, 2010 j).

Finding the sequence – even the full range of sequences – is, though, just the beginning. Then one has to do something useful with the result. Computers allow individual genomes to be compared. And not only human genomes. Cross-species comparisons are very valuable. Laboratory experiments on living beings ranging from yeast to mice can reveal the functions of genes in these species. Computer comparison then reveals which human genes correspond in DNA sequence and thus, presumably, in function, to the genes in these "model" organisms. Cross-species comparison also shows how species differ, and thus how they have diverged. Comparing DNA from individuals within a population can explain why those individuals differ from one another. And comparing the DNA from cells within an individual can show how tissues develop and become differentiated from one another, and what goes wrong in pathological conditions (*The Economist*, 2010 j).

Even before cheap sequencing became available, huge databases were being built up as indicated previously. In connection with samples from patients, physicians' reports and – most valuable of all – long-term studies of particular groups of individuals, genetic information could be linked with the phenotype, or the organism's outward expression : its anatomy,

physiology and behaviour, whether healthy or pathological. The goal of genomics and the new biology that emerges from it (post-genomics) is to understand how the phenotype emerges from the genotype (*The Economist*, 2010 j).

"Because of the diversity of the human species, there is no 'normal' sequence of the human genome"…"We are all mutants," wrote Francis Collins, director of NIH, and his colleagues in an article published on 27 May 2010 in the *New England Journal of Medicine* (Feero et al., 2010). The word "normal" applies in fact to the most frequent variants in a particular population. When a variant appears in a population with a frequence higher than 1%, the word "polymorphism" is used. The word "mutation" generally applies to genetic mutations associated with a pathology. A single mutation concerning one base (nucleotide) can cause a genetic disease; for instance, in the cystic fibrosis or in Huntington chorea, a single variation in the gene is responsible for the development of the disease. But this is far from being the case of most of the diseases where a genetic component is involved. For instance, the sequencing of the genome of cancer cells has revealed the implication of some 50 mutations associated with cancer in an individual. In addition, these mutations are not the same in all patients suffering from one particular cancer (Benkimoun, 2010 g).

Sequencing the human genome has also shown that the genetic heritage of two persons of different ethnic origin was identical in the proportion of 99.6%. Human genomics is thus revealing that "humans are really brothers and sisters under the skin." The human species is young, so there has been little time for differences to evolve. From the moral viewpoint it would be good news. On the other hand it may turn out that some differences between and within groups are quite marked. If those differences were in sensitive traits like personality, behaviour or intelligence, real difficulties could ensue. The liberal answer is to respect people as individuals, regardless of their genetic make-up (*The Economist*, 2010 i).

An important consequence of sequencing the human genome is that it is now possible to compare *Homo sapiens* with his closest relative – not the living chimpanzee, with whom he parted company perhaps 5 million years ago, but the extinct Neanderthal. That is not, of course, why the Human Genome Project started. But the announcement of the Neanderthal man's genome sequence showed the power of human genomics. Putting together some 1.3 billion fragments of 40,000-year-old DNA, contaminated as they were with the fungi and bacteria of millennia of decay and the imprints of archaeologists who handled the bones,

demonstrated how far the technology had advanced over the course of the past decade. It also showed that we may have an answer to how life has evolved and diversified over the course of time, how flexible and perfectible the human really is (nature versus nurture) [*The Economist*, 2010 i, j].

Sequencing the Y chromosome and its implications for human evolution

Of the 46 human chromosomes, the Y chromosome is one of the smallest. It bears the genetic information that induces the development of a zygote into a male foetus. David C. Page, director of the Massachusetts Institute of Technology (MIT) Whitehead Institute, is considered the world specialist of the Y chromosome. He has been working on it for more than 30 years, since 1979 when he was a medical student. He devoted most of his research endeavour to sequencing that chromosome, before the Human Genome Project was designed and implemented. Since the 1950s, the prevailing idea was that the Y chromosome bore few genes and that it was not worth studying. Paradoxically, the discovery of the *SRY* (sex region Y) gene which induces the masculinization of the embryo, was followed by a decreased interest in the Y chromosome; scientists probably thought there was nothing interesting to be discovered. A few geneticists, including D. Page, however, pursued their research on that chromosome. In 1992, his team proposed a complete map of the Y chromosome without provoking great enthusiasm from the scientific community. David Page stated that "people saw that as a technical achievement, not as a biological success." It was nevertheless the beginning of a series of discoveries. For instance, it was shown that many non-fertile men had an incomplete Y chromosome. Genes for male fertility were therefore sought on the Y chromosome; these were the only active genes present on the chromosome in addition to the gene *SRY*. Thereafter, came the discovery of the relationship between X and Y chromosomes, and in 2010 the sequence of chimpanzee's Y chromosome was deciphered and it could be compared with that of the human Y chromosome (Chaigne, 2010).

In 2003, the sequencing of the human Y chromosome was carried out and on 14 January 2010, in *Nature*, American researchers led by D. Page published a comparison between man and chimpanzee with regard to the structure of their respective Y chromosomes. They reported that this chromosome was the portion of the human genome which has been mostly modified during evolution : while the genomes of man and chimpanzee are identical in the proportion of more than 99%, the sequences of their Y chromosomes are similar to the extent of 70% only (Hughes et al., 2010).

Researchers usually thought that the human Y chromosome had almost ended its evolution. Its sequencing had suggested it contained the genes for sexuality and degenerated DNA fragments. But D. Page and his colleagues have observed that the part of the chromosome containing important genes, or *MSY* (male-specific region), differed more in the two species than other regions of the genome; in other words, the part that was supposed to be the most stable during evolution, was in fact less stable. *MSY* is made of three types of sequences : masculinization genes, the only ones that have a true role in the male outlook; sequences derived from degenerated genes of X chromosome; and palindrome sequences, i.e. that are read in an identical way in one sense or another. The difference between man and chimpanzee is mainly due to these sequences, which could exchange genetic material within the same chromosome and consequently could vary rapidly (Hughes et al., 2010).

In addition to these DNA exchanges, the American researchers have suggested three other explanations for the rapid divergence of the Y chromosomes of man and chimpanzee : the key role of some genes that prevents their loss; the preservation of certain DNA portions that are not indispensable, but are close to key genes; and a different sexual behaviour. Among chimpanzees, there is indeed a strong competition for reproduction, as several males can mate with one female when the latter is fertile (Hughes et al., 2010).

From the evolutionary viewpoint, 200 million years ago, X and Y were an ordinary pair of chromosomes; sex was determined during gestation by the environmental conditions, in the same way it is now determined in tortoises : eggs give birth to males when outside temperature is below a threshold, or to females above that threshold. A mutation may have occurred on one chromosome, giving rise to the *SRY* gene. Thereafter the two chromosomes would have evolved separately. It is likely at that point when degenerescence started. Generally each normal chromosome of a pair can exchange genetic material with the other during cell division, and this exchange can “repair” mutations. But X and Y chromosomes cannot do that : when a mutation occurs on Y, the gene is lost. By contrast, if it occurs on X, the gene can be rescued through the exchange of genetic material between the two X chromosomes in women. The X chromosome could therefore maintain its structure during evolution, while the Y chromosome lost half of its genes (Chaigne, 2010).

The Y chromosome cannot exchange genetic material with the X chromosome, but it can exchange DNA with itself. Due to the existence of palindromes, it may happen that a big Y chromosome is formed, and the latter

can break up and give rise to two chromosomes that are not always functional. Depending on the status of *SRY* gene, the person can have a male or female gender, and even a hybrid one : if, during a cell division, the chromosome breaks up and a daughter cell inherits a functional *SRY* gene, and the other daughter cell does not, the person can have a male *and* a female gonad. Mutations on the X chromosome can also cause diseases, mainly among men, because the Y chromosome cannot compensate such anomalies (Chaigne, 2010).

The Y chromosome, however, presents advantages for studying human evolution and history. As there is no exchange of genetic material between Y and X, a boy possesses the exact copy of his father's Y chromosome. It can be therefore possible to follow the filiation between male individuals. It is such property that supported the hypothesis of the African origin of modern man, *Homo sapiens*. In the same wane, a study published on 19 January 2010 in *PloS Biology* concerned the origin of agriculture in Europe 10,000 years ago. Was it born from the migration of Near East farmers, or from a simple technology transfer to the hunters-gatherers who roamed Europe at that time? The study of the Y chromosome has provided a clearcut reply : its diversity can only derive from migrations of people from the Near East, who would have been more successful to mate with women than local males (Chaigne, 2010).

Sequencing the genome of Homo sapiens' ancestors

On 12 February 2009 (Charles Darwin's 200th birthday), Svante Pääbo, director of the department of evolutionary genetics at the Max Planck Institute of Evolutionary Anthropology in Leipzig, Germany, announced at a meeting of the American Association for the Advancement of Science (AAAS) that his team had a version of Neanderthal man's DNA to compare with that of modern man, *Homo sapiens*. The comparison was not published until 6 May 2010. It showed similarities between the species (in, for instance, the *FoxP2* gene that contributes to the ability to speak) as well as differences in several genes connected with cognitive ability. These differences are the focus of attention in order to perceive the distinction between *H. sapiens* and other animals, including other types of human, and thus accounts for the extraordinary flourishing of a species that is now estimated to make use of 40% of the net primary productivity (the energy captured by photosynthesis and converted into plant biomass) of the planet's land surface (*The Economist*, 2010 j).

Johannes Krause of the same institute as S. Pääbo has studied the DNA extracted from a tiny phalanx of the finger of an individual whose bones

were found in a cave of the Altai ridge, in southern Siberia. The age of the bones was estimated at less than 40,000 years and, surprisingly, the individual belonged neither to *Homo sapiens* or *H. neanderthalensis*. This meant that a time when two human species – our direct ancestor Cro-Magnon and Neanderthal man – co-existed, a cousin was also living in Eurasia. This discovery was again complicating the phylogeny of humans as well as the history of migrations from the cradle of humankind in East Africa. The discovery in Indonesia in 2003 of the 13,000-year-old Flores man also meant for some anthropologists a fourth human species (Morin, 2010 b).

The findings concerning the Altai man were held back for a long time because the researchers wanted to eliminate any contamination by alien DNA, including that of the researchers themselves. They were finally published in the 24 March 2010 issue of *Nature* (electronic). The researchers have now the tools that allow them to identify the old DNA, often degraded; the analyzed DNA is mitochondrial DNA. By contrast to nuclear DNA, mitochondrial DNA is of maternal origin, exclusively. It is interesting because it can keep trace of many mutations over time, and this enables the geneticists to establish genetic distances between specimens. When they compared the DNA of the Altai man with that of 54 modern humans and that of six Neanderthal humans, S. Pääbo's team found that differences were twice more numerous between the Altai man and ourselves than those which separate us from the Neanderthal man. That meant if *Homo sapiens* and *H. neanderthalensis* diverged about 466,000 years ago, the common ancestor of the three human species would have existed 1 million years ago. However, S. Pääbo's team was careful about using the word "species" for the Altai man; the analysis of nuclear DNA may clarify this matter (Krause et al., 2010).

The German researchers gave more details on the implications of their discovery on human migrations. Up to then, there were three well identified migrations out of Africa. First, that of *Homo erectus*, about 1.9 million years ago. Then, that of *H. neanderthalensis*, about 500,000 years ago. And finally, that of *H. sapiens*, about 50,000 years ago. The discovery of the Altai (Denisova cave) suggests that there might be another migration of "out of Africa" (Krause et al., 2010).

Palaeogenetics may in the future lead to further surprising discoveries, particularly in Asia where there are great gaps in our knowledge about human evolution. DNA analyses could in the coming years shed light on the kinship ties between humans, particularly in the case of fossils to be found in the northern regions where DNA is better preserved. In

a first stage, the work being carried out by the Max Planck Institute's researchers in Leipzig will bring some precisions thanks to the analysis of nuclear DNA; the German researchers have already suggested that the bone found in the Denisova cave belonged to a six-year-old child, probably a girl, and they named her "X woman." The assistance of palaeontologists and archaeologists will also help to clarify the position in the human evolutionary tree of this human "species," bearing in mind that the Denisova cave – which offered a refuge against the harsh climate of the surrounding steppe – has been occupied intermittently during the last 125,000 years (Morin, 2010 b).

The Genographic Project : comparing the genomes of people alive and understanding human relationships and migrations

Comparing the genomes of people alive today allows the identification of sites where natural selection has been active in the more recent past. Whole-genome sequencing (instead of using gene chips), associated with an analysis of how *Homo sapiens* has spread out of Africa 60,000 years ago around the world, will give a rather precise picture of human evolution. This is the purpose of the Genographic Project, led by Spencer Wells, a geneticist, and run jointly by the National Geographic Society, Washington D.C., the Waitt Family Foundation – an American philanthropic association – and IBM. The project is also supported by several renowned biologists, including Luigi Luca Cavalli-Sforza, and is associated with 20 laboratories of population genetics across the world (including the Institut Pasteur, France). In a sense, the project is a type of personal genomics. Volunteers from all over the world (more than 500,000 of them up to 2010) send samples of cheek mucosa cells for genetic analysis. In exchange, they receive a detailed breakdown of the wanderings of their ancestors over the past 150,000 years or so. In fact, since 2005 the Genographic Project has set for itself the goal to reconstitute the migrations of *Homo sapiens* since he went out of Africa, 60,000 years ago (Joignot, 2010; *The Economist*, 2010 j).

Such vast enterprise, at the crossroads of biology and historic research, is the carry over of the work initiated in 1993 by the American Human Genome Diversity Project, and thereafter by the French Centre of Human Polymorphism, founded by the Nobel laureate Jean Dausset. The American geneticist Spencer Wells stated that the Genographic Project not only intended to reconstitute "the genealogical tree of humankind," but also to perform an educational and humanist task. Thanks to this research, "everyone will understand his/her ties with humans across the world, know that we are all tied to each other by a genetic thread and that our

threads have been interwoven through our ancestors' migrations," stated Spencer Wells. We are in fact all cousins, sharing more than 99% of the same genetic heritage (Joignot, 2010).

DNA is collected in two ways. On the one hand, teams are sent to remote regions where peoples or human communities are isolated; old genetic markers can be found among them, e.g. among the Sans or Bushmen of southern Africa, Hadzabes in Tanzania, Ogieks in East Africa, Peuls or Foulbé in Mali, Shuars in Ecuador, Korankos in Sierra Leone, Yagnobis in Tadjikistan. This kind of sampling provides important data on migrations of the first human communities, but it raises many ethical and political issues. For instance, the French geneticist Pierre Darlu and several associations for the defence of threatened peoples denounce a kind of "biocolonialism" and they fear that the discovery of old migrations may justify the territorial deprivation of present populations (Joignot, 2010).

On the other hand, DNA is collected from individuals who respond voluntarily to calls under the Genographic Project in order to analyze one or two samples of their DNA. The individual analysis cost US$99.95 (or €80) in 2010, and the money collected helped to finance the Legacy Fund, whose goal is to defend the survival and culture of isolated peoples who are studied by the Genographic Project (Joignot, 2010).

Genes role in human evolution and history

For Africans, or those of recent African origin, the pattern is a tree leading out of north-east Africa, where *Homo sapiens* seemed to have originated, that spreads its branches through the whole continent. For those without recent African ancestry, the journey then goes through a bottleneck 60,000 years ago, which corresponds to the pioneers who crossed the straits of Bab el Mandeb and populated the rest of the planet. And until recently there was little interbreeding between the branches, which has allowed local differences to emerge (*The Economist*, 2010 j).

The spread of humankind was followed by studying differences in mitochondrial DNA, inherited only from the mother and in DNA from the part of the Y chromosome that passes only from father to son. Now, though, it is possible to look at changes occurring throughout the genome (thanks to whole-genome sequencing). Some of these changes will be random, but some will be the product of natural selection. Pardis Sabeti of Harvard University is working on how to distinguish the two kinds of changes. The blocks of DNA studied in the HapMap and similar projects are swapped between neighbouring chromosomes during the process of ovocyte and

sperm formation. P. Sabeti realized that a gene which is being favoured by selection will be in a DNA block that will be longer than the statistics of random mixing would predict. P. Sabeti identified 200 places in the human genome that have been subject to recent selective "sweeps;" for instance, these are genes that regulate skin pigment and hair morphology, both well-known markers of geographical origin, and they had undergone significant selection. So had genes regulating metabolism, probably in response to the shift from hunting and gathering to farming as humankind's main way of life (Demogines et al., 2010).

However, several genes connected with sensory perception – hearing and balance, in particular – have altered. In this case, the changes are most noticeable in some of the Asian branches of humankind. Genes involved in the development of the sound-detecting hair cells of the ears seemed especially affected. Whether that means Asians hear things differently from other people has yet to be established, but it might (Demogines et al., 2010).

The other kind of genes that have evolved rapidly are those involved in infectious diseases. P. Sabeti has found evidence that malaria, tuberculosis, measles and poliomyelitis had all left their selective imprint as fragments of DNA that help to confer resistance were favoured. One of the greatest surprises was that Lassa fever, once regarded as a relatively rare disease, was also on that list. It is becoming established that Lassa fever is actually very common in parts of West Africa. A fifth of Nigerians, for instance, showed signs of having been infected in the past. That these people have not been killed by an illness originally thought to be up to 80% lethal suggests the protective effect of the evolutionary change has been strong (Demogines et al., 2010; *The Economist*, 2010 j).

P. Sabeti has not found any selected genes that encode controversial traits such as behaviour and intelligence. That may be because they have not been subject to recent selection. It may, though, be because few links between these phenotypes and the genes themselves have yet been made. The Beijing Genomics Institute has planned a study to look specifically for intelligence genes. And according to Nick Martin of the Queensland Institute of Medical Research, in Australia, dozens of genome-wide association study (GWAS) investigations of behavioural and cognitive links were under way. It may mean that the old arguments about the role of nature and nurture (culture) in human development would be envisaged differently. That may also be true if Fred Gage, of the Salk Institute, in San Diego, were proved right that variation in cognitive ability may be genetically determined in a way that is not inherited. F. Gage

studies LINE-1 elements, bits of DNA known colloquially as jumping genes. These make up about 20% of the human genome, and most biologists regard them as parasites. Their origin is unknown, although they may be the distant descendants of some type of retrovirus because they are able to copy themselves using an enzyme, the reverse transcriptase, the synthesis of which is controlled by a retroviral gene. If an element happens to copy itself into the wrong place it can cause an existing gene to malfunction. F. Gage showed in 2006 that LINE-1 elements were much more active in brains of mice than in other tissues (Alisch et al., 2006); and in 2009 he and a colleague, Nicole Coufal, showed the same was true in humans. Using sensitive sequencing methods they discovered that human brain cells had around 100 more LINE-1 elements in them than tissues such as the heart or the liver. That means the elements are multiplying during the process of brain formation (Singer et al., 2010).

One possible explanation of this would be that each new brain develops by a process akin to natural selection. Nerve cells grow and develop a high number of connections with each other, and then most of those connections and many of the cells themselves die. Only the fittest cells and connections survive, leaving a working network. In this context a way of generating variations on which selection can operate may actually result in better networks and thus brains. The random changes brought a bout by LINE-1 elements jumping around the genomes of neurons could yield such variation (Singer et al., 2010). But there is at least another part of the human organism – the immune system – that relies on the internal mutability of the DNA to generate the variety needed for it to recognize all the pathogens (antigens) that nature can throw at a body. Although the immune system has come up with a different solution in detail (particular cassettes of DNA have evolved to shuffle round at this one place in the genome), the general principle is the same. No one doubts that intelligence has a heritable element; studies of identical and non-identical twins confirm that. No one doubts either that upbringing and education matter too. But part of the difference between people's cognitive abilities might also literally be a lottery, if F. Gage's hypothesis were correct, because it would depend on the random movement of bits of DNA inside an individual's developing brain (*The Economist*, 2010 j).

Human migrations and family tree

Frédéric Joignot, a journalist at the French daily newspaper *Le Monde*, has sent samples of his cheek-mucosa DNA to the Genographic Project. The analysis of both his mitochondrial and Y chromosome DNA provided him with the following information regarding his ancestors. He belonged

from his father's lineage to an extremely rare group of humans in Europe, the "haplogroup G," identified thanks to "mutation 201"; 1% to 3% of Europeans belong to this group. According to population genetics, these people (M 201) have appeared in the Caucasus region, 30,000 years ago. They have left few descendants. They migrated over millennia with their flocks of sheep and goats, and settled in the mountainous areas of present Iran, Afghanistan, Pakistan and Kashmir. Later on, 10,000 years ago, some of these M 201 people reached the Fertile Crescent, i.e. the vast region between the Mediterranean and the Persian Gulf, around the Tigris and Euphrates, where they settled. Historians have dated between 9,500 and 7,000 years the development of a first civilization, based on agriculture, craftsmanship and the creation of big cities. With population growth, part of these "M 201" left the Middle East and settled in the Mediterranean islands, Turkey, the Balkans, 5,000 to 7,000 years ago, and thereafter migrated to Georgia. Later on, a small group reached southern Europe, and then northern Europe, while the majority moved towards Russia and Turkey. Nowadays, these Caucasians who became Middle Easterners represent 1% to 3% of all Europeans, while they make up 30% of the populations living in the mountainous regions of Georgia, northern Caucasus and northern Turkey. These "M 201" are still found in Sardinia (14%), northern Italy (10%), Turkey (7%), Greece (2%-3%), in Lebanon, Syria, the Middle East and even in Ethiopia. A small number can also be found in Uzbekistan, Mongolia and China (among the Ouigours) [Balaresque et al., 2010; Joignot, 2010].

F. Joignot's maternal DNA has a marker that classifies him in "haplogroup H," very common in Europe; it is found in 40% to 60% of Europe's inhabitants. This genetic marker appeared 37,000 years ago, when populations of *Homo sapiens*, coming probably from the present Russia and Turkey, reached northern Europe, as they seized the opportunity of a brief warming of the climate. This was the period of the Superior Palaeolithic, when the Cro-Magnon man – an Homo sapiens of the haplogroup "HV", and then "H" – settled across the whole of northern Europe and disseminated new techniques of stone cutting and carving. This robust man pushed the Neanderthal man down to Spain, where he disappeared 30,000 years ago for still unknown reasons. Meanwhile, the climate became colder, the north of France was occupied by the toundra, the south by the taiga, and aurochs, woolly rhinoceros, mammoths and cave lions proliferated, while human populations and their diversity decreased. However, the "haplogroup H" resisted. These humans improved their tools and weapons, and they migrated southward to Spain, Italy and the Balkans. Towards 15,000, as the climate warmed up again they moved northward, following the Atlantic coast, up to Brettany (Joignot, 2010).

F. Joignot did not receive the actual family tree of his ancestors, but only the two direct lines of his parents. And regarding the migration routes followed over 30,000 years, the Genographic Project only indicates the main itineraries followed by population groups and not by the individuals themselves. Finally, migrations described in the maps available now could evolve with new knowledge. For instance, thanks to the research carried out by Lluis Quintana-Murci at the Institut Pasteur, we know since the early 2000s the route taken by *Homo sapiens* to leave Africa, and thereafter the migration along the coastal zones of the Middle East towards India. Many other routes remain poorly defined, for instance that towards the Americas and South-East Asia. If genetics can be helpful, it is not sufficient for the identification of the complex migration routes of a human group, which depend on geological, climatic and historical events. Lluis Quintana-Murci stated that "to establish an itinerary, we must cross the results of genetics with those from linguistic studies, research on agriculture evolution, the history of domestication and craftsmanship, and climate, etc." (Joignot, 2010).

The data issued by the Genographic Project have the merit to provide the layman with the main features of the extraordinary journey made by part of his ancestors over millennia until now. They testify to the continuity of humankind and to the fact that we are all a single species with very small genetic variations. As Spencer Wells highlighted, "we are all cousins; only two thousand generations separate us from men who left Africa." That is why if a European needs an organ transplant, he may find a compatible one either near home or in Arabia (Joignot, 2010).

Controversial issues

It is true that any mention of genetic differences, even though very small, or of very far geographic origins, could generate controversy. That is why the Genographic Project should, according to F. Joignot, include an educational chapter on these issues and, for instance, question the issue of "races." In this regard, Luigi Luca Cavalli-Sforza, one of the pioneers of population genetics and the founder of genetic geography, professor emeritus at Stanford University, California, has struggled against the concept of race during all his life. He is associated with the Genographic Project. He highlighted that Charles Darwin demonstrated that it was useless to define races among humans. In fact, anthropologists do not agree on even the definition of race. At the time of C. Darwin, some thought there were two races and others considered there were over 60. Even L. Cavalli-Sforza had proposed classifications of human groups: eight, then 38 and even hundreds depending on the issue envisaged.

He ended up with the conclusion that the concept of race is useless. André Langaney, a French anthropologist who has edited one of the books on these issues, *Tous parents, tous différents* (All Relatives, All different, University of Geneva, 1997), stressed that "there is no genetic marker for race; it has never been possible to isolate one that would be present in all black people and would be absent in all white ones. As soon as one tries to define a race, seeking for classification criteria, it is an endless process, and if the process goes further it means defining a race per individual, as we are all different… The genes have no race" (Joignot, 2010).

In a sense, the promise of self-knowledge is great, but the threat is as great when this knowledge is used to speak of races and, even more, of superior races, which recalls the Delphi oracle's second admonition : "nothing is excess." People must resist the excess of racialism, nationalism and eugenics that some may be bound to propose on the basis of false interpretations of our genetic heritage. People must be respected as individuals, regardless of their genetic make-up (*The Economist*, 2010 i).

Consumer-genomics firms

Consumer-genomics firms have sprung up in the past few years. Thus 23andMe, based in Mountain View, in the heart of the Silicon Valley, that counts Google as one of its investors, was created in 2007 by Anne Wojcicki, a biologist and wife of Sergey Brin, cofounder of Google, who decided to invest US$4.4 million in the company. It seems that this investment is not alien to the launching by the Google Health Initiative, a website for the management of personal medical files, that Google wants to make available to all Americans. The company was named 23andMe, with reference to the 23 chromosomes of the human genome, and its analytical work deals with the DNA extracted from an individual's saliva. The saliva samples are sent by mail and the firm's technicians decode a tiny part of the DNA contained in the saliva, precisely 580,000 "markers" or variations that enable the identification of a series of characteristics which are specific to a individual (Eudes, 2008).

After a few weeks, results are posted on the personal, secure website of the provider. The firm's customers have formed online chat groups and blogs to share details of specific genetic mutations and exchange family and genomic histories (*The Economist*, 2009 a).

23andMe firstly informs the customer on his/her predisposition – expressed as a percentage – to develop about 30 diseases, e.g. breast or prostate

cancer, heart attack, hyperactivity, obesity, diabetes, multiple sclerosis, maniacs-depression tendency. The information provided also estimates the risk of becoming alcohol dependent if the customer does consume alcoholic beverages systematically, as well as the resilience to HIV/AIDS if he/she is exposed to the virus. To provide this kind of information 23 andMe's technicians look at single-nucleotide polymorphisms (SNPs), because variations of SNPs are correlated with the likelihood of developing a range of diseases. The firm's scientists insist they rely on the best peer-reviewed scientific studies on the matter, and recent studies, known as genome-wide association studies (GWAS), have established some correlations between a range of common SNP variants and diseases such as diabetes, heart diseases and cancers (*The Economist*, 2009 a).

23andMe's scientists attribute a mark from 1 to 4 to each result, depending on the degree of reliability of current research on the relevant genome-wide association with a particular disease. For instance, the risk of developing arthritis is given a 4 mark, because research in this area is well advanced and there is a consensus among geneticists in this respect. By contrast, the evaluation of the psychological skill of not repeating the same errors has received only a 1 mark, because research carried out in Germany is just starting (Eudes, 2008).

In September 2008, Sergey Brin, announced on his blog that "he ran the risk of developing a Parkinson's disease," after having tests at 23andMe. It was discovered that he had mutation G2019S on gene *LRRK2*. This mutation is not found in all patients suffering from Parkinson's disease, but it may increase the likelihood of developing this mental disease (Blanchard, 2009).

Regarding information about ancestry, 23andMe indicates to its client from which region of the world come his/her ancestors, while the company's Family Heritage section can determine the degree of similarity (± 0.1%) between parents and their children, and between brothers, sisters, cousins or friends. Every time a new discovery is made somewhere in the world, the team of 23andMe integrates it in order to broaden the range of its analytical work and to improve the precision of its results (Eudes, 2008).

Although rich and famous people like Rupert Murdoch, Harvey Weinstein and Ivanka Trump became customers of 23andMe and in some cases investors, two and a half years after beginning its service, 23andMe had only 35,000 customers, and at least one-quarter of them received the service free or for only US$25, instead of the hundreds of dollars that would make the business profitable (Pollack, 2010 a).

This low turnout suggests that many people have not yet adopted the genomics approach, despite the efforts of the gene-testing companies to present the technology in an understandable way. Also the economic downturn prevented customers from paying high fees (between US$300 and US$2,000) for the services. But the fundamental issue is that the current technology of DNA scanning cannot yet lead to meaningful predictions concerning the occurrence of a disease (Pollack, 2010 a).

All these factors led 23andMe to go through two rounds of layoffs in 2009, which reduced the number of employees to 40, down from a peak of about 70. The company's other founder, Linda Avey, left in September 2009, to create an Alzheimer's disease research foundation, though she remained a director. Navigenics, in Foster City, California, had among its investors, Kleiner Perkins, a venture-capital firm that was an early backer of Google, Amazon and AOL. It had fewer customers than 23andMe; in March 2010, it was on its third chief executive officer in a year and had also to trim its work force. It is now marketing to doctors and corporate wellness programmes rather than consumers (Pollack, 2010 a).

A. Wojcicki made some comments on the situation, by saying that "the business is moving ahead in a positive trajectory"…"we've hit some bumps in the road but we are learning and continuing to evolve. We did not start this company thinking it was going to be easy to create an entirely new market." Esther Dyson, a technology investor who is a member of 23andMe's board of directors, stated that the employee cutbacks were part of a necessary adjustment. "We've become a business from being a scientific adventure company," E. Dyson said (Pollack, 2010).

For now, 23andMe seemed to have a rather solid position. The company raised US$27.8 million in 2009, according to a filing with the US Securities and Exchange Commission in December 2009. A portion came from an earlier US$10 million loan from Sergey Brin of Google, which was converted into 23andMe stock. Google itself invested US$2.6 million in the round, adding to the US$4.4 million it invested in 2007. New Enterprise Associates, a venture-capital firm, was also a big investor (Pollack, 2010).

DeCODE – an Icelandic firm whose aspirations to become a full-fledged pharmaceutical company were dealt a blow when it went through a bankruptcy restructuring by early 2010 – charged US$2,000 to search a sample for 1 million SNPs predictive of 50 genetic traits, not all of them diseases. Theragen makes a similar offer from South Korea. Complete Genomics, also in Mountain View, California, planned to leapfrog the chip-

based technology by offering customers full DNA sequences. And Knome, a firm based in Cambridge, Massachusetts, offers a customized whole-genome service at US$68,500 per single operation (*The Economist*, 2010 j). YiGene, a Chinese start-up, is competing with several local rivals to provide genetic testing and counselling to Asian consumers (*The Economist*, 2009 a).

The rush of a number of firms to offer genetic tests to consumers, often raising expectations that may be questionable, has an indirect important impact on the growth of companies manufacturing and selling gene-sequencing equipment as well as the related products and services that complement the hardware. For instance, in June 2008, Invitrogen, a biotechnology firm based near San Diego, California, acquired Applied Biosciences (AB), a bigger California-based firm, in a deal at over US$6 billion. The combination will be a force in the market, offering customers affordable hardware, then would profit from Invitrogen's high-margin laboratory products. AB's president, Mark Stevenson, stated the aim was to boost sales of reagents and other consumables, which made up 75% of the combined firm's revenues (*The Economist*, 2008 c).

The deal between Invitrogen and Applied Sciences was also an important step in the consolidation of a fragmented industry. Roche, a Swiss pharmaceutical giant, controls 454LifeSciences, a gene sequencing firm, and it made a hostile takeover of Ventana Medical Systems (see p. 330). The deal also suggests, according to Drew Fromkin, chief executive of Clinical Data, a firm that uses the gene-sequencing products of AB and its rivals, that personal genomics may make its first profits in the area of medical diagnostics (*The Economist*, 2008 c).

Experts are of the opinion that as the cost of sequencing and scanning DNA continues to plummet, and as the link between genes and diseases is better understood, analyzing people's genetic makeups will become an essential part of health care. But that might take a few years and require the sequencing of a person's entire genome, not just sampling selected parts of it, as the gene-testing companies do now. That might increase the cost of the tests. One could expect a harsher competition between companies in terms of final costs, and particularly between the pioneers such as 23andMe, Navigenics and DeCODE, and newcomers. For instance, a new company, Pathway Genomics, based in San Diego, began offering service during the summer of 2009 at a lower price than others (Pollack, 2010 a).

Personal genomics offers two services broadly speaking. One is to trace the person's ancestry back through humankind's family tree to its roots in

north-east Africa – an offer like that of the Genographic Project. Indeed, a similar service is also available to pet owners, courtesy of Mars (which makes pet food as well as confectionery). The other service is predictive medicine – a list of genetic variations that might put a person at higher-than-average (or, indeed lower-than average) risk of developing particular diseases. Predictive medicine is a controversial area. Being told that one has an increased chance of illness over the course of life can be upsetting, particularly if no treatment or pre-emptive action is possible. Worse that worry may be misplaced; an increased chance is not a certainty. Conversely, it is now clear that genome-wide association studies on which many of the correlations are based have uncovered only a small part of the risk, so not showing up as being in danger does not put someone in the clear. But this sort of study can sometimes reveal the precise nature of a set of symptoms, which might affect which medicines are used to treat them. Such precision is one aspect of pharmacogenomics, which seeks to match drugs to a patient's genome. A second aspect is that genetic knowledge can sometimes predict adverse reactions to drugs that have proved safe for some people to take but dangerous to others (*The Economist*, 2010 j).

Controversy on the real benefits of genetic tests

A number of academics remain deeply sceptical about the real benefits and impact of direct-to-consumer genomics, which, they consider, has been oversold. They consider that the promised benefits of genome-inspired drugs and personalized health care have failed to materialize.

Allan Balmain of the University of California, San Francisco, questioned the scientific basis for the claims made by consumer-genomics firms, while David Altshuler of the Broad Institute, a genetics-research centre run jointly by Harvard University and MIT, thought the firms encouraged people to read too much into their results. Such "fallacies," he stated were causing a public backlash that could divert attention and resources from the worthier goal of genomics-inspired disease research (*The Economist*, 2009 a).

"With only a few exceptions, what the genomics companies are doing right now is recreational genomics" was the severe judgement of David B. Goldstein, a Duke University geneticist who was among the contributors to the 23 April 2009 issue of *The New England Journal of Medicine* that was the first public attempt by scientists to make sense of this puzzling result. "The information has little or in many cases no clinical relevance," stated D.B. Goldstein (Goldstein, 2009).

One issue of debate among researchers is whether, despite the prospect of diminishing returns, to continue with the genome-wide association studies, which cost many millions of dollars a piece, or switch to a new approach like decoding the entire genomics of individual patients.

Unlike the rare monogenic diseases, illnesses like diabetes and cancers are caused by a set of several genetic variations in each person. Since these diseases generally occur later in life, after people have had children, natural selection would not eliminate them. In the issue of *The New England Journal of Medicine*, it was underlined that many genome-wide association studies, often involving thousands of patients in several countries, had been completed for a number of diseases, and some common variants had been found. But in almost all cases they seem to carry only a modest risk for the disease. Most of the genetic association with the disease remained unravelled (Goldstein, 2009; Wade, 2009 b).

For instance, two teams of European scientists published in the journal *Nature Genetics* (7 September 2009 issue) the results of their studies on new genetic variants associated with Alzheimer's disease. The three new genetic variants have been detected by screening much larger numbers of patients and by using genome-wide association techniques (patients' DNA is scanned with devices programmed to recognize half a million sites of variation along the genome). One of the teams, led by Julie Williams of Cardiff University in Wales, scanned the genomes of 19,000 patients, the largest group so far of persons suffering from Alzheimer's disease, and found two variants that have a statistically significant association with the disease. A second team, led by Philippe Amouyel of the University of Lille, France, also found two variants, one of which was the same as that detected by the Welsh team (Harold et al., 2009; Wade, 2009 c).

The fact that two studies could result in the detection of the same gene out of three is a progress. More than 550 genes have been proposed in various small-scale studies as the cause of Alzheimer's disease, but all have failed the test of replication by others, according to P. Amouyel. The gene that has the largest effect in the disease is a variant called *ApoE4*, discovered in 1993 in the laboratory of Allen Roses of Duke University. A. Roses stated the three new genetic variants had minor effects compared with that of *ApoE4*, and that their role in the disease was unclear, despite the statistical data pointing to their involvement (Wade, 2009 c).

However, J. Williams considered that detection of the new genetic variants, which undercut the brain efforts to restrain inflammation, underlined the primary role of inflammation in Alzheimer's disease. One

of the new genetic variants is in a gene active at synapses, and the two others help reduce inflammation in the brain. Inflammation is a known feature of Alzheimer's disease, but it is often regarded as a consequence of the disease (Harold et al., 2009).

D.B. Goldstein argued that the genetic burden of common diseases must be mostly carried by large numbers of rare variants. For instance, he considered that schizophrenia would be caused by combinations of 1,000 rare genetic variants, not of 10 common genetic variants (Goldstein, 2009).

Two other geneticists, Peter Kraft and David J. Hunter of Harvard University School of Public Health, also writing in the 23 April 2009 issue of *The New England Journal of Medicine*, largely agreed with D.B. Goldstein's conclusion that probably many genetic variants, rather than few, "are responsible for the majority of the inherited risk of each common disease." But they disagreed with his belief that there will be diminishing returns from more genome-wide association studies. D.J. Hunter was in favour of pursuing these studies in order to find more common variants, whereas D.B. Goldstein still considered it was "beyond the grasp of the genome-wide association studies" to find rare variants with small effects, even by recruiting huge numbers of patients. He insisted resources should be switched away from these highly expensive studies and be marshalled to decode entire genomes (and "not in fine-tuning our analyses of common variations"). He therefore suggested that the best approach was to decode the full DNA of carefully selected variants (Kraft and Hunter, 2009).

In France, Marion Mathieu, member of the association *Tous chercheurs* ("All researchers"), had organized on 13 January 2009, at the request of the French National Health and Medical Research Institute (INSERM), a training seminar on genetic tests for patients' associations. These associations are often requested by families to give their views on the need or the advantage to rely on this form of personalized genetics or medicine. In fact they are not always in a position to give a clearcut advice, and some associations' leaders have summarized their uneasiness about the matter in the following way : "At a certain period, when it was difficult to establish a medical diagnosis, one used to say 'it was psychosomatic.' Nowadays, one says it is genetic." Marion Mathieu stated that "most multifactorial diseases have a genetic cause that is estimated at less than 30%." Having a gene for susceptibility to a disease can increase the risk of developing that disease, but it is not a fatality. Tests for predisposition or susceptibility to a disease cannot give an absolute prediction on the occurrence of a disease, but they just help evaluate the risk. For instance, an individual who bears the *HLA B27* gene

runs the risk to develop ankylosing spondylarthritis 80 times more than a person who does not bear that gene. But this is a relative risk, and in fact one should know the standard risk (i.e. in the general population) concerning this disease : it is 1 per 2,000, or 0.05%. The absolute risk, i.e. the risk for the bearer of the *HLA B27* gene to develop the disease, is 80 x 0.05% = 4%. And this figure puts things in better perspective (Blanchard, 2009).

The biophysicist Henri Atlan expressed his concern about the opinion of those who think that our future diseases or those of our children are enshrined in our genes. Such conviction, he believes, "plays sometimes the role of a real genetic superstition." Predictive medicine remains for the time being an abstraction, except in the case of rare monogenic diseases or those associated with chromosome aberrations (Blanchard, 2009).

A recent discovery would lead to a more careful interpretation of the results of blood-sample genetic analysis aimed at identifying genes that may predispose to several non-cancerous diseases, such as diabetes, coronary disease and Alzheimer's disease. It should also encourage the search for mutations in the diseased tissues, that seem to be a more relevant target than blood cells for designing therapies.

Canadian researchers of McGill University and the General Jewish Hospital of Montreal, who were working on abdominal aorta aneurysms (AAA) – abnormal widening of the wall of abdominal aorta – sequenced the gene *BAK1* of leukocytes from 31 patients. This gene was suspected to be involved in the development of that disease which affects 6% to 9% of men above 65 years. They discovered that the gene sequence was different by three precise mutations from that of the gene present in vascular cells. The difference was not only observed in the cells of patients, but also in healthy vascular cells. The results of this research work were published in the July 2009 issue of *Human Mutation*, and they contradict the dogma that all normal cells of an individual possess the same genome, while cancer cells have DNA mutations. The Canadian researchers, who were not expecting to find genetic differences between blood cells and those of the vascular tissue of the wall of abdominal aorta, came to the conclusion that "what we see in the blood is not representative of what is found in the tissues, and we should not rely only on the genetic information obtained from blood cells," as stated by Morris Schweitzer, the main author of the publication. Even though these differences are tiny with respect to the billions of nucleotide pairs that constitute our genome, they should not be underestimated when one interprets the results of genetic testing relating to disease prognosis (Gottlieb et al., 2009; Vincent, 2009 c).

To sum up, the gene-variant SNPs reported in the genome-wide association studies are undoubtedly associated with diseases, but some believe their significance is greatly overstated. Even if consumer-genetics firms uncover a handful of SNPs that suggest an increased risk of developing a disease, the customer may have a dozen other genes that lower the risk. Some consider that the common variants easily uncovered by current sequencing technologies are much less important than other, rarer variants. The issue is that the current tool of choice, the genome-wide association studies, which link variants in a given population with known diseases, cannot easily find these rare variants. But as the cost of sequencing falls down, firms hope to be able to do a more precise work in spotting these variants (*The Economist*, 2009 a).

Another camp of sceptics rejects the whole idea that medicine will ever be truly personalized. Most disorders, such as diabetes and heart disease, are linked to dozens or possibly hundreds of genes. And those genes affect only partly an individual's susceptibility to a disease. Having a balanced diet, exercising every day, avoiding smoking, and so on could have a bigger influence on preventing a multifactorial disease than a genetically-based susceptibility. That is why David Altshuler of the Broad Institute considers it unlikely that scientists will ever be able to predict diseases perfectly, never mind devise truly personal therapies (Blanchard, 2009; *The Economist*, 2009 a).

Yet, consumer-genomics firms such as 23andMe are convinced that their work and results contribute to use human genetics as a tool of preventive medicine. Anne Wojcicki of 23andMe stated that "if people learn that they may develop a heart disease, they may change their lifestyle and make periodic medical check-ups. This behaviour can reduce health costs." She added that "genetics will lead to a revolution in pedagogical methods, which will have to be adapted to the individual skills of the child. The era of standardized and uniform learning for all is about to end" (Eudes, 2008).

A. Wojcicki and L. Avey were invited to the Davos World Economic Forum, at the beginning of 2009, and they had a unique opportunity to share their views with the world economic gotha. They also collected about 1,000 samples of saliva (free of charge) from the participants, who not only knew the existence of 23andMe, but also that their genetic code was stored in a Californian database. The company has extended its service to Europe, adding an extra US$45 for the mail expenses (June 2009). A. Wojcicki realizes that the services her company is offering will encounter cultural resistance in many countries, but she is convinced

that "beyond national traditions, a new universal principle is emerging, i.e. the right for an individual to know his/her genotype and how it is made"... "Our task is to help most traditional countries to advance in this direction." It is indeed a great ambition. And even though the firm gives full assurance to its customers that their genetic information is stored in a perfectly secured database, it encourages customers to publish, share and compare their genetic code. In all fairness, customers sign an agreement format, accompanied by the following warning : "Data that you share with friends, relatives or employees might be used against your interests... Nowadays, very few enterprises and insurance companies request genetic informations from you, but this may change in the future... Sometime in the future, data that you had disseminated might be used to refuse a job or a health insurance" (Eudes, 2008).

Linking genes to diseases and disease processes

Genome-wide association studies

Hundreds of genome-wide association studies, or GWAS, have been carried out since 2005 or so. The idea behind these studies sounds sensible : collect samples from people with and without particular diseases and look for associations between those diseases and genetic mutations, in the form of single nucleotide polymorphisms (SNPs). The thinking behind GWAS was that it would expose multigenic diseases. These are conditions which seem to exist in families, but do not obey the Mendelian pathways of inheritance. While haemophilia or sickle-cell anaemia are tied to the mutation of individual genes, a blood-clotting factor and one of the genes coding for haemoglobin respectively, that is not as clear-cut in the case of the tendency in some families to suffer from heart disease, strokes, late-onset diabetes, Alzheimer's disease. Environmental factors are obviously involved, but mutations are, too – just not single, large-effect mutations like those which cause haemophilia and sickle-cell anaemia. Instead, the pattern of inheritance suggests that many mutations of small individual effect come together to produce a risk than a certainty (*The Economist*, 2010 j).

GWAS, without being a success, has revealed a large number of mutations of small effect. On average, though, these add up to only 10% of the total heritability of any given disease. Mendelian effects add about another 1%. The rest, in a phrase that geneticists have borrowed from physicists, is referred to as "dark matter." These mutations appear to be very important, yet neither Mendelian nor GWAS techniques can detect them. Mendelian mutations are noticed because they are rare and

powerful. GWAS mutations are seen because they are common. The "dark matter" lies in the middle : too rare for GWAS but not powerful enough to have a clear Mendelian signal. One response will be to make whole-genome sequencing more widely, now that it has become cheaper (*The Economist*, 2010 j).

Thus, J. Craig Venter sequenced his own full genome faster and more cheaply. He expressed his scepticism about analyzing only the SNPs and he wanted to see whole genomes sequenced because "we do not yet know which parts of the genome are medically relevant" (*The Economist*, 2009 a).

It is true, however, that over the last 20 years research on tumour transformation has advanced our knowledge of the predisposition to cancer. Nowadays, the most simple predisposition situations have been identified : they are monogenic alterations which are associated with high tumour risk and specific phenotypes. About 40 genes have been identified, which are being investigated through genetic testing. One major success has been the identification of genes for predisposition to breast and ovary cancers, *BRCA1* and *BRCA2*. In 1990, *BRCA1* has been located on chromosome site 17q21, further to the study of some 30 carefully selected families. Thereafter, a consortium of laboratories was able to identify the gene. On the other hand, alterations of genes *MLH1*, *MSH2* and *MSH6* have been associated with a high risk of colon and uterus inner lining cancers (Stoppa-Lyonnet and Lenoir, 2005).

The relevant genetic tests are more sophisticated, because the number of mutations of genes *BRCA1, BRCA2* and eventually *BRCA3* to be analyzed is very high, e.g. more than 500 for *BRCA1*. Gene chips and DNA arrays are being used. For instance, a biochip commercialized by Affymetrix for *BRCA1* cost about US$100 by mid-2009.

An interesting area of research and development is to analyze whether the mutations of some genes associated with predisposition to breast, colon and lung cancers are specific to local populations and groups, and, if it were the case, develop the relevant genetic tests.

In the case of heart disease, more than 200 mutations located on 20 different genes have been identified and are more or less closely associated with heart diseases. India, where 20% of the global population is living, is home of 60% of heart diseases that occur worldwide. The reasons for this high incidence are not well known, but there may be a clue : in the Indian population, there is a widespread genetic mutation

which heightens the risk of cardiomyopathies. This mutation would affect 4% of the whole population of the Indian subcontinent (India, Pakistan, Bangladesh and Sri Lanka), i.e. some 60 million people (Vincent, 2009 a).

Under the leadership of Kumarasamy Thangaraj of the Hyderabad Centre of Cell and Molecular Biology, Indian researchers have shown that this mutation affects the gene coding for protein MYBPC3, involved in the functioning of the heart muscle. It has been identified for the first time in 2003 in two Indian families suffering from cardiopathies. Researchers have followed the implications of the mutation in 28 Indian families, and noticed that some carriers of the deletion became ill as soon as they were around 30 years old, and that over 90% of older carriers of the mutation were affected by heart disease. According to K. Thangaraj, who published their results in the 18 January 2009 issue of *Nature Genetics*, younger patients can degrade the abnormal protein and remain in good health, but as they become older the abnormal protein accumulates and its detrimental effects appear. It has been estimated that the presence of that protein multiplied by seven the risk of heart disease (Dhandapany et al., 2009).

The disease and the correlated gene mutation are more frequent in the south of India. It has not been found in other countries than South Asia : tests carried out among 2,000 persons from 26 countries were negative, except in Malaysia (which has an important Indian community) and in Indonesia. This mutation has most likely appeared in India, some 33,000 years ago, according to Chris Tyler-Smith of the Sanger Institute (Hinxton, United Kingdom), who is one of 25 authors of the paper published in *Nature Genetics*. Thereafter, it spread among the populations, without being eliminated by natural selection, because the disease often occurs when the patients have already had children (Dhandapany et al., 2009; Vincent, 2009 a).

Whole-genome sequencing : International Cancer Genome Consortium

Correlation between mutations or variants and cancers or heart diseases is worth pursuing. There is also a future for the diagnostic use of whole-genome sequencing. But the cost of the latter has to fall from US$50,000 nowadays to US$100 by 2015 or 2020, making all kinds of things possible, but only if the value of the genetic information is far greater than it is nowadays.

In November 2008, the American magazine *GQ* published an article entitled *The Book Of Me*, whose author is one of the most renowned American writers : Richard Powers, who received the prestigious National

Book Award. In this long article, R. Powers indicated that he was the ninth man in the world to have his whole genome sequenced. Before him, J. Craig Venter and James Watson had also their entire genome sequenced. He was informed that he had no less than 248 genetic variants that predispose him to some 77 pathologies of all kinds. But he stated that, more extraordinarily, "8% of his genome contains variants that relate him to Yorubas who live in Ibadan, Nigeria." "I have become another person, somebody else than I thought to be" (Nouchi, 2008).

Cancer is at the vanguard of genomic medicine for two reasons. One is that oncologists and their patients (and also the regulators of medical practice – deontologists) are often willing to take risks that would be unacceptable if the alternative were not a horrible death. The other reason is that cancer is now considered unequivocally to be a genetic disease; its environmental correlates (smoking, for instance) act not by poisoning cells directly but by promoting mutations in those cells' DNA. Such somatic mutations (occurring in body cells and not in the gametes) can cause havoc in an individual's organs, but are not passed to his/her offspring. In the case of cancer, an accumulation of somatic mutations causes a breakdown of the regulatory mechanisms of cell division; growth and further division continue therefore unabated until the body can no longer support both healthy tissue and tumour (*The Economist*, 2010 j).

Genomics soon indicated to oncologists that cancers which look similar under the microscope (examination of tissue samples) can have completely different genetic causes and thus require different treatments. That general observation should be confirmed in detail by a project run by the International Cancer Genome Consortium (ICGC), a collaborative initiative of researchers initiated in 2008 in 11 countries. This five-year project takes advantage of the falling cost of sequencing to collect full DNA sequences from 500 people suffering from each of 50 types of cancer. Not only will the cancerous tissue be sampled, so will healthy tissue from each patient. Comparing both kinds of tissue will reveal the somatic mutations which an individual has undergone. Comparing cancerous tissue from different individuals will show which mutations are important. This is necessary because in cancer patients the genes which control the proofreading of new DNA strands often become damaged, so that mutations accumulate much faster. This means that crucial mutations are more likely to occur, but also that in any kind of cancer there is a lot of mutational "noise." This has made it more difficult to discover which mutations are important and which are merely incidental. The results of the ICGC project should be a near-complete understanding of cancer at the genetic level. That will help diagnosis and treatment (allowing physicians to choose appropriate

drugs the first time round, rather than operating through trial and error) and, with luck, should promote the development of new treatments (*The Economist*, 2010 g).

In order to avoid duplication of efforts, the ICGC members have distributed the task among them, taking account of national context and priorities. For instance, Indian researchers will focus on the cancers of mouth, caused by the massive chewing of betel nut, while Japan is studying liver cancers associated with hepatitis C, which is highly prevalent in the country. France chose to focus on liver cancers caused by alcohol consumption and breast cancers of the *HER2+* type, which represent 15% of breast tumours. "The French choice has been made because of the existence of tumour banks, very well organized and set up with the patients' agreement; also healthy tissues had been collected," according to Fabien Calvo, director of research at the National Cancer Institute (INCa) and representative of France to ICGC. In addition, France has research teams which are very advanced in the chosen cancer areas. Three other types of cancers will be selected by France. Until mid-2010 DNA from seven liver tumours had been sequenced by French researchers, and its analysis was proceeding well. Among the seven tumours, the one that had been analyzed in depth contained about 100 genetic alterations, and it will be a very complex task to determine the role of each one in tumour development, stated Jessica Zucman, who leads a team of the National Health and Medical Research Institute (INSERM- U674), specialized in the study of the hepatocellular cancer. Fabien Calvo, who is also the head of the Cancer Institute of the Alliance for Life Sciences and Health, underlined that analysis of DNA sequences of the tumours might also lead to interesting results, e.g. identification of oncogenes already detected in other cancers and for which they were drugs on the market (Morin, 2010 c).

F. Calvo reckoned that the sequencing programme was an important endeavour : €5 million for each type of five cancers, analyzed over five years, compared with the €60-million annual research budget of the INCa. But sequencing costs were falling steadily and this decrease in the cost of the overall project might encourage several associations of the civic society to financially support ICGC (Morin, 2010 c).

Identifying the genes that may cause cancer is the first step towards therapy. But all gene products are not necessarily the targets of drugs. What are therefore the roles of research institutions and biotechnology firms and pharmaceutical groups? Todd Golub of the Broad Institute, Cambridge, Massachusetts, reckoned that drug firms had not always pursued leads. For instance, many oncogenes, as those whose breakage

causes cancer are known, encode kinases. These are enzymes which are involved in intracellular signalling pathways. Small molecules proved effective in blocking their activation. Researchers with putative antikinase drugs are thus welcomed by venture-capital firms. That is in marked contrast to drugs that might control transcription factors (a failed transcriptor-factor gene is a common cause of cancer as a failed kinase gene). However, transcription factors are not regarded as "targets of drugs." No systematic way of dealing with them has yet been discovered. That is not the venture capitalists' fault. Is it the drug companies' fault? They might argue that they are not in the business of basic research. On the other hand, a breakthrough in this area would create a whole new line of business. However, if that breakthrough were a conceptual one that could not be protected by a patent rather than an individual molecule that could be patented, then other firms would be able to freeride on the discoverer's expensive research (*The Economist*, 2010 j).

T. Golub suggested that independent laboratories like the Broad Institute could act as honest brokers for general research paid for by all the big drug companies. Having paid equally, all would benefit equally. This being basic research, openly published, such collaboration would probably be permitted by antitrust laws. Something similar was tried at the beginning of SNP studies (through admittedly those have not yet led to much in the way of medicine). It seems that drug firms are not interested, but that may change as their pipelines of new drugs empty (*The Economist*, 2010 j).

Despite such obstacles, human genomics has led to some successes in cancer treatment. For instance, a molecule named PLX4032 could clear almost completely tumours from secondary melanoma, one of the most aggressive cancers known. It was designed specifically to interact with the protein produced by a particular mutated version of a gene called *B-RAF*. This mutation, called V600E, has been found to be involved in 60% of cases of malignant melanoma and, less commonly, in other cancers. PLX4032 inhibits the activity of the mutated protein and causes cells containing it to die. The protein encoded by *B-RAF* is a kinase (and therefore familiar to venture capitalists). The initial development was done by a small biotechnology firm called Plexxikon, co-founded by Joseph Schlessinger of Yale University, one of the early researchers on *B-RAF*. The molecule has been picked up by Roche, a big drug company, which is paying for phase-3 trials, the last stage before a drug is submitted to the authorities for approval. If all goes well, the drug will soon be available for those suffering from melanoma, and will also be undergoing trials in other kinds of tumour in which V600E is implicated (*The Economist*, 2010 j).

There is nevertheless a shortcoming : the protective effects of PLX4032 last only for six months or so. Presumably, further mutations bypass the V600E – precisely the sort of question that the ICGC project is supposed to address. Once those mutations are identified, the hope is that drugs against them can be developed, too. If that proved possible, all of the pathways leading to cancer could be blocked, and a cure would be achieved (*The Economist*, 2010 j).

Richard Lifton, of Yale University, is studying a number of genes that are also the subject of investigation by drug companies. One of these genes *PCSK9* encodes an enzyme involved in cholesterol metabolism, and therefore could be a target for the prevention of heart disease. Also people with mutated versions of *ROMK*, the gene for a type of potassium-ion channel, have abnormally low blood pressure, so the research is on for a drug that tweaks the unmutated version of the channel, to lower the pressure of people with hypertension. Those with mutated versions of *SCN9A*, which encodes a particular sodium-ion channel, are insensitive to pain. Tinkering with this might produce a superior analgesic. And *BACE*, the gene for beta-secretase involved in Alzheimer's disease, if its action is inhibited, the progress of the illness may be delayed (Yue et al., 2009; *The Economist*, 2010 j).

Personal Genome Project

Thinking on a larger scale, George Church of Harvard University Medical School has proposed through his Personal Genome Project (PGP) to collect samples and medical data from 100,000 people and use the newly emerging mass-sequencing techniques to record the whole genomes of each of them. In a way, the PGP will be competing with a number of commercial operations of "personal genomics." But the two differences are that it will be free to enter (the PGP) and that all the information will be publicly available. That open access is a bold idea, and PGP is even made bolder by the fact that G. Church hoped to use new techniques available to convert some sample cells into stem cells. This would enable the project's researchers to carry out investigation on a tissue-by-tissue basis. In other words, the PGP aims to identify genes in the genome, then to find out what they do, and then to understand whether that knowledge has any medical value (*The Economist,* 2010 j).

The United Kingdom's Biobank was even more ambitious. It is a government-run project that aims to collect tissue samples and medical data from 500,000 people in the United Kingdom. That will allow an analysis of long-term correlations between genes (as well as lifestyles)

and health, though in contrast to the PGP the participants' anonymity will be preserved (*The Economist*, 2010 j).

Exploiting existing data-collection projects can also yield results in terms of associating diseases with genes. Richard Lifton cited the Framingham Heart Study, which began in 1948 and had followed several generations in Framingham, near Boston, to try to disentangle the causes of heart disease and strokes. It had revealed a number of genes that might prove classic examples of "dark matter." If present as a single copy inherited from either mother or father, certain mutations of these genes protect against heart disease and strokes. Those mutations are not, however, common enough to be detected by conventional GWAS. On the other hand, individuals who inherit two copies of them, one from each parent, will be stillborn and so they cannot be detected by classic Mendelian counts either. This kind of results suggested the logical thing to do would be to apply mass-genome sequencing and compare the resulting data with everyone's medical records. The bigger the sample, the more robust the eventual result. The main objection to that approach is privacy, and the extent to which this really is an issue might be shown by the success or otherwise of G. Church's approach. The PGP began recruiting in March 2010, so all should soon be clear. If the project is a success, the view that a person's DNA is his own business may fade away (*The Economist*, 2010 j).

Even when the data from the biobanks and the personal genome projects are in, though, they will still have to be turned into knowledge that can be used for developing pharmaceuticals. Turning data into knowledge is, for instance, the task of the Broad Institute, as it involves more sequencing (of RNA rather than DNA, to see which genes are actually active in particular sorts of cells) and considerable computing work (to show up the correlations in activity which indicate that particular molecules are parts of the same biological pathway) [*The Economist*, 2010 j].

Genetics of ageing

The study of Paola Sebastiani, of the Boston University School of Public Health in Massachusetts, and her colleagues is the largest investigation conducted into the genetics of very old people. The team analyzed DNA from more than 1,000 people aged between 95 and 119 years to look for genetic markers which were not present in those who live an average span. The results of the study, published in the 11 November issue of *Science*, concerned 150 markers correlated with long life. Many sequences were associated with mental alertness, insulin regulation and

DNA stability, confirming results from smaller studies and suggesting areas for life-prolonging research (Sebastiani et al., 2010).

After determining which genetic markers were most strongly associated with lifespan, the researchers applied that knowledge to DNA samples from another set of individuals. Using their model, they were able to predict with 77% accuracy who of these people had lived for more than 100 years and who had died at a more modest age. There was no single, winning combination of genes unique to healthy ageing. Rather, 19 distinct cohorts emerged, each bearing different combinations of genetic markers – and equally distinct patterns of disease onset and physical decline. This suggested that the cumulative interactions of many genes orchestrated both overall lifespan and patterns of healthy ageing (Sebastiani et al., 2010).

Despite this strong genetic evidence, attributing longevity entirely to biological traits would be premature. Even among the very old P. Sebastiani found 30 centenarians who possessed none of the expected genetic markers. Some had a family history of prodigious old age – a pattern of heritability indicating that the study may have missed rare genetic variants. The rest, however, had no such ancestry. Maybe good luck and healthy living (exercise, moderation and a healthy diet) can sometimes overcome a genetic make-up (*The Economist*, 2010 k).

Have the promises of the Human Genome Project been fulfilled?

When President Bill Clinton shook hand with the runners of the human-genome race on 26 June 2000 at the White House, he stated that "genome science ... will revolutionize the diagnosis, prevention and therapy of most, if not all, human diseases." There was indeed the promise of a lot of new drugs as genetic targets previously unknown to biologists would become the objective of successful pharmaceutical investigation. There was talk of an era of "personalized medicine" in which treatments would be tailored to an individual's genetic make-up. And there was hope that a biotechnology boom based on genomics would occur and lots of money would be made (*The Economist*, 2010 j).

Such promises did not occur at once; the miracle drugs did not appear, nor did personalized medicine; and biotechnology firms, with a few exceptions, were consuming cash with little return. Why? Some of the reasons for this delay are technological. For instance, the machines that would sequence a whole human genome in three days and for a cost of US$2,000 would be available in the spring of 2011, stated

Philippe Froguel, of London's Imperial College and the Institut Pasteur in Lille, France. Laurent Alexandre, president of the biotechnology firm DNAVision, specialized in genome sequencing, had other explanations : "The analysis of the sequence of the DNA molecule is insufficient; one must also study how the DNA molecule is compacted, the biochemical modifications that change its interpretation by the cell machinery, what is called epigenomics; the last-generation sequencers will be able to read this second code in 2012" (Benkimoun, 2010 g).

Complexity of the human genome and genetics

In 1990, when the human-genome project began, everybody thought they knew what a gene was. Nobel prizes were awarded in abundance, and, except for a few specialized genes whose RNA was directly involved in the protein-manufacturing process, it was understood that the "central dogma" of biology (so described by Francis Crick, co-discoverer of the double helical structure of DNA) was that one gene equalled one RNA messenger molecule equalled one protein. The system made perfect sense. There were a few oddities. Most notably, the best estimate for the amount of DNA that encoded proteins was only 3% of all the DNA in the genome. But no much attention was paid to that, and the non-gene DNA was dismissely labelled "junk." But it turned out that a sizeable amount of the junk is transcribed into RNA even though it does not make proteins. Instead, the RNA itself was doing functions that were once thought to be the prerogative of proteins : regulating the transcription of other genes, protecting cells from viral attack and even keeping control of bits of DNA that really are parasitic on the whole genomic apparatus (*The Economist*, 2010 j).

There could be more than 100,000 "RNA-only" genes. Ten years ago, only a handful were known. By contrast, the current estimate of the number of protein-coding genes in the human genome is 23,000. The RNA-only genes are, moreover, medically significant : they keep popping up, for instance, in cancers (*The Economist*, 2010 j).

In bacteria, the so-called "regulator RNAs" may play an important role in their adaptation to the environment. Thus, the virulence of *Staphylococcus aureus* is strongly associated with these RNAs, as demonstrated by a team of French biochemists in June 2010. Brice Felden, the key author of the work carried out at the University of Rennes-I and published in the review *PLoS Pathogens* (June 2010), was with his colleagues the first to identify in 2005 some 15 regulator RNAs in *S. aureus*. In 2010, there were at least 100 of these RNAs in the genome of the bacterium. Some

of them are better known, such as the one called SprD, which was the subject of the recent publication by the French scientists. The association of the bacterium virulence with this RNA has been demonstrated by the following experiment : when the expression of this RNA was inactivated, in mice, the latter remained healthy after injection of *S. aureus*; conversely, if the RNA is activated, the mice fell ill after being injected by *S. aureus*. What could be expected in terms of application from these findings? First, as shown in the experiments carried out on mice, inactivation of SprD RNA (a patent has been filed on the identity of this RNA) may open therapeutic prospects in the struggle against infections caused by *S. aureus*. Secondly, in a nearer future, this RNA could be used as an early marker of the infections caused by *S. aureus*, so that clinical physicians could prescribe an appropriate antibiotic treatment more rapidly (Chabelskaya et al., 2010; Vincent, 2010 d).

The second layer of complexity is called epigenetics. This is the process by which DNA is chemically alerted by the addition of a methyl group to the nucleotide base cytosine (C). Epigenesis is yet another way of regulating transcription (the methylation stops this happening). It is, however, more permanent than the on/off switching provided by transcription factors and RNA-only genes. Indeed, it is so permanent that it can sometimes be passed down to the generations, leading to controversies about the heritance of acquired characteristics (Lamarckian evolution as opposed to Darwinian evolution). This controversy does not seem relevant, because epigenetic changes are not passed on indefinitely. Nevertheless, they may help to explain patterns of disease such as late-onset diabetes. This, some researchers assume, might be encouraged by children inheriting epigenetic patterns appropriate to the diets of their parents but inappropriate to the different, more calorific diets those children are having in modern life (*The Economist*, 2010 j).

The third layer of complexity is just starting to be explored. It has to do with the complex architecture and conformation of the DNA molecule. The latter is, indeed, a long-chain molecule, so long that if the 3 billion nucleotide pairs were linked together and pulled out straight, the result would be a meter in extent. In reality, DNA is twisted and folded up inside the cell nucleus, with the result that bits of the molecule that seem far apart on a map are actually next to each other in the nucleus or in the chromosome. The implications of such structure are still obscure. What is known is that there are often active zones of DNA transcription within a nucleus that seem to be much bigger than the width of a strand of DNA and its associated proteins. This suggests that genes apparently a long way from one another are actually, in some sense, collaborating (*The Economist*, 2010 j).

Expectations

To sum up, over the past decade research has shown the extent of our lack of knowledge as well as the complexity of what has been discovered. For instance, we discovered the wealth of the so-called junk DNA, which does not encode proteins, but plays a key role in gene expression, but this is more complex to study than what has been accomplished. It is also necessary to make comparisons with the genomes of other organisms. In fact, nothing is simple in genetics : "One single gene explains that a cow gives more or less milk, but in mice a hundred genes play a role in obesity," commented David Altshuler, one of the founders of the Broad Institute, Cambridge, Massachusetts, where he leads the Programme of Medical Genetics and Population Genetics. He also stated : "Genome science aims not to make predictions (like astrology), but to understand the genetic component of diseases. All the pathologies known to us include an inherited element which is also influenced by the environment." To be able to correlate genomic markers with pathologies, generally multifactorial, as in the case of diabetes or schizophrenia, "we need to sequence the whole genome of tens of thousands of patients, whereas we have done it for only 400 patients up to 2010," underlined Laurent Alexandre, president of DNAVision. All this explains why the promises of the human-genome sequencing did not hold true immediately (Benkimoun, 2010 g).

Francis Collins, director of the US National Institutes of Health stated that "we invariably overestimate the short-term impact of new technologies and we underestimate their long-term effects" in April 2010 in *Nature*. David Altshuler added that "the speed of technological progress has been spectacular, but that of conceptual progress has not been as fast. The same occurred in informatics or for Internet." There is a need for a renewed international collaboration that has driven the human-genome project. Some countries have pursued a policy that will enable them to remain leaders. This is the case of the United States which have a largely dominant position in sequencing technologies. "The National Institutes of Health have a strategic objective in this area and devote US$20 million per year to it. They have supported all the companies and platforms that contribute to a higher-performing and less costly sequencing. The National Human Genome Research Institute (NHGRI) has invested US$200 million in this area," stated Jeffery Schloss, in charge of the coordination of technological development at NHGRI (Benkimoun, 2010 g).

Other countries like China have decided to give a top priority to this area and made considerable investments in terms of human and material

resources. Conversely, France has lost ground. Philippe Froguel of Imperial College, London, and the Institut Pasteur, Lille, France said that "France dominated medical genetics in 1990, but it did not participate in the mass sequencing of different organisms because of lack of funding during the last ten years" (Benkimoun, 2010 g).

There is no doubt that many discoveries are expected from the research on the human genome and on the interactions that influence its expression. Eric Lander, director of the Broad Institute and co-president of the Council of Experts on Science and Technologies which assists President Barack Obama, "is convinced that we are dealing with a scientific revolution that is expected to transform the way we treat diseases. A cancer could occur in a hundred or a thousand ways; we must review all of them in order to better know this enemy. In this area as well as in others of medicine, we shall be able to treat the causes of diseases and not only their symptoms" (Benkimoun, 2010 g).

Contribution of China to advances in genomics

Important advances in genomics may happen in China, which is enlarging and equipping with the last-generation sequencing machines the once known as the Beijing Genomics Institute (BGI). In addition to the institute's Hong Kong operation, the institute itself is based over the border in the People's Republic proper, in Shenzhen. The BGI is the leading part of China's effort to show that it can be the scientific peer of the West. The institute's director, Yang Huangming, aims to make the BGI the first global genomics operation. Part of the reason for building its newest sequencing centre in Hong Kong was to attract North American and European laboratories and researchers. The BGI began in 1999, when Yang Huangming participated in the Human Genome Project by sequencing part of the tip of chromosome three (about 1% of total human genome) as the Chinese contribution to that international project. From this modest beginning, it now plans to sequence 200 full human genomes as part of an international collaboration called the 1,000-genome project. Half of these genomes will be Chinese, but the institute's researchers intended to sample the full geographical range of humankind. BGI had already sequenced the genomes of rice, cucumbers, soybeans and sorghum, honeybees, water fleas, pandas, lizards and silkworms, and some 40 other species of plants and animals, along with over 1,000 bacteria (*The Economist*, 2010 j).

BGI is also interested in cancer. According to its director, the institute will not merely compare healthy and tumorous tissues from the same individuals, as the International Cancer Genome Consortium (of which it is part) plans to do, it will actually be able to follow the pattern of mutation, in the order that it happened, within an individual that has led to his/her cancer. That may allow pre-emptive treatment to be developed for people whose tumours are not yet malignant. Indeed, as the cost of sequencing falls, this "internal phylogenetics," as Yang Huangming calls it, might be extended to trace the pattern of mutation that develops in even an apparently healthy body as cells proliferate within it. It might help to explain patterns of disease associated with ageing as cells whose ancestors were genetically identical slowly diverged from one another (*The Economist*, 2010 j).

BGI's researchers are experimenting with animal cloning. They were the first to clone pigs, and they have developed a new and more effective way of cloning mammals that might ultimately be applied to humans, if that were ever permitted. Researchers are also studying fast-growing plants with interesting structural properties, such as balsa, a lightweight South American wood familiar to generations of schoolboy model-makers, and bamboo, a traditional construction material in China (*The Economist*, 2010 j).

But BGI is involved in more controversial projects, e.g. the research on the genetic underpinning of intelligence. Two thousand Chinese schoolchildren will have 2,000 of their protein-coding genes sampled, and the results correlated with their test scores at school. Although it will cover less than a tenth of the total number of protein-coding genes, it will be the largest-scale examination to date of the hypothesis that differences between individuals' intelligence scores are partly due to differences in their DNA. BGI's director is also candid about the possibility of the 1,000-genome project revealing systematic geographical differences in human genetics, or "racial" differences (*The Economist*, 2010 j).

BGI's activity is funded mostly by regional development grants and loans from state-owned Chinese banks, but the institute's director hoped to go properly commercial. The Hong Kong operation will work partly as a contractor, and Yang Huangming hoped to persuade biologists around the world to send their samples in and have them sequenced there rather than relying on their own universities to do the sequencing (*The Economist*, 2010 j).

Epigenetics

Epigenetics versus conventional genetics

In the 1980s, Lars Olov Bygren, a preventive-health specialist at the Karolinska Institute in Stockholm, was trying to understand what long-term effects the feast and famine years might have had on children growing up in Norrbotten – the northernmost county of Sweden – in the 19th century, and not just on them but on their offspring as well. Norrbotten is underpopulated: an average of just two people live in each square kilometre. In the 19th century, it was so isolated that if the harvest was bad, people starved. For instance, 1800, 1812, 1821, 1836 and 1856 were years of total crop failure and extreme suffering. But in 1801, 1822, 1828, 1844 and 1863, the harvests were so abundant that the same people who had been hungry in previous winters were able to eat much food for months (Bygren et al., 2001).

L.O. Bygren drew a random sample of 99 individuals born in the Overkalix parish of Norrbotten in 1905 and used historical records to trace their parents and grandparents back to birth. By analyzing meticulous agricultural records, Bygren and two colleagues determined how much food had been available to the parents and grandparents when they were young. Around the time he started collecting the data, Bygren became aware of research work showing that conditions in the womb could affect health not only of the foetus, but also of adults. In 1986, for instance, the *Lancet* published the first two groundbreaking papers showing that if a pregnant woman ate poorly, her child would be at significantly higher than average risk for cardiovascular disease as an adult. Bygren wondered whether that effect could start even before pregnancy : in other words, could parents' experiences early in their lives somehow change the traits they passed to their offspring? (Bygren et al., 2001).

This was not an idea that could be accepted easily. Charles Darwin in his book *On The Origin Of Species* (in November 2009, his 150th anniversary had been celebrated) indicated that evolutionary changes take place over many generations and through millions of years of natural selection. But there has been a lot of historical evidence suggesting that powerful environmental conditions (near death from starvation, for instance) can somehow leave an imprint on the genetic material in ovocytes and sperm. These genetic imprints can short-circuit evolution and pass along new traits in a single generation. For instance, Bygren's research showed that in Overkalix, boys who enjoyed those rare overabundant winters, i.e. who went from normal eating to gluttony in a single season,

had sons and grandsons who lived shorter lives. In the first paper he wrote about Norrbotten, published in 2001 in the Dutch journal *Acta Biotheoretica*, he showed that the grandsons of Overkalix boys who had overeaten died an average six years earlier than the grandsons of those who had endured a poor harvest. One Bygren and his team controlled for certain socioeconomic variations, the difference in longevity rose to an astonishing 32 years. Later papers using different Norrbotten cohorts also found significant decreases in life span and discovered that they applied along the female line as well, meaning that the daughters and granddaughters of girls who had gone from normal to gluttonous diets also lived shorter lives. In other words, the data suggested that a single winter of overeating as a youngster could initiate a chain of biological events that would lead one's grandchildren to die decades earlier than their peers did (Bygren et al., 2001).

Bygren's data – along with those of many other scientists working separately since the early 1990s – have given rise to epigenetics, which is the study of changes in gene activity that do not involve alteration to genetic code but still are passed down to at least one successive generation. These patterns of gene expression are governed by the cellular material – the epigenome – which is outside the genome but above it (*epi* means above). It is these epigenetic "marks" that switch the genes on or off. It is through epigenetic marks that environmental factors like diet, stress and prenatal nutrition can make an imprint on genes that is passed from one generation to the next (Cloud, 2010).

For instance, there is evidence that smoking and eating too much can change the epigenetic marks in ways that cause the genes for obesity to express themselves too strongly and the genes for longevity to express themselves too weakly. It is becoming clear that some behaviours can also predispose offspring to disease and early death. But scientists are also learning to act on epigenetic marks in the laboratory, i.e. they are developing drugs that treat illness simply by silencing genes and expressing others. In 2004, the US Food and Drug Administration (FDA) approved an epigenetic drug for the first time. Azacitidine is used to treat patients with myelodysplastic syndromes, a group of rare and deadly blood malignancies. The drug uses epigenetic marks to silence genes in blood precursor cells that have become overexpressed. According to Celgene Corp., based in Summit, New Jersey, which makes azacitidine, patients with the myelodysplastic syndromes live a median of two years on the drug; those taking conventional blood medications live just 15 months. Since 2004, the FDA has approved three other epigenetic drugs that are thought to work at least in part by stimulating tumour-

suppressor genes that disease had silenced. The great hope is that genes playing a role in many diseases (including cancer, schizophrenia, autism, Alzheimer's, diabetes) could be silenced (Cloud, 2010).

Scientists have known about epigenetic marks since at least the 1970s, but until the late 1990s researchers have begun to realize that epigenetics could also explain certain scientific data that conventional genetics never could : for instance, why one member of a pair of identical twins can develop bipolar disorder or asthma even though the other is fine, or why autism affects boys four times as often as girls; or why extreme changes in diet over a short period in Norrbotten could lead to extreme changes in longevity. In these cases, the genes may be the same, but their patterns of expression have clearly been tweaked (Cloud, 2010).

DNA methylation altering gene expression; epigenome mapping

DNA methylation, i.e. the addition of a methyl group to a specific locus on a gene, can change the gene's expression, switching it off or on, dampening its expressing or increasing it. In 2003, Randy Jirtle, an oncologist of Duke University, and one of his postdoctoral students, Robert Waterland, conducted an experiment on mice with a uniquely regulated agouti gene – a gene that gives mice yellow coats and a propensity for obesity and diabetes when expressed continuously. Jirtle's team fed one group of pregnant agouti mice a diet rich in B vitamins (folic acid and vitamin B12). Another group of genetically identical pregnant agouti mice did not receive such prenatal diet. The B vitamins acted as methyl donors : they caused methyl groups to attach more frequently to the agouti gene *in utero*, thereby altering its expression. Therefore without altering the structure of DNA, Jirtle and Waterland induced agouti mothers to produce healthy brown pups that were of normal weight and not prone to diabetes (Cloud, 2010).

In another experiment, fruit flies exposed to a drug called geldanamycin showed unusual outgrowths on their eyes that could last through at least 13 generations of offspring even though no change in DNA had occurred (and generations 2 through 13 were not directly exposed to the drug). Similarly, according to a paper published in the June 2009 issue of the *Quarterly Review of Biology* by Eva Jablonka and Gal Raz of Tel Aviv University, roundworms fed with a kind of bacteria can feature a small, dumpy appearance and a switched-off green fluorescent protein; the changes last at least 40 generations. The paper published by the Israeli scientists documented some 100 forms of epigenetic inheritance (Jablonka and Raz, 2009).

Knowing the protein-coding genes has been useful, it has provided a lexicon of proteins, including many previously unknown ones. What is needed, however, is an explanation of what the proteins mean and what they are. For that, one needs to know how the genes' activities are regulated in the 220 or so different cell types a human body is made from. That is the purpose of the American government's Roadmap Epigenome programme, aimed at understanding the role of epigenetic processes in the expression of genes (*The Economist*, 2010 l).

In 2008, the National Institutes of Health (NIH) announced it would devote US$190 million into a nationwide initiative to understand "how and when epigenetic processes control genes". Elias Zerhouni, director-general of NIH at that time, stated that epigenetics had become "a central issue in biology." In October 2009, the NIH grant started to pay off. Scientists working jointly at a largely Internet-based effort called the San Diego Epigenome Center, announced from the Salk Institute, La Jolla, California, that they had mapped a portion of the human epigenome. Ryan Lister and Mattia Pelizzola of the Salk Institute and their colleagues published their results by mid-October 2009 in *Nature* (Lister et al., 2009; *The Economist*, 2009 l; Cloud, 2010).

Epigenomics studies the distribution over the genome DNA of methyl groups. These can attach themselves to cytosine. In so doing, they help to control transcription, in which a copy of a gene is transcribed into a molecule of RNA, the first stage in the translation of a gene into a protein. The presumption is that the pattern of methylation, by controlling which proteins are manufactured, helps to determine what type of cell is produced. A cell with its haemoglobin genes switched on to overdrive, for instance, will become a red blood cell. Another one that synthesizes actin and myosin, which link up to form units that can expand and contract, will become a muscle cell. R. Lister and M. Pelizzola have tested this idea by describing the first epigenomes of human embryonic stem cells, which retain the potential to turn into a variety of other cell types, and of fetal lung fibroblasts, which are the end of one line of cell specialization (Lister et al., 2009).

They studied the methylation patterns of these cells using a chemical reaction that turns methylated cytosine into uracil. This base is found in RNA, rather than DNA. Altogether, the researchers were able to read and compare about 90% of the genomes of their two types of cell. Their first discovery was that the stem cells were more methylated than the lung cells – 5.8% of cytosines, compared with 4.3%. It is a common characteristic of the so-called promoter regions of genes, where transcription begins, that they contain long, repetitive sequences of alternating cytosines

and guanines. If these areas become methylated, it tends to suppress transcription of the gene in question. A quarter of the methylated cytosines in stem cells, however, were not followed by guanines. Nor were they found in the promoter regions of genes, but rather in the transcribed parts of the genes themselves. They also had the opposite effect from methylated cytosines found in promoter regions. The regions they occurred in tended to be transcribed more than usual, not less. In particular, a lot of genes involved in processing RNA were activated in the stem cells in this way ((Lister et al., 2009; *The Economist*, 2009 l).

According to Ecker, a biologist of the Salk Institute who worked on the epigenome maps, each of the 220 or so cell types of the human body is likely to have a different epigenome. The human epigenome contains an as yet unknownable number of patterns of epigenetic marks – probably in the millions. Consequently, a full epigenome map would require major advances in computing power. When completed, the Human Epigenome Project (already under way in Europe) would probably make the Human Genome Project a relatively small task. Many scientists consider that the era of epigenetics has arrived, with all its implications for human, animal and plant physiology.

Examples of potential epigenetic applications

Apomyxis is a characteristic that enables a plant to produce seeds without going through sexual reproduction. The offspring are therefore perfect copies of the mother plant. It is an attractive idea to transform the main food crop species or varieties into apomyctic plants, but such transformation is limited by epigenetic constraints concerning either the regulation or the transmission of the trait. French scientists belonging to the Research for Development Institute (IRD) have tried to transfer apomyxis to cultivated maize through hybridization with a wild and apomyctic relative, *Tripsacum*. The research team in charge of the Seed Development Unit showed that one single chromosomal region, likely to be regulated epigenetically, was responsible for apomyxis in *Tripsacum.* The team's objective is to understand this regulation using the association maize/*Tripsacum* as a model, and through the study of chromatin structure among wild plants, apomyctic and that reproduce sexually, as well as in various mutants. Preliminary results indicated that deregulation of certain genes involved in the control of chromatin structure could be at the origin of apomyxis in sexually reproduced plants. Although the understanding of apomyxis is still very fragmentary, the work already carried out would allow the design of strategies aimed at developing apomyctic maize varieties and, more generally, at using this trait in crop biotechnology.

Clonal multiplication of oil palm, following *in vitro* tissue culture (somatic embryogenesis), has been used since the mid-1970 to expand the plantations of this tree worldwide, and particularly in Malaysia and Indonesia, the world's first and second-biggest producers of palm oil. It has been observed that the process gives rise to a small percentage of variants with abnormal flowering, which are therefore unproductive. These variants are not the result of mutations, but of epigenetic variations. This explanation has been put forward by a research team of the French Research for Development Institute (IRD) Development Biology of Palms Unit, which was searching reliable criteria or indices that allow for the early identification of the variants, so as to eliminate them before plantation. Their research has led to the selection of some 20 early epigenetic markers. All these are related to abnormal flower development and therefore to the further production of fruit. These markers are being validated through tests on plant material from various origins (palms from Africa, America and Asia). The French scientists have studied the epigenetic variations affecting chromatin structure and DNA methylation, as well as their role in the expression of potential markers. Furthermore, they are also studying the relationships between chromatin structure and small RNAs, which were discovered by the late 1990s and which seem to play a key role in the regulation of epigenetic variations.

Another team of the French Research for Development Institute (IRD) Laboratory of Plant Genomics and Development has identified the proteins encoded by the virus causing the rice yellowing disease and which suppresses the defence system of the plant. Once the defence system has been inactivated by the virus, the host plant cannot detect the invasion by the virus. Thereafter, the virus invades the plant and starts multiplying, causing the yellowing and wilting of leaves. It remains to be demonstrated that the modifications induced by the virus at the epigenetic level are found in rice seeds. In this case, the virus could modify its host in the long run via its offspring. Once the exact modalities of the virus attack are deciphered, the interaction between rice and yellow streak virus may serve as a model for other plant/virus interactions.

In the area of human parasitic diseases, such as leishmaniasis, caused by the protozoan *Leishmania,* IRD researchers are studying epigenetic factors under the project Epi-Vir. Among those factors that may be involved in adaptative phenomena, certain enzymes that modify chromatin structure could regulate the virulence of the pathogen. The researchers are checking the hypothesis concerning the conservation of these mechanisms in *Leishmania* and their role in the parasite development cycle. Genetically modified parasites are produced, and they are able

to over- or underproduce the enzymes acting on the parasite chromatin structure. It has been demonstrated that the partial inactivation of one of these enzymes results in the inhibition of the multiplication of the parasite in humans and dogs. The analysis of gene expression in these genetically modified parasites aims at identifying those which are under epigenetic control. Among those identified, some are known to be involved in virulence and therefore in host/parasite interactions. These results should be confirmed and their role in natural conditions must be assessed.

Epigenetics and evolution

Epigenetics should not be confused with evolution. It does not alter DNA. Epigenetic changes are a response to an environmental pressure. That response can be inherited through many generations via epigenetic marks, but if the environmental pressure is removed, the epigenetic marks will eventually fade, and the DNA code will over time begin to revert to its original programming. Only natural selection causes *permanent* genetic change. But epigenetic inheritance can be powerful. On 17 March 2010, the *Journal of Neuroscience* published a paper showing that even memory can be improved from one generation to the next via epigenetics. The paper described an experiment with mice led by Larry Feig, a biochemist of Tufts University. Feig's team exposed mice with genetic memory problems to an environment including toys, exercise and extra attention. These mice showed significant improvement in long-term potentiation (LTP), a form of neural transmission that is key to memory formation. Surprisingly, their offspring also showed LTP improvement, even when offspring received not extra attention (Kochlamazashvili et al., 2010).

In his book *The Genius In All Of Us: Why Everything You've Been Told About Genetics, Talent And IQ Is Wrong,* science writer David Shank stated epigenetics is helping usher in a "new paradigm" that "reveals how bankrupt the phrase 'nature versus nurture' really is." He calls epigenetics "perhaps the most important discovery in the science of heredity since the gene." Geneticists are progressively acknowledging that we may have dismissed an early naturalist who anticipated modern epigenetics – and whom Darwinists have long disparaged. Jean-Baptiste Lamarck (1744-1829) argued that evolution could occur within a generation or two. He made the hypothesis that animals acquired certain traits during their lifetimes because of their environment and choices. For instance, giraffes acquired their long necks because their recent ancestors had stretched to reach highly located, nutrient-rich leaves. The Darwinist thinking is that giraffes acquired their long necks over millennia because the relevant genes had, very slowly, gained advantage. The Lamarckian

hypothesis came to be seen as a scientific blunder, but epigenetics is now bringing scientists to re-evaluate Lamarck's ideas (Cloud, 2010).

By early 2000, Lars Olov Bygren was convinced that the feast and famine years in the 19th century Norrbotten had caused some form of epigenetic change in the population. In order to explain the way the change occurred, he was inspired by a paper published in 1996 by Marcus Pembrey, a prominent geneticist at University College London. Published in the Italian journal *Acta Geneticae Medicae et Gemellologiae*, Pembrey's paper, now considered seminal in epigenetic theory, was contentious at that time, and major journals had rejected it. Although M. Pembrey is a committed Darwinist, he used his paper – a review of available epigenetic science – to speculate beyond Darwin : what if the environmental pressures and social changes of the industrial age had become so powerful that evolution had begun to demand that our genes respond faster? What if our DNA now had to react not over many generations and millions of years but, as M. Pembrey wrote, within "a few, or moderate number, of generations?" (Pembrey, 1996). In May 2000, M. Pembrey received an e-mail from Bygren – whom he did not know – about the Overkalix life-expectancy data, and both began to design a new experiment that would clarify the Overkalix mystery. While there was a need to replicate the Overkalix findings, it was not possible to conduct an experiment in which some kids starve and others overeat, and it was not possible either to wait for 60 years for the results (Cloud, 2010). By coincidence, M. Pembrey had access to another trove of genetic information. He had long been on the board of the Avon Longitudinal Study of Parents and Children (ALSPAC), a research project at the University of Bristol, developed by Jean Golding, an epidemiologist at the university and friend of M. Pembrey. ALSPAC had followed thousands of young people and their parents since before the children were born, in 1991 and 1992. For the study, J. Golding and her staff recruited 14,024 pregnant mothers – 70% of all the women in the Bristol area who were pregnant during the 20-month recruitment period. The ALSPAC parents and children had undergone extensive medical and psychological testing every year since. All this data collection was designed from the outset to show how the individual's genotype combines with environmental pressures to have an impact on health and development. For instance, ALSPAC data had shown that baby lotions containing peanut oil may be partly responsible for the rise in peanut allergies; high maternal anxiety during pregnancy was associated with the child's later development of asthma; young little children who were kept too clean were at higher risk for eczema. A groundbreaking paper was published in February 2006 in the *European Journal of Human Genetics*, where it was highlighted that of the

14,024 fathers in the ALSPAC study, 166 said they had started smoking before age 11 – just as their bodies were preparing to enter puberty. Boys are genetically isolated before puberty because they cannot form sperm (girls, by contrast, have their ovocytes from birth). That makes the period around puberty fertile ground for epigenetic changes, i.e. imprint epigenetic marks on genes in the Y chromosome, when sperm is first starting to form (Pembrey et al., 2006).

When M. Pembrey, L.O. Bygren and J. Golding – all working together – looked at the sons of those 166 early smokers, it turned out that the boys had significantly higher body mass indexes than other boys by age 9. That meant the sons of men who smoke in prepuberty were to be at higher risk for obesity and other health problems well into adulthood. It was very likely these boys were to have shorter life spans, just as the children of the Overkalix overeaters did. In other words, one can change his/her epigenetics even when one makes a wrong decision at 10 years old. If one starts smoking then, it is not only a health mistake, but also a genetic mistake with serious implications (Pembrey et al., 2006; Cloud, 2010).

Use of genomics information

Personality and genomics

There is a general consensus that our lives are shaped by genes, environment and interactions between the two. Certain people are much more likely than others to be exposed to stressful life experiences, including specific traumas, like car accidents, industrial injuries or being a crime victim. Some of this variation is traceable to genetics. Psychiatric geneticists have been studying "heritability," the amount of the variation within a population that can be explained by genetic differences between individuals. Identical twins are more likely to both experience a variety of life events than fraternal twins, who, like siblings of different ages, share only half of their genes. About one-fourth of the variation in life experiences can be traced to genetic origins, as dozen of studies have shown (Wang and Aamodt, 2009).

A person whose identical twin is alcoholic – whether or not he himself, or she herself, has any substance abuse problems – is more likely to have been robbed or in trouble with the law than a person whose fraternal twin is alcoholic. In other words, people with similar personalities seek out similar experience and may take similar risks. What connects our genetic inheritance to environmental experiences? Most likely it is personality, which is known to depend on genes. In one study, three common

measures of personality – extraversion, neuroticism and openness to experience – were enough to explain the entire heritability of some life events. In general, neurotic people are more likely to experience negative life events, while extraverted people are more likely to experience positive and controllable life events (Wang and Aamodt, 2009).

So some of the effects that we call "genetic" (or "nature") are the indirect result of people being drawn to particular environments because of their personality. In other words, some "environmental" (or "nurture") effects are actually attributable to genetic tendencies. The debate about "genes versus environment" does in fact point to something more complex : genetic predispositions interact with circumstances to produce unique individuals (Wang and Aamodt, 2009).

For instance, major depression arises from a vicious cycle between genes and environment. A particular gene influences the sensitivity of individuals to bad experiences. One famous paper demonstrated a complex interaction between the serotonin transporter gene and negative events (the gene encodes the synthesis of a protein that removes the neurotransmitter serotonin from the synapse after a neurone releases it; the action of this protein is inhibited by antidepressants like Prozac). Persons with two copies of the high-risk variant of the gene are likely to develop depression in response to multiple stressful experiences like divorce or assault, but they are not depressed if their environment remains benign. In contrast, people with two copies of the low-risk form of the gene are resilient against depression, even when they experience environmental stressors. People with one copy of each variant fall somewhere in between. Genes that predispose people to depression, though, also influence their risk of experiencing negative environmental events. In one study, women whose identical twin suffered from depression were significantly more likely to have been assaulted, divorced, or had a serious illness or major financial problems than people whose fraternal twin was depressed (it is not known which genes are responsible for this effect) [Wang and Aamodt, 2009].

These bad events did not occur because the women were depressed, as the correlations persisted even when women who were currently depressed were excluded from the study. Thus, genes can act on the same disorder by making people more sensitive to stressful environmental events and by making these events more likely to occur. The interaction between genetic tendencies and life experiences may explain another puzzling finding : the heritability of many psychological traits – from intelligence to anxiety – increases as people mature. This observation seems odd, since

genes are most important in brain development in babies and children. As people become older, they become more able to determine their own circumstances, and they may be able to choose environments that reinforce their natural personality tendencies. Apparently those who suspect that we resemble our parents as we become older may have a valid point. To conclude, Sam Wang, associate professor of neuroscience and molecular biology at Princeton University, and Sandra Aamodt, former editor in chief of *Nature Neuroscience*, consider that we should not blame our genes for our good or bad fortunes, because only one-fourth of the variation in life events is heritable (Wang and Aamodt, 2009).

Banning genetic discrimination

Genetic testing may be used to discriminate people, because they carry a gene or genes that predispose them to a disabling disease or to an illness which would handicap their efficiency at work. But just as the law forbids discrimination against a person because he (she) is black, a woman, or an immigrant, it should forbid discrimination against a person because he (she) carries a gene that may be detrimental. And the logic is similar : why should the person be punished for something completely beyond his (her) control? (Kinsley, 2008).

In May 2008, the US House of Representatives voted 414 to 1 to outlaw genetic discrimination. The Senate passed the same bill unanimously. The Genetic Information Nondiscrimination Act prevents employers and insurance companies from using the results of genetic tests in choosing their employees and customers. But Michael Kinsley (2008) who considered that this law was a good thing asked the question : "How far should it be taken?" "We cannot outlaw discrimination on the basis of talent. We do not want to. Discrimination in favour of talent – rewarding a talented cellist over a lousy one – is how we get talent to express itself."

As writers like Richard Dawkins (*The Selfish Gene*) and Robert Wright (*The Moral Animal*) have shown, it is difficult to differentiate between genetically determined aspects of the human condition and those which are the result of free will (nature vs. nurture, see above p.122). In the context of insurance, the goal is to protect against the unexpected or unlikely. Consequently forbidding insurers to take predictable risks into account when choosing whom to insure and how much to charge is requesting them to make bets they are sure to lose. M. Kinsley considers that "not insuring people who are likely to develop a cancer, or charging them more, is not evil." But there should be some genetic justice and the law voted by the US Congress is a welcome step in the right direction,

because it is part of the struggle for equality among human beings and citizens.

Freedom of DNA information? Gene patenting?

One of the lessons of genomics and post-genomics is that it is all about information. DNA databases are a good illustration of that, and of the conflicts and paradoxes the new age of genomics is creating. There is an understandable fear not so much of what governments might do with the information now as what they might do with it in the future. DNA is more than just a reliable biometric, it is an individual's essence. Many people prefer to keep their essences for themselves. Few want their weaknesses exposed publicly. They may not even want to confront those weaknesses in private. That is what freedom of DNA information threatens. Disease susceptibility, life expectancy, personality traits, intelligence, criminal tendencies – all might be illuminated by the harsh light of free DNA information. Even if laws against genetic discrimination are passed (as in America with the Genetic Information Nondiscrimination Act of 2008), it is possible to imagine a future in which individuals are dogged by their DNA (*The Economist*, 2010 j).

Yet the benefits for medical research – and thus the health of future generations – of DNA information being free are considerable. And perhaps projects like George Church's Personal Genome Project will show that those who are allowed to volunteer their genes, rather than having them wrenched from them by the authorities, will be inclined to be generous. Once the limits of DNA-based knowledge become apparent, some of the fears are likely to disappear (*The Economist*, 2010 j).

On 29 March 2010, the Federal District Court of New York ruled on a longstanding American legal dispute. This was a claim by Myriad Genetics, of Salt Lake City, to patent protection on two human genes *BRCA1* and *BRCA2*. Some versions of these genes increase the risk of breast cancer, and Myriad Genetics sells a test that detects these versions. The patents in question, though, are not for the test but the genes themselves. The company claimed to own the intellectual-property rights to these sequences of DNA, even though they are natural and found in every human being. The court disagreed (*The Economist*, 2010 j).

J. Craig Venter, meanwhile, was seeking a patent on his DNA sequence implanted in a cell he called JCVI-syn1.0. His request is somewhat on firmer ground than that of Myriad Genetics, since the DNA in question is clearly an artefact, albeit one based on a natural sequence. Also bacteria are not

humans. However, the principle that anyone can "own" an organism's DNA in this way is disturbing to many people (*The Economist*, 2010 j).

What can and cannot be patented needs to be sorted out, for property rights lie at the heart of the business. Patent law is supposed to encourage innovators, rewarding them with temporary monopolies but requiring them to place the details of their inventions in the public domain and thus open them to competitors. In this context, patenting an artificial genome for a bacterium seems reasonable. Similar rules may apply to a crop with an artificial genome. Such things both need and deserve to be expensive. They need to be because they cost money to develop. It should not apply, though, to a natural genome. Nor, many would argue, should it apply to synthetic DNA if that DNA is then inserted into a human being. Indeed, the question of whether such insertions should be allowed at all is fraught. Since ethical norms vary from country to country, it seems unconceivable that no one would try this. Some thirty years ago the fears felt by some about *in-vitro* fertilization largely disappeared in the face of the first child born using the new technology. However, all that is technically possible is not necessarily feasible. The value of genomics can be enormous, and we ought to decide about keeping its results private or making them public for the good of humankind, and also to determine the extent of the application of intellectual-property rights to these results (*The Economist*, 2010 j).

VACCINES AND VACCINATION

The long fight towards eradication of smallpox

Smallpox attacks people of all ages, but is particularly deadly among children. The mortality rate, before the disease was eradicated worldwide, was around 30%, and those who survived were often blind and with deep scars on their faces. The viral disease dates back at least 12,000 years, and is thought to have originated in either Africa or India. References to it can be found in ancient history, e.g. a quarter of the Athenian army was killed by the disease in 430 BC during the war with Sparta, while up to 7 million Romans died in epidemics. The Spaniards took it to the Americas, and up to half of the 30 million population of the Aztec empire in Mexico died in a few months after catching the disease from the troops of Hernán Cortes in 1519 (Ariza, 2010).

The first step towards the eradication of smallpox occurred with the observation by Edward Jenner that milkmaids who developed cowpox, a less serious disease, did not develop the deadly symptoms of smallpox. In 1796, the British physician took the fluid from a cowpox pustule on a dairymaid's hand and inoculated it to an 8-year-old boy. Six weeks later, he exposed the boy to smallpox, and he noticed that the boy did not develop any symptoms. E. Jenner coined the term "vaccine" from the Latin word *vaca* (cow). His work was initially criticized, but thereafter it was adopted. By 1800, about 100,000 people had been vaccinated, most of them still in Europe (Ariza, 2010).

Hundreds of thousands of people in the Spanish colonies of the New World and around the globe were not protected against the virus, and no means had been found to safely and efficiently transport the vaccine. Shipping infected cows across the Atlantic was one option under consideration until Xavier Balmis, who was surgeon royal to the court of Charles IV of Spain, suggested taking orphans instead. On 16 September 1805, X. Balmis who had set sail since 20 November 1803 from the

Galician port of La Coruña aboard the fregate María Pita (seven years after the development of the vaccine by E. Jenner) landed on the Portuguese colony of Macau, off the South China coast. Along with him were three young orphans, who carried the vaccine against smallpox (Ariza, 2010).

X. Balmis, already past 50, had been dreaming about taking a cure for smallpox to the Spanish colonies of the Americas, and thereafter continue across the Pacific to Asia. The Portuguese vessel carrying him on the final stage of his journey had been wrecked by a hurricane, with the loss of 20 men. But X. Balmis pursued his travel and took particular care of the three children who had the precious vaccine in their bodies. Transporting vials from one continent to another on hazardous sea journeys that often lasted months, and without refrigeration, was not an easy option, and that meant smallpox was still a rife in the Americas, Africa and Asia. Under the commission of the Spanish crown, X. Balmis found the solution by taking 22 orphan boys as successive *in vivo* carriers of the vaccine (Ariza, 2010).

It was the world's first international health-care expedition. Two of the 22 orphans, Tomás Metitón and Juan Antonio, aged three and five respectively, died during the first part of the journey to Puerto Rico. Another boy was lost in the Caribbean as the Maria Pita was sailing towards Venezuela, Cuba, Yucatán and Mexico, then across the Pacific to the Philippines and on to Macau and Canton, before eventually returning to Spain. X. Balmis left behind his deputy, the surgeon José Salvany, in the New World. J. Salvany pursued the journey southward, taking vaccines with him along the way. He died in what is now Bolivia, seven years later in 1810. In addition to pirates, X. Balmis and his team had to deal with the British navy, at a time when Spain was at war with Great Britain, and also cope with storms. In the ports where he stayed, authorities would periodically take the children from him and put them in orphanages and hospitals. Many clerics also opposed the vaccination at the time (Ariza, 2010).

Young children were ideal carriers of the attenuated virus : the vaccine worked better in them. The vaccine would be injected into a small cut in the shoulder (scarification), and ten days later they would produce a small amount of pustules containing the vaccine. The scar would then heal, and the vaccine would then immediately be injected into another child, twice, to be sure. A human chain was thus set up. The use of *in vivo* carriers of the vaccine was considered by Charles Arntzen, a researcher at the University of Arizona, who works on the production of vaccines in plants, as "fascinating" for the age, and Guillermo Olagüe, a professor of the history of science at the University of Granada, described Balmis'

approach as "the most important contribution by Spain to the history of public health" (Ariza, 2010).

Nevertheless, the project faced huge logistical difficulties. The vaccinated children could only provide a single vaccine from their immune systems. This implied that X. Balmis had to constantly recruit new children as he moved on. But families were very reluctant to allow their children to be used in this way. In the same way that condemned convicts had been used to test vaccines in Great Britain, X. Balmis found equally vulnerable members of society : orphans. At each port he arrived in, he would visit orphanages, but often without success. In Cuba, he bought three young African slaves and a drummer boy. He sailed to the Philippines with 26 *in vivo* carriers (Ariza, 2010).

G. Olagüe stated that nothing is known of the fate of the orphan children. "Aside from those selected to make the trip, we have no information about them. There is a little information about those who left Mexico for the Philippines. In Mexico, Balmis went to great lengths to make sure that they were found appropriate homes, and not dumped in the local orphanage. He also made sure that they were given an education. Many of them were adopted by families in Mexico." Even less is known about X. Balmis' journey through Asia. According to some sources, Balmis failed in Macau, managing only to vaccinate 22 people with the aid of Macau's bishop. Diaz de Yraola wrote that when X. Balmis arrived exhausted in Canton with a Chinese *in vivo* carrier, the local authorities were deeply suspicious of his intentions, and refused to allow him to vaccinate on the basis that he was carrying out the "Machiavellian policies of the sons of Albion" (Ariza, 2010).

Despite the paucity of information concerning the number of vaccinated people (probably under 100,000), "Jenner himself, along with other notable scientists in Europe, were the first to recognize the importance of Balmis' work," stated Diaz de Yraola. Susana Ramírez, a professor of American history at the University Carlos III in Madrid, considered that Balmis was "the beginning of the end of smallpox; he conceived a health network that controlled epidemics from the first vaccine councils established in the early 1800s in the Spanish colonies until independence in the 1820s" (Ariza, 2010).

The fall of smallpox began with the realization that survivors of the disease were immune for the rest of their lives. This led to the practice of variolation – a process of exposing a healthy person to infected material from a person with smallpox in the hope of producing a mild disease

that provided immunity from further infection. The first written account of variolation described a Buddhist nun practising around 1022 to 1063. She would grind up scales taken from a person infected with smallpox into a powder, and then blow it into the nostrils of a non-immunized person. By the 1700s, variolation was common practice in China, India and Turkey. In the late 1700s, European physicians used this and other methods of variolation, but reported "devastating" results in some cases. Overall, 2% to 3% of variolated people died, but the practice decreased the total number of smallpox fatalities tenfold (Ariza, 2010).

Despite vaccines, smallpox continued to take a heavy toll on human populations through into the modern age, with up to 300 million people dying from the disease in the 20th century. In 1967, the World Health Organization launched a worldwide campaign to eradicate smallpox. This was achieved in ten years due to massive vaccination campaigns. The last endemic case of smallpox occurred in Somalia in 1977. On 8 May 1980, the World Health Organization's general assembly declared the world free of smallpox. At present, the virus that killed more people than any other in human history is to be found in two laboratories : one in the United States, and another in Russia. There have been calls for the samples to be destroyed; others stated that they should be kept for research. In 2010, WHO was expected to review the issue (Ariza, 2010).

Contemporary trends

Vaccines are generally considered as the most cost-effective means to prevent the occurrence of diseases and even in some cases to cure them (therapeutic vaccines). Massive vaccination campaigns, particularly of children, could eradicate diseases or at least drastically reduce their occurrence. Thus, in June 2009, the World Health Organization (WHO) approved the first rotavirus vaccine for global use. This vaccine, which in trials in Latin America, Europe and the United States, cut rotavirus infections by 85%, could become part of routine vaccination programmes and contribute to markedly reduce the number of deaths in children under five years due to diarrhoea (at least one-third of all diarrhoea deaths among young children are caused by the rotavirus, which infects the cells lining the small intestine and causes gastroenteritis). After the eradication of smallpox, vaccination against poliomyelitis, measles and other diseases affecting young children resulted in plummeting death rates due to these diseases.

For many years, however, the large-scale production of vaccines has not been attractive for big pharmaceutical groups. Profits were considered

marginal because prices have to be low. Consequently, basic vaccines mainly for immunizing infants and young children were manufactured by a small number of companies, causing sometimes supply difficulties.

There is now a renewed interest in vaccine production because of the prevailing policies throughout the world aiming at decreasing health-care costs (prevention versus therapy). In addition, to help developing countries carry out massive vaccination campaigns and thereby alleviate the burden of diseases among their populations, funds from charitable foundations (e.g. Bill and Melinda Gates Foundation) or international initiatives (e.g. International Finance Facility for Immunization or IFFIm) are being invested in both research-and-development projects and vaccination campaigns.

Another driving force of the vaccine-sector growth is the urgent need to struggle against emerging and re-emerging diseases (e.g. avian flu, swine flu; West Nile virus; chikungunya, Saint-Louis encephalitis, dengue, all three transmitted by the tiger mosquito – *Aedes albopictus*) by developing effective vaccines for both human beings and domestic animals – where the pathogens often multiply and mutate before contaminating human populations. That also explains the spurt of growth of the market of veterinary vaccines. It has been estimated that the growth of the vaccine market would be 10% to 15% for the three-year period 2008-2010, and global sales would rise from US$8.9 billion in 2005 to US$22.2 billion in 2009 (see Sasson, 2008).

Insurers and governments in the wealthy countries have started to pay higher prices : firms making new vaccines against pneumococcal disease or the human papilloma virus are receiving US$100 or more per dose of the relevant vaccines. Recent deals are also a good omen for the future of the vaccine market. For instance, Johnson & Johnson (J & J) has offered US$2.4 billion for most of the shares it did not yet control in Crucell, a Dutch vaccine firm. In 2009, J & J spent US$1.5 billion buying part of Ireland's Elan, which tried to develop a vaccine against Alzheimer's disease (*The Economist*, 2010 l).

Also the improvement of existing vaccines and the development of new ones are driving forces of vaccine production. For instance, Sanofi-Aventis – the second-biggest vaccine-producing company in the world – is developing an antiflu vaccine for old people and another for immunodeficient patients. New acellular vaccines are based on fragments of the killed pathogen (and not the whole cell); they are as efficient, but induce less secondary effects such as fever, rash, etc. Sanofi-Aventis has

developed and commercialized Gardasil, an anticervical cancer (caused by human papillomavirus, HPV) vaccine, while GlaxoSmithKline (GSK, the third global vaccine manufacturer) developed a similar one, marketed under the name of Cervarix. Other innovative vaccines are expected in the years to come, e.g. against dengue, malaria, new forms of tuberculosis, meningitis B (see Sasson, 2008).

Crucell, a Dutch vaccine firm, is working on a "universal" influenza vaccine, which might eliminate seasonal flu injections. At present, an annual endeavour to develop suitable vaccines starts as the most potent current strains are identified. But the production methods are cumbersome : typically, growing viruses in hundreds of millions of hens' eggs. Novartis has invested in production facilities in America that use cell cultures, rather than hens' eggs, to develop the vaccines. It has also teamed with Synthetic Genomics, a young American biotechnology firm, to develop a variety of "seed" viruses, which could provide a three-month head start in making seasonal flu vaccines. Joseph Kim of Inovio, an American start-up, stated his firm had devised a DNA-based method to produce a universal flu vaccine. Early trials funded by a grant from the American government had been promising (*The Economist*, 2010 l).

Finally, the accelerated growth of the vaccine sector responds to the vast market provided by emergent countries like Brazil, Russia, China and India, which set up massive vaccination programmes. Sanofi-Aventis' executives estimated that these countries would soon represent a market equivalent to that of the United States (see Sasson, 2008).

The following examples of new or improved vaccines illustrate the importance and relevance of vaccine production and use for reducing the burden of disease and health-care costs relating to expensive therapies. They also illustrate the contribution of research, development and innovation to developing new vaccines – a typical biotechnology product.

Antipneumococcus vaccine; an innovative funding mechanism for massive vaccination campaigns

Pneumonia and meningitis cause 800,000 to 900,000 deaths among children annually, mainly in developing countries. It has been estimated that about 700 million children are threatened by diseases caused by pneumococci worldwide. Vaccines against these diseases exist already and some of them have been recently improved in order to better combat the bacterial strains that prevail at regional level.

However, pharmaceutical companies are reluctant to manufacture these vaccines, because their cost, between US$71 and US$84 a dose in the United States, is considered too high for developing countries where the purchasing power is low. An innovative funding mechanism had therefore to be found. Under the Global Alliance for Vaccines and Immunization (GAVI), which includes UNICEF, WHO and the World Bank, five donor countries (Italy, US$635 million; United Kingdom, US$485 million; Canada, US$200 million; Russia, US$80 million; Norway, US$50 million) and the Bill and Melinda Gates Foundation (US$50 million) have pledged subsidies of US$1.5 billion to the companies that will manufacture the vaccine for developing countries (Faujas, 2009).

On 2 April 2009, the World Bank's governing board decided that the Bank will replace any defaulting donor, up to US$1.5 billion. For the first time in the World Bank's history, this amount of money will be guaranteed by the Bank's own capital. According to Philippe Le Houérou, vice-president of the World Bank in charge of global partnerships, a major difficulty had to be overcome : what should be the cost of the vaccine dose, on which the Bank had to evaluate its financial contribution? As vaccine-manufacturing companies keep secret their production costs, the Bank called on several economists, including Eric Maskin, a Nobel Laureate, in order to fix a minimal cost. P. Le Houérou stated that the Bank "proposed a price of US$3.50 per dose of vaccine during ten years, and that the companies which will accept it, will receive in addition to this guaranteed income, a subsidy of US$3.50 per dose for about 20% of all doses they will produce; this proposal aimed at guaranteeing a satisfactory amortization, with a possible revision in case of inflation." This innovative funding mechanism was to be finalized in three months, so as to allow the companies to start producing the vaccine in 2010 (Faujas, 2009).

Gradual vaccination of the 700 million children that are under the threat of pneumococcal diseases worldwide would save 900,000 lives from 2010 to 2015 and 7 million from 2010 to 2030. In addition, the funding mechanism developed for pneumococcal diseases could be extended to an antimalaria vaccine or to the development of food supplements. The World Bank is willing to fully play the role of an effective broker between public and private donors, and to ensure a synergy among them (Faujas, 2009).

HIV/AIDS

Current situation of disease control

At the 17th international AIDS conference, held in Mexico at the beginning of August 2008, Anton Pozniak, director of research on HIV at the Chelsea and Westminster hospital in London, summarized in the plenary session of the conference, on 7 August 2008, the progress made in the treatment of the disease since the 2006 conference held in Toronto : "We have now more efficient and better tolerated treatments; we see less toxicity in the short and the long term, and we have access to drugs that are easier to take, less numerous and which do not need to be stored in a refrigerator" (Benkimoun, 2008 b). See also *The Economist* (2008 b).

The improved efficiency and lesser toxicity of AIDS treatments lead the researchers to reflect on several hypotheses. Firstly, would it be feasible to change the number of compounds/molecules used, either by starting with three and phasing out some, or by starting with more than three drugs? In France, the Appolo study of the French National Agency for Research on AIDS (ANRS) aims at answering this question. Secondly, can we markedly reduce the amount of virus that persists in the body, while the number of copies of viral RNA in the blood cannot be detected (i.e. less than 50 copies per ml of blood)? Thirdly, with a broadened range of available drugs, when to start the treatment? Recommendations made in France and the United States advise to wait until immune defences are too much altered. Currently, anti-HIV treatment should start when the number of T CD4 lymphocytes (the target of the virus) is not superior to 350 per mm^3 of blood, or when their number goes under 500 per mm^3 in the cases where the number of copies of viral RNA is very high (Benkimoun, 2008 b).

The availability of several new drugs is a great hope for patients that have used other antiretroviral medicines, as well as for those who receive their first treatments (efficient and less toxic). But all the patients have not access to anti-AIDS drugs and even less to the most recent ones (Benkimoun, 2008 b).

The 18th international conference on AIDS, held in Vienna from 18 to 23 July 2010, has been attended by more than 20,000 researchers, physicians and members of associations against AIDS. This disease caused 2 million deaths a year. Over 30 million people across the world were contaminated by the HIV. The majority of these seropositive persons lived in the South Hemisphere, two-thirds of them in sub-Saharan Africa. Of the 15 million

seropositive persons needing a treatment in the countries with low or average income, around 10 million did not receive any in 2010. The universal access to treatment has become even more urgent because it is the pillar of the new strategy supported by the UNAIDS programme, that coordinates the action of various United Nations agencies engaged in the struggle against the disease; this strategy recommends the use of tritherapy as a preventive measure against the transmission of HIV (Benkimoun and Vincent, 2010).

The funds collected by the Global Fund for the Control of AIDS, Malaria and Tuberculosis – a public-private partnership that is the biggest funding mechanism for health assistance in the world – are helping countries with "low and intermediary income." In the case of the struggle against AIDS, one should add in terms of funding the contribution of the American President's Emergency Plan for AIDS Relief; under this plan, the funds, mainly public, are delivered through bilateral agreements. Since its creation in 2002, the Global Fund had devoted about US$11 billion (€8.5 billion) to the struggle against AIDS through projects carried out in 140 countries. The American President's Emergency Plan for AIDS Relief devoted US$26 billion (Benkimoun and Vincent, 2010).

This joint effort has led to a reduction in mortality, in the overall number of hospitalizations and new infections by the virus. However, the UNAIDS has estimated that the funds needed for an optimum control of the disease (universal access to prevention, to treatment and care) should amount to US$25 billion in 2010, i.e. US$11.3 billion more than the funds already available. The concern about fund availability was even greater beyond 2010, due to the budget austerity measures taken by the main donors, the industrialized countries. In the fall of 2010, the conference for the reconstitution of the Global Fund was expected to take place and the international community was to make commitments for the three-year period 2011-2013. Michel Kazatchkine, executive director of the World Fund, did not hide his concern : "The objective to collect US$17 billion for the period 2010-2013 may seem very ambitious, but it just aims to pursue the efforts made over recent years." If all the donors speak of crisis, the attitudes vary from one country to the other : "Some countries decrease all their budgets, e.g. the Netherlands or Spain. Others, like the United Kingdom, express the will to maintain the budget devoted to public aid for development," stated M. Kazatchkine (Benkimoun and Vincent, 2010).

Efforts are being made to persuade emerging countries to make significant contributions to the World Fund. For instance, China, to which the Fund had allocated since its creation more than US$1 billion, i.e. 30% of the

resources devoted to the struggle against AIDS in this country. China made a contribution of only US$16 million to the Fund. M. Kazatchkine stated : “China could either continue to receive assistance, or become part of the global governance of the Fund. We prefer the second option. If China does it, other emerging countries will follow its example, Brazil and India will do so” (Benkimoun and Vincent, 2010).

Early detection and treatment of the disease

Several studies have shown that the treatment of patients suffering from HIV/AIDS started too late. For instance, in France, despite the 5 million tests carried out in 2009, many of them revealed that the immunodeficiency syndrome was already advanced. The disease was diagnosed too late in one patient out of three. Generally the treatment should start when the number of TCD4 lymphocytes (the virus target) amounts to 500 per mm^3 (compared with 1,200 to 800 per mm^3 in a healthy individual). The World Health Organization recommends a threshold of 350 TCD4 lymphocytes per mm^3 to start the treatment, while 200 lymphocytes per mm^3 is considered a critical threshold. In France, in 2009, 50,000 persons ignored that they had been contaminated by the virus, and 47% of those who were diagnosed positively were considered as screened too late. Such late diagnosis resulted in a higher mortality rate, estimated at +10.9% (*Le Monde Magazine*, 4 December 2010, p.18).

It has been estimated that 6,400 to 6,900 individuals had been contaminated in 2009 in France; that 67% of seropositive individuals were men and that one-fourth of infected pregnant women had discovered their seropositivity during pregnancy. Regarding the mode of contamination, heterosexual intercourse represented 61% of total contaminations; homosexual relations, 38%; and the use of injectable drugs, 1% (*Le Monde Magazine*, 4 December 2010, p.18).

A systematic early diagnosis of HIV/AIDS will therefore lead to start treatment earlier, to improve its effectiveness and to sometimes restore the patient’s immune system. That is why the national plan for controlling HIV and AIDS presented by the French health ministry in October 2010 proposed a systematic detection of the virus and the disease for all adults between 15 and 70 years of age, i.e. about 40 million people. The average age for the diagnosis of HIV/AIDS was 38 years in 2009. The objective of the new plan was to broaden the detection of the pathogen beyond the usual populations at risk (homosexuals, prostitutes and users of intravenously injected drugs) and to address apparently lower-risk populations like heterosexuals (*Le Monde Magazine*, 4 December 2010, p.18).

Another preventive measure against HIV : anti-HIV gel

For the first time, an anti-HIV vaginal gel has been unveiled at the 18th international AIDS conference in Vienna (18-23 July 2010) as a possible means to inhibit the transmission of HIV. After two decades of unsuccessful efforts, testing of a dozen of microbicides, this was the first proof that a microbicide can offer significant protection against HIV. The anti-HIV vaginal gel is a microbicide cream which aims to protect a woman against HIV in semen. This new-generation gel does not contain wide-spectrum toxic products but a specific anti-HIV substance; it contains in fact a one-percent concentration of tenofovir, a frontline drug in the combination therapy to treat people already infected with the virus. Antiretrovirals work by preventing HIV from reproducing in CD4 immune cells. Previously tested gels, which had not used antiretrovirals, had negligible protection or even boosted the risk of infection.

The Centre for the AIDS Programme of Research in South Africa, CAPRISA, has carried out a trial called CAPRISA 004 since 2007, under the leadership of Salim and Quarraisha Abdool Karim, two physicians working at the centre. The trial was carried out in the State of Kwazulu Natal, where women, in majority Zulus, are seropositive in high proportion : about 10% for girls, less than 16 years old, and 50% among women, more than 24 years old. This is one of the highest prevalence rates in the world among this population category. The trial's objective was to test the effectiveness of a microbicide gel containing 1% of tenofovir among 889 sexually active, HIV-uninfected women living in urban and rural settings; they were between 18 and 40 years old and had a high risk of becoming seropositive. Of the 843 women who participated in the trial until the end of the experience, 422 used the gel with tenofovir and 421 used a placebo gel. All were given the instruction to use one dose of the gel about 12 hours before an intercourse, then another dose 12 hours after the intercourse. They were regularly monitored every month during 30 months with regard to both the proper use of the gel and the frequence of their sexual relations. The overall trial was framed by tough ethical guidelines, in which the women were regularly advised on the risks, counselled on safe sex, and given access to condoms. Women who became infected or pregnant were withdrawn from the trial. In the placebo group, 60 became infected, while the tally in the gel group was 38 (Abdool Karim et al., 2010).

Three years after the initiation of the trial, the results were published on line on Tuesday 20 July 2010 in *Science* and they were presented at the 18th international AIDS conference. They showed that the use of the gel

containing tenofovir had reduced HIV infection by 39%, compared with the use of the placebo gel, and more precisely : by 28% among those women who did not use the gel correctly, by 28% among those who used it more or less correctly, and by 54% among those who used it regularly. The gel was found to be safe, which historically has been a great worry in microbicide research. Also, there was no sign that a woman who became infected after using the gel, the HIV was resistant to tenofovir. This too was a relief, given concerns that on-again, off-again use could help HIV to become resistant to this important drug (Abdool Karim et al., 2010).

Specialists are awaiting the outcome of the third phase of the trial before claiming victory. Among the questions is how the gel was used. Effectiveness fell markedly over the course of trial, possibly because use of the microbicide declined among some women. Scientists also wanted to see whether different antiretrovirals or ways of using the gel – perhaps through a slow-release vaginal ring – could boost protection. Another avenue of exploration is whether the microbicide is effective in anal intercourse, where the statistical risk of infection is ten times higher than in vaginal intercourse.

Protection of 39% may not be high enough particularly in industrialized countries, where 80% would be a likelier benchmark. But, coupled to other prevention measures, like condoms and circumcision, it could be acceptable in countries in sub-Saharan Africa where two-thirds of infections occur. In South Africa alone, a gel with such effectiveness would save 1.3 million new infections and avert 800,000 deaths over 20 years, the researchers estimated.

The anti-HIV gel was not available in 2010. The results obtained about its effectiveness were only for a second stage in a long three-phase process in which new drugs are vetted for safety and efficacy. Third phase takes two or three years. Health regulators then screen the data before deciding whether to license a product. Even at the stage reached in 2010, some veterans compared the news to the advent in 1996 of the antiviral drug mixture and the 2006 discovery that circumcision more than halved the infection risk for men.

If the results of the CAPRISA 004 trial were confirmed and even improved, “it would be the first time when a prevention tool would become available to women, who could manage it by themselves; this would be very important in developing countries,” stated Jean-François Delfraissy, director-general of the French National Agency for Research on AIDS (ANRS), who recalled that until now the recommended prevention

methods (condoms and sexual behaviour) were mostly "in the hands of men." According to the executives of CAPRISA, the anti-HIV gel "would play a key role in the prevention of the infection by HIV, especially for women who are exposed to coercive sex and are unable to negotiate a mutual monogamy of the use of condom." "Women represent the majority of new infections by HIV in the world, and consequently the good results of the CAPRISA trial is an important step towards offering a prevention tool, innocuous and effective, to a population at risk," stated Anthony Fauci, director of the US National Institute of Allergies and Infectious Diseases (NIAID). The cost is also very low : 8 to 15 cents of euro per application (Vincent, 2010 f).

Will the World Health Organization and UNAIDS decide to accelerate phase-3 trials of the anti-HIV gel, due to the urgent situation prevailing in Africa? A group of experts was expected to meet in South Africa during August 2010 in order to advise both international bodies. The need to speed up the trials to be carried out on several thousands of women with a view to confirming the effectiveness of the gel, was underlined by the fact that about 60% of infected persons in Africa were women, and the youngest among them, in some countries, ran three times the risk to become seropositive than men. In the majority of cases they became infected during an intercourse with a man already seropositive (Vincent, 2010 f).

Strategy for vaccine development

In September 2007, a large-scale trial of a vaccine against HIV/AIDS carried out by Merck (known as STEP) was interrupted because of its inefficiency and of the larger number of infections among the vaccinated volunteers than among those who received a placebo. It is true that it is not easy to develop a vaccine against a virus that mutates very frequently and thus escapes the defences of the immune system. The vaccine consisted of designing an adenovirus that contained HIV genes, such as the *GAG* gene. When the vaccine was administered to mice, it triggered a response of T-cells. But in humans the protection by the vaccine was insufficient. Perhaps, the best epitopes had not been selected; maybe the solution would have been to design antibodies not so much against an HIV protein, but against the site of adhesion of the virus on a human cell.

New candidate vaccines must be found. The not-for-profit association, International AIDS Vaccine Initiative (IAVI), has presented its 2008 programme, where it indicated a road map towards an effective vaccine. Alan Bernstein, executive director of the Global HIV Vaccine Enterprise, which involves researchers and donors, emphasized that

"the development of a vaccine against HIV/AIDS is a long-duration task; failure is part of scientific research and many lessons should be drawn from the interruption of the STEP trial." Peter Piot, the executive director of UNAIDS at that time, considered that "it would be silly to stop such kind of research, where there are 7,500 newly infected persons every day in the world" (Benkimoun, 2008 a).

Scientists indeed are not surprised by the slow speed of research in this area. "Twenty years elapsed and huge investments had been made before therapeutic applications were made with cytokines (intercellular messengers synthesized by immune-system cells), and after resounding failures," recalled Jean-François Delfraissy, director-general of the French National Agency for Research on AIDS (ANRS). The IAVI 2008 programme recommended to reallocate the resources devoted to most least promising candidate vaccines to the solution of crucial scientific issues. The second strategic approach was to lay emphasis on unravelling the means to induce cell immunity against HIV (cell immunity, induced by T-lymphocytes, acts against bacterial and viral pathogens). Seth Berkley, president of IAVI, expressed his optimism, because "we know that we can protect animals against HIV/AIDS and we also know that humans can produce effective antibodies. We must therefore ensure the sustainable funding of research" (Benkimoun, 2008 a).

Fighting AIDS needs patience and a sustainable R&D effort. Not only because of the very high genetic variability of the virus, but also because there are several strains that infect people. In addition to the three known strains of HIV (the widespread HIV-1 M strain, and the much rarer N and O strains) which have been linked to chimpanzees, a new variant – discovered in August 2009 in a 62-year-old Cameroonian woman who tested positive in 2004 – appears to have come from gorillas. Researchers stated that the new strain (P) might be difficult to detect using conventional tests, but expected current treatments to remain effective (Plantier et al., 2009; *Time*, 17 August 2009, p.6).

The results of the vaccine trial against HIV, carried out in Thailand, were presented at the largest world scientific conference on anti-HIV/AIDS vaccines (AIDS Vaccine 2009, Paris, 19-22 October 2009). The trial, called RV144, was set up by the Thai ministry of public health, the US National Institutes of Health (NIH) and the US Army Walter Reed research centre. It started in 2003 and involved more than 16,000 seronegative men and women, between 18 and 30 years of age. The aim of the trial was to prove the validity of the concept that vaccination could prevent HIV infection and decrease the number of virus particles in the bloodstream of those

who would have been infected after being involved in the trial. With respect to this second aspect, the results did not show any beneficial effect (Benkimoun, 2009 g, j).

The volunteers received in a random way either the vaccine or an inactive substance. They were checked over a three-year period, and all those who had been infected were taken care of. Two vaccines have been combined : the ALVAC-HIV, manufactured by Sanofi Pasteur, injected four times over a period of 24 weeks in order to trigger an immune response, and the AIDSVAX B/E of VaxGen, injected twice at the 12th and 24th week of the trial, in order to strengthen the response. According to Nelson Michael, of the Walter Reed research centre, the ALVAC-HIV, when used alone, could trigger an immune response in only 20% of the vaccinees. In the same conditions, the vaccine AIDS VAX B/E triggered an immune response in 90% of the vaccinees, but without any protection effect. His conclusion was that "the protection effect was modest, but significant" (Benkimoun, 2009 g, j).

Lengthy debates had been devoted to the method of analysis of the data : taking account of the results concerning all the volunteers who received their first injection, or analysis of only the results concerning seronegative persons, in accordance with the protocol. Seven persons happened to be infected when they were involved in the trial and therefore they were not included in the second approach to data analysis. The latter was considered more in conformity with the conditions of real life by N. Michael, and it showed that vaccine efficiency was 31.2% : 51 persons infected among the vaccinees and 74 among those who received the placebo. With the first method of data analysis, the calculated vaccine efficiency amounted to 26.4% (Benkimoun, 2009 j). Yves Lévy of the department of clinical immunology, Henri-Mondor hospital, Créteil, south of Paris, commented that "efficiency was rather high during the first year, about 60%, and thereafter it decreased down to 30% over two years; this meant that vaccination had induced an immune response." Consequently, the RV144 trial was considered encouraging and also a starting point. Alan Bernstein, executive director of the Global HIV Vaccine Enterprise, stated that "we must try to understand the relative efficiency of the combination of two vaccines, as well as the type of induced immune response." Jean-François Delfraissy considered that the results of the Thai vaccine were promising, even though we were still far from having an efficient vaccine against HIV/AIDS. He quoted a trial carried out by ANRS that targeted dendritic cells, whose role is to present the antigen to the immune system. In this way, it might be possible to use the immune system in order to modulate its response ("biovaccine" approach) [Benkimoun, 2009 j].

Basic research should be pursued on the elucidation of the reasons for the partial protection achieved in the Thai trial, i.e. which are the immunological factors involved in this kind of protection. New strategies should be explored before having an effective anti-HIV/AIDS vaccine. All these aspects, as well as the detailed results of the RV144 trial, were discussed at the conference AIDS Vaccine 2009 (Benkimoun, 2009 g).

On the other hand, a paper by Laura Walker of the Scripps Research Institute, La Jolla, San Diego, California, and Sanjay Phogat of the International AIDS Vaccine Initiative (IAVI), in New York, published in *Science*, early September 2009, brought some hope in the struggle against AIDS/HIV. The paper describes broadly neutralizing antibodies (known as bNAbs), that can deactivate a wide range of HIV strains – which is particularly important for an effective vaccine, because HIV is so variable. Until now, those bNAbs which have been identified have been effective only against strains circulating outside Africa. Moreover, no new bNAb has been found for more than a decade (Walker et al., 2009).

The two new bNAbs reported by L. Walker and S. Phogat were the first results of a project called Protocol G, one of the several multicentre investigations into various aspects of AIDS that IAVI is supporting. This particular project is looking specifically for bNAbs in volunteers from seven African countries (Côte d'Ivoire, Kenya, Nigeria, Rwanda, South Africa, Uganda and Zambia) and from America, Australia, United Kingdom and Thailand. The antibodies were found by screening blood serum from 1,800 volunteers from these countries. The researchers focused their work on antibodies that reacted with part of the virus called its spike. This consists of two glycoproteins known as gp 120 and gp 41 that react with a receptor on the surface of CD4 cells of the immune system. The reaction allows the virus to enter the cell and explains why HIV infects immune-system cells rather than other kinds of cells (Walker et al., 2009).

With the aid of two biotechnology firms, Monogram Biosciences of South San Francisco and Theraclone Sciences of Seattle, L. Walker, S. Phogat and their colleagues tested all their serum samples for anti-HIV activity, picked the top 10%, extracted all the antibodies from each of these samples, tracked down the cells that made each antibody, identified the relevant genes, cloned them into faster-reproducing cells, made usable samples of pure antibody from each of these clones, and tested the results to see if they could stop HIV from entering CD4 cells. Two such antibodies, both from the same donor and dubbed PG9 and PG16, could (Walker et al., 2009).

Further research steps were to find out exactly which parts of the virus spike PG9 and PG16 lock onto. This knowledge is necessary to designing a chemical that resembles the relevant spike element to prompt the immune system to make antibodies that will neutralize it. That chemical might be the basis of a vaccine. Otherwise, it is hoped that Protocol G would help identify more bNAbs that could better neutralize the HIV (*The Economist*, 2009 i).

Vaccine against Ebola virus

Researchers of the French Research for Development Institute (IRD), in collaboration with scientists of the National Museum of Natural History (Paris), the health ministry of the Democratic Republic of the Congo (DRC), the National Biomedical Research Institute of Kinshasa and World Health Organization (WHO) have elucidated the source of the epidemic due to the Ebola virus that occurred in DRC from May to November 2007. Enquiries carried out by the Congolese and international teams had identified the person considered at the origin of the epidemic : a 50-year-old woman living in Ndongo, one of the ten villages of the town of Kampungu, located along the road linking Mweka to Luebo in the province of Western Kasai, in the centre of the country. Kampungu was the epicentre of the epidemic. Around 25 June 2007, this woman showed the symptoms of Ebola haemorrhagic fever (high fever, nausea, diarrhoea and haemorrhage) and died a week later. The persons who took care of her also died a few days later from a haemorrhagic fever. That was the start of the epidemic (Leroy et al., 2009 b).

How this woman had been infected when she had no contact with an ill person or a dead animal? Researchers extended their inquiries in the various villages of Kampungu in order to find out how the woman was infected. They discovered that at the end of April 2007, a villager living in another settlement used to go regularly to the weekly market of Mombo Munene in order to buy fresh fruit bats. Each year, during their large-scale migration, bats are caught and sold on the local markets; they are an important source of food for people. Further to frequent contacts with the blood of purchased bats, that man probably showed light symptoms of the disease, which was not therefore noticed. Most likely he was contaminated by a small number of virions, and that was the case of many people attending the same markets. But that man had a four-year-old daughter whom he systematically brought with him during the month of May 2007, as well as his wife, to the village located in the bush and reached after a three-to-four-hours walk. In fact, each village of the Kampungu area has two components : one which is located along the

main road and a "twin" one located in the bush. During the 1960s and 1970s, DRC's authorities requested the inhabitants of isolated villages to come close to the main roads in order to facilitate their access to medical centres, education and administrative services. Nowadays, the village located in the bush or forest supplies the markets of its "twin" village, thus providing agricultural products or hunted animals, e.g. bats (Leroy et al., 2009 b).

The daughter was probably infected by her father during the long journeys. She fell ill and passed away rapidly. Following the local tradition, the corpse of the child was washed before being buried by a close friend, the woman of Ndongo. No other person in the neighbourhood of the child had been contaminated. As the child could not likely develop a high viremia in a short time, only a prolonged contact with her body could lead to contamination. The conclusion was that the father and his daughter, and thereafter the woman of Ndongo who washed the child's body, had been contaminated by the Ebola virus from bats (Leroy et al., 2009 b).

There is a very strong correlation in space and time between the Ebola epidemic that occurred from May to November 2007 and the annual migration of bats, although there is no formal evidence of virus transmission from bats to humans. According to the villagers, bat migration has been massive during the spring of 2007. Each year, during the month of April, tens of thousands of bats used to arrive and settle for some time in the islands located in the Lulua River, near the villages of Kampungu. Two of the three species of migrating bats, identified in the area (*Hypsignathus monstrosus* and *Epomops franqueti*), are among those considered as the natural reservoir of Ebola virus. The latter has been found in the saliva and sweat of the animals. Other wild animals, such as gorillas and chimpanzees, that can be infected and killed by the virus, are not present in the region of Kampungu (Leroy et al., 2009 b).

Contrary to what was thought a few years ago, human beings could become infected by Ebola virus through direct contact with the animal reservoir : bats. For the first time, IRD scientists and their colleagues have established a direct link between Chiropterans and an Ebola epidemic. This discovery would lead to appropriate preventive measures, particularly in the villages located along the migration route of Chiropterans (Leroy et al., 2009 b).

The etiology of Ebola disease is now better known, as it has been demonstrated that the contamination of humans is not only through person-to-person contact, but also through contacts with animals (e.g.

gorillas and chimpanzees) and with natural dead reservoir animals such as Chiropterans. Preventive measures can therefore be taken and appropriate warnings can be made. See Sasson (2008).

There is not yet an effective vaccine that could control the outbreaks of lethal Ebola haemorrhagic fever which occur sporadically in Central Africa. However, an experimental vaccine has been tested on chimpanzees and it was found effective against the two main strains of Ebola virus, identified in 1976 and in 2007. It is also crucial to elucidate the process which makes the experimental vaccine effective against the new *Bundibugyo ebolavirus*. Once this is done, Nancy Sullivan, of the US National Institute of Allergy and Infectious Diseases and the main author of the research work on the candidate vaccine, stated that "a single vaccine that protects against all the virus strains could be developed" (Sullivan et al., 2010).

Dengue and chikungunya : a global threat

Dengue

During the last 20 years, both chikungunya and dengue have caused serious epidemics in several tropical countries. Their viruses have been isolated for the first time during the 1950s, in Southern Africa for the chikungunya virus and in South-East Asia for the dengue. Chikungunya means the "disease of the bent man" in makondé language, which is spoken in Southern Africa where the pathogen has been isolated in 1953; the serious forms of the disease consist of very invalidating stiffness of joints. Up to now, there are no vaccines nor effective treatment against both diseases, which constitute a global threat.

The main vectors of the viruses are mosquitoes of the genus *Aedes*, generally *Aedes aegypti*. But another species, *Aedes albopictus*, has been associated with the epidemics. The so-called "tiger mosquito" is expanding rapidly, as it lays its eggs in any place which contains water. Eggs are spread thanks to human activities and trade exchanges. While the chikungunya virus has caused many epidemics in Africa and South-East Asia during the 20th century, *A. albopictus* is now present in all continents, particularly in the islands of the Indian Ocean, Central Africa and the south of Europe. The epidemic outbreak in Italy in 2007 as well as the first case of chikungunya detected in the south of France during the summer of 2010 underline the potential of global dissemination of the virus.

Dengue symptoms are generally : high fever, headache, sore eyes, nausea and muscular and joint pain. At this stage, it is not considered a real danger for human health, but it should be treated. It is nevertheless an economic and social issue, because it causes a high level of absenteeism at school and in the workplace. The real danger is that of haemorrhagic dengue, that generally affects 10% of the population suffering or having suffered from the disease. In the case of haemorrhagic dengue, after the initial symptoms, haemorrhagic spots appear on the skin and mucosa; thereafter, if not treated, the patient can suffer from the dengue shock syndrome – water accumulates in the respiratory tract, lungs are affected and death could occur (Escobar Gutiérrez and Fonseca Coronado, 2009).

Dengue reached the American continent during the 17th or 18th century, probably through contaminated slaves from Africa. The disease is considered endemic to South-East Asia. The first description of the disease in the United States had been made in 1789 by Benjamin Rush, but the name of the disease was given in 1827-1828, when there had been an epidemic outbreak in the Caribbean. The symptomatology consisting of fever, exanthema and muscular pain, was given the name of *dinga* or *dyenga* by African slaves, from the Swahili phrase *ki denga pepo*, which can be translated as "sudden attack caused by a malign spirit." Dengue began to be perceived as a public health problem during the second world war, when Japanese troops invaded a large part of East and South Asia, as well as the Pacific islands, and disseminated the pathogen. In 1943, Japanese researchers, Ren Kimura and Sumuso Hotta, discovered that the pathogen was a virus (Escobar Gutiérrez and Fonseca Coronado, 2009).

In 1945, it was discovered that the virus had several serotypes, each one having its particular antigens which induce specific immune responses that protect only against the relevant virus. First, Albert Bruce Sabin (who developed the oral vaccine against poliomyelitis several years later) and Walter Schlesinger isolated the dengue virus strains Hawaii and New Guinea, DEN-1 and DEN-2, respectively. Then, in 1957, during the first global epidemic of haemorrhagic dengue, Hill Hammon and co-workers isolated serotypes, DEN-3 (strain H87) and DEN-4 in the Philippines (Escobar Gutiérrez and Fonseca Coronado, 2009).

It is estimated that every year there are between 50 and 100 million cases of dengue throughout the world, but 2.5 billion people are at risk. In Mexico, for instance, dengue and its vector are distributed in over half of the national territory; in 2008, 32 million cases of dengue fever and 6,000 of haemorrhagic dengue fever were reported (Escobar Gutiérrez and Fonseca Coronado, 2009). See also Kyle and Harris (2008).

The *Aedes* mosquitoes are widespread between latitudes 40°N and 40°S, and under 1,500 meters of altitude. They tend to spread beyond these limits. They are susceptible to extreme temperatures and dry climates; adults lose their activity under 12-14°C or in a very dry atmosphere. Only the female mosquito can transmit the virus because it has to feed on animal blood whose proteins are indispensable to the development of its eggs. When the female mosquitoes bite a dengue-affected patient in the stage of viremia, which starts one day before the onset of fever and extends until the sixth or eight day of the disease, they become infected and the virus multiplies in the intestinal epithelium, nerve ganglia, fat tissue and salivary glands. After seven to 14 days (incubation time), the female mosquito can infect other human beings. Each female lays about 140 eggs and it can do that twice or more times during its lifespan. Eggs can withstand desiccation during a year and hatch after one or four wet days (Escobar Gutiérrez and Fonseca Coronado, 2009).

Dengue virus transmission is only indirect through the insect vectors, there is no transmission through direct contact with an infected person, nor through contact with water or food. Protection against the vector is done with nets, repellents and insecticides. The use of pesticides in permanent water reservoirs should take into account their impact on wildlife and crops and be guided by entomological monitoring of the distribution and prevalence of the vectors. It is also necessary to drain swampy areas, cover water pipes or sewers around houses and protect water storage areas or recipients (Escobar Gutiérrez and Fonseca Coronado, 2009).

Once the virus is deposited on the skin of a non-infected person, dendritic cells "recognize" the viral envelope protein (protein E) which sticks to a cell receptor. Then endocytosis takes place and the virus is enclosed in an intracellular vesicle. The membranes of the virus and vesicle fuse, the virus capsid is dismantled and the virus single-stranded RNA can enter the host cell and start its replication to give rise to the three structural proteins of the virions and seven non-structural proteins (NS) that are involved in the replication process inside the host cell. New RNA strands are synthesized, which associate themselves with structured proteins on the internal cell membranes to form new virions which leave the host cell (Escobar Gutiérrez and Fonseca Coronado, 2009).

At the Fundación Instituto Leloir (Buenos Aires, Argentina), the laboratory of molecular virology is renown worldwide for its work on the molecular biology of the dengue virus. The mechanism of replication of the virus has been described there. Argentine researchers are focusing their work on the possible development of a vaccine against the dengue virus. To

that end, they are pursuing the study of the molecular biology of the virus, they are identifying targets for antiviral drugs and designing trials for the high-throughput screening of these antiviral compounds. A genetically modified virus has been developed in order to carry out these trials rapidly; the genetically modified virus has been transferred to public and private research laboratories, through material transfer agreements, without any patent protection or request.

Dendritic cells where the replication of the virus is taking place circulate in the bloodstream, where virions are discharged (viremia stage). The new virions can infect monocytes and T and B lymphocytes, as well as cells of the vascular endothelium. When they are infected, cells immediately respond by producing cytokines (interleukins 1 and 6, tumour necrosis factor – TNF, and interferons) that trigger dengue symptoms. When dendritic cells reach lymph nodes, they induce immune responses that are specific to the infectious serotype. In so far as the infection is controlled, a long-lasting memory, also specific, is set up and prevents the reinfection with the same serotype, but not with the other three serotypes. This means that dengue can affect several times the same person, but not with the same serotype. Adaptive protection mechanisms are well documented and they are mediated by cellular immune response (CD8+T cells which recognize and kill infected cells, and CD4+T cells which cooperate with B cells to form antibodies) as well as by humoural immune response (neutralizing antibodies IgM and IgG against E protein of the virus envelope). It is always IgM that is produced first when the infection starts, and this fact is of great utility for the serological diagnosis of the disease (Escobar Gutiérrez and Fonseca Coronado, 2009).

With respect to haemorrhagic dengue, Scott Halstead and co-workers observed till the 1970s that this serious form of the disease was much more frequent among persons with secondary infections or breastfed infants having antibodies against dengue virus of maternal origin. According to the antibody-dependent enhancement (ADE) theory, IgG subneutralizing antibodies, induced by a first infection, do not protect against another infectious serotype, but they can form virus-antibody complexes that can stick to receptors for the Fc of immunoglobulin at the surface of monocytes and macrophages, which increases the likelihood that more active viruses penetrate into more cells. However, there are also reports on undisputable cases of haemorrhagic dengue primary infections. In order to find an explanation, comparative studies have been made of the analysis of the genomes of viruses isolated from haemorrhagic cases and conventional ones, in order to identify "pro-haemorrhagic" genotypes within each virus serotype. For the time being,

there are many epidemiological and experimental evidences to support one hypothesis or the other, and it would not be surprising that in natural conditions both may occur (Escobar Gutiérrez and Fonseca Coronado, 2009). See also Halstead (2008).

Regarding the diagnosis of the dengue, a first method consists of identifying the non-structural protein NS1 of the virus circulating in the bloodstream, using an enzyme-linked immunosorbent assay (ELISA); since 2007, it has been the preferred method of virus detection, but with the shortcoming of not being able to recognize the virus serotype. The second method consists of isolating and cultivating the virus, generally on the cell line C6/36 (from mosquitoes), then identifying the expression of E protein at the cell surface using fluorescent monoclonal antibodies, which are specific to each of the four serotypes. This is the best standard for the diagnosis and serotype identification of dengue infections, but it requires a week for obtaining the final results, special equipment and highly trained staff. The retrotranscription-polymerase chain reaction (RT-PCR) is faster : it allows for the retranscription of the virus RNA into DNA, which is amplified millions of times by the DNA-polymerase, using as initiators regions of the virus genome which are distinct in each serotype; these differences can be visualized through electrophoresis, so that the virus serotype can be identified in less than 24 hours. This technique is the one recommended for the cases of haemorrhagic dengue; it needs very modern equipment and highly trained staff, which explains why it is not within the reach of any country. For the time being therefore, the ELISA technique is the most widely used, because it is efficient, cheap, easy to perform and relatively fast. It is very useful during epidemics and for the seroepidemiological vigilance, although it cannot detect the circulating serotypes (Escobar Gutiérrez and Fonseca Coronado, 2009).

There are growing concerns about a possible pandemic of dengue that would have disastrous consequences from the social, economic and political viewpoints. It is worth mentioning the epidemic of dengue that occurred in Cuba in 1981, where among a total population of 10 million inhabitants, 345,000 cases had been diagnosed in four months, and 10,300 patients were considered seriously ill and 158 deaths were recorded; the whole cost of the epidemic was estimated at US$103 million (Kouri et al., 1989). Currently, major concerns related to the increasing simultaneous circulation of the four serotypes of the dengue virus, as well as to unprecedented increase in the number of cases. In 2008, in the Americas, were reported 900,000 cases of conventional dengue and over 25,000 of haemorrhagic cases (see Kyle and Harris, 2008; Gómez-Dantés and Willoquet, 2009).

Transmission of dengue and chikungunya

The presence of both *Aedes aegypti* and *Aedes albopictus* in tropical and subtropical regions where dengue epidemics occur is also a matter of concern, as both vectors transmit the virus. For instance, researchers of the French Research for Development Institute (IRD) as well as those of the International Medical Research Centre of Franceville (CIRMF, Gabon), the Health Sciences University of Libreville (Gabon) and the Faculty of Medicine at La Timone in Marseille, have found *Aedes albopictus* in Gabon, in urban zones around houses, where *A. aegypti* was already present. They also detected both viruses of chikungunya and dengue serotype 2, when a double epidemic occurred in the capital between March and August 2007. The outbreak started in the capital in March 2007 and reached the border with Cameroon, in the North, at the beginning of July. A total of 20,000 cases were suspected, the peak of the number of patients having been reached in May 2007 (Leroy et al., 2009 a).

Researchers captured several thousands of mosquitoes belonging to the genera *Aedes*, *Culex*, *Anopheles* and *Mansonia* around the houses where cases of dengue and/or chikungunya had been detected. They constituted 20 homogeneous groups of mosquitoes, according to the species and the place of their collection. Seven groups of *A. albopictus* reacted positively to chikungunya and three to dengue. The other mosquito species reacted negatively to both viruses. These results implied, for the first time, that *A. albopictus* could transmit simultaneously both dengue and chikungunya (Leroy et al., 2009 a).

During the epidemic, the researchers also took blood samples from the patients – a total of 800. Some 35% of the patients reacted positively to chikungunya and 7% of them to dengue. Eight patients were infected by both viruses. This research work is the first to demonstrate the presence of these viruses in Gabon. In addition, it was the first simultaneous epidemic of both diseases on the African continent. The researchers discovered that some patients had been infected simultaneously by both viruses. It was the first time that cases of co-infection had been observed (Leroy et al., 2009 a).

IRD and CIRMF researchers have isolated the strains of both viruses and deciphered their entire genomes. There are four evolutionary lines of the chikungunya virus : Asia, West, Central and South-East Africa. Strain Gabon 2007 belongs to the evolutionary line of Central Africa, and is close to the strains isolated during the second half of the 2005-2006 epidemic that occurred in La Réunion island. It has a genetic mutation

(A226V). The latter is found only in the strains isolated in the Indian Ocean islands (Réunion 2006, Mauritius 2006 and Madagascar 2007) and in Italy (2007). These epidemics were mainly caused by viruses transmitted by *A. albopictus*, while former ones were due to *A. aegypti*. Mutation A226V seems to be characteristic of the virus strains transmitted by the "tiger mosquito." The appearance of the same mutation in different regions of the world suggests an adaptation to the new vector. *A. albopictus* probably exerts a positive selection pressure on the chikungunya virus. This phenomenon of evolutionary convergence is extremely rare in nature (Leroy et al., 2009 a).

Another surprising fact is that strain Gabon 2007 of dengue virus serotype 2 is genetically distinct from other strains isolated in Africa, which excludes the hypothesis of a close common ancestor. By contrast, strain Gabon 2007 belongs to a genomic group that does not include Asian and Australian strains. The most likely current hypothesis is the import – old or recent – of an Asian strain of dengue virus serotype 2 (Leroy et al., 2009 a).

While shedding a new light on the dynamics of transmission of dengue and chikungunya, research work carried out by the French scientists and their African colleagues could lead to new approaches to controlling the populations of the vectors of both viruses, which is currently the only tool for preventing the diseases (Leroy et al., 2009 a). A vaccine against dengue remains to be developed, as well as against chikungunya. See Whitehead et al. (2007).

Vaccines against cervix cancer

Cervix cancer causes the death of 260,000 women annually according to the World Health Organization, and about 500,000 new cases of cervix cancer are registered every year, 90% of them in developing countries (Santi, 2010).

In France, 940 deaths due to cervix cancer have been recorded in 2009, and this cancer is placed at the eighth rank of prevailing cancers and at the 13th rank as the cause of death due to cancer among women, according to the National Health Monitoring Institute (INVS). In 2010, 2,820 new cases were recorded; between 1980 and 2005, there has been a 2.9% decrease of the number of cases (Santi, 2010). Two vaccines against cervix cancer are available. Gardasil, produced by Sanofi Pasteur MSD, is a tetravalent vaccine which protects against human papillomavirus (HPV) strains 6, 11, 16 and 18, and against genital warts. Cervarix, produced by

GlaxoSmithKline, is a bivalent vaccine that protects against HPV strains 16 and 18. In 2010, their cost was €135.59 and €111.82 per injection of Gardasil and Cervarix, respectively; three injections are necessary for the immunization of girls (Santi, 2010).

In France, vaccination against HPV is not compulsory. However the health ministry has launched a vast campaign of prevention in June 2010. Vaccination protects against some strains of the human papillomavirus (HPV) that cause 70% of cervix cancer. Most often these viruses are eliminated by the organism, but in certain persons they can cause precancerous lesions, and the evolution into an invasive cervix cancer can take between 15 and 20 years (average) [Santi, 2010].

The two available vaccines – Gardasil, recommended since 2006 by the High Council of Public Health in France, and Cervarix – should be administered before the first intercourse in order to be effective. Health authorities recommend therefore the age of 14 years for the injection, but also target those women between 15 and 23 years who had no intercourse, or at the latest during the year which follows the start of their sexual life. "The objective is to vaccinate *before* the contamination through intercourse, and mothers should explain the usefulness of the vaccination and the necessity to regularly monitor the eventual appearance of the disease," stated Pierre Bégué, coordinator of the vaccination group of the French National Academy of Medicine. In 2008, 38% of girls between 14 and 17 years had started their vaccination (at least one dose of the vaccine was reimbursed by the social security) and 23% were vaccinated, according to a study by the National Health Monitoring Institute (Santi).

The risk is that vaccinated girls stop making the smear checks (cytological examination of the cervix mucosa), and considered they were completely protected. In 2010, only 60% of women between 25 and 65 years old, most often from privileged social classes, made these checks regularly. Henceforth the health ministry wanted to reach women that are more vulnerable socially. The vaccine is reimbursed by the social security at the level of 65%. Its effectiveness is demonstrated according to many physicians, but there is no complete consensus. Marc-Alain Rozan, president of the National Trade Union of French Gynaecologists Obstetricians (SYNGOF) thinks that girls "are wrong to raise the issue. The vaccination should take place early. We had requested the government to do it at the same time as the boost injections for tetanus and polio." By the end of 2007, the French Academy of Medicine had proposed to do the vaccination at the age of 11-12 years. But others are not so sure. Claude Béraud, former vice-president of the transparency committee of

the French Agency for the Sanitary Safety of Health Products (AFSSAPS), stated : "We are almost certain that this vaccine does not present any risk, by I remain sceptical about its usefulness. Many girls are going to think, wrongly, that they may have a sexual life without constraint." "The interest of the vaccine is much less than it is believed, and it is very low, or even almost nil, if the girl has had intercourse," estimates Jean-Pierre Spinosa, a gynaecologist, co-author with Catherine Riva of the book entitled *La piqûre de trop* ("The unnecessary injection"), published in 2010 (Santi, 2010).

What to do therefore? "The information on the vaccine is systematically given in courses of sexual education. It is recommended, but one refrains from making propaganda for the manufacturing pharmaceutical group, it is up to the physicians and parents to make the decision," stressed Sandie Cariat, a nurse in a high school and member of the national bureau of the Trade Union of Nurses. Another paediatrician has suggested that it was preferable to administer the vaccination when the girls have not yet had intercourse, i.e. when they are 11-12 years old (Santi, 2010).

New clues for developing vaccines against oncogenic retroviruses

Oncogenic retroviruses that are involved in the development of tumours have, like all retroviruses, a surface protein that has extraordinary physico-chemical properties. It plays a key role in the fusion of the virus envelope with host-cell membrane, which enables the retrovirus to penetrate into the target cell and to infect it. In addition, as shown by the work carried out by French researchers at the Institut Gustave Roussy (IGR, south of Paris) and published on 23 February 2010 in the *Proceedings of the National Academy of Sciences (PNAS USA)*, the envelope protein of the virus can suppress the host-cell immune system, so that the virus can easily multiply in the cell and in the organism affected. In order to demonstrate this new property, the French scientists have worked with a very simple retrovirus, the murine leukaemia virus (MLV), which causes leukaemias in mice. They identified the regions of the envelope protein that most likely had the capacity to suppress the host's immunity. This can be done through bioinformatics and comparing the MLV immunity-suppressing domains with those identified in other viruses. Then the MLV was genetically modified so that its membrane protein kept its fusion property with the host-cell membrane, but lost its immunosuppressive function, as stated by Thierry Heidmann, director of the unit for endogenous retroviruses (a unit linking IGR, the National Scientific Research Centre – CNRS, and the University of Paris XI), and the main author of the study published in *PNAS USA* (Schlecht et al., 2010).

The normal and mutant strains of the MLV were first inoculated to mice that were deprived of their immune system; both were invaded by the virus, which meant that the envelope of the mutant virus, although it had been modified, functioned correctly, enabling the virus to penetrate into the host cells and to infect them. But when both versions of the virus were injected to healthy mice, their respective effects were completely distinct. The normal strain of MLV infects its host and stays in. By contrast, the mutant strain could not multiply in the mouse, which meant that the virus, when it was deprived of its immunosuppressive function, was quickly inhibited by the host's immune system (Schlecht et al., 2010).

The French researchers also discovered that when the mutant virus had been inoculated to a mouse, the latter became "vaccinated" against the virus. T. Heidmann has stated that this vaccination was very effective, because the immune response was more than ten times higher when these mutated viral proteins were used instead of the non-modified ones (Schlecht et al., 2010).

Two other retroviruses that are involved in human cancers, the human T- cell leukaemia virus, HTLV, and the xenotropic MLV-related virus, XMRV, were found to have the same behaviour as the MLV. They both have on their surface immunosuppressive proteins. HTLV is endemic in Japan and the Caribbean, and causes in a small proportion of the population fulminant leukaemias, several years after the initial infection. XMRV was identified in 2006 only and is present in about 25% of prostatic tumours. It is undoubtedly involved in oncogenesis, but its role in triggering cancers has not been yet clearly established (Foucart, 2010).

This kind of research may pave the ground for revolutionary vaccination methods, including against HIV/AIDS. T. Heidmann and his colleagues are working on HIV to eventually elucidate the presence of immunosuppressive proteins on its envelope.

Influenza viruses

Types and epidemiology

Influenza or flu is a viral infection of the respiratory tract, caused by an RNA virus, of which there are three types, A, B and C. The A virus infects birds, mammals and humans, while types B and C cause the disease only in humans. Type A is the only one that causes pandemics, i.e. disease in more than two continents. There are some theories that refer to an interpandemic period of 35 to 40 years. The transmission of the influenza virus is aerial and through direct contact.

Three influenza types can therefore affect the human beings :
- bird or avian influenza, caused by type A virus, which infects wild and domesticated birds; it can cause disease in humans, especially among those who are in close contact with animals;
- seasonal flu, caused by types A, B and C, generally during the winter months, resulting in about 500 million sick people worldwide and some 500,000 deaths; seasonal flu is particularly severe among old people (3 to 5 million severe cases worldwide);
- pandemic influenza, caused by a new virus subtype that can induce a serious disease, in more than two continents.

The influenza viruses belong to the family of Orthomyxoviruses and have a diameter of 80 to 120 nanometers. Over recent years, there has been a global endeavour to isolate and sequence the genome of type A influenza viruses. As a result, more than 46 million sequences have been stored at the National Center for Biotechnology Information (NCBI) Resources for Influenza Viruses of the US National Institutes of Health. Until 25 May 2009, the NCBI database contained the genome sequences of more than 220 strains of human A influenza virus, of swine origin, isolated in many parts of the world. Sequences can be compared thanks to the NCBI Basic Local Alignment Research Tool (BLAST) – alignment of nucleotides and comparison of the sequences in order to find out the relationship. About 98% of the sequences of type A viruses stored in the NCBI database have at least 99% of nucleotide similarity among them, and 95% of them have a relative that had been initially isolated during the two years preceding its first appearance (Trifonov *et al.*, 2009).

The genome of the virus is made of eight RNA segments, that code for the following proteins : polymerases PB1 and PB2, polymerase PA, haemagglutinin, nucleoprotein, neuraminidase, matrix protein and non-structural proteins. On the virus envelope or capsid are found the haemagglutinin (HA) and neuraminidase receptors. The A type virus is subdivided into subtypes, that correspond to the surface receptors, e.g. H1N1, H3N2, H7N7, etc. HA is a polypeptide trimer, with a cylindrical form, 14-nanometer long and with a diameter of 4 nanometers, a molecular weight of 224,640. NA is a tetramer that has the shape of a mushroom; its enzymatic and antigenic sites are found in the globular head; its molecular weight is 240,000. Fifteen shapes of HA and nine of NA have been identified. In a virion, there is only one type of each protein, NA (N) and HA (H) [Ramos Jiménez, 2009].

The matrix protein (M1) gives its shape to the virus, while the M2 protein is a membrane protein that functions as an ionic transporter, thus regulating

the pH of the virion. The nucleoprotein (NP) and polymerases PB1, PB2 and PA are associated with virus RNA (this complex of RNA and proteins is also named ribonucleoproteins – RNPs). Two non-structural proteins, NS1 and NS2, are also encoded by the virus genome. The virulence of the influenza is closely associated with both HA and NA surface proteins (Ramos Jiménez, 2009).

Genomic studies have shown that the RNA segments of the 2009 A(H1N1) influenza virus had been coexisting in the strains of the swine A influenza virus for more than ten years. The ancestors of the neuraminidase have not been observed for almost 20 years. It is very likely that the host for the mixture of the current virus strains is a porcine one, but it is not known for sure (Trifonov et al., 2009).

The identification of an influenza virus strain is based on four parameters:

- type A, B, C;
- place where the strain has been isolated for the first time;
- number (order) of the strain isolated during the year;
- year of isolation.

For instance: A/PortoRico/8/34 is a type A virus strain, isolated for the first time in Porto Rico in 1934, and it was the eight strain isolated during the year 1934 (Ramos Jiménez, 2009).

Every year or every two years, small modifications may occur in the structure of HA and NA, or in both antigens of each subtype; they are known as "antigenic drift." The mechanism of this drift is an accumulation of amino-acid changes in one or more main antigenic sites of the HA protein. When comparing the genetic sequences of HA in viruses prevailing during different years, it was found that the patterns of evolution of HA was distinct among influenza types A, B and C. In general, one single strain of the A type circulates among humans and the accumulation of point mutations is linear, each new strain replacing the former one, while for the C type many strains could circulate (Ramos Jiménez, 2009).

At 20- or 30-year intervals, major antigenic changes occur and that could pave the ground for a flu pandemic. The mechanism is considered different from that of antigenic drift. The "new" viruses – antigenically speaking – are new subtypes (HxNx to HyNy) and are much more dangerous because people have no immunity. The genetic difference between two subtypes being 30%, it cannot be explained by point mutations. To explain the occurrence of new and eventually pandemic subtypes, the following factors should be taken into consideration : the

virus has a segmented genome and the A influenza viruses have a large animal reservoir (Ramos Jiménez, 2009).

Two mechanisms have been suggested for the occurrence of the genetic shifts : firstly, the recombination between avian viruses (that contain antigens unknown to humans) and human viruses that would confer to the avian virus the ability to replicate in a human host; secondly, the direct adaptation to a human host (this mechanism has been observed in swine, because these animals have both human and avian receptors (Ramos Jiménez, 2009).

In April 2009, in Mexico, in the federal district and in San Luis Potosi, the cases of human flu that were detected were caused by a new virus that was not that of seasonal influenza. The sequencing of its genome showed that it contained genes of avian, human and porcine (mostly) origin. That is why it was initially dubbed swine A(H1N1) influenza virus; later on, the World Health Organization (WHO), taking account of the global impact of such a denomination on the production, trade and consumption of pork and derived products, requested the elimination of the word porcine, so as to avoid confusion about the tagging of the new virus (this was called influenza A California H1N1, Swine Origin Influenza Virus -SOIV- or Novel Influenza A(H1N1) [Ramos Jiménez, 2009].

From the epidemiological viewpoint, the influenza viruses affect human populations in the following way : annual outbreaks that affect 10% to 30% of the population; periodic pandemics, at 20-30 year intervals, causing millions of deaths worldwide; outbreaks of influenza viruses of animal origin, as it was the case with the avian flu in Asian countries. Most isolates of human influenza A viruses are from North America, Asia and Europe. Although there are many swine influenza A viruses from North America, Asia and Europe, there is none from Africa, Oceania and South America. A(H1N1) viruses from North America and Europe show a high geographic homogeneity, while some isolates of Asian viruses contain a mixture of European and North American subtypes. Although the human influenza A viruses travel across the world with their hosts, the porcine viruses of different continents show great differentiation. Due to the close relationship between the distribution of influenza A viruses and geographical location, and because of the lack of samples in some parts of the world, it may not be surprising that the ancestors of the new H1N1 virus had not been detected for almost two decades. Consequently, only a more efficient vigilance could prevent the occurrence of similar events in the future (Trifonov et al., 2009).

The Spanish influenza pandemic of 1918-1919

The Spanish influenza pandemic occurred at the end of the first world war and apparently caused more deaths than the war itself. Although influenza epidemics had struck Europe and the Americas in 1729-1730 (it had been suggested that the disease killed more people in London than the epidemic of pest in 1665), and another influenza pandemic hit first Saint Petersburg (Russian influenza) and then spread to Europe, North America, South Africa, India and Australia, in 1889-1890, the Spanish influenza pandemic was considered the most devastating in human history : between 30 million and 50 million people died across the five continents, including about 408,000 in France. These numbers were estimates, because the statistical data were not accurate in those times of war, and most probably millions of persons died from a conjunction of the disease and other illnesses caused by pathogenic bacteria, not to mention the deadly wounds of the fighting. The influenza pandemic was christened the "great killer," as people died in a few days, everywhere from the battlefields in France and Germany, to Switzerland, the United States, Asia and even in the Arctic. It was also dubbed as "Spanish" influenza, because Spain that did not participate in the war, quickly reported its cases, while the countries in war imposed military censorship on the news. For instance, French newspapers used to mention the "Spanish flu," but not the national cases, kept secret in order to prevent the enemy from thinking that the French army was weakened by the disease (Jacot, 2009).

The disease affected more young healthy adults than children and old people, generally considered more vulnerable. That unusual situation was nevertheless accompanied by initial symptoms resembling those of a seasonal influenza : high body temperature of 40°C during 24 to 48 hours, violent headache, generalized muscle pain. Then, for those who were not to survive, breathing became increasingly difficult, followed by a rapid death. The victims of the Spanish influenza died from being choked by the fluids exuded during the infection. The luckier patients were able to recover slowly, as reported by Jay Winter, professor of history at Yale University and specialist of the first world war (Jacot, 2009).

Although the available epidemiological tools did not allow the precise determination of the disease origin, it was rapidly realized that it was not Spanish. In fact, the disease had been reported for the first time on 4 March 1918 in Kansas, where economic growth was spurred by the war, and a week later in New York. Some also mentioned the return to Boston, some time before, of an American battalion from the region of Canton, China, where an influenza outbreak had occurred at the beginning of 1918;

that influenza was benign, but very contagious. The assumption was that the virus might have mutated in North America and consequently induced pneumonia-type complications. In the United States the peak of the epidemic occurred in October 1918, with a mortality rate of 5% of infected persons (Jacot, 2009). See also Phillips and Killingray (2003).

In France, the first cases of the influenza epidemic were reported on the battlefield in April 1918, soon after the arrival of the American troops. By the end of April 1918, the disease was spreading quickly and all the armies were affected, according to the under-secretariat of health. The number of infected soldiers amounted to 14,584 on 20 May 1918, and six persons died. A second wave of the disease, much more deadly, occurred at the end of August 1918 in Brest, where American troops used to land, and then spread to the rest of the country. The third and last wave, less lethal, struck during the winter of 1918-1919 (Jacot, 2009).

It seems that the medical military authorities did not perceive the magnitude of the epidemic. Patients were returned from the battlefield to their homes, which contributed to the extension of the disease, while the soldiers benefiting from a permission transmitted the illness in regions that were still free of influenza. Behind the troops, populations lacked medical care, as many physicians were at the front. The Academy of Medicine recommended to use masks containing tissue soaked with antiseptics. As the epidemic was spreading, influenza became a military issue that had to be minimized, when the German armies launched a huge offensive in May 1918. The truth was that the disease hurt both the allies and their enemies (Jacot, 2009). See also Phillips and Killingray (2003).

In France, far behind the battlefield, there was an increasing awareness of the seriousness of the influenza epidemic. Theatres were closed down and religious ceremonies were cancelled, and rumours mentioned that the disease was due to a poisoned vaccine supplied by the Germans, or to poisoned cans also introduced by the German soldiers, or even to contaminated aspirin (discovered by the German company Bayer). The French newspaper *Le Matin* reported for the first time that people had died from influenza on 31 August 1918 (four persons died in a village located in the Allier department), when German troops started to withdraw after a counteroffensive by the allies (Jacot, 2009).

During the fall of 1918, concerns were followed by panic. In Paris, 64 persons died in a week at the end of September, and 616 at the beginning of October, according to the city's police prefecture. Between 6 October

and 9 November 1918, up to 210 deaths were recorded daily in France's capital. Specialists considered that hospitalization of the patients was ineffective and might have contributed to spreading the disease, according to J. Winter. The latter is of the opinion that "quarantine was the only effective way to control the epidemic : thus the State of South Australia simply closed its borders." Some French medical doctors, who were despaired by the sanitary situation, bled their patients, or injected terebenthin. Miracle treatments were proposed, using aspirin, caffeine citrate, tea, rum or cognac, herbal infusions, etc. The disease indeed was a great killer (Jacot, 2009).

Famous people died from the Spanish influenza. In France, the poet Guillaume Apollinaire, who participated in the war, serving in the artillery from April 1915 to March 1916, was wounded on 17 March 1916. He underwent head and brain surgery and was recovering when he was infected by the influenza virus and died on 9 November 1918. He was 38 years old. His funeral took place on 13 November and he was buried at the Père-Lachaise graveyard, in Paris, among other victims of the same disease, two days after the end of the war. The writer and dramaturge Edmond Rostand died on 2 December 1918, at the age of 50, while the Austrian painter Egon Schiele (a leader of the Secession Movement in painting), the Brazilian president Rodrigues Alves, the South African prime minister Louis Botha and S. Freud's daughter Sophie were also victims of Spanish influenza. Theodore Roosevelt and Walt Disney were luckier and escaped from the deadly illness (Jacot, 2009).

The influenza pandemic disappeared everywhere during the spring of 1919, after having killed between 2.5% and 5% of the world population (30 million to 50 million deaths, much more than the 10 million deaths caused by the Great War 1914-1918), including 549,000 American citizens, 408,000 French people, 250,000 Japanese, 220,000 Britons and 187,000 Germans. India and China would have deplored 7 million deaths each. Africa was not spared either. The US Centers for Disease Control and Prevention estimated that at least one-third of the world population at that time was infected. Spanish influenza pandemic is considered one of the most devastating sanitary disasters throughout human history (like the black pest pandemic that occurred by the middle of the 14th century) [Jacot, 2009].

Spanish influenza pandemic has fostered international cooperation in public health. The first attempt in this regard has been the creation in 1923 of the Hygiene Organization of the Society of Nations (SDN). The second was the birth of the World Health Organization (WHO) in 1948

with the objective of collecting and centralizing epidemiological data. In the meantime, in 1933, the first human influenza virus (type A) had been isolated by British researchers, while viruses belonging to types B and C had been identified in 1936 and 1950, respectively (Jacot, 2009).

In 1957, Asian influenza pandemic struck the world and in two years killed 4 million people. It started in the south of China, in the Guizhou province, then moved to Singapore, Hong Kong and the United States (in June 1957) – where some 70,000 people died, and thereafter to the rest of the world. Eleven years later, another influenza pandemic occurred : the Hong Kong flu struck the world between 1968 and 1970. It first spread to Asia and Australia, then to the United States and Europe. It killed 2 million people, half of the figure of Asian flu, partly because a vaccine was made available one month after the disease had peaked in the United States. Only in 1997 the virus of the Spanish influenza pandemic was isolated by Jeffrey Taubenberger and his American team. The Washington-based Institute of Pathology of the US Army had kept samples from the lungs of a victim of the flu. Analysis of these samples by J. Taubenberger and his colleagues revealed that the 1918 virus strain belonged to the subtype A(H1N1); the same result was obtained from the analysis of the remains of persons who died from Spanish flu and were frozen in the permafrost of the Spitzberg (Arctic). J. Taubenberger and David Morens predicted that if the present A(H1N1) strain were as virulent as that of Spanish influenza pandemic, it would kill "more than 100 million people throughout the world" (Jacot, 2009).

However, according to Patrick Zylberman, a French historian of medicine and public health, the 1918 scenario will not necessarily occur again in 2009 or 2010. It is true that modifications of the genome of the A influenza virus have resulted in the new virus A(H1N1), which is derived from two distinct porcine viruses (H1N2 and H3N2), one of them having originated from the H1N1 virus of Spanish influenza pandemic. It has also been proved that the virus of the 1957 Asian flu, A(H2N2), has evolved into that of the Hong Kong flu (H3N2) in 1968. The A(H1N1) virus of the 1918 Spanish flu is most probably the common ancestor of the viruses that caused the 1957 and 1968 pandemics, and of the 2009 A(H1N1) [fourth generation]. But we do not know anything about the antigenic structure of the viruses of the pandemics occurring before 1918 – about 15 between 1510 (Great Britain) and 1889-1890. We shall never know if before 1918 there had been a genetic shift that led to the viruses which prevailed in 1918, 1957, 1968 and 2009. Despite the epidemiological similarities between the current pandemic and that of 1918, the progress made in epidemiological monitoring and health-warning systems, in

respiratory assistance, vaccination, and the availability of antiviral drugs and antibiotics, the comparison seems anachronic to P. Zylberman. It should be recalled that the great majority of deaths that happened in 1918 was due to bacterial infections (superposed on the flu) that can be treated nowadays with the available antibiotics (Jacot, 2009). See also Bricaire and Derenne (2009), Hannoun (2009).

Origin and dissemination of the A(H1N1) virus

By the end of March 2009, Mexican health authorities were able to detect an unusually high number of patients suffering from influenza as well as of deaths among young adults. This was considered unusual because seasonal flu, that causes about 10,000 deaths annually in Mexico, generally affects very small children and old people. At the beginning of April 2009, in the district La Gloria, of Perote, Veracruz, where is located the swine farm Carroll – the biggest in Mexico, with more than a million animals – a child named Edgar Enrique Hernández was the first person to become ill from influenza, in a community of 3,000 people. About 400 persons became ill, but none of them died. On 13 April 2009, in the State of Oaxaca, a woman who was diabetic died from pneumonia (Tamez Guerra and Rodríguez Padilla, 2009).

All these events led the Mexican health authorities to alert the PanAmerican Health Organization (PAHO) about the possible outbreak of a non-seasonal flu epidemic. At the same time, samples of respiratory tract secretions of a number of patients were dispatched to the National Laboratory of Microbiology of Canada, located in Winnipeg. On 23 April 2009, it was announced that of the 51 samples, 18 were positive (i.e. contained an influenza virus) and that 12 of them contained the same genetic pattern of an influenza virus, that had been isolated for the first time in 2005 in Wisconsin, and later on in other American States. That was a subtype of the A(H1N1) virus, present for the first time in the country, and whose behaviour was difficult to predict; because, contrary to the infections observed in the States of Wisconsin, Ohio and Texas, the virus was now killing some of the patients (Tamez Guerra and Rodríguez Padilla, 2009).

The new A(H1N1) virus subtype was detected for the first time in Mexico in April 2009, but epidemiologists considered that it did not originate in that country. By mid-November 2005, in Wisconsin, a 17-year-old teenager and his brother in law, who was a butcher, slaughtered 31 pigs; later on, his family bought him a chicken, which he kept at home. During the first week of December, the teenager fell ill from influenza, but then recovered. The US Centers for Disease Control and Prevention in

Atlanta analyzed the genetic make-up of the virus and found it was a "virus mosaic," very unusual, which contained RNA segments of a human influenza virus that appeared for the first time in 1999 in New Caledonia, of two types of swine flu virus that were circulating previously in Asia and the State of Wisconsin, and of an unknown avian flu virus (Tamez Guerra and Rodríguez Padilla, 2009).

In 2006, the American Association of Swine Veterinarians (AASV) had reported that in some swine farms of the Middle West outbreaks of A(H1N1) influenza had occurred among the pigs, due to human/swine infections, and in 2007 another outbreak occurred in Ohio in swine farms, but did not affect humans. The pathogen was a virus similar to that detected in Wisconsin. The same event happened in Texas in 2008, when infections were reported in humans and swine, but were not followed by epidemic outbreaks. In other words, the virus which appeared in Wisconsin in 2005, was disseminated to other American States during the following years, and it is the same virus or virus derived from it that caused the 2009 pandemic of A(H1N1) influenza (Tamez Guerra and Rodríguez Padilla, 2009).

By mid-April 2009, when was confirmed in Mexico the presence of a new A(H1N1) influenza virus, that could be transmitted from person to person and could be lethal, the two first cases of the same influenza in children were reported in California, in two different counties. On 23 April 2009, WHO declared that the world was facing an international public health emergency and it decided to give a 3-level to the pandemic warning, which meant the human transmission of a virus that could cause a world epidemic (Tamez Guerra and Rodríguez Padilla, 2009).

On 11 June 2009, WHO raised the level to 6 (the highest), thus indicating that we were facing a global pandemic.

In Mexico, until 26 June 2009, 9,029 cases of A(H1N1) influenza had been reported and 119 deaths deplored, i.e. 1.3% of the total number of cases; 51.9% of these deaths were women and 48.1% were men; 71.3% of the persons who died had between 20 and 54 years of age. Most of the cases occurred in the federal district (Mexico City), followed by the States of Veracruz, San Luis Potosi, Jalisco, Mexico and Zacatecas. About 37% of the persons who died suffered from metabolic diseases, such as diabetes mellitus and obesity; 18.5% suffered from cardiovascular diseases, 13% were smokers, 8.3% had respiratory problems, and the rest suffered from infectious, autoimmune and cancer diseases (Tamez Guerra and Rodríguez Padilla, 2009).

In Japan, it all began at a high school volleyball tournament in Kobe, on 2 May 2009. After volleyball players were contaminated by the H1N1 virus, a wider outbreak hit Japan. On 22 May 2009, 279 confirmed cases had been detected in Kobe and the neighbouring city of Osaka in western Japan. The bouts of flu had been mild and there have been no deaths, but more than 4,800 schools had been shut down in the region, medical services were swamped and testing laboratories had to work round the clock. Thereafter Tokyo confirmed its first cases of flu. The outbreak had come as a particular shock for hygiene-obsessed Japan, where hand-washing is taught in schools and sick people dutifully wear surgical masks to keep from infecting others, and the country is one of the world's largest stockpilers of the antiviral drug Tamiflu (Tabuchi, 2009).

In the Southern Hemisphere, Australia was particularly hit : 14,037 confirmed cases and 37 deaths on 21 July 2009, and these figures were expected to raise during the winter (Vincent, 2009 b).

Saudi Arabia declared four deaths and Qatar its first death by the end of July 2009, on the eve of the annual pilgrimage to the holy places of Islam. Drastic health-security measures were taken by the Saudi authorities, while several Islamic countries reduced the number of their pilgrims.

From swine flu to human influenza pandemic

The causative agent of the recent outbreaks of human influenza is the virus A(H1N1), which is found in pigs; but animals do not die from the viral infection, they are "incubators" of the virus. There was no particular monitoring of the affection in pigs in the United States, and even less in Mexico when the first human cases were detected in April 2009. According to the American Association of Swine Veterinarians (AASV), there were no reports of unusual diseases among pigs during the month preceding the H1N1 epidemic; most likely because this affection is common in these domestic animals and it is rarely serious.

However, there should be a systematic monitoring of animal populations (wild and domestic), in order to detect any pathogen which is being incubated and which could be transmitted to humans, and thereafter from human to human. In other words, public-health monitoring and early warning systems must include the systematic monitoring of animal health. If that were the case for the H1N1 virus, the epidemiological risk would have been evaluated much better. In addition to bird flu, the A(H1N1) influenza is an example of a disease that is present among animal populations and can be transmitted to humans, either from wild

animals or via domestic livestock. These anthropozoonoses are a serious threat to human health, that exists since the dawn of human evolution. The old Roman saying *hygia pecoris*, *salus populi* ("the good health of livestock is a guarantee of human health") illustrates this concern.

Globalization explains the fast expansion of epidemics that could become global pandemics. Henceforth the need for an international monitoring and early warning system, that must be networked with national reliable epidemiological bodies. For instance, the Global Viral Forecasting Initiative (GVFI), in San Francisco, has field teams in Africa and Asia who monitor wild animals and human communities living next to them, in order to detect any new pathogen. These "sentinel populations" constitute an early warning system when a new pathogen emerges, when a dangerous disease is identified at the time it overcomes the barrier between animal and human beings. Rapid measures could then be taken. According to Peter Daszak, president of the New York-based Wildlife Trust, "the tens of millions of dollars invested in these monitoring systems could help saving the hundreds of billions of dollars of losses caused by a pandemic" (Walsh, 2009).

On 20 October 2009, health authorities of Ontario, Canada, announced that turkeys had been contaminated with the A(H1N1) virus, probably transmitted by humans. They stated that the danger for the population was very low. In August 2009, in Chile, the A(H1N1) virus had been detected in turkeys for the first time. On 19 October 2009, the US Department of Agriculture reported the virus had been transmitted to swine present in an agricultural fair in Minnesota (Benkimoun, 2009 i).

Mark Schrenzel and Bruce Rideout, two experts on wildlife diseases working at San Diego Zoo's Institute for Conservation Research, have studied the complex "receptors" for influenza viruses found on the surface membranes of host cells. For instance, the avian strain of influenza, A(H5N1), makes use of receptors containing an alpha 2,3 sialic-acid linkage, whereas A(H1N1) strains stick to receptors containing the alpha 2,6 sialic-acid bond. Any animal species, including domesticated swine, which carries receptors of both types could act as a mixing reservoir. So the American researchers tried to find out which animals had what, and they focused in particular on species likely to encounter waterfowl such as geese and ducks, which are known to be particularly vulnerable to the A(H1N1) influenza virus (*The Economist*, 2010 b).

They analyzed the receptors of 60 species and found that some which were once of considerable concern because they are often around water

birds – the European otter, the polar bear, the raccoon and the bald eagle – were unlikely to be infected because they did not carry the fitting receptors. However, several species of small carnivores, such as the arctic fox, the Chinese wolf and the corsac fox, along with the opossum, contain the receptor used by A(H1N1), making these animals potential targets for the virus. The researchers also found several animal species, including the Persian leopard, the North American striped skunk and a few other small carnivores, that carry both receptors and are therefore likely to be dangerous when one considers viral mixing. Inhibiting such mixing in wild animals is impossible, but the lesson learnt from M. Schrenzel's and B. Rideout's work is that those in charge of watching for new outbreaks of influenza would have some hints where to look for. Monitoring the health status of wildlife is considered by many epidemiologists as an important means of controlling the disease (*The Economist*, 2010 b).

Consequently, the approach to confront the risks of epidemics or pandemics consists of implementing two series of measures simultaneously :

- organization of a monitoring system aimed at following the flow of pathogens among animal and human populations, of an early warning system and quarantine measures in order to circumscribe the sites of outbreaks and protect the populations concerned;
- isolation of the pathogen as soon as possible, in both animal populations and human communities, identification of the circulating strains, sequencing of their genomes, in order to predict their genetic variability and mutation rates; and collaboration at national, regional and global levels aimed at preventing and curing the disease(s).

On 11 June 2009, WHO's director-general, Margaret Chan, declared A(H1N1) influenza virus was at the origin of the first pandemic of the 21st century. In an interview with the French newspaper *Le Monde*, published on 30-31 August 2009, she stated it was the first time in human history that we could observe a pandemic evolving under our eyes. In the past, the world was always taken by surprise, and consequently there was no sufficient time to examine the ways and means to face and control the disease. Since the emergence in 2004-2005 of the virus of the avian flu A(H5N1), which causes an extremely serious disease, with a 50% to 60% death rate among patients, health ministers have discussed with WHO the planning of the measures aimed at facing the pandemic threat, while taking as a reference the 1918 Spanish flu (Benkimoun, 2009 e). See also *The Economist* (2009 d).

M. Chan emphasized that the 2009 virus A(H1N1) was new and almost nobody was immunized against it. It spreads very quickly : in six weeks

it covers the same distance as other viruses do in six months. Although it does not seem to cause a severe disease among most infected persons, the number of the latter is unprecedented : up to 30% of the inhabitants of high-density countries run the risk of being infected (Benkimoun, 2009 e).

The data published by WHO on 28 August 2009 showed that at least 2,185 persons died from the A(H1N1) influenza. The number of people infected at that time was at least 209,438 in more than 177 countries. The virus A(H1N1)has therefore become the predominating virus worldwide, ahead of the seasonal influenza virus (Benkimoun, 2009 e).

About 60% of lethal cases occurred among people having underlying health problems, which meant that 40% of the lethal cases occurred among healthy young adults, who died after five to seven days from a viral pneumonia. This was the most serious concern, as it was very difficult to treat these patients. In many countries, therefore, hospital departments in charge of emergencies and intensive care would be overwhelmed in case the disease struck 20% to 30% of their whole population. What would happen if the influenza is even more severe than expected? A big chunk of health-care resources would be used, while other diseases such as cancer or cardiovascular diseases would receive less attention (Benkimoun, 2009 e).

On 30 October 2009, the World Health Organization (WHO) reported that the A(H1N1) influenza virus had killed 5,700 people worldwide, with a 14% increase in a week. The American continent was the most hardly hit by the flu pandemic : 4,175 deaths (+636 deaths in a week) [Benkimoun, 2009 e). See also Vincent (2009 b).

Physiopathology and transmission of A(H1N1) influenza

The A(H1N1) influenza virus attacks the bronchial mucosa, which loses its ciliae and later on the basal membrane is exposed to the virus. In patients that recover from the disease, there is a rapid regeneration of the mucosa and no complications occur. By contrast, in persons suffering from asthma and respiratory ailments, the syndrome is more severe and they could die from viral pneumopathies.

As mentioned before, all the A virus subtypes do not attack humans; in fact, human viruses belong to H1, H2 and H3 subtypes, as well as to N1, N2 and probably N8. There is also evidence that H5N1 and H7N7 viruses can cause outbreaks in humans. Thus, in 1997 in Hong Kong an important outbreak of avian A(H5N1) flu occurred in chickens. At that

time, nobody thought it could threaten human beings, until 18 persons became ill and five out of them died. Prophylactic measures were taken and 1.5 million birds were slaughtered. The virus disappeared, but not for long. It reappeared in Asian countries as outbreaks, and some of these became aggressive epidemics. That of 2004-2005 caused the death (through infection or eradication measures) of more than 120 million chickens. It also caused 74 infections in humans and 49 deaths; all of them had been in contact (direct or indirect) with infected chickens. The international scientific community warned WHO about the likelihood for this avian virus to mutate and to be transmitted from person to person. At the beginning of 2005, WHO appealed to all its member states to design national programmes aimed at facing a potential influenza pandemic (Tamez Guerra and Rodríguez Padilla, 2009).

The A(H1N1) virus is transmitted via aerosols of less than 10 μm of diameter. Large quantities of the virus are present in respiratory secretions. One single person can therefore transmit the virus to a large number of susceptible hosts, the period of transmissibility of a recently infected person is one or two days before the pathological symptoms appear, and up to seven days after the symptoms. The incubation period as well as the severity of the infection depend on the size of virus inoculation and on the host susceptibility (Ramos Jiménez, 2009).

Clinically, initial symptoms of the disease are systemic, such as fever, headache, myalgia, discomfort and anorexia. In a few more severe cases, the patient remains prostrated in bed. Respiratory symptoms include dry cough, pharyngytis, nose obstruction and rinorrhea; they also appear at the beginning of the infection, but they are less painful than the systemic symptoms; this is a clinical characteristic that distinguishes the influenza virus from other respiratory viruses (Ramos Jiménez, 2009).

Fever is the most important clinical symptom. Body temperature climbs up rapidly from 37.7°C to 40°C, and occasionally reaches 41.1°C, 12 fours after the initial symptoms. Fever is generally steady, but could be intermittent, especially when the patient is given an antipyretic drug. It lasts generally two or three days and then decreases; it may, however, last five to eight days. Other clinical observations are : warm and humid skin, weeping red eyes, rinorrhea, heated pharynx without exudate, slight swelling of cervical lymphatic nodes and slight pain of the latter on palpation. The overall clinical pattern of the influenza caused by the A(H1N1) virus is rather similar to that of seasonal influenza, although it had been noticed that among the first 643 cases reported in the United States, 25% had diarrhoea (Ramos Jiménez, 2009).

It is very important to differentiate the illness caused by the influenza A(H1N1) from other severe respiratory diseases that can be provoked by other flu viruses, or the respiratory synscitial virus or parainfluenza viruses and bacteria. All these diseases are known as influenza-like illnesses (ILIs). One way to confirm the presence of the A(H1N1) virus is an immunological test, that brings together specific antibodies against virus antigens. These tests have a high specificity (over 90%) and an accuracy of more than 70%. A limitation is that these tests cannot help to differentiate the various subtypes of the virus. Another method relies on the use of the polymerase chain reaction (PCR), which starting from a sample of the virus, can translate RNA into DNAc using a reverse transcriptase, and thereafter amplify the DNAc; then a specific gene of the virus can be identified, using specific initiators for that gene. Researchers also can inoculate a sample containing the virus into cultured cells (kidney cells of rhesus, cynomolgus or Madin-Darby monkeys) or into embryonated chicken eggs. The presence of the virus can be detected after three days of incubation in these cell cultures (75%) or after five to seven days (25%) [Ramos Jiménez, 2009).

The A(H1N1) virus seems to be very contagious and it spreads rapidly. But mortality (up to September 2009) was only 0.4%, quite less than that of other similar emergent diseases. For instance, during the epidemic outbreak of SARS (severe acute respiratory syndrome) that started on 21 February 2003, 7,761 cases had been recorded up to 17 May 2003, and 623 patients had died; the mortality rate was therefore 8%. Regarding the avian flu A(H5N1) virus, the average mortality rate in humans was 63%. It is, however, not excluded that, as occurred in the past, new outbreaks of the same disease could provoke a higher mortality among those infected with the A(H1N1) virus. That was the case with the 1918 Spanish flu : during the first week of September 1918, 68 persons died, while during the last week of January 1919 more than 125,000 died (Tamez Guerra and Rodríguez Padilla, 2009).

Two antiviral drugs are available, they both inhibit the synthesis of neuraminidase : oseltamivir (Tamiflu, manufactured by Roche) and zanamivir (Relenza of GlaxoSmithKline). The former is the most utilized because it is taken orally and stockpiles had been made by several countries in order to protect themselves against avian flu caused by A(H5N1) virus. It is also less expensive than Relenza, which requires a special device for inhalation and cannot be used by patients suffering from respiratory ailments. These drugs can also be used as a preventive measure by those who are in close contact with patients (Ramos Jiménez, 2009).

Old persons and those who suffer from chronic lung, heart or metabolic diseases, are very vulnerable to bacterial pneumonia which occurs after an influenza infection. The most frequent pathogen is *Streptococcus pneumoniae* or *Haemophilus influenzae*. Antibiotic treatment is therefore necessary in addition to antiviral drugs. It is also frequent that a person suffering from asthma, kept under control, could see its health worsened further to an infection by influenza. Also cystic fibrosis can be complicated by influenza (Ramos Jiménez, 2009).

Another aspect relating to the transmission of the disease that must be underlined is the key role played by the multiple contacts between human beings and poultry and swine farms, especially in developing countries where at the village level domestic animals and humans are living very close to each other. These contacts facilitate the transmission between humans and animals, and among animals themselves, thus generating new viruses through recombination between distinct viruses that can infect the same cell. These farms are ideal sites for the formation of new influenza viruses, which can infect birds and mammals, and new hosts, thereby increasing the pandemic potential of these viruses and their danger for human health (Tamez Guerra and Rodríguez Padilla, 2009).

Host-virus interaction, and natural defence against the influenza virus

On 24 December 2009, two studies were published in the journal *Cell*, regarding the role of some human genes that play a key role in the interaction between A(H1N1) virus proteins and host proteins, as well as in the virus replication, and the identification of a family of proteins, called IFITM that block virus replication. Researchers of the Broad Institute (Massachusetts Institute of Technology – MIT, Cambridge, Mas., and Harvard University Medical School, Boston) have shown that the 10 key proteins of the A(H1N1) virus interfere with a number of human proteins, much higher than one could expect : 87. Sagi Shapira and his colleagues stated that "the virus was in a way maximizing the diversity of functions of each protein." Another observation revealed by the study of the Broad Institute's researchers was that some human proteins were interacting with a higher number of viral proteins than expected : 24 human proteins interact with at least two viral proteins. A genetic analysis identified 1,745 human candidate genes that could be involved in the virus replication. S. Shapira and his team were thus able to assign to every gene a "specific role in the host-pathogen interaction or network" (Shapira et al., 2009).

Abraham Brass and his colleagues, also working at MIT and Harvard University Medical School, have discovered that "interferon-induced

transmembrane proteins" (IFITM) played a key role in the struggle against virus infection. When host cells detect the infection by the influenza virus through their receptors, they produce type I interferon, a glycoprotein that hinders virus replication. They are also able to respond to an attack by RNA viruses. These IFITM proteins, that can inhibit RNA expression, do confer a resistance to the A influenza virus and play a key role in triggering the antiviral action of interferon, according to the American researchers. They can also inhibit the multiplication of mosquito-transmitted flaviviruses, which include the dengue virus (that causes haemorrhagic fevers and septic shocks) and the West Nile virus (which affects the nervous system) [Shapira et al., 2009; Benkimoun, 2009 o).

Potential vaccines against common cold and influenza

The common cold has long defied treatment because the rhinovirus, the causative agent, has many strains and presents a moving target for any drug or vaccine. A research team headed by Claire Fraser-Liggett and Ann Palmenberg, a virologist at the University of Wisconsin, has brought new insights into the rhinovirus behaviour. With the help of Claire Fraser-Liggett, a leading genome researcher at the University of Maryland, they have sequenced the genomes of all 99 strains of rhinovirus and reported their results in the 3 April issue of *Science* (Palmenberg et al., 2009).

The common cold virus has a genome of about 7,000 nucleotides that encode the information to make the 10 proteins needed for the replication of the virus and the infection of human cells. By comparing the 99 genomes with one another, the American researchers were able to arrange them in a family tree based on similarities in their genomes. From this family tree, it was evident that some regions of the rhinovirus genome are mutating all the time, while others do not change at all. The fact that the latter have been so conserved over the course of the evolution means that they perform crucial roles for the virus survival. They are therefore ideal targets for drugs or vaccines, since, in principle, any of the 99 strains would be eliminated by the same drug or prevented to multiply by the same vaccine (Palmenberg et al., 2009).

C. F. Liggett considered that one such target lay at the very beginning of the rhinovirus genome, where its genetic material is folded into a clover-leaf shape. All strains of rhinovirus have much the same sequence of units in this region, and all could be vulnerable to the same drug. She also stated that the new data might lead to new vaccine approaches. Her colleague, A. Palmenberg, was not that optimistic, because vaccines do not protect the linings of the nose where the virus attacks (Palmenberg et al., 2009).

The new findings were, however, of particular interest to physicians who treat asthma. Rhinoviruses are thought to induce half of all asthma reactions. Fernando Martinez, an asthma expert at the University of Arizona, stated that it should be possible with the new rhinovirus family tree to identify which are the viruses most provocative to asthma patients. Antiviral agents could be developed against this group of viruses and would help a lot in the treatment of many asthma patients. People at high risk from rhinoviruses, such as children with asthma or adults with chronic obstructive pulmonary disease, would benefit greatly from new drugs and should be an interesting target for the pharmaceutical industry (Wade, 2009 a).

Another discovery that could lead to a seasonal flu vaccine that would not have to be changed yearly, was made by researchers from Harvard University Medical School, the Centers for Disease Control and Prevention and the Burnham Institute for Medical Research, and published in the March 2009 issue of the journal *Nature Structural and Molecular Biology*. One team leader, Wayne Marasco of Harvard university Medical School, stated that researchers began by screening a library of 27 billion antibiotics he had created, looking for ones that target the haemagglutinin "spikes" on the capsids of influenza viruses. The latter use the lollipop-shaped haemagglutinin spike to invade nose and lung cells. There are 16 known types of spikes, H1 through H16. The spike tip mutates constantly, which is why influenza shots have to be reformulated every year. But the American team found an area of the spike which apparently does not mutate, and picked antibodies that stick onto it. Once this is done, the virus can still penetrate a human cell, but it cannot unfold to inject the genetic material that instructs the cell machinery to make more of the virus (Sui et al., 2009).

The American researchers then turned the antibodies into full-length immunoglobulins and tested them in mice. The mice were injected both before and after receiving doses of the A(H5N1) bird flu. In 80% of the cases, they were protected. The team then showed that their new antibodies could protect against both H1 and H5 viruses. Most of the influenza was H1 during the 2008-2009 winter season in the Northern Hemisphere, and experts still fear that the lethal A(H5N1) bird or avian flu might start a human pandemic. But each year other seasonal influenza outbreaks are usually caused by H3 or B strains, so flu shots must also contain those. But there is always at least a partial mismatch because vaccine manufacturers must pick from among strains circulating in February since it takes months to produce the vaccine. By the time the flu returns in November, its haemagglutinin site has often mutated (Sui et al., 2009).

Therefore, other antibodies that clamp to and disable H3 and B will have to be found before researchers even think of designing a once-a-lifetime flu shot. It is also unclear how long an antibody-producing vaccine will offer protection; new antibodies themselves fade out of the bloodstream after about three weeks. Anthony Fauci, head of the US National Institute of Allergy and Infectious Diseases, stated that the work carried out by W. Marasco and his colleagues was so promising that his institute will offer the researchers grants and access to its laboratory ferrets, animals which can catch human influenza, in order to advance their work and pursue the testing of antibodies against haemagglutinin sites (McNeil, 2009).

Prepandemic vaccines against avian A(H5N1) flu

Avian flu had been described for the first in 1878 in Italy. In 1901, Centanni and Savonuzzi identified the likely pathogen causing the disease, and in 1955 this was described as a virus belonging to the influenza family.

The avian influenza virus A(H5N1), when it infects birds, can be a benign disease among wild birds, or a lethal disease in domestic poultry. With respect to the spread of that virus and the underlying factors of epidemic outbreaks, migratory birds moving from the south to the north have often been considered the vectors of the avian flu and the culprits. However, researchers of the department of ecology and evolutionary biology of the French Research for Development Institute (IRD) and of the joint research unit on genetics and evolution of infectious diseases – GEMI (IRD, National Scientific Research Centre – CNRS – and University of Montpellier) have demonstrated that the avian influenza viruses were permanently found in the aquatic environment of Camargue – a large water body and associated wetlands in the south-east of France – and they considered therefore that epidemic outbreaks of the disease were not necessarily due to migratory birds (Roche et al., 2009).

Camargue is for some 250 bird species an area where they settle during the fall and early spring on their way between Europe and Africa. Consequently this area could be for France a potential point of entry of pathogens transmitted by wild birds. In order to understand the underlying causes of epidemic outbreaks, the French researchers have gathered population and epidemiological data concerning wild birds circulating in Camargue from September 2005 to July 2006. More than 90 species, including seagulls, ducks, pink flamingos and sparrows, have been studied. Thereafter a mathematic model was developed in order to simulate the dynamics of the viruses and the host populations, i.e. the birds. It was concluded from the first analyses that epidemic outbreaks

do not result from the arrival of wild birds. Virus particles persist in the water and two main routes of transmission could play a key role in the dissemination of avian influenza : direct contact between individuals and water transmission. Birds can become contaminated and generate an epidemic outbreak when they drink water or when they filter water while eating. The French researchers are trying to understand how the pathogens which persist in the environment can have a causal effect on epidemic dynamics and also which are the specific roles of such triggering factors as the persistence of the virus in the water, migration departure, bird densities, etc. (Roche et al., 2009).

Clinical symptoms of avian flu include asymptomatic infections and respiratory ailments similar to pneumonia. The A(H5N1) strain has caused several outbreaks during recent years and has become a major public health problem, because of its high pathogenicity. The infection of humans by this strain occurred at the same time as an epidemic of bird flu, caused by the A(H5N1) virus in poultry farms in Hong Kong (Rodríguez Padilla, 2009).

Waterfowl is the natural reservoir of the bird flu virus and the infection in birds is generally asymptomatic. The virus is found in the intestine of birds (wild and poultry) and does not cause a lethal illness in general. This kind of virus has nevertheless mutated into very lethal strains causing the present epidemics. The virus mutations occur periodically, as it was the case with the 1918 Spanish flu pandemic – caused by a variant of the A(H1N1) influenza virus (see p.158). See Bahl et al. (2009) and Melidou (2009).

The first association of the virus A(H5N1) with human respiratory diseases had been noticed during an epidemic that occurred in Hong Kong in 1997 and where 18 cases had been detected. The epicentre of that epidemic was the market places where living poultry was commercialized. That epidemic had a high mortality (up to 33%), with a high incidence of pneumonia (61%) and of cases that needed intensive respiratory care because of the gravity of the disease (51%). In that mutation, all the virus genes were of avian origin, which suggested that the A(H5N1) virus had crossed the interspecific barrier. Serologic vigilance showed that there was little evidence of a transmission from human to human. See Chen (2009) and Xu et al. (2009).

The 1997 Hong Kong epidemic had been studied in a comprehensive way. However, data about this disease in human beings and its modes of transmission were not exhaustive because of the small number of cases. Investigations led to the conclusion that the origin of the human infection

was, in the 18 cases studied, the close contact with infected poultry. That is why it should be prohibited to sell living poultry directly to the consumer in the zones where outbreaks of bird flu occur (Xu et al., 2009).

In February 2003, A(H5N1) bird flu reappeared in China's Fujian province when it had been diagnosed in a man and his son. The transmission of the infection still prevailed between poultry and humans; transmission from human to human has been suspected in a few cases. That 2003 outbreak in China resulted in 21 deaths, while the world figure reached 250 (Xu et al., 2009).

On 27 January 2009, another A(H5N1) bird flu outbreak had been detected in the village of Dingdang in China. The first measure taken was to isolate the regions affected (quarantine). Although 16 Chinese provinces had been affected, no new cases were reported until 21 February 2009, and the quarantine was lifted, which suggested that the epidemic was under control in the country (Xu et al., 2009).

Reports from Vietnam and Thailand signalled new cases of A(H5N1) flu. The ten Vietnamese patients (all children or teenagers, average age : 13.7 years) had fever, coughed and could not breathe well; in addition, a significant lymphopenia was diagnosed, as well as a moderate thrombocytopenia. All the patients showed characteristic anomalies on their chest radiographies; eight of them needed mechanical ventilation to help them breathe. Pharyngeal samples were cultured and bacterial pneumonia was diagnosed in two patients. Twelve cases were reported in Thailand. All of them, but one, were in good health, and seven of them were less than 14 years old. All the patients had fever, coughed and could not breathe well; six patients had diarrhoea and pain in the muscles. Leukocyte numbers decreased in seven patients; in four of them thrombocytopenia was diagnosed and an increase in the level of liver enzymes occurred in eight patients due to the destruction of liver tissue. It was observed among the Vietnamese and Thai patients that respiratory ailments – such as cough and dyspnoea – occurred five days (on average) before the initial symptoms, but the variation was rather large. Egypt reported a case in June 2009, which brought the total number of cases to 60 in this country, compared with 109 cases in Vietnam (Rodríguez Padilla, 2009).

At the third flu conference organized in Faro (Portugal) from 14 to 17 September 2008 by the Association of physicians struggling against influenza, Robert Booy, director of Syndey's Centre for Research on Immunization, asked the participants in plenary session the following question : "How many of them thought that the pandemic of avian flu

was to become a reality in 2009?" Less than one-fourth of the scientists present raised their hand. But to the question : "How many thought that the pandemic would be a reality in five years?", two-thirds of the attendants raised their hand. Avian flu pandemic is therefore looming on the horizon (Manou, 2008 c).

In 2006, governments had stored tens of millions of doses of the antiviral drug, Tamiflu, in order to control the disease, or at least slow down its expansion across the world. In 2008-2009, the avian flu virus A(H5N1) was still endemic in two countries : Indonesia and Egypt. It also reappears from time to time in Pakistan, in one or two regions of China and India, in Thailand, Laos, Vietnam, Bangladesh and South Korea. These epidemiological data were published at the end of an international interministerial conference held in Sharm-el-Sheikh, Egypt, from 24 to 26 October 2008; at this conference, a state of knowledge of the avian flu epizootic was made. In 2008, 28 persons died from an infection by the A(H5N1) strain, less than 59 who died in 2007. A total of 245 persons died in the world until October 2008. See Sasson (2008).

In February 2008, the European Medicines Agency (EMEA) recommended the authorization of a "prepandemic" vaccine developed by GlaxoSmithKline (GSK), named Prepandrix. The European Commission was to approve the commercialization of the vaccine, while a spokesperson of GSK stated that the approval by the US Food and Drug Administration (US FDA) was to be requested by the end of 2008. Prepandrix was developed to trigger the immunization against several strains of the avian flu virus A(H5N1). The development of this vaccine is based on the following reasoning : rather than waiting for the pandemic to burst and spread, for the circulating virus to be identified and for a vaccine to be developed (at least six months are necessary to do so, starting from the circulating virus), why not prepare a "prepandemic" vaccine from the A(H5N1) virus strains already present in birds and that could become transmissible from human to human (Mamou, 2008 c).

After having obtained the authorization for commercialization, Switzerland ordered 8 million doses of Prepandrix, and Finland 5.2 million doses. In France, health experts stated that the health ministry should store the vaccine as well as Tamiflu. They thought that in case of a pandemic, the vaccination of target populations (e.g. health staff, police, army) would slow down the propagation of the disease. But all these decisions were based on the assumption that the causative agent of the pandemic would be the A(H5N1) virus. If it were not the case (e.g. strains H9N2 or H7N7), the prepandemic vaccine would be ineffective (Mamou, 2008 c).

Regarding the pharmaceutical groups that have increased their production capacities (US$2 billion or €1.4 billion have been invested by GSK), they wanted to see their efforts rewarded. Novartis also started to develop a prepandemic vaccine against A(H5N1), that was to be commercialized by the end of 2009 or early 2010 (Mamou, 2008 c). Baxter International Inc. (USA) and Commonwealth Serum Laboratories Ltd (CSL, Australia) were also developing a similar vaccine.

It has been estimated that a world pandemic of avian influenza would cause the death of 1.4 to 70 million people, depending on the degree of virulence of the pathogen. The World Bank has calculated that the economic impact of such pandemic would be around US$3,000 billion of losses. Since 2003, this threat had been considered a simple hypothesis by public-health authorities. Following the precautionary principle or approach, and with the support of the World Health Organization (WHO), many countries were taking a series of preventive and therapeutic measures which could be implemented when the first cases of transmission of the virus among humans appear.

A major concern has been expressed in case of a pandemic : the lack of cooperation between countries, not only in the health area, but also in the political, economic, social and administrative fields. A report by the United Nations coordinator of the control of avian flu (David Nabarro in New York) highlighted that most of national plans were, to a large extent, oriented towards the sanitary response, while a pandemic would have a large impact on all aspects of daily life, and this could lead to collective panic. For instance, in the European Union, for reasons of subsidiarity in the area of health, the European Centre for Disease Control, created in 2004 and headquartered in Stockholm, could only advise and support countries that requested it to do so. According to Richard Coker (London School of Hygiene and Tropical Medicine), only one-third of the European Union's member states had designed precise strategies aimed at monitoring socioeconomic activities in case of a pandemic. This expert noted that, despite some progress, there were great disparities among countries with respect to some important issues, such as : detection of the virus and ill persons at frontiers, restrictions to the entrance of persons coming from infected countries or regions, and the use (prophylactic or therapeutic) of national stocks of antiviral drugs. Another major inconsistency denounced by Antoine Flahaut, director of the French Higher School of Public Health Studies (EHESP) was the absence of massive stocks of antibiotics and of specific vaccines. See also Sasson (2008).

Development and production of vaccines against A(H1N1) influenza

Development technologies and issues

Surveillance systems and antiviral drugs will help contain the human A(H1N1) flu, but they cannot halt it the way a vaccine can do. The production of flu vaccine was developed to cope with seasonal flu, but the latter still causes the death of about 500,000 people a year worldwide. It is hard to develop a perfect vaccine against seasonal flu, because there are usually several different strains of A influenza active at any time, and genetic variations evolve. Even when flu incidence decreases at the end of the Northern Hemisphere winter, the disease merely shifts to the Southern Hemisphere, and six months later, it moves back. When the mutations are gradual, as with seasonal flu, it is known as a drift; when they are abrupt, as with the new strain of A(H1N1), one has to cope with a shift (*The Economist*, 2009 d).

As the disease had spread through at least 170 countries by August 2009, according to WHO's data, the vaccine had to be developed and produced quickly so as to vaccinate people in the Northern Hemisphere by October 2009. To help the vaccine manufacturers plan, WHO issues guidelines every six months listing the three strains of seasonal flu that appear to pose the biggest threat during the relevant hemisphere approaching winter. Then the companies prepare the "vaccine virus" which contains the specific antigens of the three strains (H and N proteins, i.e. haemagglutinin and neuraminidase) and which is multiplied inside live hens' eggs in sterile conditions. The resulting "trivalent" vaccine triggers the patient's immune system to produce antibodies, and that primes him (her) for an attack by the flu strains. The final product is commercialized in two forms. Most of the world's influenza vaccine is a killed virus, bearing on its surface the specific antigens of the major circulating flu strains, and administered as an injection. In Europe, Novartis and Sanofi Pasteur are leading producers that use this approach. In the United States, MedImmune is producing a nasal spray that contains a live virus in an attenuated form, also multiplied in hens' eggs (*The Economist*, 2009 d).

With regard to the potential global pandemic of A(H1N1) flu, WHO requested the 20 manufacturers that had an authorization for commercializing the vaccine against seasonal flu, to produce the new vaccine. The three leading producers are the British GlaxoSmithKline (GSK), the Swiss Novartis and the French Sanofi Pasteur. Despite considerable progress in life sciences and medical biotechnology, this vaccine industry relies on capital-intensive, conventional technologies,

such as producing vaccines from millions of hens' eggs. These firms can produce around a billion doses of seasonal flu vaccines every year. Some feared that companies were unlikely to be able to respond quickly enough, and even if all the capacity were switched to pandemic flu there would still be a global shortfall. Keiji Fukuda of WHO stated : "There is much greater vaccine capacity than there was a few years ago, but there is not enough vaccine capacity to instantly make vaccines for the entire world's population for influenza" (*The Economist*, 2009 d).

Switching an important part of the production to the pandemic vaccine would be risky. A lack of vaccines against seasonal flu would mean that many unprotected people would die from this disease. And a second wave of an A(H1N1) strain could be deadly. The basic issue was how fast a commercial vaccine could be developed. Five to six months are needed before a vaccine that has been authorized for commercialization, is delivered, according to WHO. The preparation of the "seed" strain of the pandemic virus, and the relevant laboratory checks, last up to two months. Thereafter, production and quality control of the first vaccine batches, used for clinical assays, take about nine weeks. Clinical assays are absolutely necessary for determining the optimal dose to be injected (i.e. that triggers a good immune response) as well as for evaluating the need or not to add an adjuvant that reinforces the effectiveness of the vaccine. Clinical assays also permit to decide whether one or two injections are necessary. This is crucial, because the same volume of vaccines or doses will serve to vaccinate a certain number of people or only half of that number (Benkimoun, 2009 d).

In August 2009, the vaccine against the influenza A(H1N1) virus was being developed and produced by at least seven manufacturers which even started the first clinical trials of the experimental vaccines. All the manufacturers use the same virus strain that was sent to them by the end of May 2009 by the collaborating centres of WHO; the latter also sent them the reagents that serve to test the effectiveness of the experimental vaccine. These are made from the surface viral proteins (H and N), associated with an attenuated flu virus. The "seed" vaccine is therefore a non-pathogenic hybrid, that bears the specific antigens of the strain A(H1N1) and against which people will be protected. The "seed" strain is multiplied in hens' eggs and in cell cultures (*The Economist*, 2009 d; Benkimoun, 2009 d).

An accelerated manufacture system for vaccines against viruses has been developed by INCELL Corporation, San Antonio, Texas. Called JITM (Just in Time Manufacture), it is based on the use of a vaccinia virus vector,

the Modified Vaccinia Ankara (MVA), which carries the genes of interest, such as that encoding the haemagglutinin of the influenza virus. The MVA has been selected because this kind of vectors carrying the relevant genes can replicate in cultured cells, and it is a safe vector that had been inoculated in more than 120,000 persons. It has a limited range of host cells, it cannot multiply in human cells and in most mammalian cells, except those of hamster (Meyer et al., 1991; Carroll and Moss, 1997; Drexler et al., 1998).

The baby hamster kidney cell line, BHK21 (C13) [CCL10 : American Type Culture Collection] grows in culture media that contain complex sera and form a monolayer. INCELL's researchers have designed a culture system in order to grow this cell line as a suspension in a well defined culture medium. Later on, cells were cloned and they could be cryopreserved, and then brought back at ambient temperature and used for the replication of MVA (Pat Moyer, 2009).

INCELL has designed a recombinant MVA (VEC) that contains the influenza genes coding for HA (haemagglutinin), for the virus internal nucleoprotein (NP), for a green fluorescent protein (GFP) – the expression of this protein allows to check the insertion and transcription of the genes of interest. The HA gene can be eliminated and replaced by a new synthetic gene. Using this system, INCELL, has been able to produce a vaccine in cultured cells in less than three weeks, while several months are needed to produce an anti-influenza virus vaccine in embryonated hens' eggs (Pat Moyer, 2009). Vaccine manufacturers are focusing their research work on the improvement of vaccine production in cultured cells, because these techniques can accelerate production, particularly in times of epidemic or pandemic, contribute to finding an optimal economy of scale and react rapidly to the appearance of a new virus strain.

Before setting up the global plan aimed at controlling avian flu A(H5N1), the world's annual capacity for the production of influenza vaccines was estimated at 450 million doses, mostly in developed countries. It was raised to 900 million doses in 2009. This is still not enough, but it had never occurred before with other pandemics. Vaccine manufacturers also promised to deliver 150 million doses of A(H1N1) influenza vaccine, free of charge, to developing countries via WHO (Benkimoun, 2009 e). For instance, Merck and Wellcome Trust – the leading British charity fund – have announced on 17 September 2009 the creation of a not-for-profit laboratory specialized in the production of cheap vaccines for developing countries. Based in India, this joint venture had a budget of US$130 million (€88.5 million) over seven years and a staff of 60 researchers. One of its

priorities was to contribute to controlling infections by A streptococci that cause over 500,000 deaths every year.

Regarding the possible recombination of the A(H1N1) virus with other influenza viruses, it seems unlikely, as shown by a study conducted by Daniel Perez and his team of the University of Maryland on ferrets that have been inoculated with influenza A(H1N1) virus, seasonal flu virus and A(H3N2) virus. The purpose of this research work was to monitor the extent of propagation of each virus and the possible recombinations between them. The American researchers concluded that the A(H1N1) virus had a higher speed of propagation in the respiratory tract than other influenza viruses; this finding explains the very fast and strong infectivity of the virus that had been observed in pandemic regions (e.g. New Caledonia in August and September 2009) [Perez et al., 2009]. The infection of the respiratory tract is fast and widespread, which would explain the higher prevalence of acute and severe respiratory ailments, i.e. viral pneumopathies. That prevalence had been estimated at 1 per 10,000 patients (compared with 1 per 1 million) at the end of August 2009 by the French epidemiologist Antoine Flahaut. When both viruses – A(H1N1) and that of seasonal flu – were inoculated simultaneously, no recombination between the two had been observed. This would alleviate the fear that such a recombination might occur before the winter season, when both viruses are circulating. But the presence of both viruses in the human body seemed to increase the severity of respiratory ailments (Perez et al., 2009).

Clinical trials

In August 2009, according to Marie-Paule Kieny, director of WHO's Initiative for Vaccine Research, seven vaccine manufacturers had initiated clinical trials of the first experimental vaccines against A(H1N1) influenza, in China, Australia, the United Kingdom, Germany and the United States. The European clinical trials were to be carried out in France and Finland by Sanofi Pasteur before the end of August 2009, according to Benoît Rungeard, communications director of Sanofi Pasteur. Similarly, GSK was carrying out clinical trials in several European countries, particularly in Germany, Spain, Belgium and France, according to Soizic Courcier, medical director in charge of regulatory affairs at GSK (Benkimoun, 2009 d).

The American government and five companies began tests by early August 2009, so as to make available an effective vaccine to the public by mid-October 2009. A total of 11,131 adults and 5,740 children were involved in these initial trials, and more assays were planned :

Novartis	: 2,380 adults and 1,190 children;
CSL Limited	: 1,540 adults and 850 children;
Sanofi Pasteur	: 1,300 adults and 400 children;
GSK	: 4,011 adults and 1,800 children;
MedImmune	: 300 adults and 300 children;
American government-sponsored	: 1,600 adults and 1,200 children (Sternberg, 2009).

Since the A(H1N1) influenza emerged in Mexico in April 2009 and until the beginning of August 2009, the disease had been linked to 6,506 hospitalizations and 436 deaths in the United States. About 36,000 people in the United States die each year of seasonal flu. The new vaccine was designed to blunt the effect of the A(H1N1) virus that, starting in the fall of 2009, could infect 100 million people in the United States and cause 30,000 to 90,000 deaths based on scenarios drawn from past pandemics, according to Arnold Monto of the University of Michigan, an adviser to the Centers for Disease Control and Prevention and the World Health Organization (Sternberg, 2009).

"Vaccine is a huge component of the public health response to influenza," stated Anthony Fauci, director of the US National Institute of Allergy and Infectious Diseases, which was sponsoring a trial at Saint Louis University and four other trials at eight federally funded vaccine research centres across the United States. Clinical trials were necessary because little was known about the pandemic vaccine, although many researchers believed it was likely to act much like seasonal influenza vaccines, according to Bruce Gellin, director of the Department of Health and Human Services National Vaccine Program Office (Sternberg, 2009).

Clinical trials are carried out in the following way : the vaccine manufacturer or the laboratory request a physician, in a hospital or a vaccination centre, to be the main researcher and responsible for the trial; he (she) is in charge of recruiting volunteers, carrying out the trials and collect the data. Volunteers are healthy people, either already registered in the files of centres carrying out clinical trials, or recruited on purpose. In France, a person participating in clinical trials cannot receive more than €4,500 a year. Sanofi Pasteur's trials were carried out on three groups : children (with the approval of their parents and not paid), adults and old people (Benkimoun, 2009 d).

There are always side-effects of vaccination. Consequently, WHO recommended a strict health-monitoring system after the vaccines had

been used. A clearcut difference should be made between the side-effects associated with the vaccine and those which are caused by other factors. Governments had requested to report to WHO any side-effects really associated with the vaccine. Margaret Chan, WHO's director-general, underlined that WHO was monitoring any side-effect and should act accordingly; she appealed to the mass communication media to fully play their role by bringing the most important informations to public opinions and by following up WHO's work (Benkimoun, 2009 e).

The number of people participating in a clinical trial, generally between 1,500 and 2,000, does not always allow the researchers to identify rare secondary effects. That is why a follow up is always planned during the vaccination campaigns. Flu vaccines have few side-effects beyond redness and soreness at the infection site. It is not easy to detect such rare ailments as the Guillain-Barré syndrome, a rare nerve disease that occurs in one of every million flu vaccine recipients (Sternberg, 2009).

A major uncertainty which clinical trials aim to unravel is how big a dose of the vaccine to give, how many doses will be needed per person for protection and whether it will be necessary to add an immune system-booster called an adjuvant. For instance, if the usual 15-microgram dose has to be doubled, the number of people who could be vaccinated would fall to 98 million in the United States. If two shots are required at double the dose, the number of people who could be vaccinated would drop further, to 50 million. Andrin Oswald, chief executive officer of Novartis Vaccines and Diagnostics, stated vaccine makers also had found that the "seed" virus needed to make A(H1N1) vaccine produced just half the yield of seasonal flu vaccine viruses. Consequently, if the yield is 100%, Novartis could deliver 10 million doses of the A(H1N1) vaccine to the American government by December 2009. If it were only 50%, it would take until March 2010. But it seemed that manufacturers had been able to develop a "seed" strain which could boost yields (Sternberg, 2009).

In order to speed up the delivery of the authorization for commercializing the new vaccine (its duration varies according to the decision to be made by the drug administration on whether one deals with a new vaccine or with a different strain to be included in a vaccine already approved), vaccine manufacturers have registered "prototypes." For instance, Sanofi Pasteur that was expecting to have the results of its clinical trials during the month of October 2009 intended to initiate production without having the final authorization for commercialization. Other manufacturers may be able to deliver the vaccine earlier, and the French health minister stated that vaccination campaigns would start at the end of September 2009 (Benkimoun, 2009 d).

WHO experts reported by the end of October 2009 that a single dose of the vaccine being used against the A(H1N1) influenza virus was sufficient for immunization; that the vaccines which had been authorized were safe and could be administered to pregnant women and could even be associated with those against season flu (Benkimoun, 2009 f).

On 25 September 2009, the European Medicines Agency (EMEA) announced that it had authorized the large-scale use of two vaccines against A(H1N1) influenza : Forcetria, manufactured by Novartis, and Pandemrix of GlaxoSmithKline (GSK). EMEA recommended a two-dose vaccination at a three-week interval for adults (including pregnant women) and children after six months of age. EMEA was waiting for more data in order to make a decision on the injection of a single dose vaccine. After EMEA, the European Commission had to decide about the commercialization of the vaccines in all the European Union member states. Other manufacturers were terminating their trials before submitting their files to EMEA. For instance, Sanofi Pasteur stated that its trials on vaccines (including one without adjuvant) were to be terminated by the end of October 2009 and that the relevant files for authorization would be submitted in November (Benkimoun, 2009 h).

For the two vaccines approved by EMEA, manufacturers had been able to rely on prototypes ("mock-up") developed against the bird A(H5N1) influenza virus and which had been authorized for commercialization; they replaced in these prototypes the antigenic elements of A(H5N1) by those of A(H1N1). EMEA stated that the experience accumulated over decades with vaccines against seasonal flu had shown this kind of substitution of antigens should not affect the safety of the vaccines, nor their effectiveness. In addition, the Agency indicated that its positive recommendation was based on the results of clinical trials had been carried out on more than 6,000 volunteers, when the mock-up vaccines were authorized; EMEA also took into consideration the data provided on the substitution of strain H1N1to H5N1. Finally, other clinical trials were being carried out on adults and children, and their results were to be available as of October-November 2009 (Benkimoun, 2009 h).

Both antipandemic vaccines that had been evaluated, contained adjuvants, aimed at boosting the immune response and reducing the quantity of antigen needed in a dose of vaccine. According to EMEA, these adjuvants are widely used in the manufacture of vaccines and are considered safe. The adjuvant of Forcetria had been used in another antiflu vaccine since 1997 in more than 45 million doses, while the adjuvant of Pandemrix had been tested in clinical trials involving several thousands of patients (Benkimoun, 2009 h).

On 9 September 2009, Novartis announced that the vaccine being tested by the company against A(H1N1) influenza triggered a strong immune reaction after the first shot among the majority of volunteers. In a trial involving 100 volunteers, 80% showed a strong immune response after the first injection, and 90% reacted strongly after the second injection (15 days later). Novartis underlined that these results had been obtained with a vaccine produced in cell cultures and not in hens'eggs – a faster process – and containing an adjuvant. This vaccine, called Celtura, was being tested in various clinical trials and could be used in a single injection instead of two (most companies were producing a vaccine to be administered in two shots). Also Novartis was working on a vaccine without adjuvant, to be used for vulnerable persons, e.g. pregnant women, patients suffering from chronic and autoimmune diseases.

EMEA had requested all vaccine manufacturers to set up plans for risk management, in order to monitor and search any side-effects of the vaccines, as soon as they were to be used in the European Union member states. Action had to take place immediately. Consequently, manufacturers committed themselves to carry out safety studies (after the authorization for commercialization) on around 9,000 patients for each vaccine. In France, the health minister who announced that the vaccination campaign against A(H1N1) influenza will start by the third week of October 2009, added that pharmacovigilance measures had been strengthened in order to detect any eventual undesirable effects of vaccination. French patients were expected to be part of the pharmacoepidemiological trials carried out by the vaccine manufacturers at their cost (Benkimoun, 2009 h).

Stockpiling

The American government ordered about 195 million doses of A(H1N1) influenza vaccine, expected to be available by the end of September 2009 (Sternberg, 2009). The estimated cost was around US$660 million.

In France, in 2007, an Establishment for the Preparation and Response to Health Emergencies (EPRUS) was created in order to alleviate the difficulties faced by public authorities in the management of stockpiles of drugs and other health-care products. EPRUS manages stockpiles of a value estimated at €845 million, including 33 million doses of antiviral drugs, 1 billion surgical masks and 537 million protecting masks (FFP2). The finance committee of the French Senate strongly criticized the management of EPRUS in a report published on 22 July 2009. EPRUS'director replied that the pharmaceutical status of his organism had been granted only in March 2009, a new software was put in place

to better follow up the situation of stockpiles and measures were taken to deal with the current health crisis. While the director-general of health at the health ministry, Didier Houssin, had said on 1 May 2009 that national stockpiles of antiviral drugs included 9 million doses of Relenza, 24 million doses of Tamiflu in the form of powder and 9 million of doses of Tamiflu in the form of pills (1 to 1.5 millions of them being off-dated), EPRUS ordered, at the beginning of July 2009, 100 million doses of vaccine against A(H1N1) influenza (for a cost estimated at up to €1 billion) to Sanofi Pasteur, GSK and Novartis, to be delivered between October 2009 and January 2010 (Vincent, 2009 b).

In China, by early September 2009, several companies had been authorized to produce the vaccine against A(H1N1) influenza. Sinovac, a biotechnology start-up, and thereafter Hualan Biological Engineering, a laboratory specialized in blood products, were among the first companies in the world that had been authorized to produce the vaccine at the beginning of September 2009. The Chinese government had requested them to produce 7.3 million doses of the vaccine. Sinovac announced that it had the capacity to produce 5 million doses of its Panflu 1 vaccine by the 1st of October 2009. That was a single-shot vaccine to be injected to persons between 3 and 60 years of age. Hualan Biological Engineering committed itself to produce 13 million doses (Pedroletti, 2009 b).

A third company, the Shanghai Institute of Biological Products, had also requested the approval of its vaccine by the State Food and Drug Administration (SFDA), in charge of monitoring the safety of foods and drugs. A total of ten Chinese companies had requested WHO to send them the virus strains (among 25 worldwide) and the vaccines were at various stages of clinical trials, according to WHO's representative in Beijing (Pedroletti, 2009 b).

It was stressed by WHO that the export of a vaccine – Mexico and Korea were interested in buying the Chinese vaccine – was a strictly bilateral issue. The authorization of a vaccine is based on national standards and it is not a prerogative of WHO to test a potential vaccine against the A(H1N1) influenza and to declare it was safe for the whole world. In China, the marketing authorization process is different from that in Europe, and although the SFDA had been criticized for misbehaviour, China had the capacity to mobilize its resources and to produce vaccines in a short period of time (Pedroletti, 2009 b).

In the case of the anti-A(H1N1) vaccine, China's endeavour was spearheaded by several small-size companies that were producing each

one a different vaccine. Thus, Sinovac – with an annual turnover of US$46 million and a staff of 354 employees – is listed on the American stock exchange (its shares had soared by 50% since the end of August 2009) and is based in a tax haven, Antigua and Barbuda. It had been created by a Chinese researcher, Weidong Yin, after the development of antihepatitis A vaccine, Healive, that supplied 86% of the company's revenue in 2008. The start-up also manufactures vaccines against seasonal flu and severe acute respiratory syndrome (SARS). In 2007, Sinovac had received €2 million of subsidies in order to increase its capacity of production of antiflu vaccines, and €10 million in June 2009 in order to struggle against the pandemic. Sinovac claimed it had a capacity of production and packaging of about 20 million doses of antiflu vaccines (Pedroletti, 2009 b).

The overall production capacity is still small. China intended to vaccinate 65 million people by the end of 2009, i.e. only 5% of its whole population. It would therefore be difficult to face a widespread dissemination of the disease. The health minister, Chen Zhu, did recognize that China's vaccine production was "limited" on 8 September 2009, and stated that "we are confronted with a serious situation." By early September 2009, nevertheless, the number of infected persons recorded was 5,592 and there had been no deaths (Pedroletti, 2009 b).

How developing countries would react to a pandemic flu? These countries are particularly exposed to the risk of major or lethal infections caused by A(H1N1) virus, as about 85% of people suffering from chronic diseases live in these countries and are therefore very vulnerable targets of the virus. WHO had estimated that some 4.9 billion doses of the new vaccine could be produced by mid-2010. Each dose would cost between €7 and €11, according to Daniel Vasella, Novartis' chief executive officer. But the cost of treatment could be higher, depending on the number of doses and injections needed to obtain the effective immune response. Regardless of the total cost, it will be too high for most developing countries. Consequently, Margaret Chan, WHO's director-general, made an appeal on 11 June 2009 to vaccine manufacturers to show their generosity towards developing countries : "This is the moment to get together on behalf of global solidarity so as to make sure that no country will be abandoned," she stated (Porier, 2009 c).

The response of vaccine manufacturers has been uneven. For instance, Novartis stated that it did not intend to make gifts of its new vaccine against A(H1N1) influenza, but would just make retail prices. On 14 June 2009, in an interview to the *Financial Times*,

Daniel Vasella indicated that "financial incentives should be created in order to make production viable." In contrast, Sanofi-Aventis' CEO, Chris Viehbacher, announced on 17 June 2009 a gift of 100 million doses of vaccines against influenza A and avian flu to developing countries. That would represent 10% of its total production and would comply with WHO's request. The American company Baxter also pledged to devote part of its vaccine production to the most vulnerable countries. GSK has confirmed its intention to convert a gift of 50 million doses of vaccines against bird flu to the poorest countries into a gift of vaccines against A(H1N1) influenza. These commendable decisions would not prevent a catastrophe in the poorer countries if a pandemic occurred, stated an expert. But it was crucial to halt the pandemic in the South, because it would affect the North a few months later (Porier, 2009 c).

Sub-Saharan Africa seemed to have been spared by the first wave of A(H1N1) influenza. Isolated cases have been detected in South Africa and Ethiopia. The population of sub-Saharan Africa is about 800 million people and 60% of the world's HIV/AIDS seropositive patients live there. It has been estimated that to stop an influenza pandemic in that region, 70% to 75% of the whole population should be vaccinated; in that case, at least 560 million doses of vaccine would be needed (Porier, 2009 c).

Vaccination campaigns against A(H1N1) influenza

United States

In September 2009, while clinical trials of the anti-A(H1N1) influenza vaccines were still being carried out and even though the US FDA authorized vaccination on 15 September, a controversy had been developing on the need for massive vaccination as well as on the potential risks of the vaccine that had been developed and produced in a short period of time. Sceptics and opponents questioned the efficiency of anti-flu vaccination in general and considered it was useless in the case of the new virus A(H1N1)-2009, as they mentioned its possible mutation. They also stressed the serious side-effects occurring when tens of millions of people are vaccinated. These fears were in particular substantiated by the events that occurred in 1976 in the United States, when a massive vaccination against an epidemic of swine A(H1N1) influenza failed (Benkimoun, 2009 f).

On 15 February 1976, a soldier stationed at Fort Dix (New Jersey) who felt tired and weak, died. Another four soldiers were hospitalized during the days following the death of their companion. Two weeks after that death,

health authorities announced the discovery of the lethal agent : a virus of swine A(H1N1) influenza, a close relative to the virus that caused the Spanish flu pandemic in 1918-1919. The American president Gerald Ford made the decision to carry out a national vaccination programme against that form of influenza; the campaign started on 1st October 1976. It was interrupted on 16 December 1976, after more than 48 million persons (22% of the whole population) had been vaccinated. Health authorities had recorded 532 cases of the Guillain-Barré syndrome related to vaccination. This rare neuropathology (1 or 2 cases per 100,000 persons), which starts as a paralysis of inferior limbs and which generally disappears after treatment, could result from vaccination with an incidence of 1 case per million. During the 1976 vaccination campaign, 25 deaths had been recorded (Benkimoun, 2009 f).

In the perspective of the massive vaccination that started by early October 2009, the pharmaceutical groups that produced the vaccines had been protected against any legal action before courts by persons who would suffer from side-effects of the vaccination. Since 15 June 2009, vaccine manufacturers had been protected by a legal immunity provided by the government, within the framework of a legislation on the preparation and public emergencies (PREP Act). On 15 June 2009, the American secretary for health and social services, Kathleen Sebelius, had signed an amendment to a 2007 "Declaration" made within the framework of the PREP Act. As the flu pandemic was considered a "public health emergency," the document extended the legal immunity to individuals (excluding professional errors made deliberately) and to institutions or bodies involved in all the steps of anti-influenza A vaccine development (trials, manufacture, distribution, prescription, administration, use) [Benkimoun, 2009 f].

The PREP Act had been adopted by the US Congress and signed by President George W. Bush in December 2005. It had been referred to during the attacks against people using spores of *Bacillus anthracis* (anthrax), or in relation with botulism or the syndrome of acute irradiation. The text signed by K. Sebelius followed the same approach, it amended the 26 January 2007 declaration that protected in the same way individuals and institutions against the side-effects of the avian A(H5N1) flu, later on extended to other influenza viruses that could cause a pandemic (Benkimoun, 2009 f).

The period of protection of individuals and institutions, in the case of A(H1N1) influenza, was 15 January 2009 to 31 March 2013. The PREP Act also authorized the health secretariat to set up an emergency

fund, supplied by the US Treasury, with a view to providing financial compensations to those suffering from side-effects of vaccination and for their treatment. During the fiscal year 2010, US$5 million were allocated to this fund while the budget for the PREP Act amounted to US$3.8 billion for the preparation to the pandemic (Benkimoun, 2009 f).

On 15 September 2009, the US Food and Drug Administration (FDA) authorized the use on a large scale of the vaccine against A(H1N1) influenza. FDA had bought 195 million vaccine doses from five manufacturers, one-third of this quantity being available in October 2009. Vaccination was voluntary and priority was given to five groups considered as the most at risk : pregnant women, medical staff, persons in contact with children, young persons between six months and 24 years of age, individuals that are less than 65 years old and suffering from other diseases. That meant a total of about 160 million people out of the whole population of 300 million people. Vaccines were free, but physicians and chemists could be paid for their administration.

The vaccination campaign started by early October 2009. On Friday 16 October 2009, health authorities foresaw a lack of available vaccines against the virus that had been detected in 41 States, and when the flu syndrome was the reason of 6% of medical consultations across the country. This was considered a high rate, especially for the month of October, by Anne Schuchat of the Centers for Disease Control and Prevention, in Atlanta. The situation was worsened by the death of 43 children since 30 August 2009, i.e. a number generally recorded by the end of an epidemic of seasonal flu. A. Schuchat predicted that this figure would raise. Since the beginning of the pandemic and until mid-October 2009, 86 children had died from A(H1N1) influenza (Benkimoun, 2009 i).

According to a poll by Harvard University School of Public Health and data from the Centers for Disease Control and Prevention, issued separately on Friday 5 January 2010, only about a fifth of all Americans had had swine flu vaccine shots. And although the disease hit children and teenagers the hardest, only about 40% of them had been vaccinated. That figure could improve because 13% of adults surveyed by Harvard University School of Public Health told the pollisters that they still intended to have their children vaccinated. Most parents who were not vaccinated stated they felt the threat had passed. The second most common reason was fear of the vaccine, even though monitoring of the injection of the first 60 million doses showed no unusual rate of side-effects (*International Herald Tribune*, 8 February 2010, p.4).

European and other countries

In France, vaccination was launched on Tuesday 20 October 2009 by the health minister, Roselyne Bachelot-Narquin, when, according to Françoise Weber, director-general of the Institute of Health Vigilance (Institut de veille sanitaire, InVS), there were no signs of outbreaks of the epidemic over recent weeks; the disease even seemed to remain stable. However, the French health minister stated that the epidemic could spread suddenly from one week to the following one during the winter. That is why the minister considered necessary to maintain the measures aimed at controlling the disease and even to take the opportunity of the epidemiological respite recorded in October 2009 to improve and complete the overall preparation process (Benkimoun, 2009 i).

The French mass vaccination programme started with that of health staff, particularly of those working in emergency services, intensive care units and neonatology departments. The vaccine used for the campaign was GSK's Pandemrix, that contains an adjuvant, and that is injected in two doses at an interval of three weeks. This was in conformity with the authorization for commercialization granted by EMEA. In October 2009, France had a stock of 1.5 million doses of that vaccine (Benkimoun, 2009 i).

Sanofi-Aventis' vaccine was to be authorized for commercialization by 20 November 2009. The vaccine was to be used for vaccinating pregnant women, because it did not contain any adjuvant. The French health minister commented on the reluctance of health professionals to be vaccinated, and she called on their responsibility; the French National Council of the Order of Physicians also recalled that national staff had the duty to be vaccinated in order to take care of patients, to protect themselves and not to spread the disease. It was underlined by Bruno Lina, director of the National Influenza Reference Centre in Lyon, that in Chile 50% of health staff that were in charge of attending patients with influenza, had been contaminated, according to a recent study. The A(H1N1) virus had the ability of infecting at least one-fifth to one-fourth of the population, and one-third in the worst case, stated B. Lina (Benkimoun, 2009 i).

Further to the statements by the French health minister, Didier Houssin, director-general of health at the ministry, announced that around one thousand vaccination centres were being set up in the French departments. Assuming that vaccines will be delivered on time and in sufficient quantities, these centres should have enough staff to vaccinate people. Henceforth the will to call on medical students and retired physicians to bolster the vaccination squads. Finally, the French health

minister insisted on the safety of the vaccine against the A(H1N1) virus, which had been tested in clinical trials over a longer period than the usual seasonal influenza vaccine (Benkimoun, 2009 i).

Italy, Sweden and Japan also launched their respective vaccination campaigns. The United Kingdom followed suit on 21 October 2009 and Germany on the 26, with the same reluctance, here and there, towards vaccination, as in France (Benkimoun, 2009 i).

In Canada, British Columbia, Alberta and Northwest Territories were seeing widespread A(H1N1) activity by mid-October 2009. Ontario, too, was seeing more of the disease, which had killed 83 Canadians (out of 4,700 worldwide). The Ontario province had received more than 200,000 doses of the vaccine of the 2 million sent across the country in October 2009, in the first shipment. Health officers requested Canadians to understand the priority for vaccinating the most vulnerable or high-risk people. The Public Health Agency of Canada set out recommendations on the use of the vaccine which included :

- Canadians 10 years of age and older should receive one dose of adjuvanted vaccine;
- children from six months to nine years of age should receive the adjuvanted vaccine in two half-doses, administered at least 21 days apart;
- infants less than six months old should not receive the vaccine;
- pregnant women should receive one dose of the unadjuvanted vaccine, which was to become available in November 2009; but if the unadjuvanted vaccine was unavailable and the flu was spreading in a particular community, women pregnant for more than 20 weeks should receive one dose of the adjuvanted vaccine. Both the anti-A(H1N1) vaccine and the seasonal flu one could be administered at the same time, as long as the needles go into different arms (Alphonso, 2009).

On 25 October 2009, the vaccination campaign started in Canada, pregnant women, people under 65 with chronic conditions and those in remote communities receiving the priority vaccines. The latter were also offered to young children, to those who care for infants and to health-care workers during the last week of October 2009. Local public health districts had been planning mass vaccination clinics and hiring more staff in preparation for Canada's largest immunization campaign. Federal health officials assured Canadians that the vaccine was safe and conferred more than 90% immunity in healthy adults, according to Canada's chief public health officer, David Butler-Jones. The head of section of Health Canada that regulates vaccines, Elwyn Griffiths, stated the regulator was

satisfied with the results of thousands of clinical trials on the safety of the vaccine and its adjuvant. One dose was considered enough for adults, six months to nine year-old children received two half doses, administered 21 days apart. E. Griffiths stated : "We firmly believe that citizens should take advantage of the opportunity to protect themselves and members of their families from this virus" (Alphonso, 2009).

However, on 24 November 2009, health authorities confirmed the existence of six cases of serious allergic reaction among persons receiving the anti-A(H1N1) vaccine, Arepanrix, produced by GSK (and different from Pandemrix used in France). These cases of anaphylactic shock were traced back to a batch of 172,000 doses of hat vaccine. One case of allergic reaction out of 100,000 doses of vaccine administered was considered normal by the World Health Organization (WHO). It was the latter which first detected these cases of allergy and it asserted that it was not changing its recommendations for large-scale vaccination campaigns in Canada and elsewhere. The persons suffering from allergy after they were vaccinated were treated and recovered completely.

How to cope with a flu pandemic?

The World Health Organization, being aware of the disparities concerning the spread of a pandemic throughout the various regions of the world, has revised the International Health Regulation (IHR) in May 2005. The new system was adopted by 194 WHO's member states and became operational on 15 June 2007. This system defines the rules aimed at reinforcing health security at national, regional and international level. Member states must notify to WHO any event that is considered a public-health emergency and that has international implications. In June 2009, WHO made an evaluation of the surveillance capacities and reactions to a health crisis, and national action plans should make these capacities operational by 2012.

To ask the question : How a country would cope with a major health crisis? implies the implementation of many tasks, such as : the identification of the persons that have been in contact with an infected person; planning of the means needed for the isolation of a high number of contagious individuals or for moving out an important population; the easy understanding by migrant populations of the recommendations issued by public authorities; taking care of thousands of non-residents trapped in airports.

It is true that we are nowadays better prepared to cope with a pandemic than formerly [in the case of A(H1N1) influenza, the virus genome had

been sequenced very rapidly, vaccines were developed and clinical trials carried out, stockpiling of antiviral drugs, and later on of vaccines, advice and assistance were provided to governments]. But the key issues remain : can the countries implement a control strategy, complying with WHO's guidelines? Do they have the necessary resources and, if this is the case, can they deploy them in the most efficient way? See Walsh (2009).

Margaret Chan, WHO's director-general, ended her interview with the French newspaper *Le Monde* (30-31 August 2009) by stating that we needed political leadership, good plans to control the disease, good coordination and good implementation of the decisions made. All this does not just concern health ministries, but the whole government, because many measures have profound economic and social implications, e.g. closing down schools or enterprises. The best investment for a government is a "pandemic communication policy," i.e. identify the most appropriate communication means in the country, so as to convey the relevant information to the whole population on time and in a transparent way (Benkimoun, 2009 e).

There are good practices. For instance, Hong Kong that has been devastated by the severe acute respiratory syndrome (SARS) outbreak in 2003, has been able to stockpile 20 million doses of Tamiflu in 2009, i.e. three times its population (while the American federal government's stockpiles of the drug could cover the needs of one-sixth of the population, plus stockpiles withheld by the States). Holiday camps around Hong Kong could be used to isolate patients, and the city has made important investments into the creation of epidemiological laboratories and the increase of hospital capacities. The World Health Organization declared that Hong Kong has become an international model or reference for the control of infectious diseases (Walsh, 2009).

Coping with a potential pandemic can be hampered by the high number of people who have no health insurance or are not protected by a social security system. These people would swamp already overwhelmed hospitals and would furthermore disseminate the disease. Henceforth solidarity within and among nations is an important factor for an effective control strategy (Walsh, 2009).

In France, both the ministers of health and interior made important statements regarding the strategy the country should carry out in the autumn of 2009. The former was under the pressure of medical experts and staff who feared that hospitals would be swamped by patients infected with the A(H1N1) virus, and she was not excluding the likelihood

of raising the level (from 5 to 6) of the national plan of prevention and control of the influenza pandemic. The latter stated that there no reason to panic, but only good reasons to prepare oneself; the minister was worried about the implications of a level 6 for the overall functioning of the country (Vincent, 2009 b).

By the end of July 2009, the French government decided to keep the level 5A of its national plan of control of the influenza pandemic. Raising it to level 5B (in case of an important circulation of the virus) or 6 (pandemic) would imply a series of heavy measures, such as : strengthening the checks made at the borders, interruption of arrivals and departures of international passengers, closing down of schools and kindergardens, interruption or reduction of certain public transport means. It should be recalled that the national plan had been set up with a view to coping with a bird A(H5N1) flu pandemic. As the bird flu virus was considered much more dangerous than the A(H1N1) virus, the above-mentioned measures could be alleviated (Vincent, 2009 b).

On 22 July 2009, Roselyne Bachelot, the French health minister, stated : "We are confronted with a virus that seems to have a strong capacity of transmission, which circulates rapidly, but has a moderate virulence." She decided that as of 23 July 2009 physicians working in the private sector, and particularly general practitioners, will have to deal with patients that may be infected with the A(H1N1) virus, while public hospitals and the service of health-emergency assistance (SAMU) will focus their work on the serious cases of the disease and on infants. Patients will be isolated at home and systematic prescription of antiviral drugs will be replaced by a case-by-case prescription, after careful evaluation by the clinician. Pharmacies will deliver, if necessary, and on prescription, antiviral drugs such as Tamiflu (Vincent, 2009 b).

Some medical experts questioned the relevance of some measures taken by the French health minister. For instance, they considered that confining ill people at home would not help, because domestic environment is not protective enough and could even contribute to the spread of the disease. They made a strong plea for the creation within hospitals of special wards for the infected persons, as it was done in the past for contagious diseases. This would be a more effective means of isolation and for providing the right therapy. Others criticized the huge amounts of funds devoted to buy stockpiles of antiviral drugs and vaccines, or masks (the efficiency of which is not well established). They recommended a more balanced distribution of funds between simple hygienic measures for the whole population and vaccination of target groups (e.g. medical staff, police, etc.).

Another controversial issue concerneds the closure of schools in case of an influenza pandemic. On 18 August 2009, the French education minister, Luc Chatel, presented to the press the plan of his ministry aimed at coping with an A(H1N1) influenza pandemic eventually. He stated that if a total pandemic occurred, the government was ready to close all schools but for the time being this was not envisaged. Pupils would be welcomed as usual by early September 2009 and parents would be duly informed. The decision to close a school or a classroom would be made on a case-by-case basis and from one day to the other. The decisions would be made by the government representatives (prefects) in close cooperation with those of the education ministry. Parents would be informed about the decision by the director of the school. Before resuming courses in September 2009, 12 million families would receive a four-page brochure, titled *Informing you about A(H1N1) influenza and your child's schooling*; in addition to the issues relating to the closure of schools, the brochure contained details about the ways to prevent contamination (e.g. washing hands several times a day, using a disposable handkerchief when one is coughing or sneezing), and the conduct to be followed in case of contamination (Floc'h and Jacqué, 2009).

If a school has been closed, it will be reopened only after six days of closure at least; when pupils and staff are not contagious any more (this period is generally estimated at seven days); and when the facilities have been cleaned and disinfected completely. Finally, in case of complete closure of schools, the education ministry intended to rely on the public radio and television to broadcast courses for all levels of primary education, while high-school pupils would be linked to "anchor" teachers who would liaise them with their teachers working from home. Also pupils and students could work through the internet thanks to the "on-line academy" developed by the ministry (Floc'h and Jacqué, 2009).

On 13 August 2009, the European Union's Health Security Committee considered that the preventive closure of schools or postponement of resuming classes in September 2009 was irrelevant at that stage (Jacqué, 2009).

In the United Kingdom, the European Union's member state most affected by the disease, no real plan had been prepared concerning schools. The government made the commitment to immunize as soon as possible 8.5 million pupils between 5 and 16 years of age, when the vaccine became available. The government adopted a pragmatic approach and had no national plan like in France. Even though several schools had been closed for a week during the month of June 2009, the British government

had reservations on the efficiency of school closure. Classes should resume at the usual dates (Jacqué, 2009).

In Germany, a federal state, each Land makes the relevant decisions. The dates for the resumption of classes vary in the different regions, but they had not been postponed. For instance, in Rheinland-Westphalen, classes started on 17 August 2009, as scheduled. There was no plan designed to close schools eventually. Here again, a pragmatic, case-by-case approach seemed to have been adopted (Jacqué, 2009).

In Spain, there was no plan either, but the education ministry launched an information campaign, that foresees the possibility to close schools for a week in exceptional cases. In August 2009, the Spanish government considered that the infection rate (33 per 100,000) was low and there was no reason to postpone the dates of resuming classes. In June 2009, two schools had been closed for a day (Jacqué, 2009).

In Belgium, the French-speaking education ministry emphasized the preventive approaches (posters and brochures to be distributed in schools when classes resume); special care was taken not to scare children unnecessarily. In case of crisis, health protection would prevail on education. Closure of schools was not excluded, but no threshold had been fixed (Jacqué, 2009).

In Switzerland, the 26 cantons should take the appropriate measures. In Neufchatel, where schools had reopened, it was decided that any pupil who showed the symptoms of the disease, should be isolated. In case of an epidemic, the canton schools would be closed first, because human contacts were more frequent there (Jacqué, 2009).

In Brazil, like other federal countries, each State was responsible for designing a plan against influenza with regard to schools. In August 2009, the closure of schools was considered irrelevant. However, it was decided to postpone the resumption of classes by two weeks in several southern States, the coldest ones, e.g. in those of Rio Grande do Sul and Paraná. In the State of Paraná, "influenza watchers" had been designated with a view to identifying ill persons (Jacqué, 2009).

In Argentina also each region or province was in charge of taking the measures to control the spread of influenza. School holidays started two weeks before the usual dates at the beginning of July 2009. On 3 August 2009, schools and universities reopened (Jacqué, 2009).

On 23 December 2009, the World Health Organization (WHO) reported that the pandemic of A(H1N1) influenza had killed at least 11,516 persons worldwide, including 176 in France, since its outbreak in April 2009. With 6,670 deaths recorded, the American continent was the mostly affected region. It seemed that by the end of 2009 the activity of the virus had reached a peak in the North Hemisphere, but its transmission remained "active and geographically extended," according to WHO. In France, where the epidemic was on the downward trend, according to the National Institute for Health and Medical Research (INSERM), the number of new cases recorded during the week of 14 December 2009 by the Regional Groups of Observation of Influenza (GROG) was still under the epidemic threshold : 593,000 in France compared with 794,000 a week earlier.

In the Netherlands, the Dutch Institute for Health and Environment had also stated on 28 December 2009 that the epidemic was over. At that date, the number of recorded cases of A(H1N1) influenza continued to decrease down to 4.4 persons per 10,000 (an epidemic meant that more than 5.1 persons per 10,000 were ill). Since April 2009, 2,156 contaminated persons had been hospitalized in the Netherlands and 53 (including four that had a mutant virus) died.

Lessons learnt from the vaccination campaign in France

Overall strategy

In France, more than 6.3 million people had been infected by the A(H1N1) influenza virus from early August 2009 to early January 2010. The GROG network had confirmed by early January 2010 that the epidemic was slowing down, but it underlined the existence of regional disparities. In France, excluding the French overseas department, the number of recorded influenza cases amounted to 307,000 during the week of 28 December 2009-3 January 2010, compared with 398,000 cases a week earlier. The network reported that the influenza epidemic was still progressing in Burgundy and in the south-east of the country.

On the other hand, about 5 million persons had been vaccinated against the virus between 21 October and 27 December 2009. According to the data published on 30 December 2009 by the French Agency for the Sanitary Safety of Health Products (AFSSAPS), 2,600 undesirable events had occurred; two deaths were recorded, but they had no relationship with the administration of the vaccine. The main vaccines used were Pandemrix (manufactured by GlaxoSmithKline – GSK – with adjuvant, 3.7 million doses) and Panenza (produced by Sanofi-Pasteur, without

adjuvant, 1.4 million doses). Regarding Pandemrix, 2,390 undesirable events (6.4 per 10,000) had been recorded, including 83 serious ones (27 during the last week of December 2009). Among these 27 cases, 12 were related to the administration of the vaccine.

On 3 January 2010, the French health ministry announced that it was trying to sell millions of doses of the vaccines against A(H1N1) influenza to countries wanting to buy them. On 4 January, it announced furthermore that it was cancelling the purchase of 50 million doses of the 94 million which were to be bought from four pharmaceutical groups in June-July 2009 : GSK (Pandemrix), Sanofi-Pasteur (Panenza), Novartis (Focetria) and Baxter (Celvapan) [Benkimoun, 2010 a; Benkimoun et al., 2010).

According to Bruno Lina, head of the National Influenza Reference Centre in Lyon, vaccine manufacturers sent a message at the beginning of the 2009 summer to the governments that producing capacities would be limited and deliveries would be made on a first-ordered first-served basis. Consequently, contracts or amendments to pre-existing contracts were signed during July 2009 with Novartis, GSK and Sanofi-Pasteur, on behalf of the French government, by the Establishment for the Preparation and Response to Health Emergencies (EPRUS) and within the rules of public purchases. In addition, on 1 August 2009, a contract was signed with Baxter for the delivery of 50,000 doses of a vaccine produced by cell cultures (and not in embryonated hens'eggs) and to be administered to persons who were allergic to eggs. Thus, 50 million doses were to be bought from GSK, 28 million doses from Sanofi-Pasteur and 16 million doses from Novartis. Total cost of the 94 million doses was €869 million. A purshase option for another 34 million doses was also envisaged : 28 million from Sanofi-Pasteur and 6 million from Novartis. At the health ministry, it was mentioned that the cancellation of these supplementary optional orders was not to be followed by penalties (Benkimoun et al., 2010).

But the whole cost of the pandemic was estimated at €2.2 billion by the French Senate's finance subcommittee. Parliamentarians of the Socialist Party and Nouveau Centre demanded the setting up of a parliament's enquiry committee on the management of the pandemic by the French government. The latter was harshly criticized by the opposition and also by several physicians and epidemiologists, who underlined that large quantities of vaccines had been bought unnecessarily and that only 5 million persons had been vaccinated by the end of 2009 (Benkimoun, 2010 a; Benkimoun et al., 2010).

Which were the reasons behind such a situation?
One should recall that since October 2004 France had had a plan aimed at reacting to an influenza pandemic. The threat of a pandemic caused by the A(H5N1) avian flu virus as well as the catastrophic consequences of the August 2003 heat wave that caused 15,000 deaths had a serious warning effect. In fact the plan that was updated several times foresaw the acquisition of the means of prevention and control and, inspired by the measures needed to react to a bioterror attack, it considered necessary to set up a mass vaccination in *ad hoc* centres. As soon as the new virus was identified, the Committee for the Control of Influenza (CLCG), created by decree on 25 July 2008, recommended the storage of vaccines against A(H1N1) virus, as well as of masks and the antiviral Tamiflu drug.

Six days after the alert of 24 April 2009 – the beginning of the pandemic in Mexico – the French prime minister called on a first interministerial meeting that was qualified as "a meeting aimed at putting on alert the state's machinery." This interministerial cell met on a weekly basis and was led by the ministries of interior and health. Such dual leadership was based on the conclusions of a white paper on defence and national security, published on 17 June 2008, which stated that "the ministry of interior coordinates in particular the management of crises on the national territory," whatsoever they are – health, terror or weather. In the case of the influenza pandemic, the interior ministry was responsible for the management and communication on the overall plan of disease control, the vaccination centres and the organization of peoples' flows, while the health ministry was in charge of the communication on health aspects (Benkimoun et al., 2010).

Very soon the interior minister considered it was too costly to involve general practitioners (GPs) in the vaccination campaign (it would have been necessary to add the cost of an individual consultation or to negotiate a lump sum), while the health ministry added that GPs should not be disturbed and prevented from their usual work, that the vaccines were delivered in multidose flasks (which should be therefore fractionated before use) and that logistic difficulties were uneasy to solve. At the beginning of July 2009 a crucial meeting took place at the prime minister's office and, while many uncertainties remained on the possible extension of the pandemic and its severity, decisions were made on the organization of a mass vaccination campaign, on a voluntary basis, on the vaccination strategy and the quantities of vaccines to be ordered. As stated by a prime minister's adviser, "all decisions were made in agreement with the head of state's office and in a collegial way among all the ministries concerned – interior, health, budget and education" (Benkimoun et al., 2010).

The strategy recommended by the experts aimed at protecting such priority groups as health personnel, the staff indispensable to the good functioning of the state, and at finding out the best means to protect the French population. For instance, it was considered useful to vaccinate children systematically because they are key vectors of virus propagation. According to Bruno Lina, head of the National Influenza Reference Centre in Lyon, the vaccination of 10 to 12 million children would have had a massive protection effect far more important than the total of vaccinated persons. Some epidemiologists, like Antoine Flahault, assumed that an early vaccination of part of the population – about 30% - would curb the epidemic. This strategy indeed was chosen by many of France's neighbour countries (Benkimoun et al., 2010).

That was not the approach adopted by the French government whose health minister justified "the political and ethical choice to make the vaccination available for everybody." On 3 January 2010, a press release by the health ministry underlined that "the purchase of 94 million doses of vaccine aimed at protecting the French population, on the basis of two doses per vaccine and of a 25% refusal rate." Indeed this strategy was asserted by the prime minister during a short visit outside Paris with the health minister. He stated that the country would not be caught by surprise and that in fact France had been preparing itself to face this risk for many years. Consequently, the decision made was in conformity with the precautionary principle and with the plan designed in October 2004 (Benkimoun, 2010 a; Benkimoun et al., 2010).

The precautionary principle and risk management

François Ewald, a French philosopher, in charge of the chair of insurances at the Conservatoire national des arts et métiers (CNAM, Paris), had founded and chaired the observatory of the precautionary principle. He has described the origins of that principle of risk management and the likely distortions of its use or application in a collective book titled *Aux risques d'innover* ("the risks of innovation").

F. Ewald recalls that the precautionary principle, in its original definition, applies to environmental management and not to crisis management. It appeared in Germany at the end of the 1990s, as the *vorsorgeprinzip*, and was meant to avoid immediate dangers, prevent mid-term risks and to ensure an optimal management of natural resources in the long term. Thereafter the precautionary principle became part of international environmental agreements, the first being the 1992 Earth Summit in Rio de Janeiro (Prieur, 2010).

F. Ewald also recalls that the concept that authorities in charge of administrative police should be guided by precaution in the implementation of their duties dated back to the 17th century. The *Traité de la police*, authored by Nicolas de la Mare in 1707 dealt with environment management (water, air and wastes). Without talking of precautionary principle, the same concept is found in the French law of 1884 on municipalities, where the duties of the mayor were defined. Old concepts are therefore reactivated with the precautionary principle applied to contemporary situations (Prieur, 2010).

In France, the 1995 law, called Barnier law, was the first legislative act that officially mentioned the precautionary principle. It did not trigger any significant debate, while when the principle was introduced in the French Constitution with the 2005 Environment Charter, France became a pioneer in the area. The principle was particularly applied to health-related crises; it was first understood as a principle of responsibility of the state, in particular when blood donations had been contaminated with the HIV/AIDS virus. It therefore became both a scarecrow and a means of protecting oneself, according to F. Ewald, who explains that there are two different ways of considering the precautionary principle : either, it is considered a deliberation process that does not make a prejudgement about the final decision (in a situation of uncertainties, all parameters are analyzed and the most appropriate solution is chosen); or it is interpreted as the suspension of any action because of uncertainty (in fact action is stopped and this is the logics of the moratorium) [Prieur, 2010].

F. Ewald highlights that the precautionary principle is always associated with the defence of a system of values. For instance, human health is a value that should be highly protected, and that explains the strategy followed by the French health minister when dealing with the A(H1N1) influenza epidemic : society must be protected so as to avoid any death. In the case of genetically modified (GM) organisms, an environmentalist organization that opposes a seed company would tend to give more weight to environment protection than to the potential benefits of GM seeds. In other words, there is a fight between values (Prieur, 2010).

At the beginning of the A(H1N1) influenza epidemic, the precautionary approach of the French health ministry was justified by the alarmist information provided by the World Health Organization. Thereafter, when it was realized that events did not occur as foreseen, the strategy should have been changed. But it should be realized that in a precautionary context, politicians not only have to manage the objective risk, which is difficult to establish due to the lack of knowledge, but also the subjective

risk, created by the people's imagination around the threat. One has therefore to deal with the fears with the appropriate communication policies (Prieur, 2010).

Regarding the fact that the French population did not adhere to the massive vaccination campaign, F. Ewald stressed that two logics were present during that crisis. The first one is a classical state logic, based on prevention and therefore on vaccination, which assumes that everyone will abide by public hygiene prescriptions. The second logic entails that people cannot be governed through obligation, because they make their decisions on the basis of the information they receive and their own system of values. The management of the crisis reveals the following paradoxical situation: the state must provide all the means needed for a massive vaccination, while these means will be used freely by each individual. The fact that there were plenty of vaccines which were not used reflects the individual freedom of choice to be vaccinated or not. Conversely, if people wanted to be vaccinated and there was a lack of vaccines, obviously the political cost for the government would have been very high (Prieur, 2010).

In the future, the application of the precautionary principle may go through the same stages, according to F. Ewald : overevaluation of the threat, and then disappointment. The French philosopher considers that the precautionary principle does not strengthen the state's authority, but weakens it and finally deprives the public decision from its legitimacy. And also, because of the exaggeration of emotions which the precautionary principle induces, it tends to put society in a situation of crisis, of permanent urgency, as it is for instance the case with the issue of climate change (Prieur, 2010).

Olivier Godard, a research director at the French National Scientific Research Centre (CNRS), who has studied the issue extensively, has underlined that "the precautionary principle is valid in situations of scientific uncertainty, where the risk is unknown." In other words, "when there is a debate among experts on the existence and nature of the risk," added Arnaud Gossement, a specialist of environmental law (Le Hir, 2010).

Maurice Tubiana, a renown oncologist and member of the French Academy of Medicine (honorary president) and Academy of Sciences, stated that he was not against the precautionary principle, but the latter had two major defects. First, "it considered only the risks and not the advantages, while any decision – especially in medicine – should be based on a benefit/risk balance." Secondly, the measures taken to reassure the

population generally reinforce its fears. He therefore proposed to create a "national committee of experts" whose mission would be to assess the reality of the risk (Benkimoun and Le Hir, 2010).

Nathalie Kosciusko-Morizet, member of the French government in 2010 and who was the rapporteur of the Environment Charter, stated in an interview (May 2010) that "a certain vision of progress does not leave space for uncertainty and precaution; that modern science itself contains a part of doubt and that the precautionary principle aims at doing everything possible to eliminate scientific uncertainty." She added that "we must put some ranking among the risks, those which are acceptable and those which are not, in order to help people make rational choices" (Kosciusko-Morizet, 2010).

In the case of three recent crises, the risk was known and identified. Regarding the cloud of ashes spewed by the volcano Eyjafjöll in Iceland, we knew that these ashes could damage plane engines (about 100 incidents had troubled long-distance flights). Regarding the Xynthia hurricane which hit France's western coast, the threat of floods was also known, as well as the danger of building houses in the areas running the risk to be flooded. With respect to the A(H1N1) flu, the virus had been identified and the potential effects of the disease were rather certain, even though the severity of the flu had been overevaluated. These three cases were in fact related to the principle of *prevention* of an identified risk (Le Hir, 2010 a).

In the case of the A(H1N1) flu, even though the ordering of vaccines must be quick, because of the time needed for their manufacture, the "proportionate" characteristic of the response would have induced the public authorities to sign revisable and reversible contracts with the manufacturers, in order to take account of the evolution of the epidemic (Le Hir, 2010 a).

Managing the influenza threat

In an interview with journalists of the French daily newspaper *Le Monde* (Clavreul and Prieur, 2010), William Dab, professor of hygiene and safety at the Conservatoire national des arts et métiers (CNAM, Paris), and former director-general of health from 2003 to 2005 at the French health ministry, asserted that an influenza threat was difficult to deal with, because of the unpredictability of the influenza virus. In the 1980s, W. Dab had created the Regional Groups of Observation of Influenza (GROG), and, as an epidemiologist, had designed the first master-plan against flu

pandemic, after the epidemic episode of the Severe Acute Respiratory Syndrome (SARS) in 2003. Regarding the A(H1N1) influenza pandemic and the way France decided to face and manage it, he reiterated that in health security the main concern is more the result of uncertainty than the level of the risk. In addition, the approach to tackle the pandemic of this kind is not purely scientific; it has to rely on the full participation of the whole society. Henceforth the crucial importance of organizing democratic debates on the uncertainties and their implications. Such debates will create the trust between the population and governmental authorities in charge of solving the crisis (Clavreul and Prieur, 2010).

Such forum exists in France : it is the National Conference on Health, created by the 2004 law on public health. This is a parliament of health whose president is elected and whose members are health associations, professionals, researchers and representatives of the pharmaceutical industry. W. Dab was of the opinion that the vaccination strategy could have been discussed there, and this would have strengthened the trust between all actors and made the decisions more legitimate. Many questions on issues about the pandemic should be discussed publicly, otherwise the population may have the impression that all possible options had not been reviewed. For instance, why France did remain at the WHO 5A level for the pandemic? Why restrain the use of Tamiflu in October 2009 and generalize it in December 2009? Why pregnant women should receive a vaccine without adjuvant if the latter is innocuous? Why vaccination carried out in *ad hoc* centres would be more efficient than in general practitioners' cabinets? Why not involving general practitioners from the outset, and using them also as good mediators for convincing the population of the necessity of being vaccinated and thus contributing to the overall trust? (Clavreul and Prieur, 2010)

The state runs the risk of triggering mistrust if it avoids discussing the issues publicly. Since the Chernobyl disaster, it is well known that hiding the doubts is counterproductive. To avoid panic, it is not advisable to transform uncertainties into certainties. It is true that the French government has applied the precautionary principle, which is mentioned in the country's constitution and which imposes a risk assessment in order to reduce the uncertainties. There was not, according to W. Dab, an overapplication of that principle. But in France, the debate was further complicated by a succession of failures in health security (e.g. blood contamination by the HIV/AIDS virus, mad-cow disease, contaminated growth hormone, heat wave in August 2003) which generated mistrust among the population and subsequently a logics of utmost precaution among politicians. Another illustrative example is that of the vaccination

campaign against hepatitis B by the end of the 1990s; over 20 million persons had been vaccinated, most of them outside the priority target. With the development of uncertainties about the potential secondary effects of that vaccine among adults, health authorities backstepped and let the general practitioners recommend or not the vaccine to their patients. The confusion was set in and it has persisted. Health professionals remain suspicious when a vaccination policy seems to them inspired by political reasons (Clavreul and Prieur, 2010).

That being said, the decisions made concerning the A(H1N1) pandemic were clear and well adapted till the end of the 2009 summer. They were also in conformity with WHO's recommendations. The plan designed to control influenza since 2005, when the threat was that of avian flu, considered that it was up to the health ministry to manage the situation in so far as the epidemic is limited to health problems. It was only when its impact became more global that the interior ministry took the leadership. Epidemiologists and health experts considered that until January 2010 the impact of the epidemic was minimal and physicians were reluctant to obey instructions from the interior ministry. In other countries, health services were sufficiently strong and equipped, so that it was not necessary to call on the interior ministry (Clavreul and Prieur, 2010).

W. Dab considered that in January 2010 the epidemic was slowing down in France and that for the second time after the mad-cow disease the most favourable scenario would prevail. But this should be an opportunity to question the national health security model and to review the strategy of risk management. Epidemics can amplify the strengths and weaknesses of social links, they may also generate the most irrational prejudices. That is why a lot of pedagogy and good communication is necessary. Health protection is not just an individual issue, but a social issue that needs a democratic debate on the anticipation of the risk and the most appropriate strategies to assess and manage it (Clavreul and Prieur, 2010).

Controversial issues

It was only in November 2009 that it became clear that a single dose of the vaccine was sufficient, as confirmed by an advice from the European Medicines Agency (EMEA). That of course halved the quantities of vaccines needed. Henceforth, the cancellation of the delivery of 50 million doses of vaccine on 4 January 2010 by the health ministry, as well as the sales of 300,000 doses to Qatar and of 2 million doses to Egypt. Negotiations were also being carried out with Ukraine and Mexico. It was decided to sell the vaccines at their cost of purchase : €7 for GSK's Pandemrix,

€6.25 for Sanofi-Pasteur's Panenza, €9.34 for Novartis's Focetria and €10 for Baxter's Celvapan. France had made a gift of 9.4 million doses of vaccine in September 2009 to the World Health Organization, in order to help developing countries. WHO had requested developed countries to donate 10% of their stockpiles to the poor countries (Benkimoun, 2010 a).

In fact, the Netherlands who had ordered 34 million doses of vaccine, also announced that they intended to sell 19 million doses to third countries who needed them. Two million doses had been sold in December 2009. Germany was also negotiating with Ukraine to sell extra quantities of vaccine. Some kind of competition might lead to retail prices, as some French parliamentarians suspected (Benkimoun, 2010 a).

Another criticism made by some epidemiologists was that no evaluation was carried out on the acceptance of mass vaccination by the French population. For instance, A. Flahault stated that "no study had been designed, for instance during vaccination against seasonal influenza." In fact, after vaccination of hospital staff that started on 20 October 2009, the campaign was launched on 12 November 2009, with about 6 million people considered as priority groups : family members of less than six-months-old infants, non-hospital health personnel, those taking care of young children and adults having health problems (respiratory diseases). At the beginning the attendance of the 1,080 vaccination centres was not heavy, then it increased due to the fast rise in influenza cases, three weeks after the opening of the centres, people queuing up for up to several hours. On 30 November 2009, the French president strongly reacted to the messy situation and requested the government to take more measures in order to offer a better service to the population; he demanded the opening of the vaccination centres on Sundays (Benkimoun et al., 2010)

In January 2010, while the number of influenza cases was decreasing it was decided to involve general practitioners and enterprises in the vaccination campaign, while the government considered that the epidemic was still there and vaccination was relevant. The initial objective was to vaccinate 75% of the French population, but in January 2010 only 8% of the population was vaccinated and vaccine stockpiles were very large. It was therefore difficult to avoid the polemic (Benkimoun et al., 2010).

Another debate and even polemic concerned the use of antiviral drugs, of which France had a stockpile of 33 million treatment equivalents. In its 12 December 2009 edition, the *British Medical Journal* (*BMJ*) published an editorial and several articles that questioned the capacity of these drugs to mitigate the complications of the influenza pandemic among healthy

persons. This was contrary to what the health authorities asserted. By the end of November 2009, Keiji Fukuda, special adviser to the director-general of the World Health Organization (WHO) on the influenza pandemic, estimated that "most information provided by clinicians across the world led us to think that these antiviral drugs, when used early and correctly, reduced the number of serious complications" (Benkimoun, 2010 a).

That was not the conclusion of the literature review on oseltamivir carried out by a team of the Cochrane Collaboration – an independent and not-for-profit international organization of physicians. After a lengthy work aimed at obtaining from Roche, the Tamiflu manufacturer, the commitment to provide data on clinical trials in the future, the Cochrane Collaboration's team published its conclusions in the *BMJ*. This antiviral drug mitigates the symptoms and reduces the transmission of seasonal influenza, but there is doubt on previous results regarding the mitigation of the complications of the lower respiratory tract by oseltamivir among healthy adults (Doshi, 2009; Ellis and McEwen, 2009).

In France, on 10 December 2009, the health ministry issued new recommendations concerning the treatment of patients suffering from influenza. These recommendations requested physicians to systematically prescribe a therapeutic antiviral treatment to all those suspected of being affected by A(H1N1) influenza, and to replace for the persons exposed to the virus the prophylactic treatment (half doses for 10 days) by a "preemptive" type of treatment (therapeutic doses for five days). This modification of treatment recommendations provoked the reactions of several medical associations and trade-unions. In particular, the Collective for an Independent Medical Training (FORMINDEP) wrote an open letter and petition to the ministry's director-general of health, Didier Houssin. The collective estimated that the new recommendations were in contradiction with reliable scientific data, and requested the provision of scientific evidence that could justify a prescription not foreseen for the commercialization of the drug. Didier Houssin observed that the recommendations addressed to physicians and health centres were based on the advice by the Committee for Influenza Control. Experts of this committee had observed that serious complications of the disease occurred among persons who had not been treated or received a late treatment. In addition, the French Agency for the Sanitary Safety of Health Products (AFSSAPS) approved the preemptive use of the antiviral drug. French health authorities had been encouraged by the first pharmacovigilance data and by the experience of British physicians who prescribed Tamiflu rather widely before the French ones, and did not find important resistance reactions (Benkimoun, 2010 a).

The legal protection granted to vaccine manufacturers by the French government also raised polemics. On 30 October 2009, three of the four contracts concluded by the French government with vaccine manufacturers were made accessible to the public by the Establishment for the Preparation and Response to Health Emergencies (EPRUS). The contract signed with the American company Baxter was not made public, due to the existence of a confidentiality clause. This contract concerned the supply of 50,000 doses of a vaccine produced by cell cultures, that can be injected to persons allergic to eggs. The contract with Baxter also dealt according to some sources, with the sale of 100,000 doses of a vaccine against meningitis, manufactured by the same company (Benkimoun, 2009 k).

Regarding the contracts signed with Novartis (16 million doses), Sanofi-Aventis (28 million doses) and GSK (50 million doses), the range of details accessible to the public was variable. For instance, in the documents made public, prices of vaccines were not mentioned. Nevertheless, these prices were given by the health ministry : from €6.25 for Sanofi-Aventis' vaccine to €10 for Baxter's (Benkimoun, 2009 k).

The issue of the liability of vaccine manufacturers in case of severe side-effects of their products, is a thorny one, especially when the innocuity of the new vaccines against A(H1N1) virus led to controversy. In the United States, the health secretary, Kathleen Sebelius, had signed a text during the summer of 2009 whereby vaccine manufacturers were protected against the threat of suing in courts and of paying high fines to those who, among vaccinees, would suffer from undesirable effects (Benkimoun, 2009 k).

In the case of France, the four contracts signed by the health ministry included a "clause of responsibility". The minister recalled that "the manufacturer is in principle responsible for any consequence due to a defective product." When the vaccine has no defect and complies with the specifications mentioned in the authorization for commercialization, "the state commits itself to protect the manufacturer against any judicial claim or action" that would ensue during vaccination. Such a protection was granted because of the "exceptional circumstances" that characterized the influenza pandemic. At the French health ministry, it was indicated that, in the absence of wrongdoing by the vaccine manufacturer, the occurrence of eventual known undesirable effects, such as those mentioned in the authorization for commercialization, would be considered as medical accident. The latter could be brought to the National Office of Indemnization of Medical Accidents (ONIAM), which is

the ruling body in charge of finding the most appropriate compensation. With respect to unexpected undesirable effects, the situation seemed less clear (Benkimoun, 2009 k).

The commitment of the state, as recognized in the four contracts signed by the French government, granted a more extended protection than that foreseen in article L3131 of the code of public health in the case of "major sanitary threats," according to some French lawyers. It was thought that the first pathological observations that might be related, rightly or not, with the vaccination against the A(H1N1) influenza virus, might lead to controversy and to legal action. History has shown that such controversy could last for years (Benkimoun, 2009 k).

Official enquiries : "a public health failure?"

During the 2009 summer, health authorities and the French government were all mobilized against the forecast invasion by the A(H1N1) flu virus. The most alarming figures were hammered by public authorities and displayed on TV screens : 20 million French people ran the risk of being infected by the pandemic virus. Spectacular measures were taken : the health ministry purchased 94 million doses of vaccine, that was being developed by big pharmaceutical groups. In hospitals, schools and enterprises control plans were being prepared.

One year later, the results were known : 312 deaths in France and 18,000 in the whole world, caused by the A(H1N1) virus; 5.3 million French people had been vaccinated, i.e. 8% of the whole population; half of the vaccine doses ordered had been cancelled, with subsequent compensation for the pharmaceutical groups; the total cost of the campaign was estimated at €500 million by the government and at €700 million by the Cour des Comptes (an independent body which reviews the expenses of public organisms and makes recommendations on the better use of funds).

The issue was therefore whether the French government had overestimated the risk, excessively dramatized the threat and badly managed the crisis, and all this on the basis of the most likely alarmist predictions of the World Health Organization (WHO). Many evaluation reports have been published in this respect.

On 24 June 2010, the Council of Europe's Parliamentary Assembly denounced "the great lack of transparency" of WHO's decisions and of national authorities, under the influence of the pharmaceutical industry. On 29 June 2010, the French Parliament's Office for the Evaluation of

Scientific Options deplored that the decisions made by WHO were less and less understandable.

On 13 July 2010, the French National Assembly's enquiry committee on the vaccination campaign against A(H1N1) influenza published its report. This committee concluded that this campaign had been "a public health failure." The committee's rapporteur concluded that the "disappointing results" were due above all to "a too rigid application" of the foreseen pattern of action, in addition to "errors in communication by the public authorities" (Benkimoun, 2010 h).

Earlier on, on 23 March 2010, the health minister, Roselyne Bachelot, who was testifying under oath before the French Senate's enquiry committee on the role of pharmaceutical firms in the management of the A(H1N1) pandemic by the government, confirmed that the former purchase of 50 million doses of vaccine had been cancelled (out of the 94 million doses to be bought through contracts signed with four pharmaceutical groups). Not only there was cancellation, but also compensation paid to the pharmaceutical firms, the minister confirmed. The amount of the compensation for each pharmaceutical firm was not expected to exceed 16% of the total value of the purchase, which was estimated at a total of €879 million billed to the French government (Benkimoun, 2010 d).

The minister considered it was a good basis for negotiations and informed the Senate's committee that discussions were being held with GSK (in order to cancel a purchase of 32 million doses of the vaccine) and Sanofi-Pasteur (11 million doses). She also added that if negotiations were not successful, the ministry would proceed to a unilateral cancellation, and the compensation would be allocated on the basis of the agreement reached with Novartis (7 million doses had been taken back by the company and the compensation amounted to 16% of the total cost indicated in the contract) [Benkimoun, 2010 d].

The health minister defended the way the crisis had been managed, and recalled that it combined "a principle of justice and a principle of reality," i.e. proposing the vaccination against the A(H1N1) flu virus to all after having estimated that one-fourth of the population would not wish to be vaccinated. With regard to the purchase of the quantities of vaccine, the minister explained that the objective during the negotiations with the companies was "to put France among the priority countries for the delivery of the vaccines." She alluded to a context "that was not favourable to the countries, in which pharmaceutical groups had a dominant position" and they decided "that the volumes of vaccine delivered initially would be

proportional to the total to be bought." R. Bachelot indicated that if France had ordered half of the volumes of the vaccine, it would have received during the first deliveries half of the volumes foreseen, and, therefore, it would have run the risk of not having enough vaccines for its vaccination campaign. Finally the health minister reiterated that "all the decision concerning the strategy of purchasing the vaccines and the organization of the vaccination campaign had been made at the interministerial level" (Benkimoun, 2010 d).

Regarding the conclusions of the National Assembly's committee, published on 13 July 2010, the members of the committee admitted that the objective of vaccinating 75% of the French population was "justified." However, they considered that the involvement of both the ministries of interior and health in the vaccination campaign was not a good approach, because it has complicated the change in the implementation pattern, once it was realized that the flu was not that dangerous. The criticism of the committee focused on the non-involvement of private physicians in the vaccination campaign, which has deprived of flexibility the implementation of the national action plan conceived against the flu pandemic due to the virus A(H5N1), much more lethal than the new virus A(H1N1). The criticism also underlined the errors in communication by public authorities (Benkimoun, 2010 h).

The communication campaign used conventional tools and did not sufficiently take account of the mindset of the population, e.g. the low intention of the population to be vaccinated. The committee stressed that the mistrust about vaccination was a challenge for the future, and if again confronted with another pandemic the full implication of health professionals was indispensable. The committee went on to state that the pandemic revealed a crisis in trusting the messages of prevention and the state's recommendations in a period of health crisis. Finally, the committee's report mentioned that the cost of the purchased vaccines might be reviewed and would amount to €382.53 million instead of €808 million indicated in September 2009 by the health minister. The amount of the compensation to be disbursed to the pharmaceutical groups would be €48.3 million, assuming that GlaxoSmithKline which questioned the sum proposed, would finally accept the deal (Benkimoun, 2010 h).

The National Assembly's committee added a note of relativity to its conclusions by underlining "a deficiency in vaccination in most other countries" and "a responsible application of the precautionary principle" (Benkimoun, 2010 h).

The Senate's enquiry committee published its final report by the end of July 2010. The report reiterated the former criticism and in particular the defects concerning the contracts signed by the French public authorities with the pharmaceutical groups during the 2009 summer. These contracts, according to the senators, showed a glaring disbalance in favour of the pharmaceutical firms, as well as a dubious legal justification of some of their clauses. "It is not acceptable that the authorities in charge of a public service of vital importance fall under the power of vaccine suppliers" and consequently are tied by too rigid contracts that do not let them many options (Benkimoun, 2010 h).

The French government has been reiterating during the crisis and since then that it had rather take too many precautions than less; that it had acted on the basis of the information supplied by the World Health Organization which stressed the gravity of the pandemic. However, the reports authored by the Council of Europe and by the French National Assembly and Senate underlined the disproportionate reaction to the real situation, the lack of capacity of adaptation to the evolution of the risk, the rigidity, inefficiency and excessive cost of the action plan, and the lack of transparency of the contracts with pharmaceutical groups.

The credibility of the World Health Organization

The World Health Organization (WHO), headquartered in Geneva, is at the service of 193 member states and employs 8,000 public-health specialists and administrators distributed in six regional offices and 147 countries. The institution was created in 1948 by the United Nations with a view to bringing all the peoples of the world to the highest level possible of public health. At its headquarters, built in 1966 in Geneva, around 2,200 persons are employed (Benkimoun and Duparc, 2010; Duparc, 2010).

Like other United Nations agencies, WHO has often set goals which are not realistic. For instance, the declaration of Alma-Ata in 1978 was very ambitious, when it stated : "One of the main social objectives of governments, international organizations and the whole international community during the next decades should be to provide all peoples of the world, from now to the year 2000, a level of health that will enable them to have a socially and economically productive life." Thirty-two years later, infectious diseases that tended to be forgotten (with the exception of pest, yellow fever and cholera, which are not found in developed countries) are coming back. In addition to HIV/AIDS, tuberculosis, malaria and emerging diseases are spreading, and pathogens resisting

to treatments are becoming more frequent. In 1996, WHO created the division of infectious diseases (Benkimoun and Duparc, 2010; Duparc, 2010).

During the 1988-1998 decade, WHO's credibility was questioned, when it was unable to impulse a strong global response to the AIDS pandemic. HIV appeared by the end of the 1970s, but WHO launched the first special programme aimed at controlling the disease in 1988, five years after the isolation of the virus at the Institut Pasteur, in Paris. In 1995, the United Nations AIDS programme was created (Benkimoun and Duparc, 2010).

In 2001, a global network for warning and response to epidemics was set up. In March 2003, WHO launched a global warning concerning the severe acute respiratory syndrome (SARS), an atypical pneumonia which appeared in China in 2002. In July 2003, the epidemic was almost controlled. WHO's credibility improved significantly and its powers were increased formally in the new International Health Regulation in 2005; the new regulation gave WHO legal coercive tools (Benkimoun and Duparc, 2010).

Having recovered its authority and credibility, WHO was confronted in 2004 with a strain of the avian flu virus A(H5N1), that could be transmitted from birds to humans and was highly lethal (50% to 60% deaths among infected people). In 2007, in order to prevent a pandemic that would cause between 7 million and 300 million deaths over three years, WHO was able to convince Japan and the United States to allocate US$2.5 million (€1.9 million) for the purchase of vaccines to be delivered to six countries. In 2008, the avian flu had killed 248 persons. It should be recalled that Margaret Chan, the present WHO's director-general, was in 1994 the health director of Hong Kong Territory, where she was born and started her career in 1978. She was confronted in 1997 with a local outbreak of avian flu, and she decided to put an end to that outbreak through the slaughtering of more than 1.5 million chickens (Benkimoun and Duparc, 2010).

Handling the A(H1N1) influenza pandemic

Margaret Chan was elected director-general of WHO in 2006, after the sudden death of her predecessor, Lee Wong-jook, from South Korea. She became the first Chinese citizen to be appointed as head of one institution of the United Nations. She had also the Canadian citizenship (she did her medical studies in Canada). Earlier on, in 2003, when Hong Kong was hit by the severe acute respiratory syndrome (SARS)

epidemic, M. Chan was in charge of the health services in Hong Kong. Her management of that outbreak was controversial : in 2004, an enquiry committee estimated that her action was "unsatisfactory", because she had been unable to detect the earlier signals of the epidemic, that had been smouldering since November 2002 in the neighbouring Chinese province of Guangdong. M. Chan defended herself by underlining that it was difficult at that time to obtain precise information from China; that was also the view of WHO's director-general Lee Wong-jook (Benkimoun and Duparc, 2010).

In 2003, M. Chan joined WHO as director of the department of protection of human environment; in June 2005, she was nominated director in charge of a department named "communicable disease : monitoring and action," and she also became representative of the director-general in charge of pandemic flu. As assistant director-general she supervised all the organization's activities in the vast area of communicable diseases. When she was elected director-general, WHO has had since 2005 a new International Health Regulation, which enabled the organization to be more demanding towards the member states in terms of monitoring diseases, information sharing and control measures. Henceforth, when on 24 April 2009, M. Chan decided to launch a global public health warning after an epidemic outbreak caused by a new flu virus in Mexico and the United States, her decision, based on the new regulation system, was taken very seriously by WHO member states. The same was true when WHO announced on 11 June 2009 the occurrence of a human flu pandemic and declared a maximum alert level of 6. On 3 June 2010, WHO maintained this maximum level; a re-evaluation of the situation was foreseen by mid-July 2010. But on 10 August 2010, M. Chan announced that the 2009-2010 pandemic caused by the A(H1N1) virus was over, following the advice of the Emergency Committee which met in Geneva the same day in the morning (Benkimoun and Duparc, 2010; Benkimoun, 2010 i).

Since the beginning of the flu outbreak in April 2009, the pandemic had caused a total of 18,500 deaths, according to WHO, compared with 250,000 to 500,000 deaths caused every year by seasonal flu. But in the case of the A(H1N1) virus, the deaths had been confirmed by laboratory tests, an unprecedented measure that will be applied in the future, as confirmed by Keiji Fukuda, special adviser to M. Chan. Has therefore WHO overreacted to the disease? It would be unfair to accuse anyone for not having foreseen well in advance how the epidemic would develop from its initial outbreaks in Mexico and the United States. However, WHO was suspected of not having been able to keep the strategic decisions

made on the level of global alert, the treatments and vaccines against the A(H1N1) influenza, outside dubious conflicts of interest. Many analysts were of the opinion that moving to the level of a global pandemic had offered an extraordinary opportunity for a few pharmaceutical groups, whose vaccine sales would have generated between US$7 billion and US$10 billion of profits.

WHO's director-general reacted by stating that the organization had not overreacted to the epidemic, but had been "consistent in its messages." It indicated that "the pandemic had a moderate severity and most affected persons would recover, but there were some acute forms among young adults and pregnant women." M. Chan has estimated that WHO had found "the good balance." She nevertheless reckoned that the post-pandemic period should enable both the member states and WHO to "review their reaction plans," and that "probably more flexibility will be needed, with optimistic, intermediary and pessimistic scenarios." "In the perspective of another pandemic, it will be necessary "to examine the stages of the pandemic, including its severity." In 2009, WHO had been severely criticized for having deleted this criterion in the definition of a pandemic (Benkimoun, 2010 i).

M. Chan also stated that the world had been "lucky" with respect to the handling of the pandemic. She admitted that "the new A(H1N1) virus had practically ceased to affect people, but this did not mean it had completely disappeared." It would adopt "the behaviour of a seasonal flu virus and would circulate for a few more years." Consequently member states should remain vigilant (Benkimoun, 2010 i).

Lack of transparency in procedures

The criticism remained about the lack of transparency and even conflicts of interest concerning WHO experts involved in preparing the director-general's decisions on the level of the pandemic as well as their relations with the pharmaceutical industry. On 4 June 2010, two reports were published on these matters. One concerned the enquiry carried out jointly by the *British Medical Journal* (*BMJ*) and the London *Bureau of Investigative Journalism*. The other was the report adopted by the Health Committee of the Council of Europe's Parliamentary Assembly (Benkimoun, 2010 f).

BMJ enquiry revealed that some experts who participated in the drafting of WHO's guidelines concerning a global pandemic, had received fees from pharmaceutical companies – Roche and GlaxoSmithKline – involved

in the manufacture of drugs or vaccines against flu viruses. The Council of Europe's report underlined a "lack of transparency" in the handling of the A(H1N1) flu crisis by WHO and national public health institutions; it accused them for having "erased part of the trust the European public has in these highly-estimated organizations," and considered that this "decline in trust might represent a risk in the future." The British member of the Council of Europe's Parliamentary Assembly, Paul Flynn, remarked that this increasing lack of trust regarding public health decisions should not be compounded by the protection of certain parts of the CVs of experts who serve international organizations, e.g. their relationship with an industrial pharmaceutical group. Such behaviour or "tradition" should not "prevail on the right of 800 million citizens to be openly and fully informed on major decisions that may have an impact on their individual health and well-being."

Since 1999, when a document presented the first guidelines of WHO concerning an action plan on a flu pandemic, recommendations concerning the international organization's strategy in this area had been drafted by four experts in collaboration with the European Scientific Working Group on Influenza (ESWI). The British journalists Deborah Cohen and Philip Carter revealed that "ESWI was entirely funded by Roche and other flu vaccine manufacturers." In addition, "two of WHO experts, René Snacken and Daniel Lavanchy (the latter was at the time a WHO employee) had participated in events funded by Roche, one year earlier, according to the marketing documents reviewed by *BMJ* and the London *Bureau of Investigative Journalism*" (Benkimoun, 2010 f).

Both British journalists have also quoted several other experts who participated in the drafting of WHO's strategic documents on pandemic flu and who had received fees from pharmaceutical groups, or/and wrote articles supporting the effectiveness of antiviral drugs. Both journalists also deplored the secret on the composition of the Emergency Committee, set up by WHO's director-general, which advised her on the moment when a pandemic should be announced. WHO's spokesperson, Gregory Hartl, did not convince both *BMJ* and the London *Bureau of Investigative Journalism* when he stated that the composition of the Emergency Committee will be made public once it has terminated its work; this measure aimed at "avoiding that the Committee's members be subjected to pressures, bearing in mind the enormous implications of the decisions made" (Benkimoun, 2010 f).

The Council of Europe's report, as adopted by the Health Committee of the Parliamentary Assembly on 4 June 2010, underlined that "it was mainly the

decision made by WHO to move to level 6 of the pandemic, when it was realized that the flu symptoms were rather moderate, in addition to the change in the definition of pandemic levels just before the announcement of the A(H1N1) pandemic, that raised major concerns and suspicion from the scientific community." The report was submitted to the Parliamentary Assembly (47 member states) on 24 June 2010 (Benkimoun, 2010 f).

Confronted with all these criticisms, at the beginning of June 2010, M. Chan appointed a group of experts who were requested to scrutinize the response to the pandemic and how the International Health Regulation had been applied. And after declaring that the 2009-2010 human flu pandemic caused by the A(H1N1) virus was over, WHO had revealed the list of the 16 members of the Emergency Committee, as well as the declaration of interest made by six among them. Until then, only the name of the Committee's president had been made public, Australian John McKenzie. Among the six members who had made a declaration of interest, some of them led research centres that had received funds from the pharmaceutical industry. However, WHO had estimated there was no such conflict of interest that would have prevented the experts from participating in the committee. But the University of Michigan professor Arnold Monto's file, already strongly criticized by the *British Medical Journal* in June 2010, was worth mentioning : he declared consultant activities, past and present, to GlaxoSmithKline, Roche, Baxter and Sanofi-Aventis, with fees not exceeding US$10,000 each time. These companies were the four main producers of antiflu vaccines, but also two manufacturers of antiviral drugs (Roche and GSK) [Benkimoun, 2010 i].

The overall conclusion is that, although WHO claimed that it had not overreacted to the A(H1N1) flu pandemic and that it applied the precautionary principle in some way, there is an urgent need for the international organization to update its procedures and to set up a clearcut transparency in its operations. In particular, its decisions should be made out of reach of any influence, particularly of private interests.

Surveillance of A(H1N1) swine flu

Although the strains of A(H1N1) virus that caused the panic did not prove particularly deadly outside Mexico, officials warned that this might change. It is common for viruses that originate in animals and go on to infect humans to evolve by recombining their genetic material with that of other strains. This could make them more virulent. In the case of A(H1N1), however, the virus remained mostly benign, so popular attention had faded. That is a mistake, argued a study published in June 2010 in *Science*.

As part of a long-running research project (beefed up since the 2009 outbreak), a group at the University of Hong Kong has been monitoring the viruses of pigs slaughtered in the territory's main slaughterhouse. The Hong Kong researchers sequenced viruses they found by swabbing snouts of pigs coming from all over southern China. That testing, supported by an American government grant, has gone on for 12 years (McNeil Jr, 2010 b).

No dangerous new strain has emerged, stated several experts who saw the report issued on 14 June 2010. But in January 2010 the researchers found a new strain with one of the pandemic flu virus surface proteins. That was a reminder of how easily another swine strain capable of spreading among people could emerge. "Just because we have just had a pandemic does not mean we have decreased our chances of having another," stated Carolyn B. Bridges, an epidemiologist in the flu division of the Centers for Disease Control and Prevention in the United States. "We have to stay vigilant" (McNeil Jr, 2010 b).

That is of crucial importance because pigs can catch both human and bird flu viruses. The latter easily swap their genes, and any new combination might be able to spread among pigs and reach another human. Pigs in the huge hog-raising barns of the United States and Western Europe are tested regularly, but the millions of pigs on small farms and in big operations in Asia and Latin America seldom are. Among the globe-circulating flu viruses that pigs could, in theory, catch are six swine flu viruses, several human seasonal ones and at least two avian ones. The latter include the feared A(H5N1) virus, which has killed 60% of the 500 people known to have caught it since 2003 but so far almost never spread from person to person, and an A(H9N2) strain, which has been found in a dozen humans but caused only mild disease so far. The pandemic in 2009 was originally dubbed a "swine flu" because the eight genes in its makeup had been seen before in American or Eurasian pigs during the previous ten years, though never in the exact combination that made people sick in Mexico (McNeil Jr, 2010 b).

Malik Peiris, a flu expert at the University of Hong Kong and one of the authors of the study published on 18 June 2010 in *Science*, stated that he and his colleagues found strong evidence that the A(H1N1) virus afflicting humans was indeed recombining in pigs. They saw the mingling of A(H1N1) with two other types of swine influenza virus. These were the North American triple-reassortant viruses and the Eurasian avian-like swine viruses. They did not, however, observe reassortment with human seasonal influenza viruses, something they had worried might occur. "The message from our paper is not an inevitable disaster around the corner, but the need for continued vigilance," stated M. Peiris (Vijaykrishna et al., 2010).

In fact it is possible that new recombinations will make the virus even less dangerous. But the reverse is also possible. It would make sense, therefore, to maintain a strict surveillance on pig populations around the world. "The implication of the Hong Kong study is that we have to be very careful," stated Peter Palese, a flu researcher at the Mount Sinai School of Medicine in New York (McNeil Jr, 2010 b). See also *The Economist* (2010 a)

Cold storage of vaccines : a great challenge for developing countries

Nowadays, vaccines must be stored at temperatures between 4°C and 8°C maximum, and thermosensitive capsules are means to make sure that the cold chain is not interrupted during the storage and transport of vaccines. Matthew Cottingham of Oxford University Jenner Institute has summarized the current situation in the following way : "You need a health-care centre with a nurse, a refrigerator, electric supply and refrigerated trucks for the distribution." He added that "if vaccines could be transported at air temperature, costs would be decreased considerably and the access to vaccination would be improved markedly" (Morin, 2010 a).

Since the early 1990s, in developing countries, the proportion of vaccinated people has been stabilized at between 75% and 80% of the whole population. The reason for the lack of progress is the difficulty to maintain a cold chain in the remote areas, in order to store vaccines and preserve their efficiency. A new process, developed by a team of Oxford University and the company Nova Bio-Pharma Technologies, may change the situation. The British researchers published the results of their work in the journal *Science Translational Medicine* (*STM*) on the conservation of two samples containing live poxviral and adenoviral vaccine vectors during six months at 45°C, without losing their capacity to trigger an immune response. These vaccine vectors were in a sense vitrified in a sugar solution, which solidifies when it dries up. Thereafter, this solidified solution is mixed with water and both viral vaccine vectors become active and the efficiency of the vaccine is restored. A patent has been filed to protect this innovation which is particularly simple : the vaccine stored in its solid basis or support is encapsulated in a plastic container, which is placed between the needle and the syringe before carrying out the vaccination (Alcock et al., 2010).

According to Souleymane Koné, in charge of the "cold chain" at the vaccination division of the World Health Organization (WHO), this new process "would drastically change many things in the field, because the managerial costs of refrigeration are huge." He recalled that breakdowns of the cold chain led to a loss of vaccines estimated at between 20% and 50%,

depending on their packaging type. Loss is smaller when the storage concerns individual doses of vaccines. In developed countries, about US$200 million were being spent annually (2009) for the maintenance of the cold chain, which increases the cost of vaccination by 14% to 20%, according to WHO's estimates (Morin, 2010 a).

The research carried out by Oxford University's team has been supported by the Wellcome Trust and fellowships funded by Bill & Melinda Gates Foundation under the programme "great challenges for global health." The technology of vaccines embedded in sugar had been envisaged more than ten years ago. In 2001, it was mentioned during a meeting convened by WHO; it is based on the property of some sugars to stabilize bioproducts. Christine Rollier of the Jenner Institute and co-author of the publication in *Science Translational Medicine*, recalled that these sugars enable some desert plants to dry up and then resuscitate. The company Nova Bio-Pharma Technologies is in charge of the following step, i.e. that one can move to the industrial stage and demonstrate that the process works for current and future vaccines (Alcock et al., 2010; Morin, 2010 a).

C. Rollier underlined that the preservation of vaccines at tropical temperatures is a crucial issue for the next-generation vaccines. For instance, in the case of malaria, the candidate vaccines use genetically modified viruses to introduce parasite's genes, without being infectious; thus, the immune system can recognize the parasite. But the viruses must remain alive so that the immunization could be effective. Hence the need for an efficient cold chain (Morin, 2010 a).

Controlling malaria

Malaria has been known about since ancient times and has been given many names, e.g. tertian fever, quartan fever, paludism. Nowadays, it kills 850,000 people a year worldwide, and debilitates hundreds of millions more. The World Health Organization (WHO) estimates that a child dies from malaria every 30 seconds, mainly in sub-Saharan Africa (*The Economist*, 2009; Benkimoun, 2009 c).

Malaria is mainly caused by two protozoan species : *Plasmodium falciparum*, very widespread and lethal in Africa; and *Plasmodium vivax*, prevailing in Asia and South America. Epidemiological data collected up to now showed that *P. vivax* could not infect persons whose erythrocytes had not a surface protein called Duffy. Consequently, those populations belonging to a Duffy-negative blood group were considered as naturally protected against the infection by *P. vivax.* Unfortunately, on 11 March 2010,

an international team involving researchers from the United States, France (Institut Pasteur) and Madagascar, published their results in *the Proceedings of the National Academy of Sciences USA* (*PNAS USA*) demonstrating that *P. vivax* could infect populations that were considered protected against the parasite because of their blood group. Such discovery might lead to redesigning some vaccination strategies, but not that of the current vaccine being tested (see below). But surely the discovery is a warning about the possible spreading of *P. vivax* into regions of the world when it used to be absent (Ménard et al., 2010).

Current approaches to control the disease are : insecticide-impregnated nets designed to prevent people from being bitten by infected mosquitoes, and spray of insecticides in households; new drug combinations containing artemisinin (extracted from the Chinese sweet wormwood, *Artemisia annua)*. And researchers and drug manufacturers are trying to develop and test an effective vaccine.

Fighting malaria : at last some good news

On Monday 19 April 2010, almost one week before the world day against malaria (Sunday 25 April 2010), the United Nations Children's Fund (UNICEF) and the Rock Back Malaria (RBM) partnership published their report that confirmed that malaria was causing less damage in Africa than before. Even though the World Health Organization (WHO) deplored every year 250 million cases of malaria and 850,000 deaths (in 2008), 90% of which occurred in Africa – and most of them young children. The international community had decided that in 2010 the mortality due to malaria should be halved thanks to a universal access to prevention, diagnosis and treatment of the disease. In fact, the number of malaria cases recorded worldwide in 2009 fell to 225 million, after having increased from 233 million in 2000 to 244 million in 2005.

In Africa, 40% of public health expenses are devoted to the control of malaria, amounting to some US$12 billion per year. However, in 12 African countries where these expenses were considered "reasonable," RBM partnership's report indicated that more intensive preventive measures had saved several hundred thousands children. Nevertheless, the funds available to this international partnership covered only 25% of global needs (Benkimoun, 2010 c; Vincent, 2010 a, c).

Global funding aimed at controlling malaria comes to a very large extent from the World Fund against AIDS, tuberculosis and malaria, as well as from the more recent commitments by the World Bank, the American President

Malaria Initiative (PMI) and the Bill and Melinda Gates Foundation. Global funding had increased from around €220 million in 2003 to €1.25 billion in 2009, according to RBM partnership's report. Such budget allowed for the support of the production, purchase and distribution of the main antimalaria drugs; consequently a significant improvement was recorded in several African countries (Vincent, 2010 c).

The global production of insecticide-impregnated antimosquito nets has been considerably increased between 2004 and 2010. In 26 African countries that had the relevant data and included 71% of the population of children under five years, the average use of these antimosquito nets has risen from 2% in 2000 up to 22% in 2008. RBM partnership's report indicated that by the end of 2009 many African countries had received enough antimosquito nets to protect more than half of their population at risk, and that seven countries (Burundi, Liberia, Madagascar, Namibia, Central African Republic, Senegal and Sudan) had received enough nets to be able to protect 80% of households (Vincent, 2010 c). According to the annual report of the World Health Organization (WHO), published on Tuesday 14 December 2010, about 289 million antimosquito nets had been delivered to sub-Saharan Africa by the end of 2009, so as to meet 76% of needs.

A similar situation concerned the spraying of walls inside houses with remanent insecticides. In the zones where this preventive method was appropriate, the efforts made had resulted in 2008 in the protection of almost 25 million people in 14 countries, compared with 2.1 million people in three countries in 2006 (Vincent, 2010 c).

Progress, however, was less striking with regard to the most effective treatment of the disease in case of infection : a therapeutic combination called ACT, containing artemisinin. There are several reasons for that : first, many health centres have not yet the necessary equipment and expertise to make blood smears which is the most effective method for the detection of the malaria parasite; secondly, ACT treatment is expensive. Since 2004, the purchases of ACT had been increasing rapidly worldwide (160 million doses in 2009, compared with 5 million in 2004), but surveys carried out since 2007 had shown that a small percentage of African children were treated with ACT, most of them still receiving inappropriate treatments (chloroquine sulfadoxine-pyriméthamine) [Vincent, 2010 c].

A frequent problem is due to the interruption of supply of antimalaria drugs. To mitigate this problem, Tanzania had set up a pilot project in 2009 for a five-month duration in three districts (i.e. 135 villages) with a total population of over 1 million people. With the support from Novartis,

Vodafone, IBM, RBM partnership and the health ministry, this initiative relied on mobile telephone, SMS technology and cartography software. This system allowed the follow-up of drug supply to health centres and the management of deliveries, including to remote places. The principle is simple : every week, the focal point in each health centre receives automatic SMS requesting them to check the level of the antimalaria-drug stock and to communicate the relevant information, also through SMS, to a central system of information. If deemed necessary, this central system orders the delivery of drugs to those centres which need seem. RBM partnership experts reported that the 47 health centres of one of the three districts had enough drugs to treat their patients thanks to the system put in place and during the first weeks. This was unprecedented in a country where half of the 5,000 clinics, hospitals and health centres could until then have simultaneously a breakdown in their drug supplies. Tanzania is now extending this pilot project to other districts, while a number of countries have shown interest in trying it. If successful, this kind of programme could be extended to a wider range of products and other basic drugs (Vincent, 2010 c).

The overall results of the efforts made to control malaria with the present available means are already significant. The authors of the RBM partnership's report, taking account of the estimates of the numbers of antimosquito nets available in 35 African countries (which represent more than 95% of infant mortality due to malaria), have concluded that without the use of these nets the number of deaths caused by malaria between 2000 and 2010 would have reached more than 10 million among children less than five years old. During that period, the use of antimosquito nets had saved 908,000 lives in these countries and consequently avoided 8.8% of deaths due to malaria. This percentage was expected to raise to 20% in 2010. These estimates had been strengthened by the data provided at the beginning of 2010 by nine African countries, which had recorded a significant decrease (30% to 95%) of morbidity and mortality indicators regarding malaria. The situation was improving even in countries where, a few years ago, it was far from satisfactory; for instance, Nigeria was planning to distribute 60 million antimosquito nets before the end of 2010 (Vincent, 2010 c). Also at the end of 2009, 11 African countries were delivering enough antimalaria drugs to meet more than 100% of their needs.

Another good news, while waiting for an effective and affordable antimalaria vaccine, is that the African heads of state are making commitments to achieve the objectives of malaria control, which they set for themselves. Hopefully, one of the eight Millennium Development Goals which was to control malaria (and other major diseases) by 2015

could be reached, or at least African countries would be on the right path to do so (Vincent, 2010 c).

Origin of the malaria parasite

In 1958, Frank Livingstone, an anthropologist, suggested that *Plasmodium falciparum* (which is the deadliest of the four or five protozoan parasites that cause human malaria) had been transmitted to *Homo sapiens from* chimpanzees, which harbour *Plasmodium reichenowi.* F. Livingstone speculated that the transmission to humans might have occurred during the rise of agriculture, when human communities encroached on forests and woodlands. Other experts suggested that *P. falciparum* was a variant of *P. gallinaceum*, a parasite found in chickens (*The Economist*, 2009 h).

A paper published on 1 September 2009 in the *Proceedings of the National Academy of Sciences USA* (*PNAS USA*) by Stephen Rich of the University of Massachusetts at Amherst, Nathan Wolfe of the Global Viral Forecasting Initiative in San Francisco, and their colleagues, contains genetic evidence to confirm that *P. falciparum* is indeed an offshoot of *P. reichenowi.* The American researchers suggested that the parasite might have been transmitted from chimpanzees to humans some 10,000 years ago, i.e. when, as F. Livingstone argued, human hunters-gatherers were settling down on the first farms. To reach that conclusion, S. Rich and N. Wolfe studied the DNA sequences of malarial parasites collected from nearly 100 wild or formerly wild chimpanzees born in Central and Western Africa. They identified eight distinct versions of *P. reichenowi* and found that the DNA of *P. falciparum* was well correlated with this genetic variation. Furthermore, it seems that the transmission from chimpanzees to people occurred only once. All *P. falciparum* parasites alive today appear to derive from an individual example of *P. reichenowi* (Rich et al., 2009).

As mentioned earlier (see pp. 173 and 218), understanding how parasites cross species barriers is crucial to knowing how new infections or emerging diseases start in humans. S. Rich has underlined that because *P. reichenowi* is "of greatly diminished pathogenicity" in its chimpanzee hosts (i.e. it causes them few adverse symptoms); it may provide a basis for the development of new vaccines that are naturally attenuated (*The Economist*, 2009 h).

However, new data have been published on the physiology of *Plasmodium falciparum* in the 23 September 2010 issue of *Nature* by a French research team led by Eric Delaporte of the French Research for Development Institute (IRD) and the University of Montpellier. The

researchers have been carrying out a vast study aimed at reconstructing the phylogenic tree of *P. falciparum* and they were able to demonstrate that African gorillas were a reservoir of the parasite and malaria. They also came to the conclusion that the infection was very common among some gorilla populations and that it was the ape which infected humans and not the reverse (Liu et al., 2010).

In August 2009, another research team had isolated from chimpanzees living in Cameroon and Côte d'Ivoire a *Plasmodium* whose genome was sufficiently close to that of the parasite which infects humans, and could therefore be considered as its ancestor. Doubts have been wiped out after it was demonstrated that the human and simian parasites were completely similar, thanks to the use of advanced genetic-analysis techniques (Vincent, 2010 g).

Until these techniques became available it was not possible to compare both kinds of parasites, the human one having been identified more than a century ago by the French bacteriologist and physician Alphonse Laveran (Nobel Laureate of Medicine in 1907). Another major hurdle is associated with the difficulty to gather epidemiological data on animals in their natural environment. Taking blood samples from wild animals is almost impossible; it is fraught with the risk of wounding or even killing them. Researchers, some of them co-signing the paper published in *Nature* of 23 September 2010, had the idea, some ten years ago, to collect the faeces of apes and anlyze them, instead of taking blood samples. Among them, the American biologist Béatrice Hahn of the University of Alabama, Birmingham, Alabama, and Martine Peeters, research director at HIV/AIDS and associated diseases unit, IRD, Montpellier, have been studying the prevalence of the Simian Immunodeficiency Virus (SIV) – the equivalent of HIV in monkeys – among the primates of Central Africa. They quickly realized that their approach could be extended to other diseases and pathogens, such as malaria and *Plasmodium*. In fact, most of the researchers who co-signed the paper published in *Nature* are not malaria, but HIV/AIDS specialists. Both the collection of samples and molecular-biology techniques used to study them have been developed with a view to studying the animal origin of HIV. Jean-Fransois Delfraissy, director of the French National Agency for Research on AIDS (ANRS) and of the National Institute for Health and Medical Research (INSERM) Institute of Infectious Diseases, has underlined that the AIDS epidemic was such a threat to public health that its funding has allowed the costly and risky research carried on the apes living in Central Africa's forests. He also stated that he was happy to see that this approach could be useful to the control of other diseases. In fact the collection of faeces made by the

French and American team in order to study the relationships between SIV and HIV could also be used to improve our knowledge on other pathogens, e.g. Ebola virus, yellow fever and influenza viruses, which all circulate between animals and humans, and *vice versa* (Liu et al., 2010).

The first stage of the research work consisted of collecting the faeces on the forest sites where the apes were living; the animals were not easily accessible and researchers must be assisted by local guides who knew these environments and could detect gorillas' faeces (i.e. distinguish them from thosee of chimpanzees or bonobos). The sites of sampling were localized by GPS, so as to be able to follow the primate populations over time. All this meant years of patience and hard work, leading to thousands of tubes containing the animals'faeces and stored in the freezers of the University of Alabama and Montpellier's laboratory (Liu et al., 2010).

The second stage consisted of extracting the DNA from the faeces, which comprised the apes'DNA, as well as that of the plants eaten by the apes and the DNAs of bacteria and viruses present in their digestive tract. And if the animal were affected by malaria, the *Plasmodium* DNA was also present in the faeces. Using a very powerful DNA sequencing technique, called single genome amplification (SGA), the researchers could identify the living species to which belonged the analyzed DNA (Liu et al., 2010).

Thus, the researchers discovered that gorillas could be infected by several strains of *Plasmodium*, of which one was very close to that which affects humans. This demonstrated, according to E. Delaporte, that "it is the gorilla which infected the human being and not the reverse, and this event occurred once at a date still unknown" (Vincent, 2010 g).

Another conclusion that may have important implications for public health is that the analysis of 2,700 faecal samples, collected in 57 sites distributed in Central Africa, has shown that the infection by *Plasmodium* was frequent in West African gorillas, with a prevalence rate of 32% to 48%. The prevalence rate was probably higher, because the analysis of faeces was less accurate than that of blood samples in terms of parasite identification. It remained to check whether the presence of *Plasmodium* caused the disease among apes, but these animals were by no means potential reservoirs for the contamination of humans. As the contacts between apes and human populations were becoming more frequent in Central Africa, further to massive deforestation and the subsequent human colonization, the dissemination of malaria could become another big challenge for the control of the disease (Liu et al., 2010).

Edward Holmes, an epidemiologist of the US National Institutes of Health (NIH), commented in *Nature* (23 September 2010) that although the studies of the biodiversity of pathogens were often carried out with a view to predicting and eventually preventing another future human pandemic, it was oversimplistic to think that describing what actually exists in natural conditions would enable the precise forecast of the emergence of a disease. However, this kind of research provided a very valuable genetic catalogue, which was of great help to understand the origins of human diseases (Holmes, 2010).

In this respect, sequencing the genomes of both *Plasmodium* and its vectors could be very useful. The genomes of *Anopheles gambiae*, vector of *Plasmodium falciparum*, and of *Aedes aegypti*, vector of the dengue virus, have been sequenced in 2002 and 2007, respectively. In 2010, the 18,883 genes of *Culex pipiens quinquefasciatus*, the vector of West Nile Virus and of the worm causing elephantiasis, were sequenced by an international consortium.

Killing the mosquitoes

Chemical insecticides, either sprayed directly in households or in any site where mosquitoes would breed and multiply, or impregnating bed nets, are generally effective at the beginning. But later on, mosquitoes develop resistance; in other words, they provoke an evolutionary response from the insects they are supposed to kill. How therefore to retain the effectiveness of insecticides without triggering that evolutionary response?

Andrew F. Read of Pennsylvania State University and his colleagues observed that it is old, rather than young, mosquitoes that are infectious and transmit the parasite. Only females can transmit *Plasmodium* through sucking blood (males suck plant juices only), but they are not born with the parasites inside their bodies. They have to acquire it from already infected humans. Once a female *Anopheles* feeds on infected blood, the ingested parasites require 10 to 14 days to mature and migrate to the mosquito's salivary glands, from which they can be transmitted to another host during a blood meal. In theory, therefore, killing only the oldest female mosquitoes, i.e. those at significant risk of being infectious, could stop the transmission of the disease. Since these females would have had plenty of time to reproduce before they died, the evolutionary pressure imposed by killing them would be much lower (Miller et al., 2010).

To test this hypothesis, the American researchers constructed a mathematical model of the mosquito life-cycle. They then introduced

the data collected from malaria-infested areas in Africa and Papua New Guinea, that described the insect life-span and egg-laying cycles in those parts of the world and the way that malaria parasites grow inside mosquitoes. The model revealed that selectively killing elderly mosquitoes would reduce the number of infectious bites by 95% and that resistance to such a control method would spread very slowly, if it spread at all, because mosquitoes vulnerable to a post-breeding insecticide, would have had a chance to pass on their vulnerable traits to future generations (Miller et al., 2010).

Is an insecticide that selectively kills the elderly available? One option is to use existing chemicals, but at greater dilutions, because older mosquitoes are more vulnerable to insecticides than younger ones. Another option is to use an entomopathogenic fungus. The American researchers are working on fungi that take 10 to 12 days to become lethal that is short enough to kill parasite-infected mosquitoes before they can transmit the pathogenic protozoan, but long enough to allow them to breed. A trial was being carried out in Tanzania in April 2009 : it consisted of spraying fungal spores onto bed nets and house walls. If it works, it will illustrate that the control of insect vectors could be based on an evolutionary approach. In the case of malaria, it would mean the bites of mosquitoes will merely be irritating, not life-threatening (Rivero et al., 2010).

Antimalaria artemisinin drugs

Since the year 2000, the Swiss drug manufacturer Novartis has been selling Coartem, one of the most effective antimalarials on the market, to public-health bodies in developing countries at a loss totalling more than US$253 million – not counting the millions spent on research and development. According to the firm, that allowed to save more than 550,000 lives. In late January 2009, the company marketed the first paediatric dose of Coartem – less bitter and easier to swallow than the version for adults – which is expected to help to reduce the deaths among children (more than 700,000 children under five years die each year). Coartem is at the core of Novartis' programmes aiming to prevent and cure diseases such as dengue fever, tuberculosis and leprosy. The company was spending more than US$1 billion a year on ensuring better access to medicines in developing countries (Kingsbury, 2009).

Developed in 1994, Coartem pills combine artemisinin, derived from sweet wormwood (*Artemisia annua*), with lumefantrine, designed by Chinese researchers, which does not kill parasites as quickly but lingers in the blood longer to help prevent resistance. While historically all the

antimalaria drugs have been developed for prevention, Coartem is a cure. Its high US$2.40-a-dose price was criticized by public-health officials and associations. In 2001, Novartis signed an agreement with the World Health Organization (WHO) to bring the price down to US$1 per dose, or just about the cost of making the drug. There was indeed no point to try to sell a medication to people who could not afford it, as stated by Daniel Vasella, chief executive officer of Novartis. Thereafter, the drug manufacturer slashed the price again, to UScents80, in other words taking a 20% loss. Meanwhile, it scaled up production, subsidizing *Artemisia* cultivation in China and Kenya in order to be able to supply 100 million doses of Coartem a year throughout Africa and Asia (Kingsbury, 2009).

Synthetic biology is another tool for producing artemisinin, not from the plant or tissue cultures, but from a re-engineered yeast cell where the genes encoding the metabolic pathways for artemisinin biosynthesis have been introduced. At the University of California, Berkeley, under the auspices of One World Health, a not-for-profit company that develops drugs for neglected infectious diseases, Jay Keasling, research leader, has extensively re-engineered yeast cells, so as to achieve mass production of affordable artemisinin in a "microbial fermenter." This production was expected to start in 2010, in collaboration with the French drug company Sanofi-Aventis, which, like Novartis, manufactures, in a factory located in Morocco, an antimalarial pill containing artesunate derived from artemisinin and amodiaquine, an older antimalarial drug; the drug, called ASAQ, is not protected by a patent and costs less than US$1 for adults and less than UScents50 for children. Production of artemisinin via synthetic biology aimed to improve the supply of the bioactive compound, which depends on the cultivation of *Artemisia annua*. Jay Keasling's slogan is : "With the tools of synthetic biology we do not have to just accept what nature has given us" (Cookson, 2009).

But making the antimalarial drug cheaply available is not the whole answer. Distribution, which is largely the task of health officials and non-governmental organizations (NGOs), has proved particularly difficult. The problem lies in how to successfully monitor the supply chain while still minimizing costs, and so far, no optimal solution has been found. There is also the issue of drug resistance, which makes finding the next new breakthrough antimalarial all the more vital, and designing an effective vaccine as also crucial (Kingsbury, 2009). See also Sasson (2008).

The Glaxo-Gates antimalaria vaccine

Researchers have been trying for more than 70 years to develop a vaccine against the elusive malaria parasite (*Plasmodium*) without notable success. Two studies carried out in East Africa showed that the most advanced candidate vaccine, developed by GlaxoSmithKline (GSK) Biologicals – RTS, S – with the assistance of Bill and Melinda Gates Foundation, cut illnesses in infants and young children by more than half and could safely be given with other childhood vaccines that are already routinely administered throughout Africa.

This antimalaria vaccine had been tested for the first time in 1992 in the United States and Belgium on adult volunteers. It had been developed since 1987 in the laboratories of GlaxoSmithKline Biologicals, the Belgium-based subsidiary of GSK. The vaccine combines the protein RTS, S of the malaria protozoan and the hepatitis B surface antigen. In 1995, the vaccine was tested in Gambia and Kenya. But it was in 2001 that the vaccination project started after the setting up of a partnership with the Malaria Vaccine Initiative (MVI) [Vincent, 2010 a].

MVI was launched in 1999 with the help of a subsidy from Bill & Melinda Gates Foundation. It is one of the programmes of the not-for-profit international health organization PATH, whose goal is to accelerate the development of antimalaria vaccines and to make them available in the countries which need them most (Vincent, 2010 a).

GSK Biologicals had been funding the development of a vaccine for military personnel and travellers, but was unwilling to undertake paediatric studies without a financial partner. Bill and Melinda Gates Foundation came to rescue and made a contribution of US$107.6 million up to mid-December 2008. GSK stated it had spent about US$300 million and expected to invest US$50 million to US$100 million more to complete the project (*International Herald Tribune*, 16 December 2008, p.6).

In 2004, the results of a vaccination campaign carried out in the south of Mozambique and including more than 2,000 children between 1 and 14 years of age, have shown that the vaccine had a 49% efficiency over a period of 18 months in the case of severe forms of the disease. By mid-December 2008, in the *New England Journal of Medicine* (*NEJM*) were published the results of three other vaccination tests carried out in Kenya and Tanzania. The vaccine was injected to young children as well as to infants less than one-year old, at the same time as the usual vaccines recommended by the World Health Organization (WHO) for infants and young children; it

was found that over a period of eight months, the RTS, S vaccine reduced by 53% the risk of malaria clinical periods. This efficiency had never been obtained before in the case of an antimalaria experimental vaccine (Bejon et al., 2008). The editorial of the issue of *NEJM* where the results of the vaccination campaign were published called the vaccine performance a "hopeful beginning towards prevention of the disease" (*International Herald Tribune*, 16 December 2008, p.6).

In May 2009, phase-3 clinical trials of the vaccine were initiated in Bagamoyo, Tanzania, under the aegis of PATH, on a group of infants between 5 and 17 months of age. They have been followed by similar trials on six-months-old infants and they were being pursued in 11 vaccination centres distributed in seven sub-Saharan countries (Tanzania, Burkina Faso, Gabon, Ghana, Kenya, Malawi and Mozambique). The trials were to be carried out for two years on a total of 16,000 young children. The total cost of these clinical trials amounted to US$210 million (Benkimoun, 2009 c; Vincent, 2010 a).

According to Christian Loucq, the MVI director, "these trials were expected to confirm the level of effectiveness of earlier trials, that was 53% among infants between 5 and 17 months of age, as well as the decrease in the number of severe malaria cases and the general impact of the vaccine on the population's health." The candidate vaccine RTS, S is the result of 20 years of research and development at GSK Biologicals. "This would be the first human vaccine against a parasite," said Joe Cohen, one of GSK researchers who worked on the development of RTS, S (Benkimoun, 2009 c).

The phase-3-trial promoters underlined that they will be conducted in accordance with the good practices adopted internationally and that the children's parents will be requested to give their agreement after receiving all the necessary information. C. Loucq stated that "the first results were expected after 12 to 18 months; thereafter the vaccine will be submitted to the European Medicines Agency (EMEA) and if the Agency's scientific advice is favourable, the vaccine will be submitted to WHO for the prequalification procedure; if WHO authorizes the commercialization of the vaccine, it could be sold to UNICEF with funding from the Global Alliance for Vaccines and Immunization (GAVI)" (Benkimoun, 2009 c).

The Malaria Vaccine Initiative (MVI) had received US$175 million for the experimental vaccination with RTS, S in 2008-2009, while GSK had announced that, when the vaccine is marketed, its profit margin would be just 5% of the production cost. Taking due account of the final results of the experimental vaccination trials and of the time necessary for the

authorizations to be delivered by WHO, the industrial production of RTS, S might begin in 2015 approximately. However, in order to increase the efficiency of future vaccination campaigns (it should be recalled that it took about 15 years to make the antihepatitis B vaccination an effective one), WHO and MVI were working in partnership with the health ministries of several countries with a view to designing an adequate decision-making framework (Vincent, 2010 a).

Chagas' disease

In 1909, Carlos Chagas, a Brazilian physician, described the disease that bears his name, the pathogen and the vector's cycle. He carried out an outstanding research work that established at once the linkage between the American trypanosomiasis or Chagas' disease, the flagellated protozoan named *Trypanosoma cruzi* (as a hommage to Oswaldo Cruz, the founder of the Oswaldo Cruz Institute in Rio de Janeiro), and the triatomid insects that transmit the pathogen through their bites.

In ten years, the occurrence of the disease – the fourth in terms of numbers of infected persons in South America – fell by 70%. This is mainly due to the control of the vector, but these efforts are not equally distributed throughout the subcontinent. *Trypanosoma cruzi* infects all mammals including humans. Traces of infections by *T. cruzi* have been found in tissues of Andean mummies dating back to 7,000 years BC. This shows the very long coexistence between the pathogen and humans. The human host is mainly contaminated by the dejections left on the skin by the insects when they bite. After a short acute phase, the disease remains invisible for years. It becomes a chronic disease, affecting above all the functioning of heart and digestive tract. It is not easy to cure and it is sometimes fatal.

Prevention is considered the best way to control the disease. Since 1991, the countries of the South Cone (Brazil, Chile, Argentina, Uruguay, Paraguay and Bolivia) have been participating in regional programmes under the World Health Organization, aimed at controlling the insect vectors. The latter live in the holes of walls made of dried mud as well as in the thatch roofs of rural dwellings in Latin America. Insecticide sprays and a better management of the habitat had an outstanding impact : there is not a single case of transmission of Chagas' disease in Uruguay, Chile and in the majority of the States of Brazil. By contrast, the endemic disease persists in Argentina, Paraguay and Bolivia, where wild relatives of the main insect vectors and resistance to insecticides make the efforts of health workers more difficult.

As the antivector control has shown its limitations, other research tracks are being followed, particularly by the scientists of the French Research for Development Institute (IRD). Thus, in Bolivia, under the Tibo project, François Noireau and Frédérique Brénière were trying to understand the role of wild populations of *Triatoma infestans*, the main vector of the disease, in the epidemiology of the illness, in order to evaluate the risk they represented for human health; subsequently, recommendations were to be made to local health authorities. IRD researchers also focused their attention on the insecticide resistance of the vectors and on their capacity to adapt to new constraints. These insects vectors show a morphological plasticity when they move from the natural environment to that transformed by humans. Domestic insects are generally smaller and more asymmetrical than the wild relatives of the same species. This "domestication" process, and its implications for the evolution of the vectors were being studied (Noireau, 2009).

Population genetics is another research area. IRD researchers have learned that the genetic variability of the parasite is considerable and that genotypes were subdivided into six groups that could be used for the study of the epidemiological tracking, i.e. monitoring parasite strains in space and time, as well as for clinical studies, development of vaccines and new drugs (Noireau, 2009).

In addition to studies on the populations of the parasite and its vectors, monitoring of human populations may also lead to improvements in the health situation. There is another mode of contamination that is rising in the statistics about Chagas' disease : the transmission of the parasite from the mother to the foetus. The study of this mode of transmission includes the diagnosis of the disease in the pregnant mother, then in the newborn and the treatment of the latter in case of contamination. Among 600 pregnant women in Bolivia, 33.9% were diagnosed as harbouring the parasite (Noireau, 2009).

Regarding the diagnosis of the disease, an IRD team has developed and patented a technique aimed at identifying proteins secreted by *Trypanosoma cruzi*. One of these proteins causes strong immune reactions in humans and could be used in a diagnostic kit. In 2009, the recognition of this protein by the serum of infected patients was being tested. Currently, there is a dearth of reliable and very sensitive diagnostic tools. With respect to treatment, several years were needed before new drugs against the disease would be commercialized. Multinational drug corporations did not consider that the search for new drugs against Chagas' disease was a priority, and the World Health Organization

had put the illness on the list of neglected diseases. However, several bioactive compounds were being tested in preclinical stages. For instance, an alkaloid, extracted from a medicinal plant in Paraguay, has shown antiparasite activity, and IRD and the Drugs for Neglected Diseases Initiative were working together in order to optimize the pharmacological properties of the compound (i.e. efficiency, synergy or antagonism with other drugs) [Noireau, 2009].

Chagas' disease affected 9 million people in endemic regions of Latin America in 2009 and 60 million people (i.e. 25% of the whole population of Latin America) were considered at risk of being infected. There were 300,000 new cases annually and 13,000 persons died from the disease every year (2009).

INTERNATIONAL AID FOR HEALTH : WHICH PRIORITIES?

Streamlining international aid for health

In 2000, the international community made an unprecedented commitment to halve poverty rates in the world by 2015. Such commitment has been formulated through eight Millennium Development Goals (MDGs). The United Nations Secretary-General, Ban Ki-moon, organized a summit in New York from 20 to 22 September 2010, in order to renew the 2000 commitment and accelerate the progress made to achieve the MDGs (while about 15 countries were able to meet them, many will not and consequently needed more assistance from the international community).

With respect to health, the fourth Millennium Development Goal aimed to reduce child (under five years old) mortality by two-thirds. In 1990, the global mortality rate amounted to 89 deaths per 1,000 births (alive newborns). In 2009, this rate fell to 60 deaths per 1,000 births. During the same period, the number of deaths among children under five years also decreased : 8.1 millions in 2009, compared with 12.4 millions in 1990 and 16 millions in 1970, worldwide.

United Nations bodies considered that this progress was due to advances in maternal education. Four diseases against which treatments exist– pneumonia, diarrhoea, malaria and HIV/AIDS – were the causes of 43% deaths among children under five years that occurred in 2008. More efforts aimed at controlling these diseases would lead to a decrease in infant mortality. In several regions, at least a 50% decrease in that mortality has occurred, e.g. in North Africa (-68%) and East Asia (-58%). In Bangladesh, Erythrea, Laos, Madagascar, Nepal and East Timor, the decrease in mortality has been at least 60%. While, in 2009, 31 countries had a child (under five years) mortality rate of 100 deaths per 1,000 births, compared with 52 countries in 1990, sub-Saharan Africa was still the region where mortality rates were the highest : one child out of eight died before its fifth anniversary in that region, this rate being twice the average in developing countries.

The fifth Millennium Development Goal aimed to reduce the mortality of mothers by 75%. According to the United Nations' data, 1,000 women die every day in the world because of diseases associated with pregnancy. However, progress has been made : the number of women dying further to complications of their pregnancy or during newborn delivery has decreased by 34%, from 546,000 in 1990 to 358,000 in 2008 worldwide. However, international institutions noted that the annual rate decrease was twofold lower than the rate that would allow to meet the Millennium Development Goal. To achieve this objective, an annual decrease of 5.5% would be needed, while the 34% decrease recorded since 1990 corresponded to an average annual decrease of just 2.3%. It was estimated that the delay in achieving the fifth Millennium Development Goal was the largest in comparison with other MDGs.

It should be recalled that the main causes of mortality of pregnant women are well known : serious haemorrhage during delivery, infections, hypertension and risky abortion. Out of 1,000 women dying from these causes every day in the world, 570 were living in sub-Saharan Africa, 300 in South Asia and only 5 in high-income countries. The risk to die from an ailment associated with pregnancy is 36 times higher for a woman living in a developing country than for a woman from a developed country. In addition, the reduction in the number of pregnancies among teenagers was at standstill, which heightens the risk of death for young mothers.

The sixth Millennium Development Goal, which aimed at stopping the spread of HIV/AIDS and malaria, was the one which, according to the United Nations, has recorded encouraging results. Thanks to unprecedented funding efforts and also to the political commitment of several developing countries, the struggle against HIV/AIDS pandemic has made significant progress over the last decade. The number of persons dying from HIV/AIDS or newly infected individuals was on the decreasing trend, while the number of patients having access to anti-HIV/AIDS treatments was increasing. However, the objective to achieve in 2010 the universal access to treatment has not been reached : of the 15 million seropositive people who needed treatment in low- or intermediate income countries, about 10 million did not receive any. The pandemic seemed to be stabilized in most regions, even though the prevalence of disease was rising in Eastern Europe, Central Asia and other regions of Asia, because of a high rate of new infections. Sub-Saharan Africa remained the most affected region, with 72% of new infections by HIV in 2008.

Regarding malaria, the sixth MDG foresaw in 2000 to reverse the trend in 15 years. This has been achieved, as in more than one-third of African

countries stricken by this disease, the number of sickened persons had been reduced by more than 50% between 2000 and 2008.

The results were less optimistic in the case of tuberculosis : 1.8 million people died from the disease in 2008, including one-third of them infected by HIV. Fostering research on tuberculosis has become even more urgent because of the very fast development of pathogens that are multiresistant to current treatments.

For a number of experts, there is a need to streamline international aid for health in order to achieve the relevant Millennium Development Goals, taking account of priorities. Thus Mickey Chopra, chief of health at UNICEF, which has been trying to put diarrhoea back on the global health agenda, stated : "All the attention has gone to more glamorous diseases, but this basic thing has been left behind; it is a forgotten disease." This observation is at the heart of a wide-ranging debate over whether the United States and other wealthy nations are spending too much on AIDS, which requires lifelong treatments, compared with diarrhoea and pneumonia, both leading killers of children and which can be treated rather inexpensively (Dugger, 2009).

Recent data have documented remarkable progress in reducing child mortality and treating patients suffering from AIDS. Foreign assistance, despite its shortcomings, is helping save millions of lives. But the economic crisis has heightened the need to streamline foreign assistance. The American president Barack Obama has proposed a 2% increase in AIDS spending for 2010 and a 6% rise for maternal and child health, according to the Global Health Council, but the disparity in American spending on AIDS and the major child killers remained important. For instance, in Africa's two most populous nations, Nigeria and Ethiopia, the total number of people who died from AIDS in 2007 – 237,000 – was far less than 540,000 children under five years of age, who died from diarrhoea and pneumonia. But in 2009, the US$750 million the United States had been spending on AIDS in both countries not only dwarfed the US$35 million it was spending on maternal and child health, but was also more than the US$646 million it was spending on maternal and child health worldwide (Dugger, 2009).

"AIDS is still underfunded, no question," stated Jeremy Shiffman, a political scientist at Syracuse University who has studied global health spending patterns. "But maternal, newborn and child mortality is a tremendous tragedy and gets peanuts," he added. Ezekiel J. Emanuel – a bioethicist, White House official and brother of Rahm Emanuel, American president's

chief of staff at that time – has contended that international aid for health was limited and would save more lives if future increases focused on maternal health and diseases that kill young children. He wrote in the April issue of the *Journal of the American Medical Association*, that such choices were necessary "if the United States is going to shoulder the burden of choosing which lives to save in the developing world" (Dugger, 2009).

However, Jeffery Sachs, of Columbia University, countered that wealthy donors still spent far too little on global health and rejected what he called the wrong-headed idea that "we need to make a terrible and tragic choice between AIDS or pneumonia." "Rather than tearing down what is working, we should continue to invest in what is needed," he added (Dugger, 2009).

If enacted, the American president's pledges to increase global aid for health would ensure AIDS remain the United States' dominant global health priority, constituting over 70% of its global health spending. International commitments to fight HIV/AIDS rose at an average annual rate of 48% from 1998 to 2007, reaching US$7.4 billion and making up almost half of all donor financing for global health, according to J. Shiffman's analysis of data from the Organisation for Economic Cooperation and Development (OECD). Yet, more than half the people suffering from the disease who needed drug treatment still were not receiving it, according to UNAIDS (Dugger, 2009).

The number of women and children who die of easily preventable or curable conditions is even higher. Pneumonia alone killed 2 million children under five years of age, and diarrhoea an additional 1.5 million – more than AIDS, malaria and measles combined – out of the almost 9 million young children who perished in 2008. There were 360,000 maternal deaths. Only four in ten of those who needed the oral rehydration salts that could prevent death received it. Public health experts are in agreement that there is tremendous potential to substantially lower child deaths from diarrhoea and pneumonia. For an extra US$3.4 billion in coming years, children in poor countries could be vaccinated against pneumonia and the rotavirus that caused about a third of diarrhoea deaths, according to Global Alliance for Vaccines and Immunization (GAVI) [Dugger, 2009].

Prevention of diarrhoea caused by rotaviruses

In 1963, a rotavirus was described as one of the main causes of diarrhoea among infants and young children. Ten years later, Bishop and co-workers could identify the virus in the mucosa of six children showing the symptoms of gastroenteritis. In 1974, Flewett who observed the virus on electron micrographs gave it the name of rotavirus, because of its wheel appearance. Since then with the improvement of laboratory techniques designed to identify the virus, it was agreed that rotaviruses were a main cause of diarrhoea among young children (Vargas Duarte and Deschamps Blanco, 2009).

About 25% of deaths due to diarrhoea throughout the world are caused by rotaviruses. Before the vaccines against these viruses were used on a large scale, 600,000 to 900,000 children died from diarrhoea annually, i.e. 6% of all deaths of children below five years of age (Vargas Duarte and Deschamps Blanco, 2009).

Gastroenteritis due to rotavirus is an inflammation of the intestinal tract, where the virus infects mature enterocytes. In the latter, the replication of the virus is very high and the virions that are liberated in the intestine lumen can be excreted in the faeces at a concentration of 10^{12} particles per gram during a period of four to ten days. Excreted rotaviruses are quite resistant to environmental conditions, which increases the risk of transmission among children (Contreras Cordero et al., 2009).

About 75% of infants become infected by rotaviruses before they are one year old. Some 50% of children show a respiratory syndrome at the beginning of the disease, with moderate fever. Although there is consensus that rotaviruses are transmitted via the faecal-oral route and through contaminated vomited liquids, the improvement of sanitation services does not drastically reduce the infection rate; the respiratory tract may be therefore involved in virus transmission. After a short incubation period, that lasts 24 to 48 hours, patients start having a watery diarrhoea and vomiting; these symptoms lead to dehydration and put the children's life at risk. During the active stage of the disease, an enterotoxin encoded by the virus is produced and induces the loss of water and electrolytes. Patients may be forced to evacuate faeces and water up to 30 times a day, with vomiting at a variable periodicity (Contreras Cordero et al., 2009).

Rotaviruses belong to the Reoviridae family that includes nine genera. All these share the common feature of having a segmented genome. Rotavirus genome is made of 11 segments of double-stranded RNA, which

encodes six structural proteins and six non-structural proteins. Structural proteins are organized in three concentric layers that protect the viral genome. The external layer is made of two proteins called VP4 and VP7 (i.e. viral proteins 4 and 7), which trigger the production of neutralizing antibodies in the host. Fifteen serotypes of VP7 and 13 serotypes of VP4 have been identified. Serotypes defined by VP7 are known as G serotypes (because it is a glycosylated protein), while those defined by VP4 are known as P serotypes (because the protein is susceptible to a protease). A binominal system has been set up for identifying the strains circulating during an epidemic outbreak (Contreras Cordero et al., 2009).

Consequently, 19 G genotypes and 28 P genotypes have been identified among the strains of rotaviruses. In human beings, the most frequent ones are G1-G4 and G9, and genotypes P8, P4 and P6. The combinations which are most frequently isolated from infants are: G1P8, G2P4, G4P8 and G9P8 (Contreras Cordero et al., 2009). See also Santos and Hoshino (2005).

There are two vaccines against rotaviruses. Rotarix™ is a monovalent vaccine, developed from a strain of rotavirus isolated in a child with gastroenteritis in 1992, in Cincinnati, Ohio. The pathogenic strain was called 89-12 and has a dual genotype, G1P8. This strain was cultivated in Vero cells (kidney cells of the African green monkey). Rotarix is used in many countries throughout the world. In Mexico, for instance, where circulate genotypes G1-G4 and P8, this vaccine has been used since 2006. Due to the presence of the P8 genotype, the vaccine triggers the production of neutralizing antibodies by children who are not only infected by G1P8 rotavirus (homologous to the vaccine strain), but also by those who are infected by strains belonging to genotypes G3P8, G4P8 and G9P8. Genotype P8 is therefore important in inducing crossed immunization against strains with different G genotypes. Rotarix is administered twice, each time with 10 million cellular culture infectious doses ($CCID_{50}$). Firstly, breastfed infants, 6 to 12 weeks old, receive the vaccine; then, one month later, they receive the second shot. Before use, the vaccine must be stored in a refrigerator in order to maintain the efficacy of the vaccine strain (Bernstein, 2007).

The other vaccine is RotaTeq; it is used in several countries, including the United States. It is a pentavalent vaccine, that is developed from a bovine rotavirus strain (WC3) and four strains of human rotavirus (genotypes G1P8, G2, G3 and G4). These strains are the most important ones from the epidemiological viewpoint. The pentavalent vaccine is therefore made of the most important G genotypes and of genotype P, which is found in 80% of rotavirus strains. This vaccine is administered in the form

of three oral doses, starting between the sixth and 12th week after birth, and thereafter the second and third doses are given at one or two months intervals (Offit and Clark, 2006).

Contreras Cordero et al. (2009) underlined that in Mexico and the United States the use of monovalent Rotatrix and pentavalent RotaTeq vaccines respectively, had considerably reduced the number of cases of gastroenteritis.

Cholera

Cholera is still a scourge in many countries, where poor sanitation and the lack of sufficient safe drinking water are the main causes of the spread of the disease, caused by the bacterium *Vibrio cholerae*. Cholera outbreaks or even epidemics often occur after natural disasters such as floods or earthquakes, when the systems of water distribution and sewage are destroyed.

Cholera has spread worldwide during the 19th century from its original reservoir : the Ganges delta in India. The following six pandemics have killed millions of people throughout all continents. The 2010 pandemic, the seventh one, started in South Asia in 1961. According to the World Health Organization (WHO), cholera causes 100,000 to 120,000 deaths per year (Caroit, 2010).

In 2010, an epidemic has been stricking Central Africa where more than 1,900 deaths were recorded by early October 2010. In 2008 and 2009, an outbreak of cholera killed 4,300 persons in Zimbabwe (Caroit, 2010).

In Haiti, cholera has been spreading since mid-October 2010, including in the capital Port of Prince. According to the information released by the health ministry on Monday 6 December 2010, 2,071 deaths due to the cholera outbreak had been recorded until that date. More than 90,000 cases have been registered since mid-October and the progression of the disease did not seem to slow down. According to the World Health Organization (WHO), the number of cases could reach up to 400,000 during the following 12 months. The non-governmental organzation *Médecins sans frontières* ("Medical doctors without frontiers") indicated it had treated 47,000 persons suffering from the disease since the beginning of the outbreak. Two-thirds of the deaths had been recorded in the north of the capital, where the epidemic started, before spreading to the whole country.

On 12 November 2010, the United Nations made an appeal to set up an emergency fund of US$163.9 million (€120 million) in order to stop the spread of the disease. The emergency programme was to be carried out by five United Nations agencies, as well as by the International Organization for Migrations and 42 non-governmental organizations, which will support Haiti's health ministry.The programme was designed on the basis of WHO's forecast that at least 200,000 persons would be affected by the epidemic. Production of safe drinking water, sanitation and waste treatment were the most important items of the programme (US$89 million), followed by medical treatment (US$43 million) [Caroit, 2010].

The last major cholera epidemic in Latin America occurred in 1991, starting in Peru; it caused the death of 4,000 persons and made ill about 400,000 people in16 countries. It would be difficult to determine the origin of the epidemic in Haiti, where cholera has been absent for more than a century. Neighbour countries were increasingly concerned about the risk of contagion, starting with the Dominican Republic which shares with Haiti the island of Hipaniola. A cholera outbreak in this country where more than a million of Haitian immigrants are living would dramatically affect tourism, a major economic sector, particularly during the high season starting in December 2010 (Caroit, 2010).

The spread of cholera and the origins of its regional epidemics or global pandemics underline the important issue of the geographic origin and history of pandemics that have affected humankind. They are the subject of recent studies which rely on research on the genomics of pathogens, as well as on DNA analysis of skeletons of people who died during pandemics. This is the case, for instance, of pest, which had appeared 2,600 years ago in Asia before spreading to other continents. An international study (Morelli et al. in *Nature Genetics*, 31 October 2010) has analyzed 17 complete genomes and 286 strains of *Yersinia pestis* from different regions of the world. The phylogenetic tree of the bacterium, that has been reconstitued from its genetic mutations and their rhythm, shows that its origin is in China or near China. Thereafter, the bacterial pathogen spread to Western Europe through the Silk Road, and then to Africa and finally to North America by the end of the 19th cntury. One of the co-authors of the study, Thierry Wirth of the French National Scientific Research Centre stated that "this evolutionary study was the broadest ever carried out on a bacterial disease."

Another example is that of exanthematic typhus, caused by *Rickettsia prowazekii*. DNA has been extracted from the tooth pulp of the skeletons exhumated in northern France (Douai); its analysis showed that the

persons were not killed during the battles which took place around the city between 1710 and 1712, during the war of succession of Spain; they were killed by bacteria such as *Bartonella quintana* and *R. prowazekii*. This supported the assumption that typhus and trench fever (*B. quintana*) were introduced into Europe by Spanish conquistadores at the beginning of the 16th century (Nguyen-Hieu et al., *PLoS One*, 27 October 2010).

Vibrio cholerae can remain dormant for long periods and thereafter proliferates. This means that even in the absence of epidemic outbreaks of the disease, rivers, lakes and oceans can still contain this bacterium, which infects three to five million people every year and causes about 120,000 deaths. Thanks to the pioneering work of Rita Colwell, a microbiologist and emeritus professor of the University of Maryland and Bloomberg School of Public Health (Johns Hopkins University), scientists can now better predict the environmental conditions that induce the pathogen to evolve from the dormant to the infectious stage, and therefore to correlate the changes in the natural environment with disease propagation. Rita Colwell's work was carried out during the 1960s. She has worked for many years in South Asia, Latin America and Africa. She was the first to try to elucidate the impact of climate change on the spread of infectious diseases, and to develop satellite models with a view to locating and anticipating their outbreaks (Vincent, 2010 b).

Rita Colwell, 76 years old in 2010, has been awarded the Water Prize of Stockholm 2010. This award is given every year to persons or institutions that have accomplished a major contribution to the preservation of water. In the case of Rita Colwell such prize, awarded to her in Stockholm on 9 September 2010 by King Carl XVI Gustaf of Sweden during the Water Week, rewarded a career devoted to the control of water-transmitted diseases. Rita Colwell also chaired the US National Science Foundation (NSF) from 1998 to 2004, and played an important role in the support of basic scientific research, not only in her country but also at international level (Vincent, 2010 b).

When she was informed about the award, she stated that "the Water Prize of Stockholm was recognizing the merits of a research career devoted to water safety and health, and particularly the studies on cholera, initially controversial, but were finally recognized as correct." She added that "safe drinking water is a critical factor for economic and social stability, and even for a country's national security," and recalled that progress remained to be achieved in this area (Vincent, 2010 b).

Tuberculosis

In 2009, 9.4 million new cases of tuberculosis have been recorded worldwide, including 1.1 million cases of persons co-infected with HIV. Since 2004, the number of tuberculosis patients per inhabitant has been decreasing slowly. In 2009, about 1.7 million persons died from tuberculosis, including 380,000 that were co-infected with HIV; this figure meant 4,700 deaths per day. In 2008, about 440,000 new cases of the disease that were multiresistant to the usual antituberculosis drugs had been recorded, and 150,000 of them were fatal.

In 2009,the geographic distribution of tuberculosis patients was : 35% in South-East Asia, 30% in Africa, and 24% in the Western Pacific region.

Two articles published in *Science* on 14 May 2010 were dealing with these aspects. In one of them, Christopher Dye of the World Health Organization (WHO) and Brian Williams of the South African Centre for Epidemiology, Modelling and Analysis (Stellenbosch) have stressed that progress had been real : more than 36 million persons had been successfully treated worldwide and 8 million lives had been saved over the period 1995-2008, according to WHO. The objective to treat 85% of patients had been more than achieved in 2007-2008. One of the Millennium Development Goals concerning the reduction by 50% of the 1990 levels of prevalence and mortality of tuberculosis should be reached in four of the six regions identified by WHO. But the situation was not satisfactory in the two other regions : sub-Saharan Africa and South-East Asia. C. Dye and B. Williams underlined that "80% of the 9.8 million new cases of tuberculosis expected in 2010 will be recorded in the 20-25 countries most stricken by the disease and one-third will be recorded in India and China" (Dye and Williams, 2010; Benkimoun, 2010 e).

Regarding the diagnosis of tuberculosis, two methods are currently used : direct microscopic examination of spittings of ill or supposedly ill persons, and bacterial culture. But "direct examination of spittings only detects 10% to 25% of carriers of the Koch bacillus, which means that 75% to 90% of those carrying the bacterium are undetected," stated Monique Guéguin, a physician and biologist at *Médecins sans frontières* – MSF ("Medical doctors without frontiers"). The second method is more reliable, but an average two months are needed in order to obtain a clearcut result; this period would be reduced to about three weeks with a higher-performing technique. Consequently, the treatment is generally initiated on the basis of the likely occurrence of tuberculosis. In addition, the forms of the illness resistant to the main pharmaceuticals are discovered even later (Benkimoun, 2010 m).

After 18 months of testing, the World Health Organization announced on Wednesday 8 December 2010 its support for a new rapid diagnosis test for tuberculosis. The test is carried out with a machine dubbed GeneXpert, which detects the Koch bacillus DNA in spittings of ill or supposedly ill persons in about 100 minutes. The test has been developed by the Californian company Cepheid, which is also located in France, near the city of Toulouse (Benkimoun, 2010 m).

The development of a new *fast* diagnosis test for tuberculosis, and affordable in developing countries, has been launched by the Foundation for Innovative and New Dignostics (FIND), created in 2003, and involving a wide range of public and private partners, including the Bill and Melinda Gates Foundation and New Jersey Medical University. The company Cepheid was also involved in the project and FIND was able to negociate a 75% deduction on the price of the machine so as to reduce its cost to US$17,000 (€13,000) as well as that of the test : less than €13, compared with €15-23 for the diagnosis based on bacterial cultures. FIND estimated that the cost of the test would decrease even more when the new detection method, now officially recommended by WHO, is widely used (Benkimoun, 2010 m).

These cost regulations will apply to the public sector of 116 low- or intermediate-income countries, that are heavily hit by tuberculosis. Large humanitarian organizations such as MSF will also benefit from these lower costs, as well as local non-governmental organizations recognized by the health ministries. The GeneXpert machine, of the size of a coffee machine, entirely automated, will deliver the result of tuberculosis tests in less than two hours and will also allow the detection of Koch bacilli resistant to rifampicine – the main drug used in the current treatment of the disease. Other resistances could be detected in the future. According to Mario Raviglione, director of WHO's department "Halt to Tuberculosis", "the new test has a high sensitivity, even though it is slightly lower than that of bacterial culture; it reaches 99% when it is compared with the detection of the Koch bacillus through direct examination of spittings and over 70% when the search is negative" (Benkimoun, 2010 m).

The new diagnostic has two constraints : first, the need to have an electric supply and, secondly, the cost of the machine and tests. "The method could be used in peripherical laboratories and not just in a reference laboratory located in a country's capital," underlined M. Raviglione. But it cannot be used in remote places. "This is a fantastic tool," stated Cathy Hewison, a medical adviser on tuberculosis at MSF, "but it will not solve all our problems." In fact, despite the predictable commercialization of new drugs against

tuberculosis by 2013, all experts agree that the real breakthrough would be the development of a new antituberculosis vaccine that will replace the current BCG vaccine with limited effectiveness (Benkimoun, 2010 m).

In addition to a more efficient diagnosis, other factors explain the difficulties to control tuberculosis. The first one concerns the limits of the epidemiological model. This "standard model" distinguishes a rapid stage, followed by a slow phase of the infection. The model is reductionist; it makes easier the epidemiological calculations and facilitates the treatment of patients. Its drawback, however, is to rely on the evolution of tuberculosis in developed countries and it was not very successful in developing countries. C. Dye and B. Williams also mentioned that some strains of *Mycobacterium tuberculosis* were transmitted more easily than others and that individuals were not equal versus tuberculosis, due to genetic characteristics. The treatment of tuberculosis is also compounded by the simultaneous presence of chronic diseases such as diabetes or AIDS (Dye and Williams, 2010).

In the second article published in *Science* on 14 May 2010, David G. Russell (Cornell University), Clifton E. Barry (National Institutes of Health) and JoAnne L. Flynn (University of Pittsburgh) have insisted on the need to design "new intervention strategies applicable in countries that most needed them." They made some assumptions regarding the variable immune protection of the BCG vaccine : the fact that it has become too attenuated to induce the expected immune reaction, or the role played, in India, by the exposure of children to other mycobacteria in their environment. They suggested new vaccine strategies while awaiting a new vaccine : improvement of the BCG through the addition of antigens that induce a stronger immune response; use the tuberculosis pathogen after withdrawing the genes responsible for its virulence, instead of using *Mycobacterium bovis* like in the BCG; amplification of the initial immune response (prime boost) [Russell et al., 2010].

Regarding the treatments presently followed during six months, D.G. Russel and his colleagues stressed "the urgent need in countries with low income for drugs acting rapidly and effectively." These drugs should also be used to attack the mycobacteria when the infection is latent and not only when the pathogens are multiplying, which is presently the case. Finally, D.G. Russel and his colleagues insisted on "the complete lack of biomarkers indicating the status of the disease," that would enable the physicians to predict its evolution towards an active form, and on the crucial importance of having "a system of health care with appropriate and well managed resources" (Russell et al., 2010).

On 13 October 2010 a study published in *Science Translational Medicine* revealed that early-stage trials showed a candidate antituberculosis vaccine might work against multi drug-resistant *Mycobacterium tuberculosis* strains (Bertholet et al., 2010; *The Economist*, 2010 l).

Emerging diseases in sub-Saharan Africa

Sub-Saharan Africa is a region where emerging diseases have been and are being noticed and followed through. For instance, in 2009, researchers of the French Research for Development Institute (IRD) and their colleagues and partners of the Mediterranean University have discovered that the bacterium *Tropheryma whipplei* was often found in the faeces of Senegalese children. It has been detected in 44% of 2-to-10-years old children living in two villages of the southwest of the country. These results could be extrapolated to the rural regions of Senegal and even to the whole of sub-Saharan Africa. Children are contaminated at a very young age (less than two years). This bacterial pathogen could therefore be emerging in developing countries and might become a major threat to public health. Not only it can induce gastroenteritis and several infections, but it also causes Whipple's disease – a lethal infection if not treated (Fenollar et al., 2009).

By mid-2009, some 1,000 cases of Whipple disease have been reported. The disease was considered a rare one, but its prevalence (i.e. the percentage of infected persons in a population at a certain period) seems to be increasing. IRD researchers have analyzed faecal samples of 150 children in good health, two months to 10 years old, and living in the villages of Ndiop and Dielmo in the southwest of Senegal : 44% of the children were tested positive, and 37% of those who were 8 months to 2 years old, and 11% of infants who were less than 8 months old. These prevalence rates were much higher than those recorded in Europe, Asia and America. In addition, in developed countries, very few children of less than 2 years of age carry the pathogen (Fenollar et al., 2009).

Researchers also analyzed the water of eight wells that supplied drinking water to the two villages. All the water samples did not contain any pathogen. However, contamination through the water stored in clay recipients was not excluded. Contamination would not be only due to the oral-faecal route. Recent studies have shown the presence of *T. whipplei* in sewage water, and 12% to 26% of workers employed in the collection of waste-waters and sanitation are healthy carriers of the bacterium. Tests aimed at detecting the pathogen in saliva are also being carried out (Fenollar et al., 2009).

While it was thought a few years ago that *Tropheryma whipplei* was rare, recent studies have shown that in Europe 1% to 11% of adults carry it. In addition, many children (2-4 years old) with gastroenteritis harbour the pathogen in their faeces. The IRD study has brought evidence that about half of the children living in rural zones of Senegal, and most likely in sub-Saharan Africa, carry the pathogen (Fenollar et al., 2009).

Tropheryma whipplei, still badly known, can cause gastroenteritis as well as other serious illnesses such as endocarditis (inflammation of the inner membrane of the heart), meningoencephalitis, and Whipple's disease, a severe chronic infection that is lethal if not treated with antibiotics. The disease can affect any organ and its major symptoms are joint pain, chronic diarrhoea, neurologic sequellae. However, all the persons carrying the pathogen do not necessarily become ill. Many of them are healthy carriers. On the other hand, Whipple's disease seems to affect white males, about 50 years old (Fenollar et al., 2009).

Large-scale studies are to be carried out in Senegal, and IRD scientists expect to find prevalence rates of the disease of the same order as those recorded in the villages of Ndiop and Dielmo. Due to the frequence of the pathogen, Whipple's disease could become an emerging disease in sub-Saharan Africa. The research work carried out by IRD aims at identifying the diseased persons and at treating them better (Fenollar et al., 2009).

Vaccines as the most cost-effective global public health measure

Endorsing vaccines as the world's most cost-effective public health measure, the Bill and Melinda Gates Foundation announced on Friday 29 January 2010 that it would more than double its spending on them over the next decade, to at least US$10 billion. Vaccines were already receiving more funding from the Foundation than any other cause. B. Gates stated no money would be shifted away from other projects, like improved crops, assistance to small businesses, and schools and libraries. Instead he and Warren Buffet will increase their annual gifts to the Foundation, and about 30% of all spending, up from 20%, will be for vaccines (McNeil Jr, 2010 a).

The decision made by the Foundation could save the lives of as many as eight million children by 2020, B. Gates calculated. "Vaccines are a real success story," stated B. Gates in an interview before the announcement, which he made at the World Economic Forum in Davos, Switzerland : "The cost is tiny, and yet it saves more than any other component of a health care system." In calculating that eight million lives could be saved,

B. Gates cited a computer model developed for the Foundation by public health specialists at Johns Hopkins University. Whether such a prediction comes true depends on several factors. Firstly, 90% of the world's children should be vaccinated against childhood diseases like measles, diphtheria, whooping cough and poliomyelitis. Almost 80% now are vaccinated, but with 134 million children born each year, it is a constant struggle to keep abreast. Secondly, B. Gates assumed that two new vaccines against rotaviruses and pneumococcal disease, which are major killers of malnourished children, were adopted as routine immunizations in most poor countries and reached 80% of all children by 2020. The model also assumed that a malaria vaccine currently in development by GlaxoSmithKline would be approved and would by 2014 reach at least some of the one million children, mostly in Africa, who die annually from the disease (McNeil Jr, 2010 a).

Julian Lob-Levyt, the executive secretary of the Global Alliance for Vaccines and Immunization (GAVI) stated : "If other donors follow the lead of the Bill and Melinda Gates Foundation and set up their funding for vaccines," GAVI has the ability to immunize millions of children against the world's two biggest childhood killers, pneumonia and diarrhoea" (McNeil Jr, 2010 a).

DRUGS OF THE FUTURE

Genomics and drug discovery; pharmacogenomics

Big pharmaceutical companies have seen their innovation pipelines dry up over recent years, but rapid advances in genomics can help change this situation. For instance, in cancers, biomarkers, which are genetic indicators different from SNPs, are emerging as one of the most effective means of improving the efficiency of drug discovery, according to an OECD report.

For example, during trials of an apparently unsuccessful drug for lung cancer made by the British company AstraZeneca, David Agus of the University of Southern California (USC) discovered that the drug worked well in some of his patients of Asian descent. Similarly, some people of African origin seemed to respond well to a heart drug, Bi-Dil, whereas those of other ethnic groups or stock reacted less well to the medicine. It is surprising, however, that the drugs industry seems indifferent to genomics applications, except in cancer research. Russ Altman of Stanford University explained that the industry rather believed in the "blockbuster" model with its huge potential market, i.e. in standard remedies that work for everyone. That was understandable when there were fewer tools to evaluate the likely risks and benefits of new drugs, but advances in pharmacogenomics make this model unsustainable (*The Economist*, 2009 a).

R. Altman's team published a study in *The New England Journal of Medicine* (*NEJM*) on how to dose warfarin, a drug widely used to prevent clots that could lead to strokes or heart attacks, but the correct dose can vary widely from patient to patient. Too high a dose can cause bleeding, whereas too low a dose can lead to deadly clots. The study showed that dosing decisions that took account of variations in just two specific genes in addition to factors like age, weight and race produced much better results than decision based only on the latter conventional factors (The International Warfarin Pharmacogenetics Consortium, 2009).

The NEJM and *The Lancet* also published by early January 2009 three articles concerning the individual variations of the response to a drug, clopidogrel, used as a blood-thinner and to prevent the formation of new clots in patients who suffered from a heart attack. These studies showed that those patients who carried at least a copy of a variant of a gene involved in the metabolism of clopidogrel (up to 30% of the population examined), did not respond well to the treatment and ran a higher risk of new heart ailments. The US Food and Drug Administration (USFDA) intended to update the information sheet on this drug – the world's second mostly sold drug – in order to mention the likely interference of genetic factors (Benkimoun, 2009 a).

The French team whose work was published in *NEJM*, including Philippe-Gabriel Steg of the department of cardiology of Bichat hospital in Paris, has been working on the data contained in a register created by Nicolas Danchin of Georges-Pompidou hospital in Paris, and which records the treatments and evolution of patients that suffered from a heart attack and were admitted in intensive-care units across France. Thus the researchers have monitored more than 2,200 patients and looked into the association between the presence of a variant for different genes involved in the intestinal absorption, metabolism and biological activity of clopidogrel, as well as the risk of death during the year following the registration of the patient on the national register. It was found that 30% of the patients carried one or two variants of gene *CYP2C19*, which controls the transformation of clopidogrel into active metabolites. Among 2.6% of the patients examined, both copies of gene *CYP2C19* varied and the outcome was a decrease in the gene functional capacity. In this smaller group, heart ailments (after the heart attack) were twofold more numerous than in the population not showing genetic variation. The frequence of cardiovascular ailments occurring after an angioplasty carried out to enlarge the diameter of coronary arteries, was multiplied by 3.6 among the carriers of two copies of the genetic variants. Consequently, two of the authors of the study (Céline Verstuyft of Bicêtre hospital and Simon Tabassome of Saint-Antoine hospital in Paris) concluded that the medical treatment will have to increasingly rely on genetic tests (Wallentin et al., 2009).

In another study published in *NEJM*, a team of Harvard University Faculty of Medicine and Eli Lilly company has monitored 1,500 patients treated with clopidogrel for an acute coronarian syndrome. The risk of dying from a heart failure was increased by 53% among carriers of at least one variant of gene *CYP2C19*. Among the patients who had been operated to receive a stent (a tiny spring which maintains the artery diameter), the risk of thrombosis was three times higher for those carrying at least one

variant of the same gene. These results were in agreement with those of a French team led by Gilles Montalescot of the Pitié-Salpêtrière hospital in Paris, that were published in *The Lancet*. They monitored 259 patients, who were less than 45 years old and who suffered a heart attack, and they found three times more coronary ailments after the heart attack, due to the presence of just one variant of gene *CYP2C19*. This presence was considered by the French physicians a major determinant of the prognosis among young patients taking clopidogrel after a heart attack (Montalescot et al., 2009).

According to G. Montalescot, genetics shows its great relevance in the treatment of coronary diseases. It does not necessarily mean that genetic tests will become systematic for patients who suffered a heart attack. The results of the above-mentioned studies should be validated among larger populations of patients, but in the years to come genetic information should be taken into account. This trend will have an economic impact and will impinge on the cost-effectiveness of treatments. In the short term, generic versions of clopidogrel will become available, while costly competitors of the drug will be marketed. To reserve the latter for the minority of patients among whom clopidogrel would be less effective, and to prescribe the generics to the majority where clopidogrel works well, would result in substantial savings (Montalescot et al., 2009).

Daniel Vasella, Novartis' chief executive officer, while rejecting the concept of personalized medicine ("it would be economic folly for firms to develop a special pill for every patient"), reckoned that linking individual genetics (or genomics) with specific therapies was a major challenge for the pharmaceutical industry nowadays, but he was still looking for a suitable business model. According to *The Economist*, perhaps a better way of describing how genomics will change the drug business model would be mass customization. Clayton Christensen of Harvard Business School suggested a useful phrase to describe the point where pharmacogenomics and personalized medicine meet : "precision medicine" (*The Economist*, 2009 a).

Pharmacogenomics, then, is a type of predictive medicine that could be useful for patient and company alike. It allows prescriptions to be safer and more effective, and enables firms that want to test molecules through clinical trials to restrict the tests to people who are likely to respond well. That makes trials less costly and more likely to succeed. It also ensures that the drug, once approved, is prescribed only to those who will benefit from it (*The Economist*, 2010 j).

Approval of a drug produced by genetically engineered goats

Many of the newer protein-based (or monoclonal antibody-based) drugs, like the anticancer drugs Avastin and Erbitux, or the anti-arthritis drugs Enbrel and Humira, are produced in genetically engineered Chinese hamster ovary cells (CHOC) grown in stainless-steel bioreactors. A cell-culture factory costs hundreds of millions of dollars to build, while using livestock shrinks the investment to tens of millions of dollars, according to Geoffrey Cox, chief executive of GTC Biotherapeutics, Framingham, Massachusetts (Pollack, 2009). In addition, if the production of the pharmaceutical must be raised, the number of livestock heads is incremented, while with cell-culture bioreactors their increase is much more costly.

The US Food and Drug Administration (USFDA) had approved by early 2009 the first drug produced by genetically engineered livestock, potentially opening a new era in pharmaceutical production ("pharming"). Made by GTC Biotherapeutics, the drug is produced by a herd of 200 goats living under quarantine in a high-security farm in central Massachusetts. The goats have been bred to contain a human gene encoding the synthesis of an anticlotting drug in the mammary gland of the animals. The human protein can be extracted from goats' milk and processed into the anticlotting drug. The human gene for antithrombin was linked to goat DNA that normally controls the production of a milk protein, so as to ensure that the antithrombin would be produced only in the milk (Pollack, 2009).

The gene was injected into a goat embryo, which was then implanted in the womb of a surrogate mother. A more reliable technique would be to introduce the gene into the nucleus of a skin cell of a desired animal and then fuse it with an enucleated ovocyte to make an embryo and clone. Dolly the sheep, the first cloned mammal, had not been created when GTC Biotherapeutics initiated its work in the early 1990s. Once a goat with the human gene coding for the production of antithrombin in its milk had been produced, that "founder" animal was mated with others through conventional breeding to start a herd (Pollack, 2009).

Antithrombin is sometimes in short supply or unavailable for pharmaceutical use because of a shortage of human plasma donations. People with antithrombin deficiency are vulnerable to blood clots. They generally reduce the risk by taking blood thinners. But during surgery or childbirth, blood thinners are usually not used because they increase the risk of excessive bleeding. GTC Biotherapeutics stated one genetically

engineered goat could produce as much antithrombin in a year as could be obtained from 90,000 blood donations (Pollack, 2009).

A similar drug, which had been approved in 2006 in Europe but had not been widely adopted, was the first to have been cleared by the USFDA under guidelines the agency adopted in January 2009 to regulate the use of transgenic animals in the nation's drug and food supply. Sales of the new drug, ATryn, were expected to be modest, but GTC Biotherapeutics' major goal was to show that approval of a drug produced by transgenic animals could be obtained. ATryn was sold in the United States by Ovation Pharmaceuticals. The drug was approved mainly for people born with a rare hereditary deficiency of antithrombin to prevent blood clots while they undergo surgery or childbirth. The FDA determined that ATryn was as effective as antithrombin derived from human plasma in preventing clots. However, the protein derived from plasma lasts longer in the body than the one from goats, probably because the sugars associated with the protein are different (Pollack, 2009).

Other drugs produced in animals are under development. The Dutch company Pharming planned to apply in 2009 for approval in the United States of a drug produced in the milk of transgenic rabbits to treat hereditary angioedema, a protein deficiency that can lead to dangerous swelling of tissues. Another company, Pharmathene, working under a US Defence Department contract, is developing a treatment for nerve-gas poisoning in the milk of transgenic goats (Pollack, 2009).

Turning animals into pharmaceutical factories has its obvious advantages for the health-care system, particularly for decreasing costs of treatment, but some animal-rights activists and environmental advocates have raised objections. For instance, the Humane Society of the United States stated that "it is a mechanistic use of animals that seems to perpetuate the notion of their being merely tools for human use rather than sentient creatures," in its position paper on the practice. Other concerns are that the animals could be harmed, that animal germs might contaminate the drug, and that the milk or meat from genetically engineered drug-producing animals might enter the food supply. Such transgenic animals might escape and breed with other animals, spreading the human gene with unpredictable consequences (Pollack, 2009).

Yet, the USFDA's stance on GTC's drug, issued on Friday 6 February 2009, had eliminated one obstacle to producing drugs in animals : companies' uncertainty over whether the FDA would ever approve such drugs (Pollack, 2009).

It is true that the newly appointed head of medicine approvals at the USFDA has insisted that science rather than politics or pricing is behind the growing difficulties that face new pharmaceuticals in reaching the market. Janet Woodcock, head of the Center for Drug Evaluation and Research, one of the most senior jobs at the FDA, rejected claims from drug companies that the centre was rejecting new medicines because the agency had become too risk-averse, partly in response to politicians and consumer groups which have attacked it for approving medicines and failing to identify dangerous side-effects, such as with the painkiller Vioxx of Merck (Jack, 2008).

J. Woodcock acknowledged that the rate of success in bringing new drugs to market had declined in a way that was putting pressure on pharmaceutical executives and diminishing investor interest in the sector as existing medicines came off patent. She stated, however, that when the FDA rejected new drugs, it did so based on an assessment of their safety and efficacy, not as a result of lobby or political pressure, or greater conservatism. She conceded that the size and length of clinical trials had grown in recent years, but argued that was a result of better understanding by regulators how best to identify risks (Jack, 2008).

She denied industry claims that the FDA was turning down experimental drugs simply because they were more expensive or less cost-effective than existing treatments. While endorsing a move towards greater post-marketing monitoring studies – with continued obligations on companies to study safety risks even after a drug has been launched – she allayed concerns about their extent (Jack, 2009).

J. Woodcock defended the use of the FDA's gold standard of placebo-controlled trials, where a new drug is tested against a sugar pill rather than an existing approved medicine. "If you compare with a treatment that is not proven to be effective, you do not know if either drug is effective," she stated (Jack, 2009).

Pharmaceuticals and vaccines produced by genetically engineered plants

After several years of research on the production of antibodies (plantibodies), vaccines and pharmaceuticals by genetically engineered plants, the current trend is to focus on tobacco for that production instead of edible plants, e.g. tomato, potato and banana. The main reason is the possibility to produce large volumes of biomass and to use plant viruses as vectors of the genes encoding the vaccine or the drug. For instance,

tobacco mosaic virus (TMV) has a high expression rate : in two weeks, large quantities of a monoclonal antibody could be produced.

In 2006, Charles Arntzen of the University of Arizona, a renowned scientist who has been working for many years on the production of bioactive compounds by genetically engineered plants, has been able to produce a vaccine against poultry Newcastle virus disease in tobacco plants. The vaccine was commercialized in the form of tablets, which allowed for the delivery of optimal doses.

The first drug produced by genetically engineered tobacco plants was likely to be the enzyme glucocerebrosidase, used to mitigate the ailments due to Gaucher disease. The plant-derived enzyme was expected to be commercialized in 2011-2012, after the completion of phase-3 clinical trials.

The Norwalk virus or norovirus, also dubbed the "cruise virus," causes a very infectious disease that has a profound impact on the rhythm of life, working hours and economic activities; it can also lead to the closure of hospitals, geriatric centres and medical polyclinics. Charles Arntzen redesigned phytoviruses in order to produce nanoparticles with a diameter of 25 nanometers (nm), i.e. the same size as that of the norovirus, in tobacco plants. These particles are only made of the surface protein of the virus, i.e. the virus component that is recognized by the human immune system, i.e. the antigen. At the 2009 238th meeting of the American Chemical Society, C. Arntzen reported that these particles can be used as a vaccine and administered every 12 or 18 months in order to control new strains of the norovirus; this virus, like the influenza virus, can mutate frequently, and the production of a vaccine in plants could be a fast and cheap method to control the virus and the associated disease.

The Owensboro-based Kentucky Bioprocessing was working on the industrial scale production of an antibody-vaccine against non-Hodgkin's most common variety of lymphoma. A B lymphocyte evolves into cancerous cells in this disease that multiply in swollen lymph nodes, instead of producing antibodies. To control such a disease, it is necessary to develop a specific immunotherapy for each patient, because every B lymphocyte is distinct in each human being. The antibody sequence could be transferred to a plant virus, which multiplies in a plant (tobacco) for about six weeks to produce sufficient quantities of antibody. The latter would be used in a new therapy instead of the conventional chemotherapy. Kentucky Bioprocessing has licensed the technology to Bayer AG which was to build a US$50-million factory to produce the antibody.

Another example of the flexibility and velocity of the production of vaccines and pharmaceuticals by genetically engineered plants is that of the company Vax & Inc., Arizona that develops vaccines against Norwalk virus. This company has estimated the cost of vaccine production in plants could be reduced by US$10 million before testing the vaccine in clinical trials.

The key advantage of vaccine or drug production by genetically engineered plants is the reduction of costs, which is precisely a crucial objective of health-care systems. New vaccines produced in this way would greatly contribute to the Global Alliance for Vaccines and Immunization (GAVI), which estimated the global needs for vaccines at US$1.6 billion a year in 2008.

THERAPIES OF THE FUTURE

Gene therapy

Gene therapy, conceived at the beginning of the 1970s, aimed at curing diseases caused by the mutation of a single gene (monogenic diseases) through the introduction of the non-defective version of the gene into the affected cells or tissues, using a vector. Such gene therapy, if successful, could provide a long-lasting solution instead of providing continuous medical treatments, either to control the disease, or to avoid rejection in the case of bone-marrow transplants. Despite the scepticism shown by some geneticists, trials were carried out, with the help of patients' associations and media. For instance, in France, the French association against myopathies fully supported these innovative endeavours (Benkimoun, 2009 l).

Unfortunately, most trials failed. However, in France and Italy, gene therapy was applied to treat a severe immune deficiency caused by the lack of the enzyme adenosine deaminase (ADA) which forced the children affected to live in a sterile bubble. In France, Alain Fischer and Marina Cavazzana-Calvo (Necker hospital in Paris) had to interrupt their clinical trial further to the detection of a form of leukaemia that caused the death of one of the children under treatment (Benkimoun, 2009 l).

Since then, however, there have been good news. Not only for the gene therapy of ADA deficiency (undesirable side-effects were due to the vector used to transfer the non-defective version of the gene), but also for the treatment of Parkinson's disease and Leber's congenital amaurosis (causing congenital blindness in infants), and for the treatment of adrenoleukodystrophy associated with chromosome X in November 2009. The results with gene therapy were equivalent to those obtained with bone-marrow transplants (Benkimoun, 2009 l).

Adrenoleukodystrophy

Leukodystrophies affect one newborn out of 2,000 in France. This group of diseases affect the cells of the central nervous system that synthesize the myelin sheath surrounding the nerves. In adrenoleukodystrophy, the X chromosome bears a mutant of gene *ABCD1*. Only male newborns are affected by the disease, which gradually evolves between 5 and 12 years and can be detected by brain imagery, without clinical symptoms. Later on, the patient becomes seriously affected in terms of mobility and intellectual activity. In 1993, the gene involved in the disease had been identified by Patrick Aubourg (French National Institute for Health and Medical Research, INSERM) and Jean-Louis Mandel.

P. Aubourg conceived a gene-therapy trial in order to treat those patients for whom there was no available compatible bone marrow donor. This clinical trial gave encouraging results and was followed during 24 and 33 months respectively for the two children who were treated. The final results of the clinical trial, carried out by Nathalie Cartier and P. Aubourg (INSERM), were published in the 6 November 2009 issue of *Science* (Cartier et al., 2009).

The gene-therapy trial consisted of collecting bone-marrow stem cells from each child from peripheral blood. These cells that can differentiate into the wide range of blood cell lines, were exposed to a lentiviral vector derived from HIV, which can penetrate into cell nuclei. This vector bore the non-defective version of the *ABCD1* gene. Once reinjected in the bloodstream of both children, stem cells with the normal gene could multiply and some of them were found in the brain. It was also shown that the process of demyelinization of nerves was stopped after a year. Neurological and cognitive functions remained stable during the follow-up period, and the overall results were comparable to those of a successful bone-marrow transplant, without the drawback of providing lifelong medical treatment against the reject of the transplant (Cartier et al., 2009).

This work has mobilized a large team including M. Cavazzana-Calvo and A. Fischer, and was financially supported by the European association against leucodystrophies. The first lesson drawn was the confirmation that it was possible, using a lentiviral vector, to transform bone-marrow stem cells. According to P. Aubourg, an important transformation, estimated at about 15%, has been achieved, while this proportion was only 0.01% in the case of gene-therapy trials applied to “bubble children.” The French team is trying to increase the percentage of transformed cells

containing the normal gene. The lentiviral vector could be used in other genetic diseases, such as beta-thalassaemia or sickle-cell anaemia (Cartier et al., 2009).

The second lesson drawn concerned the safety of the technique. The integration of the lentiviral vector into the patient's genome did not result into harmful secondary effects, because, once integrated, the vector was self-inactivated. With the assistance of Christof Von Kalle, of the German Cancer Research Centre in Heidelberg, the various sites of the vector integration could be monitored, and P. Aubourg concluded that the vector was harmless, although one should continue to monitor the process (Cartier et al., 2009).

The third lesson drawn was that this clearcut success was expected to give a new impetus to gene therapy, as stated by Luigi Naldini of the San Raffaele Telethon Institute for Gene Therapy in Milan, whose comment was published in the same 6 November 2009 issue of *Science*. There are of course queries about the long-term efficiency and innocuity of gene therapies, the means to control the precise insertion of the normal gene into the patient's genome. In this regard, several techniques are being tested in order to make a real "gene surgery" and target the site of the genome where the normal gene will be inserted (Benkimoun, 2009 l).

Leber's congenital amaurosis

An international team of scientists, led by a group at the University of Pennsylvania, used a genetically engineered virus to introduce the correct version of a gene called *RPE65* into six patients suffering from a retinal disease known as Leber's congenital amaurosis. In four patients, the vision improved. Katherine A. High, of the Howard Hughes Medical Institute in Maryland and one of the directors of the study, reported in the *New England Journal of Medicine (NEJM)*, reckoned the treatment could be used more widely. It offered hope for correcting any of the ten genetic defects that could cause Leber's disease, as well as some forms of *retinitis pigmentosa*, another genetic eye disease (Maguire et al., 2008).

Viruses that carry the gene can trigger strong immune reactions – which has led to the death of Jesse Gelsinger, and 18-year-old American who had a faulty gene that prevented the synthesis of a liver enzyme that breaks down ammonia. In 1999, he was the first person to be publicly identified as having died in a clinical trial for gene therapy. Viruses can also cause genetic mutations when they integrate themselves into human DNA. Of the 27 people treated for a rare and severe immunodeficiency disease,

known as SCID, worldwide four have died from leukaemia, according to L. Seymour, although this needs to be balanced against the fact that children with SCID are completely lacking a normal immune system and die in early childhood (*The Economist*, 2008 a).

Research is therefore focused on the viral vectors. One way of doing this is to create viruses that lose their activity to activate local genes when they are integrated into their host's genome. Another way, used in Leber's disease trial, is to rely on viruses that integrate themselves only into the cell, rather than the cell DNA. At Oxford University, L. Seymour is working on "stealth viruses," which are coated in a polymer that hides the virus from the immune system. This allows the modified virus to circulate for a longer time in a patient's bloodstream and thus have a better chance of hitting tumours disseminated around the body. Worldwide, several research teams are trying to develop synthetic polymers to deliver genes, thereby avoiding the need to use viruses (*The Economist*, 2008 a).

Beta-thalassaemia

On 16 September 2010, *Nature* published the outstanding reults of a French and American team, led by Philippe Leboulch of INSERM, University of Paris-South and the French Atomic Energy Commissariat, concerning the cure of beta-thalassaemia, a serious blood genetic disease. The patient who has been treated successfully did not receive any blood transfusion for almost three years (which is the case of patients suffering from this disease, who need to receive blood every month). In addition, the cured patient had a normal professional activity (Cavazzana-Calvo et al., 2010).

Thalassaemia affects 5% of the world population and is particularly prevalent in the Mediterranean Basin, the Middle East, Asia and sub-Saharan Africa. This disease is due to one or two defective copies of the gene coding for beta-globin, a key protein component of haemoglobin. Severe forms of the disease cause anaemia, which demands regular blood transfusions. This was the case of the patient treated by the French research team; of Vietnamese and Thai origin, he used to receive blood transfusion every month since his childhood. His health status was monitored by Françoise Bernaudin, a physician and professor at the Intercommunal Hospital of Créteil, south of Paris. He could have received a bone-marrow transplant, as an alternative solution to blood transfusions, but the difficulty to find a compatible donor and the risks of bone-marrow transplant generally lead to the application of this technique to only about 20% of patients (Cavazzana-Calvo et al., 2010; Benkimoun, 2010 j).

The research team therefore decided to carry out a gene-therapy experiment, after having received in 2006 the authorization to do so by the French Agency for the Sanitary Safety of Health Products (AFSSAPS). The first stage consisted of taking a sample of bone marrow of the patient, and of isolating haematopoietic stem cells, which give rise to all blood cells, including erythrocytes or red cells. These stem cells were modified using a viral vector – derived from the HIV that was made innocuous – carrying the correct gene for beta-globin. The viral vector was produced by the biotechnology company Bluebird Bio, created by P. Leboulch. The HIV has the advantage of penetrating into the host-cell nucleus and integrating into its genome. Before the transformation of the haematopoietic stem cells by the viral vector carrying the normal gene, a chemotherapy treatment helped to eliminate all the cells carrying the defective version of the gene encoding beta-globin.This initial stage was carried out by Marina Cavazzana-Calvo and Salima Hacein-Bey-Abina of INSERM – Hospital-Necker-Enfants malades and University Paris Descartes (Cavazzana-Calvo et al., 2010).

The following stages, including the intravenous injection of healthy cells into the patient, the monitoring of his health status and analysis of his blood cells, were implemented by Eliane Gluckman (Hospital Saint-Louis, Paris), Françoise Bernaudin and Frédéric Bushman (University of Pennsylvania). It was found after a few months that blood cells contained sufficient beta-globin so that no blood transfusions were needed for about two years. These good results led AFSSAPS to authorize the treatment of a second patient in January 2010. Like the first experiment, the second one will receive funds from the French association against myopathies (AFM) [Cavazzana-Calvo et al., 2010; Benkimoun, 2010 j].

Gene therapy and cancer

A Chinese company, Shenzhen SiBiono GeneTech, has tried to replace faulty tumour-suppressor genes with their correct versions. In 2003, the treatment for head and neck cancers, which accounted for about 10% of the 2.5 million new cancer patients in China every year, obtained the first approval of a gene-therapy treatment. Some experts outside China have criticized the quality of the data used to support this therapy, while L. Seymour of Oxford University reckoned that when used with chemotherapy in some situations, it could be effective (*The Economist*, 2008 a).

Another approach is not exactly gene therapy, but rather “virotherapy,” i.e. the use of viruses that selectively attack cancer cells. In 2006,

researchers of the Hebrew University, Jerusalem, isolated a variant of Newcastle virus disease, a highly contagious and deadly disease in birds. That variant could target cancer cells in humans. Trials on a form of aggressive primary brain tumour have shown one complete regression out of 14 treated patients (*The Economist*, 2008 a).

Katherine A. High, of the Howard Hughes Medical Institute in Maryland, has expressed her optimism about the future of gene therapy. She highlighted that progress in gene therapy should be put into context : the development of bone-marrow transplantation or monoclonal antibody treatments took several decades. Because these treatments are based on biological processes they are more complicated than the use of chemicals of the pharmacological "armamentarium" (*The Economist*, 2008 a).

Gene therapy to cure depression?

In the Antiquity the origin of depression was believed to be in the black bile (hence the word melancholy). For Anglo-Saxons it was the spleen, this word also meaning the depressive humour of poets during the romantic period. The French poet Baudelaire wrote a poem entitled *Spleen*, where he described the lengthy periods of anxiety that afflicted the mind. Siegmund Freud in his assay, *Sorrow and Melancholy* ("Deuil et mélancolie"), related depression with the loss of a dear person, and more broadly of an ideal, which generated a feeling of culprit. Modern medicine is still seeking the profound causes of depression (Le Hir, 2010 b).

In France, a survey made between 2005 and 2008 by the National Institute for Prevention and Health Education and involving 6,500 persons has shown that 18% of French people had undergone at one stage of their life "a major depression period." Every year, 2 million patients suffer from the illness, the number of women affected being twice that of men. And 10% of those suffering from depression commit suicide (Le Hir, 2010 b).

Depression has many facets and physicians prefer to speak of "forms of depression." Symptoms range from sadness to morbid ideas, through loss of interest, memory dysfunction, disturbance of sleep and appetite, demeanor of personality or culprit feeling. "All psychiatric pathologies are multifactorial," stated Monica Zilbovicius, research director at the French National Institute for Health and Medical Research (INSERM) : "genetic vulnerability, involving several genes, interacts with the person's environment." The fact that a direct relative of a depressive individual is at risk of being affected three times more frequently is not a proof of heredity of the illness. More significant is the risk, four times higher

among real twins than among false ones, to show similar symptoms of the disease. But lonely persons are more exposed to depression, as well as those who are jobless or in precarious conditions (Le Hir, 2010 b).

Most forms of depression are treated with the available drugs, with psychotherapy if deemed necessary. Gene therapy may be a solution, but not in the near future, as shown by a study published in the 20 October 2010 issue of *Science Translational Medicine* and cosigned by 13 biologists, led by Michael Kaplitt, associate professor at Weill Cornell Medical College, and cofounder of the American biotechnology company Neurologix Inc. This company, listed on the stock exchange, had acquired the license of a patent filed by Cornell University on gene therapy of severe depression involving gene P11 (Alexander et al., 2010).

The experiments carried out by Brian Alexander (Weill Cornell Medical College, New York) on six young male mice, consisted of inactivating in a tiny region of the brain called *nucleus accumbens* the gene P11, using a virus. The protein encoded by gene P11, called protein p11, regulates the signal transmitted to brain cells by serotonin – a neuromediator involved in the regulation of humour, sleep and memory. The researchers have then observed the movements of the rodents when they suspended them by the tail or put them in a recipient full of water and from which they could not escape. They observed that animals gave up more rapidly swimming or moving – a form of resignation that is classically observed in animal models of depression. This observation was confirmed by decreased appetite for a sweet beverage, which recalled anhedony (lack of sensitivity to pleasure) of depressed people (Alexander et al., 2010).

The American-Swedish team then made the reverse experiment : they reintroduced in the brain of mice, with the help of another virus, the normal gene P11, in order to restore protein expression. They noted that the animals recovered normal movements as well as their attraction for the sweet beverage. Also the researchers examined the brain tissues of 34 human corpses, half of them having suffered from depression and the other half not, and they discovered in the *nucleus accumbens* of the former a lower concentration of protein p11 (Alexander et al., 2010).

They concluded that in both mice and humans the *nucleus accumbens* and gene P11 played a key role in the illness, and that a gene therapy could be envisaged for "patients showing a severe form of depression, that is resistant to the usual antidepression treatments" (Alexander et al., 2010).

Specialists have reckoned the importance and relevance of the study, while others, including three researchers of the American pharmaceutical firm Johnson & Johnson, underlined that several issues must be tackled before supplying gene therapy to cure severe depression. First, the extrapolation of the animal model to humans may be inappropriate, because the experiment has been carried out on 11-week-old mice, which would mean an early treatment in humans – teenagers, i.e. probably before the appearance of the symptoms of depression. On the other hand, the observation, *post mortem*, of the lack of protein p11 in the brains of depressive patients, raises the well-known issue whether this anomaly is the cause or, conversely, the consequence of the illness (this is also the case of other psychiatric pathologies like schizophrenia or autism). The American-Swedish study has focused on one gene (P11), while depression is most probably related to several genes. It remains to be demonstrated that gene P11 plays a more important role than any other one, and in all forms of depression. Finally, the study suggests a strictly biological approach to a dysfunction of behaviour, while physicians consider that depression results from a range of personal, social and environmental factors relating to trauma, stress and life conditions (Le Hir, 2010 b).

However, despite all these observations, the results of the study have the merit of establishing the importance of a very specialized region of the brain in various forms of depression, and of confirming its role, because attempts of deep stimulation of the *nucleus accumbens* with electrodes have already been carried out in cases of severe depression resistant to drug treatment. Broadening the treatment of this illness through gene therapy would have a considerable impact, because depression is becoming the second cause of invalidity after cardiovascular diseases, according to the World Health Organization, and because it resists to antidepression drugs in one patient out of three (Le Hir, 2010 b).

Cancer therapy : a multiple approach

Over 7.5 million people die from cancer in the world every year, and more than 12 million new cases had been identified in 2007. Without major improvement in therapies, the forecasts were 17.5 million deaths and 27 million new cases in 2050.

In the United States, 1.4 million persons were diagnosed with one form of cancer in 2008, and it was estimated that 565,650 would die from the disease that same year, according to the American Cancer Society. Cancer is thus overtaking heart and cardiovascular diseases as the first killer in the United States. Because the incidence of cancer increases with

age, the nearly 80 million baby boomers reaching their 60s by 2008 will probably drive the number of cancer patients and deaths even higher. At current rates, 1 in 2 men and 1 in 3 women will eventually have some form of cancer diagnosed (gender disparity is explained by the fact that men smoke more than women) [Saporito, 2008].

Understanding the disease forms and mechanisms

Cancer is not one disease, it is many of them, each with different mechanisms that make the therapies so difficult. The most pernicious forms of cancer – among them, pancreatic, lung and brain – are still invincible. Survival rates in rare forms of cancer are also very low. Even though progress has been made in the prevention and early detection of cancer, overall the death rate from cancer in the United States dropped just 5% from 1950 to 2005. During the same period of time, deaths from heart and cardiovascular diseases fell 64% (Saporito, 2008).

There is therefore a greater sense of urgency among researchers, patients and social actors and decision-makers, with a view to speeding up the transformation of knowledge into therapies that should be made available to the cancer patients more rapidly. Some groups in the United States demand radical changes in the funding of research designed to deliver breakthroughs rather than the incremental improvements that are more typical of mainstream science. Physicians and scientists understand the frustration and "something needs to be done differently," stated Tyler Jacks, director of the David H. Koch Institute for Integrative Cancer Research at the Massachusetts Institute of Technology (Saporito, 2008).

But requesting a radical change should not mean denying the progress made in the understanding and treating the disease. According to Harold Varmus, chief executive officer of Memorial Sloan Kettering Hospital in New York City, a former director of the National Institutes of Health and a Nobel Laureate (research on lung cancer), "the rate of discovery has been phenomenal"… "we feel we understand some of the basic principles. We understand the tissue environment" (Saporito, 2008).

For instance, some of the latest anticancer drugs result from that understanding. For instance, Gleevec, a monoclonal antibody – imatinib – is used to treat one form of leukaemia by acting on the Philadelphia chromosome (Ph), which directs bone marrow to keep making abnormal white blood cells. Because of drugs like Gleevec and therapies such as bone-marrow and stem-cell transplants, there were around 12 million people in the world in 2008 who were classified as survivors (Saporito, 2008).

Unravelling the relationships between cancers and genomics

One important approach to better understanding the disease is to unravel the relationships between cancer and genomics. A key initiative in this regard was the creation of the International Cancer Genome Consortium on 29 April 2008 (see p.102).

The goal of the consortium is to gather and share the most precise data on the genome of about 50 most common types of cancer during the forthcoming decade. Each consortium member was expected to make a thorough analysis of genetic modifications in at least one type of cancer. Each cancer project will deal with samples from about 500 patients and would cost US$20 million approximately. The consortium will make the data available, rapidly and free of charge (the results will not be patented), to the world scientific community. All national research bodies concerned were invited to participate in the work of the consortium.

A team of human genome scientists led by Peter Campbell and Mike Stratton at the Sanger Institute near Cambridge, which is funded by the Wellcome Trust, the world's biggest medical research charity, announced on 17 December 2009 that they had sequenced a "cancer genome" for the first time. They are part of the International Cancer Genome Consortium. The Sanger Institute's scientists analyzed cells stored from two patients who had died from cancer. One was a 55-year-old man with small-cell lung cancer and the other was a 45-year-old man with malignant melanoma, the most lethal form of skin cancer. The researchers took a cancerous cell and a healthy cell from each patient and sequenced the full genome of all four cells. They repeated the exercise many times to make sure they had a correct final sequence, consisting of 3 billion nucleotides of the full human genome (Connor, 2009).

The researchers found that the lung cancer cell had 22,910 DNA mutations that the healthy cell from the same patient did not possess. These mutations in the lung must have accumulated during the lifetime of the patient, many as a result of exposure to cigarette smoke. The same would be true for the 33,345 mutations identified in the cancerous skin cells of the 45-year-old man with malignant melanoma, although most of the mutations here are presumed to have been caused by exposure to sunlight. Both of these "cancer genomes" showed where the mutations occurred and in which of the chromosomes of the cells. Some of them involved quite big changes to the DNA molecule, such as rearrangement of hundreds of thousands of nucleotides. But others were the smallest change possible, a shift for instance in one nucleotide base to another,

such as cytosine (C) to thymine (T) and vice versa, or an adenine (A) to guanine (G) and vice versa (Connor, 2009).

Some of these mutations were already known from previous studies to be linked with certain environmental mutagens. Tobacco smoke, for instance, often results in the mutation of G to T, whereas ultraviolet light tends to cause the mutation of C to T. But not all the mutations would have been involved in triggering the cancer. Most of them would have been harmless mutations, but some of them would have been "drivers" within the genes that are in some way involved in cancer development. In order to identify these "drivers," it would be necessary to extend the sequencing effort into other patients suffering from the same cancer, perhaps as many as 500 people to achieve statistical significance. By comparing all mutations in all patients with the same cancer, scientists will be able to identify those that appear to be common to them all, and hence likely to be involved in triggering that particular disease. Scientists have already identified more than 30 genes that play some kind of role in cancer development. This gives them a lead in terms of knowing where to search for the likely driver mutations that are probably involved in cancer etiology (Connor, 2009).

There may be as many as 200 different types of cancers, and many more subtypes. But each and every cancer involves damage to the DNA template that controls cell division. In this respect, cancer is a genetic disease, indeed it is said to be the most common genetic disease since, in the industrialized countries, it affects one in three people over a lifetime, killing as many as one in five. In the United Kingdom, for instance, it has been estimated that every 2 minutes someone was diagnosed with cancer and there were more than 293,000 new cases of cancer diagnosed each year in the United Kingdom (2008) [Connor, 2009]. By understanding the nature of these genetic mutations in a cancer cell, it should be possible to design drugs that specifically target the mutations or the outcome of the mutations, instead of developing drugs largely by trial and error. It could also lead to new methods of diagnosing cancer in the earliest stages of the disease before it becomes apparent to the patient or the physician; or new ways of finding secondary cancers lurking in the body that have evaded earlier anticancer treatment (Connor, 2009).

Malignant melanoma is one of the most aggressive cancers. If caught early, when it is a small, visible tumour on the skin, melanoma can be excised completely. Once cells have broken away from the original tumour, though, and colonized other parts of the body, the outlook is bleak: death within months and no effective treatment. That is why specialists

gathered in Chicago for the annual meeting of the American Society of Clinical Oncology (June 2010) were very interested in the presentation by Stephen O'Day from the Los Angeles Clinic and Research Institute, and Steven Hodi of the Dana-Farber Cancer Institute in Boston. These two researchers have been running a full-scale clinical trial of ipilimumab, an antibody that stimulates patients' immune systems to destroy tumours. Their hope was those tumours would include the secondaries spun off by melanoma. Although the prolongation of life offered by the antibody was not huge – three and half months on average – it was real and it was a first. Small-scale trials of other drugs have suggested such life extension, but this was the only time, though, that randomized double-blindness trails on several hundred people, have demonstrated a life extension beyond doubt for advanced melanoma (*The Economist*, 2010 g).

To demonstrate that their new drugs worked, S. O'Day and S. Hodi randomly assigned 676 people with advanced melanoma to one of three treatments : ipilimumab alone, ipilimumab plus an experimental vaccine, or the experimental vaccine alone. They found that those treated with ipilimumab or ipilimumab plus vaccine lived 50% longer than those treated with the vaccine alone – surviving an average of ten months compared with six months and half. That was, however, just an average. Almost a quarter of those treated with ipilimumab were alive two years after treatment started. That compared with 14% of those treated just with the vaccine (*The Economist*, 2010 g).

Ipilimumab is the first in a new class of drugs, called T-cell-targeted antibodies, that enable the patient's immune system to destroy cancer cells. The drug works by binding to, and disabling, a protein called CTLA4, found on the surface of the immune system's T-cells. A second T-cell-related antibody, which binds to a different immuno-suppressing protein, PD1, was being tested in smaller clinical trials. That antibody could eventually displace ipilimumab, if clinical trials showed it was equally effective and less toxic. Or, more likely, it could be combined with ipilimumab in order to be twice more effective (*The Economist*, 2010 g).

Researchers also wanted to combine ipilimumab with other drugs that interact with a protein produced by a particular mutated version of a gene. A promising one is PLX4032, a molecule which inhibits the action of the protein produced by a specific mutation of a gene called *B-RAF* (see p.105). Initial trials of PLX4032 suggested it might be as good as, or even better than, ipilimumab at improving survival of patients with advanced melanoma. A full-scale double-blind trial was under way (*The Economist*, 2010 g).

Another group, led by Bang Yung-jue of Seoul National University College of Medicine, showed that a drug aimed at a particular genetic mutation dramatically shrank the tumours in people suffering from one type of lung cancer. Two other studies, meanwhile, confirmed that a similar approach to chronic myelogenous leukaemia continued to be successful (*The Economist*, 2010 g).

A similar approach of identifying one of the key mutations that has led to a cancer arising, and attacking it, is behind the treatment of a cancer known as non-small-cell lung cancer – actually the most common form of lung cancer counting for about 80% of cases. In 2007, a group of researchers discovered that the fusion of two normally independent genes, *EML4* and *ALK*, into one aberrant gene seemed to induce the formation of tumours. Since then it has been found that fused *EML4-ALK* caused 3%-5% of all non-small-cell lung cancers (about 10,000 cases a year in the United States alone). A drug called crizotinib, which blocked the fused gene's activity *in vitro*, was already in development. At the meeting of the American Society of Clinical Oncology (June 2010), Bang Yung-jue reported the first clinical trial of crizotinib, on 82 patients whose tumours had the *EML4-ALK* fusion gene. Of those given the new drug, almost 90% showed shrinkage of their tumours and in more than half the tumours shrank at least by a third. Moreover, three-quarters of those involved were still responding to treatment after six months (*The Economist*, 2010 g).

That is significant, for one difficulty with cancer treatment in that it is constantly changing. New mutations arise that multiply the effect of existing drugs. Which is precisely what happens in the case of chronic myelogenous leukaemia. There is already a targeted treatment for this disease. Gleevec, probably the most remarkable anticancer drug of the past decade, disables the protein encoded by another fused gene, *BCR-ABL*. But Gleevec, too, can be bypassed by further mutations of *BCR-ABL* which allow the tumour cells to grow in the presence of the drug. By studying where exactly in the DNA sequence those mutations occur, and examining how Gleevec binds to the tumour-promoting protein, chemists have designed new compounds to avoid these difficulties. Two of these compounds, Sprycel (dasatinib) and Tasigna (nilotinib) have been used for several years to treat patients whose disease had progressed even though they were taking Gleevec. However, trials reported at the meeting of the American Society of Clinical Oncology (June 2010) showed that the newer drugs also worked better than Gleevec in recently diagnosed patients. Both induced faster and stronger responses than Gleevec did, although it was too early to conclude if the new molecules actually prolonged patients'

survival. The ultimate goal was to block all of a tumour's ways of genetic escape by developing drugs that deal with all the mutations that would allow such a breakout. That is the aim of the International Cancer Genome Consortium (*The Economist*, 2010 g).

Elucidation of metastasis

Another key area of research concerns the elucidation of the mechanisms that lead cancer cells to become invasive, i.e. metastatic. Robert Weinberg, professor at the Massachusetts Institute of Technology (MIT), discovered the first human oncogene – a gene that triggers the transformation of normal cell into a tumour cell – and the first tumour-suppressor gene. At the Whitehead Institute, Cambridge, Mas., he is working on cell interactions that are at the origin of cancers as well as on the processes that enable cancer cells to invade other tissues and thus become metastatic (Benkimoun, 2009 b).

It is generally agreed that at least five signal routes are disrupted when a normal cell transforms itself into a cancer cell. In contrast, an extremely complex process makes cells of a primary tumour invasive and capable of giving rise to metastases in different kinds of tissues and far away. And 99% of cancer deaths are due to metastases.

The invasion of a tissue that is distant from the primary tumour site is the result of a series of events, called "metastasis cascade," explained R. Weinberg. First, the cancer cell must have a local invasive capacity, then it has to pass into the bloodstream. In this way, it can migrate to a distant site. Once inside a new tissue it forms micrometastases, which evolve into macrometastases, to go through all these stages of the cascade. The result is not always fully successful. For instance, in breast cancers, there could be 100 or 1,000 micrometastases in bone marrow, but in 50% cases there will not be a macrometastase (Benkimoun, 2009 b).

How a cancer cell escapes from the tumour and moves into the bloodstream? Migration of cancer cells has been compared to that of embryonic cells which will initiate the differentiation of cells and tissues. When this migration occurs, embryonic cells express certain transcription factors (i.e. proteins that are necessary for the initiation and/or regulation of DNA transcription into RNA). Cells of very aggressive cancers also express these transcription factors, and this conversion is called the epithelium-mesenchyme transition (EMT). Transcription factors are activated by signals that may originate from mutated genes in the genome of cancer cells, but also from non-cancer cells of the connective tissue around the

tumour, or stroma. These non-cancer cells are recruited by cancer cells that have migrated to form new capillaries, necessary for tumour growth. When cancer cells arrive, the stroma becomes reactive and its cells return signals to cancer cells that have recruited them (Benkimoun, 2009 b).

Stem cells and cancer cells share many common traits. There are stem cells inside tumours, but it is not known when they appear. Normal epithelial cells also share many characteristics of mesenchyme cells. For the time being, there are no effective pharmaceuticals to kill cancer stem cells. For instance, the treatment of some leukaemias with Gleevec eliminates most of cancer cells, but spares stem cells. In other words, even though it increases life expectancy, it does not cure leukaemia; the main objective of the treatment is to keep the disease under control (Benkimoun, 2009 b).

Cells of cancer metastases look like epithelial cells. This leads to think that the epithelium-mesenchyme transition may be a reversible phenomenon : cells would recover some of their initial traits when they do not receive any more signals that induced the EMT. The former dogma was that to understand the transformation of a cancer cell, one should look at its genes. We now know that we must also decipher the signals it receives from its tissue environment (Benkimoun, 2009 b).

Researchers of Princeton University and New Jersey Cancer Institute have identified a gene that plays a key role in the dissemination of metastases in breast cancer. This gene is located on chromosome 8 and facilitates the adhesion of tumour cells to blood vessels in other organs than breast (Benkimoun, 2009 b).

During the formation of potentially lethal metastases, starting from a localized tumour, several genetic and epigenetic (interactions between genes and their environment) alterations occur. Several genes associated with the formation of metastases have been identified. A gene called *MTDH* (metadherine) which plays a key role in both the formation of metastases and resistance to chemotherapies, is overexpressed in more than 40% of breast cancers and is generally associated with a bad prognosis. Therefore this gene could become a target for future therapies (Benkimoun, 2009 b).

Classification of tumours

At present, oncologists assess how advanced a cancer is by taking a biopsy and examining the concentration within it of specific receptors,

such as epidermal growth-factor receptors (EGFRs), that are known to help cancers spread or making metastases. Peter Parker of the Cancer Research UK's London Research Institute had the idea of employing a technique called fluorescence resonance-energy transfer (FRET), used to study interactions between individual protein molecules, to see if he could find out not only how many receptors there are in a biopsy, but also how active they are. The technique uses two types of antibody, each attached to a fluorescent dye molecule. Each of the two types is selected to fuse with a different part of an EGFR molecule, but one will do so only when the receptor has become active (*The Economist*, 2009 j).

Pointing a laser beam at the sample causes the first dye to become excited and emit energy. With an activated receptor, the second dye will be attached nearby and so will absorb some of the energy emitted by the first. Measuring how much energy is transferred between the two dyes indicates the activity of the receptors. P. Walker's concept was applied by his colleague Banafshe Larijani, who, with her colleagues, used FRET to measure the activity of receptors in 122 head-and-neck cancers (which have an abundance of EGFRs). They found that the higher the activity of the receptors they examined, the more likely it was the cancers would return quickly following treatment. The technique was found to be a better prognostic tool than conventional visual analysis of receptor density (*The Economist*, 2009 j).

Engineers in the same group have created an instrument that automates the analysis. Tumour biopsies are placed on a microscope slide and stained with antibodies. Then the tool points the laser beam at the samples, records images of the resulting energy transfer and interprets those images to provide FRET scores. Results are available in an hour, compared with four or five days using standard methods. Having set up the standard technique with head-and neck cancer, the team hoped to extend it to breast cancer biopsies, and eventually to all solid tumours, i.e. cancers other than leukaemias and lymphomas (*The Economist*, 2009 j).

If the British researchers succeed, it would help oncologists to classify cancers biochemically and not anatomically, i.e. not by whereabouts in the body they occur, but by their molecular origin. Most cancer specialists indeed think that patients with tumours in different parts of the body that are triggered by the same genetic mutation may have more in common than those whose tumours are in the same organ, but have been caused by different mutations. The new technological approach could help make such classification routine. That could, in turn, lead to a new generation of therapies and help physicians decide which patients should receive them, and in which combinations and doses (*The Economist*, 2009 j).

Cancer treatments with nanoparticles

Packaging drugs such as Doxil (to treat ovary cancer) or taxol (to treat breast cancer) into nanoparticles (i.e. having a size of nanometers or billionths of a meter) has been used in cancer therapies for some time. This packaging (in the case of taxol, with naturally occurring blood proteins) aids the delivery of the drug and reduces its toxic side-effects. But a second generation of nanoparticles is being tested in clinical trials (*The Economist*, 2008 h).

For instance, Jennifer West at Rice University in Houston, Texas, has designed gold "nanoshells" that either absorb light or scatter it, depending on their design. The shells are built on a core of silica whose shape can be adjusted to produce the desired effect. Then a layer of gold, 15 to 20 nanometers thick, is painted over the silica core. When the nanoshells absorb infra-red light, they heat up and they would cauterize any nearby cancer cells. To deliver the nanoshells to tumours, trillions of them are injected into the patient's bloodstream, and they end up in the tumour, because the latter has abnormal blood capillaries. The pores in these vessels are larger than those in healthy tissues. Consequently, if they have the right size, nanoshells pass through the capillary pores and lodge in the tumour, but not in a normal organ. Twelve to 36 hours later, when enough shells have accumulated, an optical fibre is inserted into the tumour, and delivers an appropriate blast of infra-red light. That heats the tumour and cauterizes it. This so-called photothermal ablation has undergone six years of animal testing. In mice, tumours disappeared in 10-14 days and the animals remained cancer-free thereafter. Dogs were also tested and the results were very positive, according to Donald Payne, the executive officer of the company Nanospectra Biosciences, which was conducting trials at three medical centres in Texas on people with head-and-neck cancers (*The Economist*, 2008 g).

One advantage of photothermal ablation is the lack of a chemical agent, and therefore of toxic side-effects. However, safety issues about the particles themselves remain, but J. West and D. Payne both stated that surplus particles are cleared away by the liver, spleen and lymphatic system, or are mistaken for bacteria and phagocytated by macrophages. They may remain in those macrophages for a long time. In fact we know very little about the possible toxicity of nanoparticles that remain in the body (nanotoxicology). At the University of Texas, San Antonio, work is being carried out on nanoparticles that have been detected in the liver, kidney and even in the brain (*The Economist*, 2008 h).

MagForce Nanotechnologies, based in Berlin, has conducted three clinical trials of anticancer nanoparticles made from iron oxide and was carrying out another five in 2008. These particles were injected directly into the target tumour, rather than relying on the leaky walls of tumour capillaries to deliver them to the right place. Once there, they can be heated using alternative magnetic fields that are easily tolerated by patients. According to Andreas Jordan, the company's chief scientist, this method had no side-effects and showed particular promise in the treatment of glioblastomas and against prostate cancer (*The Economist*, 2008 h).

Other researchers prefer to use drugs delivered via nanoparticles. For instance, Azaya Therapeutics, based in San Antonio, Texas, used 100-nanometer liposomes (compared with 2,000 nm, the diameter of red cells) to deliver FDA-approved drugs, e.g. Sanofi-Aventis' Taxotere encapsulated in liposomes has a 100% effectiveness against prostate cancer and 75% effectiveness in the treatment of mesothelioma, caused by asbestos.

CytImmune Sciences of Rockville, Maryland, had begun in 2008 a study of the efficacy of Aurimune, a solid-gold nanoparticle that carries a dose of tumour necrosis factor (TNF). It attacks the blood capillaries that feed a tumour, but also healthy capillaries. With natural TNF, that does not matter, since the substance is produced only in the tumour itself. But if TNF is directly injected into a patient's bloodstream, the latter will suffer massive organ failure brought on by a drastic fall in blood pressure. The alternative option is therefore to concentrate the TNF in the tumour, via surgery (complicated and risky) or via gold nanoparticles that reach the tumour site. According to Lawrence Tamarkin, CytImmune Sciences' executive officer, the only side-effect in Aurimune safety trials was a transient fever that can be treated with over-the-counter medicines (*The Economist*, 2008 h).

Calando Pharmaceuticals, of Pasadena, California, was also using nanoparticles containing camptothecin, a substance previously too dangerous to use. The nanoparticle is made from a strand of sugar molecules and when camptothecin is attached to the sugar moiety, the strand folds up into a sphere, hiding the drug inside. The resulting particle called Cyclosert by the company was being tested to see if it could prevent the progression of ovarian cancer. Some 150 patients who had completed standard chemotherapy for this cancer, which would normally then be followed by a period known as "watchful waiting," were instead given Cyclosert. Since ovarian cancer often recurs, that trial would, by comparison with a control group remaining under watchful

waiting alone, allow the researchers to assess whether Cyclosert could reduce recurrence (*The Economist*, 2008 h).

Like Nanospectra Biosciences and CytImmune Sciences, Calando Pharmaceuticals relied on the leaky walls of tumour capillaries to deliver its products. But in the future nanaoparticles could be attached to antibodies that have a specific affinity to those proteins found on the surface of tumour cells. When infra-red light is applied to gold particles or nanoshells associated with antibodies that stick to tumour cells, only the latter would be killed. Working on particles that both scatter and absorb light, they would reveal the extent of any tumour if illuminated with a low-power beam. That would guide surgeons where to point a high-powered killing beam so that no tumour tissue is missed (*The Economist*, 2008 h).

The University of Texas, Austin, has licensed to Abbott Laboratories the technology consisting of encapsulating nanoparticles in a polymer matrix, so as to be administered orally to the patient. This was considered a good model of collaboration between basic research and clinical research, within the framework of the *Collaborative Model in Texas*.

New approaches to combat cancer

At the symposium held in the Salk Institute, La Jolla (California), from 27 to 29 October 2010, in partnership with the journal *Nature*, with a view to celebrating the 50th anniversary of the Institute, several speakers focused on cancer which is one of the priority research areas of the Salk Institute : knowledge of the processes leading a normal cell to become a tumoural one, as well as new approaches to treating the disease. David Baltimore, Nobel Laureate of Medicine and Physiology in 1975, currently professor at the California Institute of Technology (CALTECH), gave the keynote address of the symposium on the role of micro-RNAs (miRNAs) in the immune system response and tumour evolution (Benkimoun, 2010 k).

MicroRNAs are small sequences of non-coding RNA which regulate gene expression : they can inhibit the activity of messenger RNAs, which transfer the information contained in DNA to the cell machinery that synthesizes proteins. MicroRNAs can thus silence a gene or part of it whose sequence is complementary to their own sequence. If the relevant protein plays a negative role, miRNA will counter it through silencing the expression of the corresponding gene; miRNAs are therefore closely involved in a complex network of regulation of protein synthesis. The first microRNA was discovered in 1993 in *Caenorhabditis elegans*, a small

worm particularly studied by geneticists, but it was only since 2000 that it has been realized that miRNAs could be found in other living beings, including humans. Computerized models have led to an estimate of around 1,000 miRNAs in the human genome. A few hundreds have been identified up to 2010 (Benkimoun, 2010 k).

On 10 August 2010, the team of David Baltimore published in the *Proceedings of the National Academy of Sciences USA* (*PNAS USA*) an article describing the role of miRNAs in haematopoietic stem cells (HSC) which divide and differentiate into blood cells involved in the immune response and the anti-inflammatory reaction. When during the experiments the level of miRNAs was artificially increased in HSCs, the function of the cells were slowed down, or, conversely, accelerated. The moderate increase of one of these miRNAs accelerated the production of differentiated blood cells from HSCs. Conversely, a much higher expression of the relevant miRNA led to the development of a cancer in six months in mice, i.e. a very aggressive leukaemia (O'Connell et al., 2010).

The study of the expression of 200 miRNAs in blood cells (monocytes) by David Baltimore's team has shown that many of them corresponded to genes activated by a protein, called NF-kappaB. This protein is present in cells in an inactivated form; but when activated by several agents (viruses, messengers, etc.), it penetrates into the nucleus, where it triggers the transcription of certain genes, including those encoding miRNAs. "Two of these miRNAs have opposed effects," explained D. Baltimore at the La Jolla symposium. "The first one represses the inflammatory response and, if it is absent, the response will be amplified and this would favour the development of a cancer. The second one represses the inhibitors of the inflammatory response; if it is inactive, the response will be weaker and the risk of cancer would be lower." When the first gene called *miR-146a* has been desactivated, mice showed cell proliferation in the spleen and ganglia, which led to a lymphoma. Conversely, it was possible to suppress the development of the tumour, when the activity of the cell protein NF-kappaB was hindered (O'Connell et al., 2010).

But if some miRNAs are involved in the development of some cancers, it does not mean that they are the causative agents. Medical applications, in the long term, could derive from the interference with miRNAs (inhibition) or the activation of their functions. Difficulty in potential applications is due to the fact that it is not easy to introduce nucleic acids into cells; there will be therefore a problem of distribution of this therapy throughout the organism. If the product is injected into the blood, it will go mainly

into the liver. This organ could be in fact targeted. A biotechnology company, Regulus, founded by D. Baltimore, is designing the means to interfere with miRNAs. Big pharmaceutical groups like Sanofi-Aventis or GlaxoSmithKline (GSK) have signed collaborative contracts with Regulus (Benkimoun, 2010 k).

Kinases are enzymes that play a key role in cancers and could be a therapeutic target. Tony Hunter, director of the Salk Institute Cancer Center, stated that "about 30 kinases can be affected bymutations and play a causative role in human cancers. Some of them, which play a role of tumour suppressors, have an activity that is attenuated or even disappears in case of mutation." Brian Druker of Oregon University of Health and Sciences has targeted kinases in a new approach to treating cancr. He designed the substance imatinib, commercialized as Gleevec, for the treatment of chronic myeloid leukaemia. In this disease, an exchange of genetic material between two chromosomes results in an overactivated tyrosine kinase. Imatinib targets this abnormal enzyme and subsequently destroys leukaemic cells, without affecting healthy cells. "Imatinib has also given good results in the treatment of other cancers, such as gastro-intestinal stromal tumours (GIST) or certain melanomas. Other kinase inhibitors have been developed, but the response to the treatment is not as sustainable as desired. The major problem is resistances to treatment : they are primary in 20% of cases (metastatic melanoma and colon cancer), or acquired." The failure of anticancer treatments may be related to "the existence of a resistant reservoir that would preexist in the stem-cell population," recalled Brian Druker (Benkimoun, 2010 k).

Irving Weissman, director of Stanford University Institute of Stem-Cell Biology and Regenerative Medicine (Palo Alto, California), who was one of the first biomedical researchers to isolate stem cells, has estimated that "tumoural stem cells should be the privileged targets of anticancer treatment. Those medicines that kill these cells induce the degeneration of tumours, while those which kill tumour cells without eliminating tumour stem cells reduce the size of the tumour in a first stage, but later on the tumour grows again. Likewise, radiotherapy reduces the size of the tumour without destroying all tumour stem cells." I. Weissman has underlined the role of a membrane protein, called CD47, which is expressed abundantly in young cells and which enables them not to be destroyed by blood white cells. This overexpression of protein CD47 is a kind of signal "telling" the white cells not to phagocyte them. That is why red cells (erythrocytes) are not engulfed by macrophages. The same phenomenon occurs with haematopoietic stem cells when they migrate. According to I. Weissman and colleagues, anti-CD47 antibodies

counteract this camouflage behaviour of malignant stem cells and enable their phagocytosis in mice suffering from acute myeloid leukaemia. Many other cancers, such as breast, colon, non-small cell lung, kidney, ovary and melanoma cancers, are characterized by the overexpression of protein CD47, and anti-CD47 strategies could be tested in their treatment. It remains to be seen whether the results obtained with laboratory animals might apply to humans (Benkimoun, 2010 k).

Another promising area in cancer treatment is the research on the role of metabolic changes at the cell level in the origin and evolution of tumours. Craig Thompson (University of Pennsylvania), president of the Sloan-Kettering Cancer Center, is a pioneer in this area. He demonstrated the role of a mutation of genes involved in the metabolism of nutrients in 30% of acute myeloid leukaemias (Benkimoun, 2010 k).

Research strategy and political decision-making : example of the United States

The long-standing criticism concerning cancer research and its application to decrease the prevalence of the disease in the United States is that the National Institutes of Health (NIH) and National Cancer Institute (NCI) are necessarily structured for caution, for limited returns based on individual scientists working in their laboratories with grants that are generally allocated on the demonstration of a reasonable expectation of success. Writing research proposals is a lengthy process and a rejection means the loss of a scientist's productivity as well. And the overall funding of research has not increased; it remained flat at about US$4.8 billion during the last three years of President G.W. Bush's administration. In addition, the cost of research had outpaced inflation, so there was a double hit (Saporito, 2008).

Lack of funds and the shortcomings of research grant allocation, some argue, are causing a brain drain to Singapore and other regions that are actively developing their medical biotechnology and health-care system. Consequently, there were many endeavours aimed at convincing parliamentarians and politicians to adequately fund cancer research. For instance, in 2007, Lance Armstrong persuaded the advocate community in Texas to support a referendum to spend US$3 billion fighting cancer over the next ten years. The approval of the proposal was a great victory in a spend-wary State, and perhaps it was a model for others. The programme makes cancer prevention and screening key components, which saves the State money in the long term. The world renown cyclist and cancer survivor also convinced Senator John McCain to attend the

Livestrong Cancer Summit at the beginning of 2008, and the candidate of the Republican Party to United States'presidency, also a skin-cancer survivor, committed to increase spending on fighting cancer, but not to a specific amount. President Barack Obama has committed to doubling the budget for fighting cancer as part of the reform of health-care system (Saporito, 2008).

But is the increase of funds the real answer to the problem? Senator Bill Frist, a heart and lung surgeon before he entered politics, voted to double NIH funds in 1998 but he would not recommend it again without a better road map, i.e. a strategic plan. There are numerous federal agencies that cover cancer, for instance, but better coordination among them is necessary. B. Frist stated that scientific and advocacy communities needed to agree to a five-year "business plan" with specific targets and measurable results. The Kennedy-Hutchinson cancer bill, submitted to the US Senate in the fall of 2008 proposed a complete overhaul in cancer policy. Senator Ted Kennedy, who died from brain cancer in August 2009, had stated in a Senate hearing in June 2008, that "we need to integrate our current fragmented and piecemeal system of addressing cancer. Front and center in our current system are the troubling divisions that separate research, prevention and treatment" (Saporito, 2008).

Those are precisely the kinds of challenges that gave rise to Stand Up to Cancer (SU2C), an advocacy group which aims to assemble teams of scientists across disciplines and institutions, working collaboratively on projects designed to deliver a product within a defined time period. To choose the projects, SU2C has recruited a scientific advisory committee chaired by Phillip Sharp, a Nobel Laureate and cancer researcher at the Massachusetts Institute of Technology. The selected projects were then monitored by the American Association for Cancer Research. P. Sharp stated : "What I hope to do is identify areas where we could accelerate progress, particularly in areas where there is need – ovarian, pancreatic, glioblastoma." Additionally, 20% of the funds raised were allocated to higher-risk projects with potentially greater paybacks (Saporito, 2008).

The Multiple Myeloma Research Foundation (MMRF) used a pay-for-results funding model to support research that in four years achieved four new treatments to patients – Thalomid, Velcade, Revlimid and Doxil. That was about six years faster than the decade it usually takes for such drug development and rollout. Multiple myeloma is a rare cancer of the bone marrow that sickens about 20,000 Americans each year. The MMRF benefited from the aggressive work of founder Kathy Giusti, a multiple-myeloma survivor and former pharmaceutical executive. The

MMRF made it sure it obtained the most from its grants by adopting an enforced-collaboration model in 2004, linking work at four cancer centres into a consortium managed by PricewaterhouseCoopers and providing them all with patients, tissue samples and a set of targets and goals. In 2008, the MMRF had 30 drugs in clinical trials, and the average lifespan of multiple-myeloma patient had been extended by three years, to seven (Saporito, 2008).

On the other hand, MIT was building a US$100 million research centre that will bring together biologists and chemists with engineers skilled in nanofabrication. "We are going to breed a group of people who are totally aware of the cancer problem and totally aware of the modern tools and computational powers of engineers," stated P. Sharp. One of the strategies is to manufacture nanomolecules or nanoparticles that could bind to tumour cells and deliver therapies directly to them; or to construct nanomolecules that could locate abnormal genes and silence them (Saporito, 2008).

Fighting cancer to be successful obviously needs more concerted research on many fronts, as well as a road map to attack the disease from several angles, because it is an enemy with many faces. But all cancers may have common roots and knowing them could lead to more powerful and cost-effective treatments.

Stem cells : paving the ground for lifesaving breakthroughs

A decade of discoveries and conflicts

In 1998, James Thomson of the University of Wisconsin isolated human embryonic stem cells. In 2001, President G.W. Bush restricted federal funding for research on embryonic stem cells – i.e. cells extracted from an embryo that could turn into any of the human body's 200 tissue types. In 2004, Douglas Melton, co-director of the Harvard Stem Cell Institute (HSCI) and one of the leading figures in the search for cures for presently incurable diseases, created more than 70 embryonic stem-cell lines using private funding and distributed thousands of free copies of the cells to researchers across the world. The research objective was to use these stem cells in regenerative medicine, i.e. to replace tissues that have been injured or cells that are not accomplishing their function any more, such as cardiomyocytes (cells of the heart muscle) or insulin-producing cells of Langherans islets.

But research on embryonic cells was confronted with legal hurdles, many countries, like the United States or France, prohibiting it for bioethical reasons. The use of adult stem cells, found in adult tissues, was not subject to the same restrictions, and in fact since the late 1970s adult stem cells have been used in bone-marrow grafts to treat leukaemias and aplasias. It remains nevertheless that the access to adult stem cells is not easy, and their amplification is fraught with difficulties. In the case of embryonic stem cells, a technical problem was resolved : they can be cultured without using animal sera, thus enabling their possible use in humans. But, according to Hervé Chneiweiss, director of the laboratory of glial plasticity (National French Institute for Health and Medical Research, and Sainte-Anne and Broca hospitals in Paris), there is still the problem of their immune tolerance by the host body that receives embryonic stem cells. Secondly, the differentiation of these cells into specialized cells must be fully mastered, and the *in vitro* stability of the specialized cells' characteristics should be proven. Once transplanted in the host body, the stem-cell-derived specialized cells should not die, give rise or cause a tumour. Clinical trials were being carried out on the best animal models before conducting these trials on humans (Nau, 2008 d).

In 2006, Shinya Yamanaka of Kyoto University and his colleagues were able to convert mouse skin cells into the first induced pluripotent stem cells (iPS), i.e. stem cells without the use of embryos. Four genes were inserted into the skin cell genome using retrovirus vectors. All specialized laboratories are using this technique that has been patented (see Sasson, 2008).

These induced pluripotent stem cells (iPS) were initially considered incompatible with a medical use, because of the cancer risks associated with the four genes used to create them. But according to Marc Peschanski, who leads one of the most important France teams working on stem cells and their use in regenerative medicine, this obstacle could be overcome. Research is being carried out with a view to replacing the genes by the proteins they encode; thus, the same effects could be obtained and the drawbacks could be avoided, stated M. Peschanski (Nau, 2008 d). Also scientists have found a way to replac the genes with RNA, that does not integrate into the cell's genome. In addition, creating iPS cells with the new process is up to 100 times more efficient. If these results hold, iPS cells-based treatments in human trials could be introduced in four to five years (*Time*, 18 October 2010).

In 2007, S. Yamanaka and J. Thomson separately created the first human iPS cells. In July 2008, Kevin Eggan at Harvard Stem Cell Institute (HSCI)

generated the first patient-specific cells from iPS cells – motor neurons from two elderly women. Douglas Melton, who remained at the vanguard of stem-cell research, despite the decision made in 2001 by President G.W. Bush to restrict federal funding for the study of human embryonic stem cells (government funds could be used only to study the dozens of embryonic cell lines already in existence), bypassed stem cells altogether and transformed a type of mouse pancreatic cell that did not produce insulin into one that did (Park, 2009).

In September 2008, Konrad Hochedlinger at HSCI created iPS cells in mice using the common-cold virus rather than retrovirus vectors – an important step in making the technology safer for human use. In October 2008, D. Melton's team made human iPS cells by replacing two of the four genes, known to cause cancer, with chemicals. All four genes must be swapped out before iPS-generated cells could be transplanted into people. Also in October 2008, S. Yamanaka and his team created more iPS cells using safer plasmids of DNA instead of retrovirus vectors. There are hints that the iPS cells' short-circuited development makes them different in some ways from their embryonic counterparts. In mice, embryonic stem cells can generate a new mouse clone; iPS cells from the animals had stopped short of the same result, aborting in mid-gestation suggesting that some development requisites could be missing. But if iPS do not prove as stable and as versatile as embryonic stem cells when they are transplanted into patients, they remain a powerful research tool. And they make possible new ways of thinking about repairing and replacing damaged tissue. In this regard, D. Melton stated : "Everything we learned about stem cells tells us this was a really powerful approach. It would be a great shame if we let it wither and just go away" (Park, 2009).

On 23 January 2009, after nearly a decade of preparation, the US Food and Drug Administration (USFDA) approved the first phase-1 trial of an embryonic stem-cell therapy for a few patients paralyzed by spinal-cord injuries, to be carried out by the California biotechnology firm Geron. On 9 March 2009, President Barack Obama signed a decree authorizing federal funding for research on human embryonic stem cells, as he committed himself to do so during his electoral campaign. The restriction decided in 2001 by his predecessor was therefore lifted (Park, 2009; Nau, 2009).

A new era of research and development

A few months before the decision made by the United States'president, GlaxoSmithKline (GSK) announced it would invest at least US$25 million over five years on a collaborative agreement with HSCI, "leading to the

development of new medicines." At the time the GSK-HSCI deal was made (July 2008), the pharmaceutical corporation was using stem cells to help screen, identify and develop drugs; it was not thinking of treating patients directly with stem cells. HSCI, a network of 700 scientists based at Harvard University and its affiliated hospitals, claimed to have the world largest concentration of stem-cell researchers. The collaboration with GSK was expected to help HSCI to keep ahead of California's fast-growing stem-cell centres, which are benefiting from considerable State funds (Cookson, 2008).

HSCI had previously raised US$70 million from philanthropic and public sources over four years, but as a State Massachusetts had nothing that matched California's US$3 billion stem-cell funding. The first six projects planned for the GSK-HSCI collaboration covered : muscle regeneration, fat stem cells and obesity, heart disease, cancer and pain relief. The work was expected to include both stem cells derived from human embryos and those extracted from adult tissues, as well as iPS cells (Cookson, 2008).

In 2007, GSK had announced two, much smaller stem cell collaborations. One is the UK Stem Cells for Safer Medicines consortium, also involving AstraZeneca and Roche, which was developing techniques to use stem cells for safety testing of new drugs. The other was an alliance on cancer stem cells with the US OncoMed Pharmaceuticals, the objective being to stop the proliferation of aberrant stem cells (Cookson, 2008).

At the ceremony organized at the White House for the signature of the president's decree, B. Obama underlined that research on embryonic stem cells should be carried out within a regulatory framework. The decree gave four months to the National Institutes of Health for the elaboration of new rules on the federal funding of researchers involved in this area. While Republicans continued to oppose that kind of funding, many representatives of the American scientific and medical community expressed their satisfaction about the president's decision and recalled the potential applications of stem-cell research. Douglas Melton, the HSCI's co-director, who in 2004 used funds received by HSCI from the Juvenile Diabetes Research Foundation and the Howard Hughes Medical Institute, as well as from Harvard alumni, to develop a more streamlined method for generating stem-cell lines from embryos (70 new ones), remained convinced that the federal restrictions simply could not survive. He continued to insist that "the science is so significant that it will change the policy." He stated after the president's decision : "I think that patients worldwide are going to thank us and will request us to work more rapidly and harder while putting all our skills at the service of the discovery of new treatments" (Park, 2009; Nau, 2009).

Harold Varmus, a Nobel Laureate in medicine and physiology and head of the science and technology advisers at the White House, reminded that "embryonic stem cells seemed to be the most promising tools, but there were other means to produce stem cells that look like the embryo-derived ones, such as the iPS cells." The best candidates for cell therapies should be identified. The trial authorized by USFDA to be carried out by Geron on a dozen of patients suffering from spinal cord injuries, somewhat surprised the American specialists, who thought that the first authorization would be granted to the repair of heart muscle cells and of insulin-producing cells (Nau, 2009).

Douglas Melton, trained as a molecular biologist in amphibian development, began the work he is pursuing today : trying to find a way to make insulin-producing cells by using stem cells, as a cure for type-1 diabetes. According to Alan Trounson, president of the California Institute of Regenerative Medicine, the organization charged with dispensing State money for embryonic stem-cell research, "Doug drew a line in the sand, he turned the tables on an Administration that was incredibly negative toward stem cells and showed (it) we are not going to tolerate being put out of this field by ideological views that we do not think are correct." D. Melton who obtained his bachelor's degree in 1975 at the University of Illinois, his Ph.D. at Cambridge University, studying under Sir John Gurdon – the first to clone a frog – has taught an undergraduate course on science and ethics at Harvard University. He is motivated both professionally and personally. His son and daughter are suffering from type-1 diabetes. He wanted to learn more about how diabetes (and other diseases) develop, and iPS cells make that possible. For the very first time, he could watch the evolution of the disease *in vitro* as patient's cells develop from their embryonic state into mature pancreatic cells (Langherans islets). "There is a good reason we do not have treatments for diseases like Parkinson's," stated D. Melton. "That is because the only way science can study them is to wait until a patient appears in the office with symptoms. The cause could be long gone by then, and you are just seeing the end stages." Now the major steps in the disease process can be exposed, with each one a potential target for new drugs. "This is a sea change in our thinking about developmental biology. I consider it a real transformation moment in medicine," stated Arnold Kriegstein, director of the Institute for Regeneration Medicine at the University of California, San Francisco (Park, 2009).

There is no doubt that the United States president's decision to support stem-cell research will foster the work of Douglas Melton and many American researchers, and accelerate the search for new treatments of

presently incurable diseases. Some European scientists consider that it will cause a huge brain drain towards the United States, whose government has allocated US$21.5 billion in 2009, including US$10 billion for the National Institutes of Health, in addition to the US$29.6 billion already approved research budget (Nau, 2009).

Regarding the applications of stem-cell research in neuron differentiation, a team of Belgian biologists led by Pierre Vanderhaeghen and Nicolas Gaspard (Free University of Brussels), working in collaboration with Afsaneh Gaillard of the University of Poitiers, France, have been able to produce brain cortex cells *in vitro* from mouse embryonic stem cells. They published their results in the 18 September 2008 issue of *Nature*. Furthermore, these cortex cells have been grafted in newborn mice and made the appropriate connections with the host central nervous system (Gaspard et al., 2008).

In humans, the cortex is the most complex structure of the brain and its cells are involved in the most frequent diseases – neurodegenerative, neurovascular, neurological and psychiatric. Consequently, the availability of a reliable source of specific cortex neurons can help in testing new drugs and in modelling the development of brain diseases. The *in vitro* production of these cells could lead to grafts into the brain in order to replace damaged or non-functioning tissues (Nau, 2008 c).

The culture medium used for the differentiation of the cortex cells is different from the usual media used in the *in vitro* culture of stem cells that generally contain calf foetal serum and molecular growth factors. It is a medium called N2, developed in 1979 by Gordon Sato to grow mouse foetal neurons. Hervé Chneiweiss, director of the laboratory of glial plasticity (National French Institute for Health and Medical Research, INSERM) expressed his admiration for the work carried out by the Belgian team, and particularly for the graft experiments : “It is surprising to observe the growth of axons on such distances over four weeks only.” He warned that these outstanding results had been obtained in mice, and, for many technical reasons, could not be immediately transposed to human beings (Nau, 2008 c).

Nowadays, three weeks are needed to culture the skin cells of a person that has been heavily burnt, before undertaking an autograft; during that time, the patient is exposed to dehydration problems and risks of infection. Consequently, researchers are trying to reconstitute a human epidermis in unlimited quantities that would be used to treat heavily burnt persons. A French team led by Marc Peschanski and Christine Baldeschi

of the National Institute for Health and Medical Research (INSERM), have published in the 21 November 2009 issue of *The Lancet* the results of their work initiated by Gilles Waksman (who passed away in 2007), and concerning the production of skin from human stem cells. *In vitro* and *in vivo* demonstration of the functional nature of the tissue thus produced has been shown on mice that were grafted with the skin produced from stem cells (Guenou et al., 2009).

The French scientists used human embryonic stem cells (HESC), which are totipotent cells and can therefore differentiate into all kinds of cell lines, including keratinocytes that make up 90% of skin cells. It is possible to obtain an unlimited quantity of daughter cells from a few HESCs. In addition, "these cells express a low level of antigens of the HLA immune system, which would mean that the likelihood of rejection is very low," according to C. Baldeschi. But the tissue produced from the HESCs should be functional and contain the four layers existing in a normal epidermis (Guenou et al., 2009).

The first stage of the research work consisted of orienting the HESCs towards a differentiation into epidermic cells, after culturing them for more than 40 days with nourishing cells and various ingredients, including insulin and ascorbic acid. The keratinocytes that were obtained had the same morphology as those found in the basal epidermis or germinative layer. *In vitro* culture of these cells layered on an artificial matrix led to the construction of an epidermis with its different constitutive layers. The second stage consisted of grafting the artificially obtained human skin on immunodeficient mice (in order to avoid the rejection of the graft); 12 weeks later, the skin had the same architecture as human adult skin (Guenou et al., 2009).

The next stage, according to C. Baldeschi, would aim at designing a new rigorous protocol in order to carry out a clinical trial and to screen many molecules, with a view to re-establishing the normal functions of skin in dermatoses of genetic origin. The protocol would consist of testing cells *in vitro* and in mice, and should include devices that enable the withdrawal of the artificial skin in case of appearance of tumour cells (Benkimoun, 2009 n).

Production of blood by haematopoietic stem cells

Blood donations are not meeting medical needs, such as transfusions, surgery, anaemia or certain cancer treatments. Consequently several research teams worldwide are trying to produce blood from cells which

do so in natural conditions. For instance, the team led by Luc Douay (hospital Armand-Trousseau in Paris) is working on haematopoietic stem cells which can produce all blood cells lines. The French researchers were able to produce almost illimited quantities of red cells; their objective is to create red-cell banks from stem cells belonging to a few dozens of donors, while relying on the possibility to reprogram adult cells into induced pluripotent stem cells (iPSC) [Benkimoun, 2010 l].

Canadian researchers at McMaster University, Hamilton, Ontario, have proceeded differently in order to produce from skin cells stem cells of several blood cell lines. Mickie Bhatia and his colleagues published their results in the 25 November 2010 issue of *Nature*. They considered that the mechanisms of reprogramming adult skin cells (fibroblasts) into pluripotent cells (iPSC) were still unknown; furthermore, the pluripotence of these cells can be unstable and the process of their specialization is not completely elucidated. Consequently, they have tried to produce blood without going through the reprogramming of fibroblasts into iPSC (Szabo et al., 2010).

During the process of reprogramming, human fibroblasts overexpress a protein called OCT4, which is specific of pluripotent cells, a well as markers of differentiation towards a type of blood cell line; one of these markers is called CD45. Contrary to haematopoietic cells derived from reprogrammed cells, fibroblasts possessing the CD45 marker, used by the Canadian team, show the characteristic features of a programme oriented towards the differentiation into blood cell lines. The data from the Canadian study indicate that "the OCT4 protein was sufficient to induce the emergence of CD45 cells from multiple sources of fibroblasts reacting to stimulation by haematopoietic growth factors," stated M. Bhatia et al. (Szabo et al., 2010).

Avoiding the pathway through reprogrammed pluripotent cells may have several advantages. First, it eliminates the risk of tumoural evolution of undifferentiated cells. Secondly, the process adopted by the Canadian researchers would be more efficient than going through entirely reprogrammed cells. Finally, the attempts to produce different blood cell lines from embryonic stem cells or iPSC were not always successful. Research work by the Canadian scientists therefore showed that it was possible to create many blood cell lines from fibroblasts. The process could become in the future a new source of blood cells produced from the skin cells of a patient, thus avoiding to relying on a donor. McMaster University's researchers have tested their approach for two years, using the skin of newborn infants and of adults; it seems therefore that the age

of the donor does not influence the success of the technique. Clinical trials on humans may be carried out in 2012 (Szabo et al., 2010).

Genetic selection of embryos to cure disease

When in October 2010 Robert Edwards, who perfected *in vitro* fertilization (IVF) more than 30 years ago, was awarded the Nobel Prize of Physiology and Medicine, researchers at Stanford University announced they had found a way to film the development of embryos in the first 48 to 72 hours after *in vitro* fertilization. This finding may be crucial in embryo selection. Currently, IVF technicians must play a guessing game when picking out the best embryos to transfer to a woman's womb; they generally wait until the fourth or fifth day to choose them, on the assumption that those embryos that can survive as long *in vitro* are healthier and more likely to result in pregnancy. But waiting too long can also be harmful, because the longer embryos are kept outside the body, the more likely they are to develop abnormally (*Time*, 18 October 2010).

After observing movies of 242 embryos undergoing their first cell divisions, the scientists identified three factors that helped distinguish the most viable embryos : the time it took to cleave from a single fertilized egg into two cells and the time between each of the next two cell-division cycles (faster is better). These features determined with 93% accuracy which embryos would survive to the fourth or fifth day, allowing physicians to transfer them as early as two days after fertilization. The imaging technique could improve the current success rate of IVF, which is about 30% in the United States. It may also reduce the need to transfer several embryos at a time, which can result in risky multiple pregnancies (*Time*, 18 October 2010).

In families where there is a high risk of transmission of a genetic disease, the preimplantation genetic diagnosis (PGD) is increasingly being used to replace amniocentesis (carried on foetal cells extracted from the amniotic fluid, and may lead to a medical interruption of pregnancy). In the case of PGD, one cell from an eight-cell embryo is analyzed in order to eventually identify the gene(s) causing the disease; only those embryos which do not bear the genetic defect are implanted in the uterus of the mother.

In France, since 1994 and the first laws on bioethics, PGD had been strictly regulated and applied to embryos derived from *in vitro* fertilization (IVF). It is therefore carried out far ahead of prenatal diagnosis (amniocentesis). French law clearly indicates that PGD only concerns the search for genes related to "particularly serious diseases and incurable at the time of the

diagnosis," i.e. mainly cystic fibrosis, Huntington chorea, haemophilia and certain forms of myopathies and mental retardation. These diseases will certainly affect newborns having the defective gene(s). Only three centres specialized in medically assisted procreation and reproduction biology have been authorized to carry out PGD, leading to a few dozens births per year. The 2004 bioethics law foresaw that PGD could be carried out in order to give birth to a healthy child who could help curing his/her ill elder brother or sister using the cells from the umbilical cord blood (Nau, 2008 a, e).

In Spain, four families had relied on PGD and attended either the Free University of Brussels or the Chicago Reproductive Genetics Institute (RGI) for that purpose, because their country did not authorize PGD until mid-2006, when the law on assisted human reproduction had been approved, or because they considered the bureaucratic process too long and therefore threatening for the health of their children. In one of the families, of six children the first three suffered from Duncan syndrome – a lymphoproliferative disease associated with X chromosome – and the last three were born via *in vitro* fertilization and PGD was carried out to make sure they were disease free; their haematopoietic cells had been used to cure their brothers and sisters. Not only the embryos were disease free, by they also had the same histocompatibility (HLA) as the diseased children. Another couple attended the RGI in Chicago in 2003 (i.e. before Spanish law authorized PGD) and their daughter was born in August 2005, after three attempts of IVF and embryo transfer. In September 2006, a bone-marrow transplant was made to her 14-year-old sister in order to relieve her from beta-thalassaemia. This was done in Barcelona's Sant Pau hospital, and two years later there was no sign of the disease. In the case of the third family, one of the children was suffering from Franconi's disease – a blood disease the occurrence of which is one per 500,000 births and that causes a lack of haemoglobin and lower concentrations of leucocytes and platelets (symptoms of great fatigue and high sensitivity to injection and major risk of haemorrhage). The disease was diagnosed in 2003 in Barcelona's Vall d'Hebron hospital. The only therapy is a bone-marrow transplant; with a donor that is not related to the patient, the success rate is 30%, while if the donor is a compatible brother or sister, the success rate could rise up to 60% or even 80%. In December 2005, at Chicago's RGI, the birth of another child took place (using IVF and PGD), and the transplant was done in November 2007, almost five years after the disease had been diagnosed. In October 2008, the 12-year-old girl was recovering rather well and ready to go to school (Prats, 2008).

The four Spanish families have underlined the need for the health administration to speed up the bureaucratic processes required by the law in order to enable the couples to be treated in Spain and not in foreign countries. They requested that IVF and PGD leading to the birth of a child who can cure his/her brother or sister should be subsidized; most of Spanish centres that can carry out PGD are private clinics. One of the families that had to attend Chicago's RGI spent around €60,000 to €70,000 for the whole process of medically assisted procreation, using PGD to screen the non-diseased embryos. The families also responded to the criticism made by the Spanish Episcopal Conference that had expressed its strong opposition to PGD and renewed it when the first child was born in Spain in October 2008, with the help of this technique (Prats, 2008).

The Spanish law voted in mid-2006 has put in place strict ethical controls. In each autonomous region, the couple or family should request the authorization from the health administration to have an IVF and PGD. Two advices are requested from the National Organization for Transplants and from the National Committee for Assisted Reproduction. This means that patients have to wait for six to seven months, while experts consider that the reply should not take more than 15 days or one month. Shortening the time of reply is important because of the threat to the health of the diseased child and of the fact that the mother's fertility falls markedly after 39 years. Julio Martin, head of the department of PGD of the Valencia Institute of Infertility, considers the bureaucratic process should be faster, but it is not easy to do so due to the fact that we are dealing with an experimental treatment which should be strictly controlled. Since the law had been promulgated and up to the end of October 2008, the health ministry had received 31 requests, of which only eight had been approved; in addition, 17 files had been on stand-by because more clinical information on the couples was needed (Prats, 2008).

New indications for the preimplantation genetic diagnosis

In October 2006, the French Biomedicine Agency and National Cancer Institute requested Dominique Stoppa-Lyonnet of the Institut Curie, Paris, to conduct an enquiry about the possible broadening of the preimplantation genetic diagnosis (PGD) to the detection of genetic mutations that could predispose to diseases with a likelihood of 70% to 80% for the occurrence of the illness. In other words, the selection of embryos using PGD would aim not just to eliminate incurable diseases whose occurrence is certain, but to screen embryos that do not bear certain genetic markers for the most dangerous forms of some diseases,

e.g. colon, breast and ovary cancers. On 9 April 2008, the report by D. Stoppa-Lyonnet was published and revealed that, despite the usual interpretation of the bioethics law concerning PGD, 22 PGD tests having a broader objective had been carried out in France between 2000 and 2007; six infants were born without the gene defect. The author of the report believed that there was no need to modify the bioethics law in order to carry out the "broader" PGD, and that a series of technical precautions should be taken by the medical teams, and the couples concerned must be informed and involved in the decision-making process. The Biomedicine Agency supported the report conclusions and considered the proposals as temporary measures that responded to practical issues raised by the application of current law. The health minister, Roselyne Bachelot-Narquin, stated in April 2008 that all the ethical issues raised by PGD will be dealt with during the national bioethics gathering in 2009 (Nau, 2008 a).

The extension of PGD was authorized in Belgium, Spain and the United Kingdom. The British prime minister, Gordon Brown, proposed the human fertilization and embryology bill, which had wide support among scientists, and aimed at helping those seeking cures for ailments ranging from cancer to Parkinson's disease. Cardinal Keith O'Brien, leader of the Scottish Catholics, has described the bill as "monstrous," while Peter Smith, archbishop of Cardiff, wrote to the prime minister warning that the issues go to the "sacredness of human life, its meaning and purpose." Labour parliamentarians were compelled to back the bill, but they were allowed to vote with their conscience on three of the most controversial elements at the committee stage of the bill : the creation of hybrid embryos by crossing animal eggs and human nuclei; the production of "saviour" siblings to help treat brothers or sisters with genetic defects or diseases; and IVF research. The other two main parties – the Conservatives and Liberal Democrats – had promised a free vote on all aspects of the bill (Pickard, 2008).

The British government believed the legislation was needed to take account of scientific progress since the last relevant legislation was passed in 1990. The prime minister claimed that "the ethical framework in the United Kingdom was one of the best if not the best in the world," and the bill would give not only new powers, but "new responsibilities" to the Human Fertilization and Embryo Authority (Pickard, 2008).

On 24 October 2008, *The Times* revealed that a team of British researchers, led by Alan Handyside of London Bridge Centre, had developed a technique that permitted the determination of multiple

genetic characteristics which predispose to many diseases from the genome analysis of one embryonic cell. The British researchers used the most recent techniques of high-throughput DNA sequencing, and claimed that PGD could be extended to identifying in the embryo genetic markers associated with diabetes, cardiovascular and neurodegenerative diseases, and not just with family cancers. The new technology was submitted to the British relevant regulation authorities, in order to obtain the authorization of marketing it at the beginning of 2009 at a cost of €1,900 per analysis (Nau, 2008 e).

The Human Fertilisation and Embryo Authority was expected to rule on the commercialization of A. Handyside and colleagues' technology. It was nevertheless underlined that there was a practical hurdle : the limited number of embryos that could be produced *in vitro* by a couple, i.e. about a dozen for each attempt. "When you start searching for two or three genetic markers or traits, there is very little chance to find the relevant embryo," stated Alan Thornhill, the scientific director of the Bridge Centre. The solution would be the production of human ovocytes from stem cells, but in the meantime the low number of embryos produced by IVF was limiting the use of embryo screening based on the identification of a very high number of gene markers (Nau, 2008 e).

Besides PGD and its extension, American researchers have gone further in their endeavours to better understand genetic diseases and to advance knowledge of human biology. At Cornell University, Ithaca, New York, researchers led by Nikita Zaninovic presented in a scientific meeting the results of their work on the transfer into a human embryo of a gene encoding the synthesis of a fluorescent protein. Thereafter, they published their work in the journal *Fertility and Sterility*. The team that is familiar with techniques of gene therapy, had developed the method on mice before applying it to human embryos, without any direct therapeutic goal. The experiment was carried out on a human embryo produced by IVF within a programme of medically assisted procreation. Using a retrovirus as a vector, they were able to introduce the alien gene into the embryo's genome (Zaninovic et al., 2007). As revealed by the British *Sunday Times*, on 11 May 2008, the transgenic embryo was not transferred to a womb and was destroyed after five days of embryonic development (Nau, 2008 b).

Similar experiments could be carried out in the United Kingdom, where the Human Fertilisation and Embryology Authority had authorized the creation of hybrid (animal/human) embryos. It had refused to authorize the genetic modification of sexual cells. It is indeed possible to obtain the same result as that of researchers from Cornell University (a transgenic

human embryo), through the artificial modification of the genome of male or female gametes before carrying out an *in vitro* fertilization. This kind of experiment had been conducted successfully on chicken gametes several times in 2007 in the United States. American researchers had also obtained stem-cell lines derived from embryos of transgenic mice (Nau, 2008 b).

Some experts have warned that these experiments on human embryos were fraught with high risks. They highlighted that the techniques available now or very soon could allow not only the repair of genetic defects, but also the modification (without therapeutic goal) of performances of the human body, i.e. certain physical and cognitive abilities of human beings (Nau, 2008 b).

Replacing body parts

Presently, a patient may wait months, sometimes years, for an organ from a suitable donor. During that time his/her condition may worsen, and he/she may even die. The ability to make organs as they are needed would therefore save lives, and that possibility may be closer with the commercialization of the first 3D bio-printer for manufacturing tissue and organs. The new machine, costing around US$200,000 in 2010, had been developed by Organovo, a company in San Diego specializing in regenerative medicine, and Invetech, an engineering and automation firm in Melbourne, Australia. One of Organovo's founders, Gabor Forgacs of the University of Missouri, developed the prototype on which the new 3D bio-printer was based. The first production models were to be delivered to research groups which, like that of G. Forgacs, are studying ways to produce tissue and organs for repair and replacement. At present much of this work is being done by hand or by adapting existing instruments and devices (*The Economist*, 2010 d).

Initially, only simple tissues, such as skin, muscle, and short stretches of blood vessels, will be made, as stated by Keith Murphy, Organovo's chief executive, and these will be for research purposes. But the company expected that within five years, once clinical trials had been completed, the printers would produce blood vessels for use as grafts in bypass surgery. Because the machines have the ability to make branched tubes, the technology could, for instance, be used to create the networks of blood vessels needed to sustain larger printed organs, like kidneys, livers and hearts (*The Economist*, 2010 d).

Organovo's 3D bio-printer works in a similar way to some rapid-prototyping machines used in industry to make parts and mechanically functioning models. These work like inkjet printers, but with a third dimension. Such printers deposit droplets of polymer which fuse together to form a structure. With each pass of the printing heads, the object takes shape. Complex shapes are supported by printing a "scaffold" of water-soluble material. Once the object is complete, the scaffold is washed away. Researchers have found that something similar can be done with biological materials. When small clusters of cells are placed next to each other they flow together, fuse and organize themselves (*The Economist*, 2010 d).

In 2006, Anthony Atala and his colleagues at the Wake Forest Institute for Regenerative Medicine in North Carolina made new bladders for seven patients. These are still working. The process starts by taking a tiny sample of tissue from the patient's own bladder (so as to avoid rejection by the patient's immune system). From this sample precursor cells are extracted and they can form muscle on the outside of the bladder and the specialized cells within it. When more of these cells have been cultured in the laboratory, they are laid onto a biodegradable bladder-shaped scaffold which is warmed to body temperature. The cells then mature and multiply. Six to eight weeks later, the bladder is ready to be transferred into the patient (*The Economist*, 2010 d).

The advantage of using a bio-printer is that it eliminates the need for a scaffold. The Organovo's machine uses stem cells extracted from adult bone marrow and fat as the precursors. These cells can be coaxed into differentiating into many other types of cells by the application of appropriate growth factors. The cells are formed into droplets 100-500 microns in diameter and containing 10,000-30,000 cells each. The droplets retain their shape well and pass easily through the inkjet printing process. A second printing head is used to deposit scaffolding – a sugar-based hydrogel. This does not interfere with the cells or stick to them. Once the printing is complete, the structure is left for a day or two, to allow droplets to fuse together. For tubular structures, such as blood vessels, the hydrogel is printed in the centre and around the outside of the ring of each cross-section before the cells are added. When the part has matured, the hydrogel is peeled away from the outside and pulled from the centre like a piece of string (*The Economist*, 2010 d).

The bio-printers can also use other types of cells and support materials. They could be employed to place liver cells on a pre-built, liver-shaped scaffold or to form layers of lining and connective tissue that would grow

into a tooth. The printer fits inside a standard laboratory biosafety cabinet, for sterile operation. Invetech has developed a laser-based calibration system to ensure that both print heads deposit their material accurately, and a computer-graphics system allows cross-sections of body parts to be designed (*The Economist*, 2010 d).

Some researchers are of the opinion that machines like Organovo's 3D bio-printer could one day print tissues and organs directly into the body. Indeed, A. Atala is working on one that would scan the contours of the part of a body where a skin graft was needed and then print skin onto it. As for bigger body parts, G. Forgacs thinks they may take many different forms at least initially. A man-made biological substitute for a kidney, for instance, need not look like a real one or contain all its organelles in order to accomplish its physiological function and clean bloodstream. Those waiting for transplants are unlikely to worry too much about what replacement body parts look like, so long as they work and make them feel better (*The Economist*, 2010 d).

Creation of bioartificial organs

On 2 November 2010 a laboratory for the creation of bioartificial organs was inaugurated at the Gregorio Marañon hospital in Madrid. Its aim was to become the first bank of organs of this new type in the world. The laboratory with an area of 260 m^2 will be able to supply to patients that are waiting for an organ transplant "scaffolds" that have been depleted of their cells and that could be filled with stem cells of the receiver before making the transplantation. This would be a means to solve both the problem of lack of donor and that of transplant rejection. This project, coordinated by Francisco Fernández Avilés, head of the cardiology service of Gregorio Marañon hospital, and christened Scaffolds and Bioartificial Organs for Transplantation (SABio), was funded by the Spanish ministry of science and innovation for an amount of €600,000. The project resulted from the interaction between F. Fernández Avilés and DorisTaylor of the Centre for Heart Repair of the University of Minnesota. D. Taylor has been working for many years on the regeneration of organs taken from animals. She was able to revive the heart of a dead rat in 2008, after eliminating all its cells and then replacing them with the cells of another animal; she therefore created for the first time a new heart that could contract efficiently (Morel, 2010).

F. Fernández Avilés travelled to the United States in May 2010 and worked under the guidance of D. Taylor in order to carry out the first stages leading to a human bioartificial heart. The Spanish team was able to eliminate

all the cells of a human heart using detergent substances administered through the coronary arteries. "The organ becomes an inert tridimensional scaffold, without cells, but with arteries, veins and vessels, ready to receive stem cells and to become a new organ," stated F. Fernández Avilés. If deemed necessary, in order not to damage the organ, the degeneration process can be carried out in three to four days, and the scaffold, once depleted of all its cells, could be conserved for some time without any harm and be used at the right time (Morel, 2010).

"Before the end of 2010, we shall succeed in obtaining a piece of the heart muscle, completely regenerated and capable of beating. But in order to recreate a whole heart, several years will be needed," stated F. Fernández Avilés, who considered that five to ten years will have to be devoted to laboratory experiments. "In the medium term, when a patient will need a transplant, we shall choose the scaffold that best suits him/her, depending on his weight and length," he said. For the research to be carried out, the National Organization for Transplantation (ONT) supplies the organs that cannot be used for transplantation (about 40% of donated organs), while stem cells, produced from cells of fat tissue and bone marrow, come from the cell production unit of Gregorio Marañon hospital, the main one in Spain. The Spanish laboratory would be able to create, in addition to bioartificial hearts, kidneys, livers and skin tissues, on which several international teams are working (e.g. regeneration of bioartificial functional lungs and livers in rats) [Morel, 2010].

Obesity : the hope for a cure?

Obesity in the United States

In the United States, at 33.8% obesity rate is ten times higher than Japan's. In all, 68% of Americans were either obese or overweight in 2009. Some studies give lower numbers, but since they typically ask people how much they weigh, rather than weighing them, scepticism should apply. People put on weight when they consume more calories than they burn off. But it is difficult to explain why obesity in the United States has trebled since 1960. One explanation is as people become richer, food becomes relatively cheaper. Time becomes more precious, hence the attraction of fast food that saves time. Also desk work burns fewer calories than physical work, and labour-saving devices contribute to that. Barry M. Popkin, author of the book *The World Is Fat*, stated that if we still washed dishes and clothes by hand, we would burn off five pounds of body weight each year. But all this does not explain why Americans are fatter than people in other rich countries, nor why a study published in

January 2010 in the *Journal of the American Medical Association* (*JAMA*) reported that the Americans appear to have stopped being fatter. The *JAMA* study found that American women were no more likely to be obese in 2008 than they were nearly a decade before. For men, there was a small rise in obesity over the same period, but no change over the last three years. Among children, too, there was no change in obesity rates except among the very heaviest boys, whose numbers increased slightly (Flegal et al., 2010).

If the obesity rate really has stopped increasing, that would be a blessing for the American health-care system. Each obese American racks up medical bills 42% higher than an American of normal weight, according to Eric Finkelstein and Justin Trogdon, writing in *Health Affairs*. One should also add the indirect costs of obesity, such as lost productivity due to sickness or premature death. In 2008, Youfa Wang of the Johns Hopkins Centre for Global Health projected that 100% of Americans would be overweight by 2048. By 2030, his model showed health-care costs attributable to excess weight approaching a trillion dollars a year (*The Economist*, 2010 c).

Kathleen Sebelius, the American health secretary, stated that "fighting obesity is at the heart" of health reform. But Americans are suspicious of the nanny state. However, some regulations help : forcing restaurants to post calorie contents of dishes, for instance, prompts diners to choose less calorific meals. But politicians are reluctant to attack consumers' eating habits. A proposal to tax sugary drinks, for instance, went nowhere. Opponents argued that it would disproportionately affect the poor. But the poor are disproportionately likely to be overweight (*The Economist*, 2010 c).

The efforts made at school to advocate for balanced meals may have raised awareness of the need for a healthier diet. And popular pressure has prompted many fast-food outlets to offer salads and other wholesome fare. But even if healthy food were freely available, losing weight is difficult. Every year, 25% of American men and 43% of American women attempt it. Failure rates are high, but studies suggest that people eat more if they have overweight friends and relatives, and less if they do not (*The Economist*, 2010 c).

Obesity in developing countries

According to the estimates by the World Health Organization (WHO), 1.6 billion adult people were overweight in the world in 2005. Their

number could rise to 2.3 billion in 2015 and to 3.3 billion in 2030, 80% of them living in developing countries where this health problem did not almost exist two generations ago. This change was at the heart of the discussions at the international congress on obesity, held in Stockholm from 11 to 15 July 2010 (Vincent, 2010 d).

In Mexico, the country most affected after the United States and the first one for child obesity, the health implications of this "epidemic" are already obvious : diabetes is in Mexico the second cause of mortality after hypertension. In China also the change is impressive : overweight affects almost one-fourth of the whole population and has become a real problem of public health. As shown in several surveys, there were more than 200 million people overweight and 90 million obese in 2010, an increase of 39% and 97% respectively compared with 1992. Young city dwellers are the most affected : a recent study, carried out on a cohort of 80,000 children living in an urban environment, has shown a 156% increase in the number of obese children between 1996 and 2006 (Vincent, 2010 d).

Obesity was recognized as a disease by WHO in 1997 and it had been considered for a long time as a scourge of rich countries, particularly in North America. It was nevertheless observed that the number of obese people had been increasing in developing countries, but experts hesitated to draw attention on the negative implications of overweight in countries where so many people were still starving. Until the publication by the Worldwatch Institute in 2001 of data confirming that, for the first time, the number of overweight people in the world was equal to that of underfed people; and that the proportion of obese people and the correlated diseases (diabetes, cardiovascular diseases and cancers) were rising very quickly across the developing world (Vincent, 2010 d).

The reason for such a change is nutritional transition, i.e. a drastic modification of food diets in the big cities of emerging countries, associated with a critical decrease in physical activity among the inhabitants of these cities. For instance, Barry M. Popkin, an obesity specialist at the University of North Carolina, stated that "the diet of poor populations in rural or urban areas of Asia during the 1960s was very simple and rather monotonous : rice with small quantities of vegetables, beans or fish"…while "the inhabitants of these regions consume nowadays more complex meals regularly, in numerous places where prepared indigenous or Western meals are sold" (Vincent, 2010).

The faster is the urbanization in these developing countries, the greater is the threat of overweight. A study on African migrants in Australia has

shown that the proportion of obese children varied between 6% and 30%, according to their degree of aculturation. Another study carried out in the Pacific Islands showed that populations living in the coastal areas were more affected by overweight than those living on the high plateaux. Arnaud Basdevant, in charge of the department of nutrition of the hospital Pitié-Salpêtrière, Paris, stressed that "an Indian study had shown a few years ago that the children of underfed mothers were also those who would become obese or diabetic more easily." This effect, called "genetic imprint," was still badly understood. However, this observation has been confirmed in Asia, Africa and Latin America; wherever populations had suffered from serious nutritional deficiencies, the impact of diabetes and hypertension among overweight persons occurred more rapidly than in Western populations (Vincent, 2010 d).

There is also a genetic component. In Mexico, where 80% of the population is the result of crossings between Europeans and Amerindians, the first results of a vast genetic study have shed light on the role of a gene involved in obesity and the early development of type-2 diabetes. The presence of this gene has been estimated at 33% among the Mayas, the Purepechas and Tarahumaras (Vincent, 2010 d).

In sub-Saharan Africa, it is paradoxical to observe that overweight is progressing in a region where hunger has not been yet eradicated. In the rural areas, under- and malnutrition still prevail, whereas in the cities overweight and obesity are progressing. A study carried out by the French Research for Development Institute (IRD) and its African partners showed that in Burkina Faso, where urban population has been multiplied by seven since 175, one female urban dweller out of three was overweight, compared with 4% women in rural areas. This disbalance can also be observed at the level of an individual, who could suffer from both obesity and a deficiency of micronutrients (Bolis, 2010).

It is in the cities where nutritional transition occurs, as a result of social change, stressed Francis Delpeuch, a researcher at IRD. In his view, this change in eating behaviour leads to a "globalization of obesity." At the origin of this nutritional transition is the rise in the standard of living of city dwellers and the relative abundance of food in the cities; supermarkets and food marketing entice people to consume industrial foodstuffs that contain more salt, sugar and fats. A survey of 1,000 inhabitants of Ouagadougou, the capital of Burkina Faso, and of their eating habits, has shown that those who consumed "modern" foodstuffs were more prone to overweight. In addition to traditional cereals and vegetables, imported foods like bread, pasta and soft drinks are consumed. On the other hand,

urban lifestyle dominated by transport, tertiary jobs and television, does not encourage exercise. All these habits break up the balance between consumed calories and burnt ones. As urban population in Africa is rising steadily, WHO estimated that among women, who were more susceptible to overfeeding than men, 41% of those who were more than 30 years old would be overweight by 2015 (Bolis, 2010).

In the Western world, obesity affects more the poor than the well-off, but in developing world it is the reverse. And this is even more true in Africa, where overweight is "often associated with a higher social position and with good health," underlined Mathilde Savy of IRD. "In Mauritania, for instance, young girls are being overfed in order to comply with traditional aesthetic codes." Some politicians perceive positively the increasing overweight of their populations, they view it as a sign of spreading wealth. This is obviously a hindrance to any policy of obesity prevention (Bolis, 2010).

It must be stressed that overweight is a major risk factor for the development of type-2 diabetes, cardiovascular diseases, some cancers and chronic diseases. All these illnesses have a huge cost, with which Africa health-care systems cannot cope (Bolis, 2010).

Obesity and intestinal microflora

Obesity aetiology is complex, as it can be caused by many factors such as too rich food, lack of physical exercise and genetic predisposition. It also seems that a disbalance in the intestine bacterial flora could lead to overweight, as shown by a study carried out by Jeffrey I. Gordon and his colleagues of Washington University School of Medicine, Saint Louis, Missouri, published in the 11 November 2009 issue of *Science Translational Medicine*. The American scientists have shown that when transferred to mice that had no intestinal flora, the microflora from human faeces was rapidly modified by a diet overloaded with sugars and fat. This bacterial population could later on make obese mice that had a normal diet (Turnbaugh et al., 2009).

Peter J. Turnbaugh and Jeffrey I. Gordon had published an article in *Nature* (21 December 2006) which showed that in both humans and mice the intestinal flora was distinct in obese and slender individuals. Two families of bacteria are predominant in the intestine: bacteroids (that include about 20 different genera) and firmicutes (more than 20 genera including lactobacilli, mycoplasms and clostridia). In obese persons, the balance is disrupted towards the dominance of firmicutes, and this

results in a greater capacity of this microflora to extract calories from food. J.I. Gordon and his colleagues carried out an experiment whereby they transferred the intestinal flora of obese mice to non-obese ones. They observed that both the capacity to extract calories from food and weight increased among the latter (Ley et al., 2006).

In their new study, J.I. Gordon, P.J. Turnbaugh and their colleagues aimed at recreating the human intestine ecosystem, using mice that were deprived of an intestinal flora. This type of transgenic mice, when fed with a diet rich in fats and sugars, did not become obese. The American researchers introduced into the intestine of these mice the flora present in human faeces, and they observed that the rodents acquired an intestinal flora quite similar to that of the human intestine. Furthermore, they transmitted this type of bacterial population to their offspring. Mice with a microflora of human origin had a larger body fatty mass than those deprived of microflora (Turnbaugh et al., 2009).

First fed with a diet poor in fats and rich in fibers, the "humanized" mice were later on fed with a very fatty and sweet diet. One single day of this diet, qualified as "Western" by the researchers, was sufficient to cause a modification of the composition of the microflora. The following stage consisted of transferring the microflora of mice submitted to that diet into the intestine of mice deprived of microflora. It was observed that in the latter, even when fed with a low-fat diet, the body fatty mass increased. It was concluded that the microflora of "humanized" mice eating a hypercaloric diet had the capacity to make obese mice that were initially deprived of intestinal flora. Jeffrey I. Gordon and his colleagues concluded that the history of intestine colonization by bacteria "had an impact on the initial structure of bacterial population, but these effects could be rapidly altered by the diet" (Turnbaugh et al., 2009).

Also in *Science Translational Medicine*, Jeffrey S. Flier and John J. Mekalanos of Harvard Medical School, Boston, made the following comments : is there a real difference between the bacterial genome (microbiome) of the intestinal flora of obese individuals and that of normal weight individuals, in humans and rodents? If that difference exists, is it a cause or a consequence of obesity? If it were a cause, what is the mechanism of change in the energy balance sheet, because the gap between energy input and energy consumption is responsible for weight variations (Flier and Mekalanos, 2009). Jeffrey I. Gordon and his team are in fact proposing a practical model of study, using mice that have been colonized by an intestinal flora and relying on the current advances in sequencing the genomes of intestinal bacterial flora. This model may help disentangle the

relations between different bacterial populations and certain physiological features. This model will also enable the researchers to follow over several generations the transfer of a type of microflora to the offspring (Benkimoun, 2009 m).

It should be recalled that a human being's intestine contains 500 to 1,000 different bacterial species. Thanks to these bacteria, calories are extracted from ingested food in order to meet energy needs. The progress in sequencing techniques has made possible the study of the metagenome, i.e. the combination of the human genome and the genomes of the few thousand billions of bacteria present in the human body – the microbiome. Human genetic material represents only 1% of the whole metagenome (Turnbaugh et al., 2009).

Several projects of sequencing the metagenome have been launched worldwide. The European project, Metagenomics of the Human Intestinal Tract (Meta-HIT), has been launched on 1 January 2008 and is coordinated by the French National Agricultural Research Institute (INRA). Its objective is to better understand the relations between the genes of bacteria of the intestinal flora (microbiome) and human health and diseases. Emphasis is laid on inflammatory diseases of the intestinal tract and obesity. Within this framework, France and China cooperate in the project "MicroObes." Meta-HIT is funded by the European Commission under its 7th Framework Programme for a duration of four years, with a view to stetting up a catalogue of microbial genes and genomes present in the human intestinal tract. The second objective is to identify the types of genes and genomes present in different individuals and at which frequence. The third stage of the project is to compare these genomes in healthy individuals and sickened ones (Benkimoun, 2009 m).

International collaboration is in place like in the case of the human genome. On 16 October 2008, the representatives of institutions or projects of eight countries (Australia, Canada, China, South Korea, United States, France, Ireland, Japan) and the European Commission met in Heidelberg (Germany) and set up the international consortium of human microbiome (IHMC). The consortium should receive US$250 million of international funds. Data produced by the consortium will be published simultaneously by a centre of the US National Institutes of Health and by the European Molecular Biology Laboratory in Heidelberg. These data will be communicated to the main public databases (Benkimoun, 2009 m).

Intestinal microflora transplants and biotherapy

Biologists consider the 10,000 billions of bacteria which colonize the human intestinal tract, as an organ *per se*. When this complex machinery (microbiome) is not functioning well and causes intestinal illness or major discomfort, would it be possible to repair it through transplants? For instance, among the thousands intestinal bacterial species *Clostridium difficile* can take advantage on many other species as a consequence of antibiotic treatment. This disbalance causes diarrhoea and colitis which are sometimes difficult to stop; unless the microflora is reconstituted through a transplant from another individual (Morin, 2010 d).

Recent clinical research has shown this kind of "bacteriotherapy" can be effective; introduced from one or the other end of the digestive tract, the alien fecal microbiome can in at least 90% of the cases rapidly reestablish the normal functioning of the host's digestive system. In the May-June 2010 issue of the *Journal of Clinical Gastroenterology* (*JCGE*), Khoruts et al. of the University of Minnesota Center for Immunology (Department of Medicine), Minneapolis, have described the "dramatic impact" of this kind of transplant on the composition of the intestinal microbiome of a patient suffering from a disease caused by *C. difficile*. While the analysis of bacterial DNA had revealed a deficiency of firmicutes and bacteroids, the bacterial fecal composition of the patient had become almost identical to that of the donor (a relative of the host) fourteen days after the transplant. The disease symptoms had disappeared. In the September 2010 issue of *JCGE*, studies were published that showed that colonization by the transplanted microflora was effective and sustainable, 24 days after the initial transplant (Floch, 2010).

On the other hand, Chaysavanh Manichanh of the University Hospital Vall d'Hebron in Barcelona, Spain, has carried out with Spanish and American colleagues DNA analyses of the intestinal microflora of rats after a biotherapy, preceded or not by the administration of antibiotics. Published in the 20 October 2010 issue of *Genome Research*, these DNA analyses showed that "it was possible to introduce new species into the intestinal microflora without having to previously eliminate endogenous bacteria using an antibiotic treatment (Manichanh et al., 2010).

In addition to treating intestinal affections caused by *Clostridium difficile*, biotherapy could be used in other illnesses of the digestive tract. Dusko Ehrlich of the French National Agricultural Research Institute (INRA), in charge of the Metagenomics of the Human Intestinal Tract (Meta-HIT) programme, which aims at characterizing the intestinal microflora

(microbiome), made a prospective statement : “We may think of autotransplants, with samples taken during youth and used later on to reestablish the balance of the intestinal microflora in case of disease.” According to this researcher, the approach to public health might be changed by microbiome metagenomics, because an increasing number of studies are showing that the composition of intestinal microflora has something to do with diabetes, obesity or intestinal chronic inflammatory diseases – even though we often do not know whether the modifications of this microflora are the cause or the consequence of disease. It also remains to be seen how this biotherapy will be socially accepted (Ehrlich et al., 2010).

STRATEGIES OF THE PHARMACEUTICAL INDUSTRY; CONTRIBUTION TO HEALTH-CARE EFFECTIVENESS

Pharmaceutical companies and biotechnology firms, often working in close collaboration, play a key role in bringing useful innovations to the health-care system in the areas of diagnosis, prevention and therapy. The convergence of biology and engineering is at the origin of a major change in the detection and cure of disease, with the help of information technology, imaging, nanotechnology, and sophisticated modelling and simulation. This trend and other factors have a profound impact on the companies' strategies, concerning in particular their innovation policy, their management and strategic alliances. They must also face the fact that health-care systems throughout the world have to better evaluate the costs and benefits of new technologies before they are adopted on a large scale. In other words, the pharmaceutical industry and bioindustry ought to contribute to the cost-effectiveness of the health-care system.

The reasons for change

The pace of successful drug innovation has slowed over recent years, as progress in the life sciences has proved slow to translate into new medicines, while increasingly risk-averse regulators have delayed decisions and demanded larger, longer and more costly clinical trials. On the other hand, patent expirations and ever more aggressive generic drug company challengers have threatened long-standing sources of revenue. Health-care systems have negotiated drug prices harshly and started to refuse reimbursement of newer and more expensive medicines which do not demonstrate significant additional therapeutic benefit over older drugs (Jack, 2009).

These pressures have increased sharply since the fall of 2008, with the economic downturn restricting credit and spurring fresh cost cuts.

Impact of economic downturn on biotechnology companies

Credit crunch, risk-averse investment, layoffs and bankruptcy threaten innovation

Lack of access to credit as well as risk-averse investors had a profound impact on small biotechnology companies that became far more vulnerable to takeover or licensing deals on unfavourable terms to survive. As biotechnology companies are big consumers of capital – a biotechnology firm should bring in money every two or three years and cannot expect commercialize a new drug before ten years of research –, they have been the first victims of the global financial crisis and credit crunch. According to a study published on 7 October 2008 by France Biotech – the French association of biotechnology companies – investments in the sector (mainly medical and pharmaceutical biotechnology) during the first half of 2008 have decreased by 79% in Europe, and 62% in the United States, compared with the first half of 2007. During the same period, the number of companies becoming public (i.e. listed on the stock exchange) decreased by 82% in Europe and 93 % in the United States (Mamou, 2008 d).

The "Life sciences panorama," a survey carried out every year by France Biotech among 170 biotechnology enterprises has shown that in 2008-2009 (at the peak of the financial and economic crisis) scientific research was pursued. Eleven biomedicines were commercialized in France in 2009 and another 84 were submitted to clinical trials, including ten in phase-3 trials and four in the phase of registration (access to market). Between 2008 and 2009, the number of new biomedicines in clinical development stage had even increased by 20%, from 55 to 66 (Mamou, 2010 b).

Regarding the financial side, the shock has been strong. Venture-capital investments fell down by 56% between 2008 and 2009 (€65 million in 2009 compared with €151 million in 2008). The number of starting operations remained stable (18 in 2008 and 17 in 2009), but the average amount of investments (€1.3 million) went down. Even worse, the funding of the second and third rounds has crashed, from €115 million in 2008 to €43 million in 2009 (Mamou, 2010 b).

The overall losses recorded in 2008 (€239 million) and the decrease in sales (-2%, down to €194.2 million in 2008 compared with 2007, and +67% between 2006 and 2008) were probably of the same order in 2009, but the annual turnover of the 13 companies listed on the stock exchange has remained stable in 2009 (€740.2 million, i.e. -0.75% compared with

2008). Four companies listed on the stock exchange (Cellectis, ExonHit Therapeutics, Nicox and Innate Pharma) were nevertheless able to collect a total of €122 million in six operations. Public assistance has been crucial : 383 biotechnology projects have been supported in 2009 through the assistance to small-and-medium sized enterprises group Oseo, for a total amount of €91 million (Mamou, 2010 b).

Jean-Luc Bélingard, president of Ipsen group, stated that "among the main 800 biotechnology companies across the world, half of them might be taken over or disappear before the end of 2009." At least 10 American companies have gone bankrupt since November 2008, and the San Francisco-based venture-capital firm Burrill estimated that of 360 American companies listed on the stock exchange, 120 had cash for only six months (instead of 12 months one year earlier) [Porier, 2009 b].

SemBioSys Genetics in Calgary, Alberta, Canada, which was working on the production of insulin and other drugs by genetically engineered safflowers, stated in October 2008, that it would lay off about 30 workers, or more than 40% of its work force. Even so, the company's cash might last only until the middle of 2009, said Andrew Baum, its chief executive. Many other biotechnology companies were cutting their work forces and even eliminating research and drug development projects. Some might have to sell themselves at bargain prices, as Avalon Pharmaceuticals did at the end of October 2008 to Clinical Data for US$10 million in stock (Pollack, 2008).

Biotechnology companies accounted for 86, or 25%, of the 344 companies that, as of 9 October 2008, were in danger of being delisted by Nasdaq because their share prices were less than US$1 or they had failed to have an adequate market valuation (Pollack, 2008).

One of those in DeCODE Genetics which has regularly made headlines for discovering genes linked to cancer, heart attacks and other diseases. By combining advanced gene-sequencing technologies with privileged access to the genetic data of Icelanders, the company pioneered the field of personal genomics. Iceland indeed is an ideal place to study the link between genetic variations and diseases, as its population is ethnically homogenous and immigration has been limited. The company was threatened to be ousted from the Nasdaq by early November 2008 if its market value did not climb back above US$50 million. The company's cash was running low and it had trouble paying a US$230 million debt that was to come due in 2011. It has not helped that DeCODE Genetics was based in Iceland, which suffered a financial collapse and that it lost

millions of dollars in auction rate securities. DeCODE Genetics, however, had produced a series of innovations that reconfirmed its status as a global leader in its field. The firm's researchers had unveiled several genetic mutations linked to schizophrenia, made advances on a drug targeting Alzheimer's disease and identified genes linked to basal cell carcinoma. Despite cutting its work force by 30% in 2008, and reducing its cash burn rate by half, the firm's access to credit had dried up in September-October 2008. That was a severe blow for a small biotechnology company without blockbuster revenues. In addition, some US$30 million of its money had been allegedly mismanaged by Lehman Brothers, an investment bank which went bankrupt in September 2008. Kari Stefansson, the firm's founder, stressed the bad investment of DeCODE Genetics' funds in risky American auction-rate securities, which added to the firm's cash squeeze (Pollack, 2008; *The Economist*, 2008 g).

On Thursday 21 January 2010, DeCODE Genetics announced that it had emerged from bankruptcy and would carry on its genetics research and continue to develop its gene-based diagnostics as a private company. In its new form, the company will be run by a new chief executive, Earl Collier, and by its co-founder, Kari Stefansson. E. Collier, a lawyer, was previously a vice-president of Genzyme and a member of DeCODE Genetics' board. K. Stefansson will give up the post of chief executive and become head of research of the company (Wade, 2010).

The new DeCODE Genetics is owned by Saga Investments, an alliance that includes two leading life science investment companies, Polaris Ventures and ARCH Venture Partners. Terrance McGuire, a general partner at Polaris, stated DeCODE Genetics had been recapitalized because its research and database, formed from medical records of the Icelandic population, were a valuable asset. K. Stefansson stated that DeCODE Genetics' research in Iceland would carry on just as before and that the company would "continue to outperform" its mostly university-based rivals in the United States and United Kingdom. The commercial operation will be led by E. Collier in the United States from Boston (Wade, 2010).

Most biotechnology companies – several hundred publicly traded ones ad thousands more in private hands – are unprofitable and can sustain themselves only with periodic infusions of cash from willing investors or pharmaceutical groups. About 113 biotechnology companies, up from 68 in the first quarter of 2008, had less than a year of cash at their current spending rates, according to Rodman & Renshaw, an investment bank. That was about one-third of the publicly traded biotechnology companies it tracked (Pollack, 2008).

Credit crunch was not the main problem for small biotechnology companies, considered so risky that even in favourable times they cannot borrow much money from banks. Some, nevertheless, have issued securities convertible into common stock, which might have to be paid back in cash if stock prices fall below conversion rates. That occurred to AtheroGenics after its drug for heart disease failed in a clinical trial. Paying off US$30.5 million in notes that came due in September 2008 would have left it with little cash to test its drug as a treatment for diabetes. So it defaulted and entered bankruptcy. It has been trying to sell itself or the drug (Pollack, 2008).

Some hedge funds pulled out of biotechnology investing, while others have had to sell shares to cover losses elsewhere or to return money to their investors. It is true that bioindustry had been through two major crises and funding droughts in 1998 and 2002. And most companies survived. But the 2008 crisis came as other factors were already souring investors on biotechnology : pressure to cut drug prices, drug development becoming longer and more costly. Up to October 2008, public and private companies had raised US$5.6 billion, according to the publishing company FDC-Windhover's Strategic Transactions database. That was only one-third the amount in all of 2007 and likely to be the lowest amount since 2002 (Pollack, 2008).

It has been virtually impossible for biotechnology companies to go public in 2008. That deprived venture capitalists of one of the main ways of realizing a return on their investment. And it also meant they had to keep financing their companies longer. Those factors, in addition to the fact that some venture capitalists were investing in publicly traded biotechnology companies the shares of which had become so cheap, resulted in the low availability of money for starting new companies. And when they invested, investors wanted quick returns. For instance, Robert Blum, chief executive of Cytokinetics, a publicly traded company based in South San Francisco, indicated that hedge funds had constantly requested him to spend money only on the company's drugs that were already in clinical trials and to leave aside earlier-stage research aimed at discovering new bioactive molecules. Cytokinetics bowed to that pressure in September 2008, cutting some of its early research and laying off 45 employees, or 29% of its work force. There was therefore a high risk to dry up innovation (Pollack, 2008).

Exceptions

There were exceptions to that bleak situation. The big biotechnology companies, including Genentech and Amgen, had products on the

market and were highly profitable. The biggest companies were in such strong financial situation that their shares were roughly flat for the year, far better than the situation for stocks as a whole (Pollack, 2008). Other smaller firms should be able to find new owners or partners, be they big pharmaceutical groups or private-equity investors. They might therefore emerge stronger from the crisis. DeCODE Genetics' founder, Kari Stefansson, was designing a strategy for a "smaller, leaner" company that, if properly financed, could one day grow into another Genentech (*The Economist*, 2008 g).

In France, despite the gloomy situation in 2008 and at the beginning of 2009, biotechnology companies were not completely exhausted. The period before the crisis had been very favourable : between 2003 and 2007, the number of companies listed on Paris stock exchange rose from 4 to 13, and most of those already publicly traded had increased their capital; they had an average two to three years of cash, according to Sylvain Goyon, analyst at Natixis Securities (Porier, 2009 b). For the less privileged companies, those possessing medicines in an advanced development stage could find some funding, whereas those which had been created more recently and whose research-and-development projects were just starting would probably disappear. It also seemed that potential investors would screen the areas where they wished to invest : biotechnology companies working on cancer research, diabetes and ophtalmology would draw the interest of big pharmaceutical groups. The latter will be tempted to acquire many of these companies, as part of their development strategy; half of the new drugs are derived from biotechnologies (Porier, 2009 b).

France Biotech acknowledged with great appreciation the decision made by the Strategic Council of Health Industries (CSIS) and announced on 26 October 2009 by the president of the French Republic : the creation of an investment fund, InnoBio, amounting to €139 million. Although France Biotech considered that more important means should be allocated to biotechnologies, it drew the attention of the French government to the careful governance of InnoBio, because conflicts of interest might arise with pharmaceutical companies, which were subscribers to the investment fund.

The financial crisis did not change the disbalance between Europe and the United States in medical biotechnology : in 2007, the global annual turnover of the sector amounted to US$95.1 billion, of which 72% in the United States. Based on the data provided by Ernst & Young, the 2007 turnover of European biotechnology companies has been estimated at

US$20 billion, i.e. four times less then in the United States. Although in 2008-2009, there were as many biotechnology companies in Europe as in the United States, the European ones were smaller, less mature, they had less equity and they invested three times less in research and development (US$9.5 billion in Europe, compared with US$30 billion in the United States). British biotechnology companies attracted 37% of the investments made in 2007 in Europe (Mamou, 2008 d).

There are nevertheless many examples of very successful European biotechnology companies. Thus, the French Laboratory of Fractioning and Biotechnologies (LFB, Laboratoire français du fractionnement et des biotechnologies) is the world's sixth biggest company specialized in the production of drugs derived from blood plasma, and the first company of its kind in France. It is a public enterprise, founded in 1994 as a result of the restructuring of blood-transfusion institutions. It has been transformed into an anonymous company in 2005-2006. The whole equity is held by the state. This is considered a very satisfactory situation, according to LFB's chief executive, Christian Béchon, because it does not demand the maximum profitability requested by the private stakeholders in the pharmaceutical sector and it enables the reinvestment of the surplus into research and production. Even more, the Strategic Council of Health Industries authorizes LFB to seek external investments in order to fund the company's development in the area of monoclonal antibodies. In fact, on 26 October 2009, LFB has been authorized to open up the capital of its biotechnology subsidiary to private investors in 2010 (Mamou, 2009).

LFB has four international subsidiaries in Brazil, Germany, the United Kingdom and the Middle East. It is one of the European leaders in the development of monoclonal antibodies and therapeutic proteins derived from biotechnologies. Its annual turnover (2008) amounted to €350 million, the profits reaching €10 million. LFB employed 1,500 technicians and scientists and invested 19% of its annual turnover into research and the strengthening of its production tools (Mamou, 2009).

LFB uses the plasma delivered by the French Blood Establishment (Etablissement français du sang). It bought over 750,000 litres in 2008 for more than €60 million. Drugs derived from blood plasma belong to three categories: haemostatic products used to treat haemophilia for instance (€108 million of annual turnover); intensive care products (€80 million) such as albumin, used to treat heavily burnt persons; immunological products (€156 million) that include many antibodies, e.g. those used to control major immune deficiencies (Mamou, 2009).

In addition, LFB is specialized in the production of drugs for the treatment of rare diseases. For instance, one of these drugs is used by 22 patients in France. LFB is one of the very few laboratories that manufacture drugs for such diseases as the deficiency of blood-clotting XI, that of Willebrand factor – a rare disease of haemostasis – and that of alpha-1-antitrypsin – a genetic disease which affects the lung system. A total of 16 patented products were being commercialized by LFB in 2009; they were delivered to hospitals, and each one of them should be prescribed and administered at the right dose by a physician (Mamou, 2009).

LFB, according to its chief executive Christian Béchon, is a biotechnology company because it develops therapeutic proteins and monoclonal antibodies. At the end of 2008, its two monoclonal antibodies have been injected to patients for the first time. One would treat a Rhesus incompatibility between the foetus and the mother – about 150,000 women were potentially concerned. The other antibody is used in the treatment of a rare from of leukaemia; in France, a few hundred persons would benefit from such treatment. In October 2006, LFB became the major stakeholder of the American company GTC Biotherapeutics, a world leader in animal transgenesis, and in 2007 it purchased Mabgène, a French company based in Alès (Gard, south-east of France), that has an expertise in the production of monoclonal antibodies (Mamou, 2009).

In 2009, 8% of LFB's annual turnover was made outside France and the figure was to reach 13% in 2011. In addition to creating subsidiaries when the economic conditions are favourable, LFB signs licensing agreements with large pharmaceutical groups (Mamou, 2009).

The Asia-Pacific market, that had been for a long time limited to Japanese and Australian companies, had expanded since the opening up of China in 1997 and the creation of Special Economic Zones in India. In 2007, there were only 764 biotechnology companies in Asia, with an annual turnover of US$4 billion, but the growth rate (+21% compared with 2006) was very high and the investments in research-and-development (R&D) were rising by 25%. The attractivity of the region is linked to the political will of governments who wanted to develop biotechnologies, and also to the quality of scientists and low production costs. Many European and American biotechnology and pharmaceutical companies have been multiplying their cooperation activities with China, India, Singapore, Malaysia (Mamou, 2008 d).

Regarding the agreements between big pharmaceutical groups and biotechnology companies, their number (48) had been almost halved in

2007, compared with 2005 and 2006. And in 2008, despite the global financial crisis, there had been an improvement : 64 agreements had been concluded, according to Alcimed, a consultancy. Even though that increase was moderate compared with 2007 and lower than the 2005 and 2006 figures (125 and 87 agreements, respectively), Alcimed showed that the amounts paid by the big pharmas "remained very attractive" : the average amount was US$409 million in 2008, compared with US$435 million in 2006 and US$382 million in 2007. The agreements concluded in 2008 were focused on R&D (83%) and much less on services. Another striking observation concerned the increasing trend for the American biopharmaceutical companies to seek agreements with European biotechnology enterprises.

Reactions of the large pharmaceutical groups

The large pharmaceutical companies, with strong cash flows and generally little immediate decline in sales, have fared better than small biotechnology companies. But Merck's shares have underperformed those of its rivals in 2008. Investors were worried that the group's efforts to find new sources of growth were not as vigorous as those of its peers, which have been buying biotechnology firms and generics-makers, replenishing their product pipelines and moving into new markets. The economic recession in the United States resulted in slow growth of the drug market – the world most important. It seemed that financially squeezed patients without insurance, or with big co-payments, were cutting back even on their medicines. Many drug firms responded by reducing spending on sales and marketing by 10%-20% (*The Economist*, 2008 i).

The overall reaction of the big pharmaceutical groups has been accelerated cost cutting, intensifying rounds of job cuts and outsourcing of production. Part of this trend was Pfizer's purchase of Wyeth, and Merck's of Schering-Plough, because this kind of combination could lead to large and rapid synergies. Roche was better placed than its rivals because of its portfolio of specialty medicines such as for cancer. These drugs are costly, require relatively small sales forces and meet the criteria sought by reimbursement organisms : they are innovative and demonstrate clear value over existing products. Such product specialization is another strategy adopted by companies such as Shine, and increasingly by larger companies like AstraZeneca (Jack, 2009).

In fact the economic downturn accelerated the on-going rethinking by the large pharmaceutical groups of their development strategies. For many years, large pharmaceutical companies have relied on a few high-

price, mass-market "blockbuster" drugs that used to generate billions of dollars a year in sales. But as patents expire on drugs such as Lipitor, Pfizer's anticholesterol medicine that was the biggest selling medication in history, the large pharmaceutical groups need to rethink their business model. As John Lechleiter, head of Eli Lilly, stated : "At a time when the world desperately needs more new medicines … we are taking too long, spending too much and producing far too little. Repowering pharmaceutical innovation is an urgent need." On his side, Nils Behnke, a partner with Bain, the consultancy, estimated that US$100 billion (€79 million) in sales from medicines would be lost over the five-year period 2010-2014 as intellectual property protections expire, while the value of drugs in development in the industry's collective pipeline that could be launched during that period were worth just US$30 billion (Jack, 2010).

Consequently, most big pharmaceutical companies have adopted four principal strategies to diversify. First, expand the range of products in the research and development (R&D) pipeline and the use of external as well as in-house scientists to discover them. Secondly, expand geographically, especially into emerging markets. Thirdly, increase sales of products other than patented prescription medicines. Fourthly, experiment with greater flexibility in pricing in different countries and with ways to ensure drugs provide value for money (Jack, 2010 a).

Improvement of research and development

Chief executives of pharmaceutical groups have suggested the adoption of a more entrepreneurial approach, stimulating in-house the incentive-based, smaller-scale, science-based environment of biotechnology companies, finding external partners and broadening their portfolios. For instance, Merck had started to tap research conducted by other companies and academies. In 2009, it helped to fill holes in its own pipeline with the purchase of Schering-Plough. GlaxoSmithKline in 2009 spun off its HIV/AIDS work entirely into ViiV Healthcare, a joint venture with Pfizer to share the expertise, costs and benefits of new product development. As Jeff Kindler, Pfizer's chief executive, summarized it at the time : "The new company can reach more patients and accomplish much more for the treatment of HIV globally than either company on its own" (Jack, 2010 a).

Most companies have also diversified their research portfolios, shifting from their traditional reliance on chemical-based drugs to biological ones including vaccines. AstraZeneca moved in this direction with its purchase of biological medicine-focused companies such as CAT and Med-Immune.

Roche's full takeover of Genentech in 2009 re-emphasized its focus on biomedicines to treat cancer (Jack, 2010 a).

On 17 September 2010, the pharmaceutical group Johnson & Johnson (J&J) reported that it was negotiating with the Dutch biotechnology company Crucell (specialized in vaccine development) the purchase of the rest of its equity for an amount of US$1.75 billion or €1.33 billion.

Perhaps the biggest trend in 2009-2010 has been removing a taboo on cutbacks in R&D, which had previously been largely spared when marketing and other costs were trimmed instead. For instance, one rationale behind Pfizer's takeover of Wyeth in 2009 was defer the impact of patent expirations. That has allowed it to pare back in-house research, freeing up resources to forge partnerships with external researchers instead. But even if overhauling R&D boosts innovation, the benefits for some companies will not come fast enough (Jack, 2010 a).

New markets

Another means of diversification has been to expand into new geographical regions to boost income from existing medicines. Thus Novartis and Roche Holding had generated nearly 25% of sales from emerging markets in 2008. Merck hoped to achieve this by 2013, and AstraZeneca by 2014. However, the figures remained relatively small, compared with the profits made by these pharmaceutical groups in the American and Western European markets. The United States and Western Europe account for roughly two-thirds of global health-care spending despite accounting for less than 15% of the world's population. Total emerging-market spending was a little more than US$200 billion but is forecast to catch up swiftly. By 2020, developed and emerging markets could be almost equal in size, driven by an expansion of state health-care coverage. China, Brazil and Russia all have made clear commitments to expand coverage (Plumridge, 2010).

Six big countries (Brazil, Russia, India, China, Mexico and Turkey) accounted for only half of emerging-market pharmaceutical sales in 2009, according to IMS Health data. The resulting need for a presence in many countries is complicated by the challenge of diverse operating environments. In Brazil and China, advertising over-the-counter drugs is forbidden, which means pharmaceutical firms require huge sales forces. In India, three-quarters of all health-care spending is by individuals, meaning business need to be adept at selling drugs in small quantities. Not surprisingly, all that is reflected in markedly lower margins on emerging market sales. For

instance, in 2009, GlaxoSmithKline reported operating margins of 36% in emerging markets, compared with 60% and 68% in the United States and Europe, respectively (Plumridge, 2010).

Pricing decisions are important : Roche chose to restrict its market by trying to maintain prices globally. Others opted for volume over profitability, risking price arbitrage across borders. Meanwhile, local players including Russia's Veropharm and India's Lupin and generics makers such as Israel's Teva Pharmaceutical Industries were expanding fast, adding to the competitive pressure. Language and culture, not to mention regulators, can favour domestic rivals (Plumridge, 2010).

Emerging markets will be the primary driver of pharmaceuticals' forecast 4.5% global annual sales growth until 2020, but that in itself would not restore the strength and profits of big drug firms. For that, they need to persuade investors they really can develop innovative drugs (Plumridge, 2010).

Product diversification

Pharmaceutical groups are expanding the width of their own activities, and buying up or forging partnerships with others in different niches. In 2009, Sanofi-Aventis acquired Zentiva in Slovakia, Medley in Brazil and Kendrick in Mexico. Pfizer, by contrast, has so far opted for partnerships, agreeing to license medicines from Aurobindo and Claris Life Sciences in India, as did GSK with Aspen in South Africa. Novartis has moved into eyecare products with its acquisition of Alcon; GSK has strengthened its position in consumer healthcare by purchasing Stiefel for dermatology; and Sanofi-Aventis and Merck have invested in an expanded version of Merial, their joint venture for animal health (Jack, 2010 a).

Such businesses provide a more stable, long-term flow of income than patented medicines. But the markets are often smaller, the margins lower, the competition harsher and the techniques beyond the traditional expertise of the pharmaceutical companies. Geographical expansion compounds these challenges in fragmented, uncertain markets. Daiichi Sankyo of Japan found that out of its cost, when it bought Ranbaxy in 2008 just ahead of intensifying troubles when American regulators forced the Indian generic-drug maker to post large losses (Jack, 2010 a).

Andrew Baum, pharmaceuticals analyst with Morgan Stanley, cautioned that even in a country like China, which currently reimburses many innovative drugs at "Western" levels, the situation could deteriorate. "I wonder about the sustainability of prices," he stated (Jack, 2010 a).

Value and pricing

A fourth option for drug companies has been to diversify their commercial approaches. Some, such as Novo Nordisk and Roche, remain focused on a single global price for their drugs to maximize returns and reduce the risk of "leakage" from lower to higher priced markets. Others continue to justify high prices by stressing their high development costs. But as health-care systems increasingly seek ways to save money, the emphasis is shifting. GSK and Pfizer have sought to boost volumes in poorer countries by offering larger discounts. Novartis, Sanofi-Aventis and Merck have negotiated prices in Europe and North America linked to the performance of their drugs. Others are forging partnerships with physicians, nurses and other specialists to encourage better compliance among patients for whom their medicines do work, but are not taken as prescribed (Jack, 2010 a).

In the long term, as Gary Pisano of Harvard Business School stated : "Real innovation is the source of true long-term advantage in the industry." In the short term, diversification by product and region may at best provide the testing ground to develop new skills that are applicable globally. At worst, it may be one of the few ways for those with the greatest pipeline problems to buy themselves some time (Jack, 2010 a).

Example of Novartis

On 26 January 2010, in Basel, Switzerland, the presentation of Novartis' results for 2009 was superseded by the unexpected announcement of the departure of Daniel Vasella, chief executive officer of the company for the last 14 years, as operational executive of the world's third-biggest pharmaceutical group. Since 1 February 2010, he has been fulfilling the function of chairman, while the post of director-general had been given to Joe Jimenez, an American citizen, 50-years old, who joined the group only in 2007 as director of the pharmaceutical division. D. Vasella who, for 11 years, had been both chairman and executive officer, was one of the last Swiss managers to assume both responsibilities. After having successfully carried out the merger between Ciba Geigy and Sandoz in 1996, he was able to build up a big diversified group in health care. Firstly, by selling out the chemistry activities of Ciba, agrochemistry assets (Syngenta), baby food (Gerber) and medical nutrition activities. Secondly, by transforming Sandoz into a world leader in generic drugs and by moving into the vaccine market in 2006 thanks to the purchase of the American company Chiron (Porier, 2010).

For many observers, the nomination of D. Vasella's successor was a surprise, because J. Jimenez made most of his career in the area of products of large consumption and did not belong to the pharmaceutical world leaders; until 2006, he has been leading the food company Heinz in Europe. Another reason for the surprise was that Jöng Reinhardt, the closest collaborator of D. Vasella, was considered the most likely successor (Porier, 2010).

In addition to the nomination of J. Jimenez, a younger and streamlined directorate was put in place; the members of the administrative board of Novartis will be nine instead of twelve. These changes were considered very appropriate by D. Vasella, just after the group acquired Alcon, the world leader in ophtalmologic products (Porier, 2010).

D. Vasella left the executive position of the group when Novartis had recorded a profit amounting to US$10.3 billion (€7.3 billion) in 2009, an 8% increase over 2008 and much higher than analysts expected. Its annual turnover (2009) rose 7% to US$44.3 billion. The A(H1N1) influenza pandemic had also boosted Novartis' performance : the sales of vaccines generated an additional €1 billion to the annual turnover (2009). Novartis had delivered more than 100 million doses, but according to Andrin Oswald, in charge of the vaccine and diagnostic division, the sales of anti-influenza vaccines in 2010 were expected to amount to only one-third of those recorded in 2009 (Porier, 2010).

On 15 July 2010, Novartis announced that its net profit during the second quarter of 2010 had increased by 19% : US$2.4 billion (€1.9 billion), compared with US$2 billion during the second quarter of 2009. These results were considered "excellent" by the Swiss pharmaceutical group which rose its growth predictions for 2010.

Novartis was considered as the best performing company in the pharmaceutical industry. The group was granted 30 authorizations for the commercialization of new drugs, a record figure. Its portfolio of products in the development stage contained 145 products, and was considered one of the most promising in the sector (Porier, 2010).

The Swiss group, however, and his new director is facing several challenges. The first one concerned the follow-up to the purchase of Alcon; Novartis had to find US$16 billion (€11.4 billion) in order to acquire the majority stake of Nestlé. The second was to mitigate the implications of the loss of patents on some drugs that were widely consumed as well as the competition from generics. Thus, Novartis' antihypertension drug Diovan

(dubbed as Tareg in France) which had generated US$6-billion sales in 2009, was facing the competition from generics in 2010 (Porier, 2010; Smith, 2010).

Another challenge was to find ways and means to respond to the concern regarding the reduction in health-care expenses in most countries. In this respect, D. Vasella had been very conspicuous when he chose to purchase Alcon and thus to have an opportunity to diversify the activities of the group and to benefit from new growth potential. It was possible that Novartis' new director-general would target other acquisitions, but the group's debt capacity was close to saturation; this would mean in case of acquisition that Novartis would be forced to propose shares as a counterpart (Smith, 2010).

The quest for scale

Mergers and acquisitions among the large pharmaceutical companies is a strategy aimed at reaching a sufficient scale at global level in order to better compete with rivals, broadening the portfolio of innovative and patented drugs (as the pipeline of products dries up and blockbuster drugs are losing their patent protection) and at facing the growing competition from generics-makers.

1. Thus, on 26 January 2009, **Pfizer** unveiled a US$68 billion (€53 billion) takeover of **Wyeth**, reasserting its position as the world's largest pharmaceutical group and paving the way for a new bout of consolidation across the sector. This acquisition – to be paid for using equal amounts of cash, equity and debt – has created a group with US$71 billion in sales from a broad range of products; it planned to save US$4 billion in annual operating costs by cutting 15% of its combined workforce (Jack, Saigol and MacIntosh, 2009).

This deal was the eighth-largest merger and acquisition (M&A) involving an American target and it was estimated that the seven advising banks could earn as much as US$150 million in fees. It was also the third-largest globally in the sector after Pfizer's previous US$89 billion acquisition of Warner Lambert in 1999 and the US$79-billion GlaxoSmithKline merger in 2000 (Jack, Saigol and MacIntosh, 2009).

Pfizer had been under growing investor pressure to boost its performance as a series of top-selling drugs come off patent (in 2011, Lipitor, the cholesterol-lowering medicine that contributed a quarter of annual sales in 2009, will lose exclusivity). The company reported a 4% fall in 2009

fourth-quarter sales to US$12.4 billion and a 90% drop in net income to US$266 million after a US$2.3 billion settlement with the District Attorney of Massachusetts probing its marketing practices for painkiller Bextra (Jack, Saigol and MacIntosh, 2009).

Jeffrey Kindler, chief executive of Pfizer, who had, in March 2008, expressed scepticism to financial analysts in New York over the wisdom of "mega-mergers" such as the ones that created his company, conceded that Wyeth's takeover was different. He stressed that Pfizer had been transformed over two years under his leadership, with less bureaucracy, more accountability and a pivotal role for science and innovative medicine in its enlarged combined pipeline of experimental drugs. The takeover offered medium-term advantages for Pfizer. Wyeth's vaccine business allowed it to expand rapidly in biomedicines, a fast-growing, profitable niche in the industry. The consumer health-care division helped it diversify revenues away from prescription medicines. For Wyeth's shareholders, the substantial premium offered was expected to help compensate for much of the loss in the value of their shares in recent years (Jack, Saigol and MacIntosh, 2009).

2. Six weeks after Pfizer's merger and acquisition of Wyeth, **Merck** announced on 9 March 2009 the acquisition of **Schering-Plough** for US$41.1 billion. The new merger created a company with an annual turnover of US$47 billion, the world's second-biggest pharmaceutical group behind Pfizer, and ahead of the Swiss Roche (US$43.1 billion sales in 2008) [Porier, 2009 a].

Merck had to withdraw its painkiller Vioxx from the market in 2004 and to settle many complaints in court. Dick Clark, the chairman of Merck, won praise for his handling of that crisis and for being quicker than his rivals to start restructuring his firm in preparation for leaner years. He was also appointed head of the Pharmaceutical Research and Manufacturers of America (PhRMA), the industry's lobbying arm (*The Economist*, 2008 i). The logics behind the merger with Schering-Plough was clearly defensive. Several blockbuster drugs of the company were coming off patent (in 2010, Cozaar, a drug against hypertension; in 2012, Singulair against asthma; both drugs had about US$8 billion in sales in 2008, a third of total sales). Merck was not able to produce enough innovative drugs to compensate that loss, and consequently the merger was expected to double its portfolio of molecules that were in their last development phase. In addition, the merger would save about US$3.5 billion per year beyond 2011, while shares would rise in 2009-2010 (Porier, 2009 a).

3. After the mergers/acquisitions of Wyeth and Pfizer in January 2009, and of Schering-Plough and Merck in March 2009, the Belgian company **Solvay** announced on 28 September 2009 that it was selling its pharmaceutical business to **Abbott Laboratories** for €4.5 billion. Additional payments up to €300 million were to be made by the American company if commercial objectives were reached in 2011 and 2013 (Porier, 2009 d).

Solvay's pharmaceutical division made a 2008 turnover of €2.7 billion, with a profit margin of 18.8%. But the portfolio of innovative products was too thin to compensate the loss of patents of several drugs, which have been replaced by generics. The merger/acquisition will enable Abbott to speed up its development in Eastern Europe and Asia. The group will not have to pay license fees to Solvay for the commercialization of its anticholesterol drug Trilipix in the United States. Another advantage is that Abbott will have access to several drugs being developed against hypertension and Parkinson's disease, as well as to Solvay's know-how in vaccine production. All these factors were expected to decrease the high dependence of Abbott Laboratories on the sales of its anti-arthritis drug, Humira, which reached 18% of total sales of the company in 2008 (Porier, 2009 d).

4. Some analysts considered that the American **Bristol-Myers Squibb (BMS)** – the world's tenth-biggest pharmaceutical company, with a 2008 turnover of US$20.6 billion – and the British **AstraZeneca** – the world's seventh-biggest firm, with a 2008 turnover of US$31.6 billion – were potential targets for future mergers and acquisitions (Porier, 2009 a).

Lamberto Andreotti, in his first interview since taking over from James Cornelius in May 2010 as Bristol-Myers Squibb's new chief executive, explained that BMS would pursue and intensify his predecessor's plans to "not give up the big pharma legacy, and combine it with what is good in biotech." He said he would also pursue "selective integration" and continue to forge partnerships to share risk, cost and expertise in the development and sale of primary care medicines. In other words, BMS was to continue focusing on innovative prescription drugs and not follow its peers by diversifying into new areas (Jack, 2010 a).

Also among L. Andreotti's priorities were accelerating preparations for product launches – five of which could take place by the end of 2012 – and developing ways to price and provide information on drugs in its pipeline. "This is the company with the most significant changes in the industry in the last few years. In 2007, we were at the point where we had good products and good people but very ugly finances," L. Andreotti stated. BMS embarked on a series of divestments, selling many of its

non-pharmaceutical activities and closing all but a small administrative office in New York's Manhattan, so as to focus on the best of pharma with the best of biotechnology (Jack, 2010 a).

L. Andreotti was examining "appropriate ways" of dealing with pending patent expiry of Plavix, its blood-thinning blockbuster drug, marketed with Sanofi-Aventis. He added that BMS was prepared for an environment in which patients and doctors sought "more data, information and understanding" on a treatment safety, efficacy and cost. He also expressed continuing concern with pricing pressures, notably in Europe, cautioning that "governments are so eager to find quick money, they risk cutting access to innovation" (Jack, 2010 a).

5. **Merck**, of Whitehouse Station, New Jersey, and **Sanofi-Aventis** formed the **Merial Ltd.** joint venture in 1997, and it became one of the industry's biggest players, selling Frontline flea and tick protection for pets, among other products. Merial had sales of US$2.6 billion in 2008. But in September 2009 Merck sold its 50% stake in Merial to Sanofi-Aventis for US$4 billion, giving Sanofi-Aventis full ownership, to gain antitrust clearance for the Schering-Plough deal. However, the sale included a call option for Sanofi-Aventis to subsequently combine Merial with Merck's Intervet/Schering-Plough unit in a new joint venture to be equally owned by Merck and Sanofi-Aventis. The latter had about 100 days from the November 2009 close of the Merck merger with Schering-Plough to exercise the option (Loftus, 2010).

Merck's chief executive officer, Dick Clark, in 2009 stated the company looked forward "to the potential opportunity" to establish a new joint venture, and spokeswoman said it was up to Sanofi-Aventis to decide whether to exercise its option. So by early February 2010, Sanofi-Aventis was expected to renew an animal-health joint venture with Merck. The call-option provision fixed Merial's value at US$8 billion. The minimum total value to be received by Merck for contributing Intervet/Schering-Plough would be US$9.25 billion and could be revised upward by the two parties (Loftus, 2010).

If both companies were to pursue the deal, they probably would have to sell certain animal-health assets to comply with American antitrust regulation, because the companies' combined market share in some segments may be considered as harmful to competition. Merck would also need prior approval from the US Federal Trade Commission, under a provision of the FTC's clearance of Merck's US$41 billion takeover of Schering-Plough Corp. in 2009 (Loftus, 2010).

The main reason behind these huge mergers and acquisitions was that in Europe and the United States two-thirds of drugs used in the treatment of common diseases (hypertension, diabetes, high cholesterol, etc.) were available in their generic form. In addition, public authorities are exerting an increasing pressure on the pharmaceutical companies to reduce the price of drugs, because their overall policy is to decrease health-care costs. Consequently, PricewaterhouseCoopers' experts stressed that "during the next ten years, the model based on the mobilization of numerous marketing teams, on budgets amounting to billions of dollars devoted to the distribution of free drug samples, on costly television advertisements, and on lobbying physicians and patients, were to be seriously questioned." "The blockbuster drug era is over," predicted Thierry Verrechia of Raymond James Euro Equities; "in the future, pharmaceutical groups will not want to depend on a few top-selling drugs, they will try to widen the range of their product portfolio" (Porier, 2009 a).

That explains the numerous acquisitions of biotechnology companies by the big pharmas, which the latter consider a major approach to their innovation strategy. Roche, for example, has clearly chosen to bet on biotechnologies rather than on blockbuster drugs for its future development. On 9 December 2008, at the annual business review conducted at Merck's headquarters in New Jersey, i.e. before the merger with Schering-Plough, D. Clark, the chairman of the company, announced a bold US$1.5 billion plan to enter the emerging market for "biosimilars," that are the biotechnology equivalent of generics. This would put Merck in direct competition both with generics firms, such as Teva Pharmaceutical Industries of Israel – the world leader – and with big biotechnology companies, such as Amgen, which makes the expensive products that biosimilars hope to replace. The reason why Merck may succeed is that it has found a technique to make biosimilars by genetically engineered yeast cells. This could be much cheaper and more reliable than the usual method, using mammalian cells (*The Economist*, 2008 i).

Japanese deals overseas

In Japan, hefty regulations and price controls result in the fact that many new drugs appear elsewhere before they are marketed in this country. The Japanese also spend rather less on their health care than most rich countries – around 8% of GDP – particularly profligate America. To speed up growth, Japanese drug companies have been buying overseas ones. On 17 May 2010, the latest takeover took place, when Astellas, Japan's second-largest drug firm, paid US$4 billion in cash for OSI, the American maker of Tarceva, a lucrative cancer drug, and several promising

treatments for diabetes and obesity. This deal came in addition to the more than US$20 billion that Japanese firms had spent in 2008-2009 on foreign acquisitions. In 2008, Takeda Pharmaceutical, the industry leader, paid US$8.8 billion for Millennium Pharmaceuticals, a big American biotechnology drug company. The same year, Daiichi Sankyo, the third-largest Japanese drug firm, paid US$4.6 billion for Ranbaxy, the Indian generic drugmaker (*The Economist*, 2010 f).

Japanese firms have cash and the national currency, the yen, is strong, making overseas assets relatively cheap. The deals also allowed firms to move in new areas and build a presence in new markets – particularly in America, which accounted for half of global drug sales. Many drugs by Japanese manufacturers were expected to loose their patent protections shortly. As a result, Takeda, Astellas and Daiichi Sankyo all expected a steep drop in operating profits in 2010 (*The Economist*, 2010 f).

The flurry of dealmaking also coincided with a new pricing system, which went into force on 1 April 2010 and made it more attractive to sell new drugs. Products are now exempt from biannual price cuts mandated by the government if they are considered "innovative" (meaning that they have no rivals or generic equivalents) and the companies fulfill other obligations. The new policy was meant to overcome what is known as "drug lag." Foreign companies have been slow to bring new drugs into Japan, because that required expensive and cumbersome trials often duplicating those already carried out in other countries. Moreover, the mandatory price reductions made for small profits. In April 2010, most drugs on the market saw their prices slashed by 6.5% on average, according to Macquarie, an investment bank (*The Economist*, 2010 f).

After so many overseas deals, Japanese drug companies are beginning to resemble foreign ones. Takeda and Astellas have both moved their global research-and-development hub to America. As the new owner of Ranbaxy, Daiichi Sankyo now shares interests with other foreign generic drugmakers. Yet, Japanese firms have not really become global : most of their revenues come from home. The big issue is whether they will be able to integrate smoothly and manage their new acquisitions, i.e. a global workforce of vastly different cultures (*The Economist*, 2010 f).

With three large-scale deals unveiled in less than two months at the beginning of 2009 and totalling about US$150 billion, a series of smaller deals were cast into obscurity. That was the case of Gilead Sciences' US$1.4-billion bid for CV Therapeutics on 12 March 2009. The American

CV Therapeutics, had been fighting Astellas' bid since the Japanese company had agreed in January 2009 to Gilead Sciences' US$20 per share offer (total bid amounted to US$1.1 billion). CV Therapeutics asked its shareholders to tender their shares to Gilead rather than Astellas, which launched a hostile US$16 per share offer for CV Therapeutics' shares on 27 February 2009. Both companies were eager to control CV Therapeutics' two key cardiovascular products – Ranexa, which treats chronic angina, and Lexiscan, an injection used to boost blood flow during heart tests. On 12 March 2009, CV Therapeutics' shares rose above Gilead Sciences' offer to US$20.45 as some investors bet Astellas would come back with a higher price (Jack, 2009).

On 3 September 2009, Dainippon Sumitomo Pharma of Japan announced that it would buy the American drugmaker Sepracor for US$2.6 billion, in order to increase its sales in the United States as well as the products in its pipeline. The acquisition, approved unanimously by Sepracor's board, was to be made with a cash tender offer for Sepracor common shares valued at US$23 each, or almost 28% more than their closing price on Monday 31 August 2009. Sepracor, based in Marlborough, Massachusetts, develops drugs to treat disorders of the central nervous and respiratory systems, notably Lunesta for insomnia, which generates the bulk of the company's revenue, and Xopenex asthma (Nicholson, 2009).

While the strong yen may be hurting Japanese exporters, it has helped Japanese companies go on a buying spree overseas, with takeovers worth US$75 billion in 2008, according to Bloomberg. The pharmaceutical industry has been particularly active in the United States : Takeda Pharmaceutical acquired the Massachusetts-based biotechnology company Millennium Pharmaceuticals for US$8.8 billion in 2008, and Eisai bought MGI Pharma of Minnesota for US$3.9 billion. Dainippon Sumitomo Pharma expected its overseas revenue to climb to 40% of its total as a result of the acquisition of Sepracor. For the year ended 31 March 2009, Dainippon reported a revenue of about US$2.7 billion (Nicholson, 2009).

For the quarter that ended 30 June 2009, Sepracor reported that revenue increased 11% from a year earlier to US$326.2 million, but net income plunged to US$44.9 million. The company expected revenue of US$1.29 billion in 2009 (Nicholson, 2009).

Acquisition of biotechnology companies by pharmaceutical groups

By early 2009, it looked more attractive for the leading pharmaceutical companies to acquire biotechnology companies rather than negotiate complex licensing deals. The severe financial distress that many small biotechnology companies were facing in 2008-2009 forced several into takeovers to survive (Saigol and Jack, 2009).

In fact, in 2007, by one estimate, biotechnology deals in the United States amounted to US$60 billion and to US$34 billion in Europe. Roger Longman of Windhover, an industry consultancy, noted the total value of biotechnology acquisitions by pharmaceutical companies had risen dramatically in 2007 and 2008. The usual explanation was that the big pharmaceutical groups resorted to buying innovative companies in order to overhaul their business model. Another reason related to the regulatory risk : acquiring big biotechnology firms with proven drugs in the market place, rather than cheaper but more speculative start-ups, allowed the pharmaceutical groups to overcome the difficulties of winning regulatory approval for a new drug. Pharmaceuticals also had a lot of cash, which they had decided to redirect into acquisitions that could boost their innovation system. Finally, the weak dollar made it cheaper for foreigners to take over American firms (*The Economist*, 2008 e).

Acquisition of Genentech by Roche

In April 2008, Severin Schwan was appointed as the new chief executive of Roche. His comments to the *Financial Times* reflected the fact that although Roche had performed strongly on the back of specialist treatments, such as cancer drugs, its pipeline of experimental medicines included a number that will be prescribed by general practitioners, such as those for diabetes. He expressed confidence that, in spite of broader industry gloom, his company will bring new drugs to market. It will continue to focus on pharmaceuticals and diagnostics, the division he previously ran before being named to lead the company. Roche concluded the bid S. Schwan coordinated against Ventana, the American diagnostics company (Jack and Simonian, 2008).

As debate intensified over the rising cost of drugs, including a recommendation in the United Kingdom by the end of June 2008 not to reimburse Roche's Avastin for colon cancer (see p. 28), S. Schwan, while recognizing that the price pressure will be tougher, stressed that the best response was not to cut prices but to justify the launch price of

medicines with new clinical data that extended the use of a drug to a broader number of illnesses and patients, in the process shifting "the cost benefit" (Jack and Simonian, 2008).

That may have been a major reason for the buy-out of minorities at Roche's subsidiary Genentech. On 12 March 2009, Roche claimed victory in its protracted takeover of Genentech, winning backing from the American biotechnology group's independent directors after raising its bid for the 44% of the company it did not own to US$47 billion (€36.4 billion) [Simonian and Jack, 2009].

Although Genentech had long been a subsidiary of Roche in financial terms, the American biotechnology company had kept a cultural independence from its Swiss majority shareholder. Merging the two cultures after the takeover was an important challenge. Roche has become the third-biggest pharmaceutical group, just behind Pfizer (and Wyeth) and Merck (and Schering-Plough), with an annual turnover (2008) of US$43.1 billion. The company did not hide its ambition to become the leading health-care group in the world (Simonian and Jack, 2009).

Analysts believed that cost savings – many of which were expected to come from Roche's own operations in the United States, rather than those of Genentech – were likely to exceed the claimed synergies of US$800 million a year, with a direct benefit on future earnings. It was also suggested that the low corporate tax rate reported by Novartis, Roche's Swiss peer and the world's fourth-biggest pharmaceutical group, as well as a shift in the location of intellectual property rights, manufacturing and other activities could reduce Roche's overall tax bill. Such views were shared by Morgan Stanley, which on 12 March 2009, argued in a research note that the cost savings alone should justify the deal to take over Genentech at its final price (US$95-a-share bid), even if clinical data on Avastin proved disappointing (Simonian and Jack, 2009).

Acquisition of ImClone

ImClone, an American biotechnology company had only one product on the market, Erbitux, an anticancer drug jointly marketed with Bristol-Myers Squibb (BMS) that brought in revenue of US$1.3 billion in 2007. Carl Icahn, a legendary corporate raider, owned a stake and was the chairman of the firm. BMS which owned a 17% stake in ImClone was widely considered by analysts to be the most logical purchaser of ImClone. The interest in acquiring the company stemmed from its product-development pipeline,

particularly its next-generation anticancer drug, the rights of which were under dispute in 2008-2009 (Wang, 2008).

BMS' offer, disclosed on 23 September 2008, increased the value of the potential deal to US$4.7 billion. Since then ImClone's stock had been trading in the US$64 to US$65 range, leading many on Wall Street to believe BMS should raise its bid. James Cornelius, BMS' chief executive, made public a letter to C. Icahn acknowledging that ImClone considered BMS' first bid to be "inadequate" and raising its tender offer to US$62 a share for the 83% of ImClone it did not own (Wang, 2008).

C. Icahn, rather than selling the entire company to BMS, wanted it split into two parts : one containing Erbitux and the other inheriting ImClone's future drug-pipeline. He stated a break-up would enhance the firm's total value. BMS insisted ImClone would be most valuable if fully integrated into BMS. J. Cornelius, in a letter to C. Icahn, stated that BMS will solicit "written consents from ImClone stockholders to remove all existing members of ImClone's board of directors and replace them with five highly qualified nominees proposed by BMS. Bristol-Myers is taking this action to ensure that ImClone's board of directors does not prevent the ImClone stockholders from having a direct voice in the process by refusing to satisfy the conditions to our offer." (Wang, 2008; *The Economist*, 2008 e).

This story showed how difficult it could be to strike a deal between a biotechnology company and a pharmaceutical group. Another example was that of Biogen Idec, another American biotechnology firm in which C. Icahn also owned a stake. He was convinced that the firm would be much more valuable as part of a drug giant than an independent firm. So he pushed it to find a buyer. Biogen Idec put itself for sale in 2007, but the recalcitrant management found ways to make the bidding process so onerous and unattractive that nobody made a bid for it (*The Economist*, 2008 e).

Other takeovers

1. When announcing **AstraZeneca**'s annual results on 29 January 2009, the chief executive of the United Kingdom-based pharmaceutical group David Brennan declared that it definitely did not "need a merger or significant acquisition," like those made by Pfizer, Merck or Roche. Maybe the good revenue of AstraZeneca in 2008 justified that position : the full-year revenues for 2008 increased by 7% to US$31.6 billion, once adjusted for exchange-rate fluctuations. The firm also posted a 9% increase in profits over 2007, to US$6.1 billion. Even so, AstraZeneca was facing the same difficulties as the rest of the pharmaceutical industry, e.g.

a drying out of product pipeline and competition from generic versions of its patented drugs.

However, D. Brennan, unlike many chief executives of big pharmas, who are scientists, has a background in sales and he learnt a lot from his experience as a junior sales manager. His strategy was based on three pillars : reducing costs at home, increasing sales in developing countries and foster innovation.

With regard to reducing costs, AstraZeneca was planning to shedding some 15,000 jobs over five years, with a view to cutting costs by US$2.5 billion a year. The company was also shifting manufacturing to developing countries, despite the concerns about quality control and intellectual property protection. In January 2009, a factory of AstraZeneca was expanded in Wuxi, a city a few hours outside Shanghai; it will sell drugs not only in China but throughout Asia; the cost of production was there less than half that in the industrialized world. By 2012 or 2013, the firm planned to export to Europe, and in ten years AstraZeneca expected that as much as a quarter of its global output might come from China.

The second aspect of the firm's strategy was to pursue growth in big emerging markets, especially in China. It was competing with Bayer AG – the German pharmaceutical and chemical company – for the biggest market share among foreign drug corporations in the country's US$19 billion market for modern pharmaceuticals. AstraZeneca's sales were growing faster than the industry average. This was due to the company's sales push into China a decade ago, but also to the trust in local sales managers. D. Brennan has defended the expenses made in sales conferences and promotional visits to physicians, because they helped to educate medical practitioners in countries that lacked a tradition of modern medical education, and to promote a shift towards "evidence-based" medicine. Industry observers have credited AstraZeneca's diligent execution of old-fashioned sales and marketing in remote places of China, as well as the impact of medical education on doctors, for its booming sales.

The third pillar of D. Brennan's strategy consisted of sparking innovation in-house, by making targeted acquisitions of biotechnology companies, including MedImmune and Cambridge Antibody Technology. AstraZeneca had also set up a research centre outside Shanghai focused on drug discovery. For instance, local researchers were working on a drug against lung cancer that seemed to be effective in many Chinese patients, while it was ineffective on Caucasians.

On the other hand, a study the results of which were reported at the European Society of Cardiology's Congress in Barcelona, on 30 August 2009, revealed that AstraZeneca's thrombolytic (anti-clotting) medicine, Brilinta, was more effective than Plavix, a similar drug marketed by Sanofi-Aventis and Bristol-Myers Squibb. Plavix was in 2008 the world's second most sold drug with sales amounting to €6 billion in 2008. Its patent was expected to come off in 2011 in the United States and in 2013 in Europe and Japan. AstraZeneca was expecting to commercialize Brilinta in 2010; the medicine seemed to better prevent heart attacks, without increasing the risk of haemorrhage.

2. When he became chief executive of Sanofi-Aventis in December 2008, Chris Viehbacher had made clear that he wanted to multiply partnerships and to open up the group's research activities. In doing so, he intended to beef up the company's pipeline of innovative drugs, especially when several medicines of Sanofi-Aventis were losing their patent protection. For instance, its anticancer drugs, Taxotère (used against breast, lung and prostate cancers) and Eloxatine (against colorectal cancer), were expected to lose their patent protection in 2010. In 2008, both drugs represented a turnover of €3.4 billion, i.e. 12% of Sanofi-Aventis total annual sales (Porier, 2009 e).

In March 2009, Sanofi-Aventis purchased the American biotechnology company, Bipar, based in San Francisco, for US$500 million (€343 million); the biotechnology company was developing an anticancer drug that looked promising. The substance called BSI-201 had been in phase-3 trials since July 2009. Its mode of action was to prevent the DNA repair in tumour cells that were damaged by chemotherapy. The molecule was being tested in patients suffering from breast cancer called "triple negative," for which there was no efficient treatment; it affected 20% to 30% of patients suffering from breast cancer. It was shown in phase-2 trials, carried out on small groups of patients, that the lifetime of half of the patients was extended by 4.5 months. These results were comparable to those obtained 10 years earlier with Avastin, the anticancer drug developed by Genentech for the Swiss pharmaceutical group Roche. Phase-3 trials generally last three years and aim to test the efficiency of the drug on a large scale. If these trials were successful BSI-201 could be commercialized by mid-2012 or earlier (Porier, 2009 e).

On 11 December 2009, the American health authorities decided to apply the fast track approach to BSI-201; this is an accelerated procedure for the examination of the drug file (the time lapse is shortened by six to ten months), and it generally concerns drugs that have been developed for

diseases for which there are no or very few solutions. This decision might mean that in the best case the drug would be commercialized by early 2011 in the United States and by the end of 2011 in Europe, according to Marc Cluzel, vice-president for research and development of Sanofi-Aventis.

Jean-Jacques Le Fur, analyst at Oddo Securities, stated that if the drug were effective against breast, ovary and lung cancers, it could generate up to €5 billion sales a year, while Béatrice Muzard, analyst at Natixis Securities, was less optimistic and estimated the potential annual sales at €1.5 billion (Porier, 2009 e).

On 10 November 2009, Sanofi-Aventis announced the extension of its partnership with the American biotechnology company Regeneron: from 2010 to 2017, the French corporation was planning to invest US$160 million annually with a view to developing four to five monoclonal antibodies per year with Regeneron.

Also on 23 December 2009, it was reported that Sanofi-Aventis intended to take up to 19.9% of the capital of the Danish biotechnology company Zealand Pharma. The investment of up to €100 million would enable the French pharmaceutical group to have access to the products being developed by Zealand Pharma against diabetes, obesity, etc., and for which there were already partnerships between the company and the French group.

Generics manufacturers and their quest for scale

In 2009, generic drugs represented one-third of total drugs consumed in developed countries and two-thirds of those consumed in countries with low income. During the same year, generics represented a market of €68 billion, while the world drug market was estimated at €670 billion. The generics market would reach €122 billion in 2015 (Bobin, 2010).

IMS Health, an industry research firm, indicated that US$130 billion of prescription medicines would go off patent by 2012, creating a huge opening for generics. In fact generics manufacturers consider themselves as the best ally of governments that strive to mitigate the increase in drugs expenses – about 20% of total health-care expenses in industrialized countries (Mamou, 2008 b; *The Economist*, 2008 d).

There have long been two very different kinds of generics markets: genuinely competitive ones, like those found in the United States, the

United Kingdom, the Netherlands and Scandinavia, and coddled ones, like those of Japan, the rest of continental Europe and much of the developing world. The competitive markets are now becoming "hyper-competitive," in the words of Robert Coury, vice-chairman of Mylan, a big American generics firm. Generics made up nearly two-thirds of the American drug market by volume in 2008, but only 13% by value. Customers, ranging from pharmacy chains to middlemen known as "pharmacy benefits managers," are rapidly consolidating and so gaining greater power over prices (*The Economist*, 2008 d).

Competition is also spreading to places that used to protect domestic firms from foreign rivals, allowing them to preserve much higher margins. Concerns about the soaring cost of health care have prompted a change in Germany, for instance, once one of the world's most closed (and expensive) markets for generics. The European Commission is encouraging other European governments to follow suit. Japan is also opening up. Viren Mehta, an industry expert, highlighted that the country's health-care system paid local pharmaceutical firms some US$30 billion a year for drugs that were no longer protected by patent, and would cost US$3 billion in the United States. But the Japanese government decided to cut its bills by making life easier for foreign competitors (*The Economist*, 2008 d).

Harsh competition is also prompting generics firms to expand, in order to achieve economies of scale. The German generics manufacturer Ratiopharm – the world's fourth-biggest – was a target for acquisition. Eight months after the suicide of the company's founder, Adolf Merckle, both the Royal Bank of Scotland and Commerzbank were in charge of finding a buyer by mid-September 2009. The price would be around €3 billion for a group of which the annual turnover amounted to €1.7 billion in 2008. Among the potential buyers were Sanofi-Aventis, Pfizer and Mylan, Teva Pharmaceutical Industries and Novartis.

On 18 March 2010, Teva Pharmaceutical Industries announced that it had disbursed €3.7 billion to take control of Ratiopharm. Teva therefore brought in twice the annual turnover of Ratiopharm in the deal and thus won the battle with Pfizer and the Icelandic Actavis. The new entity's 2009 turnover amounted to US$16.2 billion (€11.8 billion), with a staff of 40,000 worldwide (Mamou, 2010 a).

Teva Pharmaceutical Industries, already the biggest generics manufacturer and supplier in the United States, became also the leader in Europe, and its share in the global generics market reached 19% in 2009-2010. In

2008, Teva had purchased the American company Barr as well as another one, Ivax, for US$7.4 billion in 2006. On the German market, the biggest in Europe (€6 billion), Teva was strongly competing with Sandoz, the subsidiary of Novartis (Mamou, 2010 a).

After the purchase of Ratiopharm, Teva was not only present in the generic and biogeneric (biosimilar) sector, but also in the princeps drug sector with its "blockbusters" Copaxone (drug used in the treatment of multiple sclerosis) and Azilect (for treating Parkinson's disease) [Mamou, 2010 a].

1. **Teva Pharmaceutical Industries**, the world's leading generics manufacturer, based in Israel, targeted 2012 for doubling its annual turnover up to US$20 billion. In addition to the United States, Teva intended to be a top player on the big European markets, e.g. in France, Spain and Germany. The Israeli group expected to treble the number of marketing authorizations for its drugs over the five-year period 2009-2013, making big efforts in Southern Europe (Italy, Spain and also in France), where generics have a smaller share of the market than in the United Kingdom or the United States. Harsher competition has prompted Teva to reduce its production costs markedly in order to survive in a market that is being "commoditized" (the word commodity is used to designate the generic as a raw material with no real added value, even tough it must keep all the characteristics of the original medicine) [Mamou, 2010 b].

Teva's objective for 2012 was to produce 100 billion drug units (pills, capsules, etc.), compared with 41 billion in 2008. Production capacities had been increased in Hungary, the Czech Republic, the United Kingdom, France, as well as in Ireland and Israel (Mamou, 2008 b).

Teva made a takeover bid for Barr, an American rival, valued at US$7.5 billion. The bid for Barr was in fact the latest (July 2008) in a string of deals upending the generics business. On 23 July 2008, GlaxoSmithKline (GSK) announced it would enter the generics market through a joint venture with Aspen, a South African firm. Barr and Mylan, another American generics company, had been busy acquiring smaller firms (*The Economist*, 2008 d).

2. In 2009, **Sanofi-Aventis** generated US$2 billion in OTC (over-the-counter) revenues with little presence on the American market. The company had been looking therefore for new areas of growth as more of its drug patents began to expire and competition from generic drugs was growing. By

the end of December 2009, the French group agreed to buy Chattem, an American consumer health-care company, in a US$1.9 billion cash deal that was expected to strengthen its diversification away from prescription medicines. Chris Viehbacher, Sanofi-Aventis' chief executive, stated : "The acquisition of Chattem will be a significant milestone in Sanofi-Aventis' transformation strategy and will provide us with the ideal platform in the US consumer healthcare market" (Jack and Rappeport, 2009).

The French company was to pay US$93.50 a share for Chattem, which sells products such as Gold Bond powder, Icy Hot cream and Selsun Blue antidandruff shampoo, pushing Sanofi-Aventis from the sixth to the word's fifth-largest consumer health-care group by revenues. The over-the-counter products division was one of five growth priorities alongside emerging markets, diabetes, vaccines, and new products. The deal helped Sanofi-Aventis gain a foothold in the large American market for consumer health products, while it could also use its extensive international network to expand sales of Chattem's brands in other markets where it was far less present. The French group had made smaller acquisitions in 2009 in the Netherlands, Australia, France and Argentina (Jack and Rappeport, 2009).

Sanofi-Aventis stated its first move would be to convert its prescription Allegra antihistamine brand into OTC product. Chattem would help the group convert other prescription brands for sales directly to consumers. The deal was expected to close in the first year. Sanofi-Aventis was expected to keep the Tennessee-based Chattem's two manufacturing plants in the United States and build a third (Jack and Rappeport, 2009). Sanofi-Aventis also took control of Zentiva, a Czech generics firm in which it previously had a minority stake.

Mylan's vice-chairman, Robert Coury, considered that further consolidation in the generics business was inevitable and insisted that survival for generics companies now depended on "scale, scale, scale and scale!" (*The Economist*, 2008 d)

3. That was the reason behind the purchase in June 2008 of India's biggest generics firm **Ranbaxy** by Japan's **Daiichi Sankyo** for US$4.6 billion. Daiichi Sankyo's Y490-billion acquisition was the largest foreign takeover of an Indian company. But Ranbaxy's share price more than halved since the takeover was agreed in June 2008. Under Japanese accounting rules, companies were required to post valuation losses on their stockholdings if the share price fell 50% or more. Daiichi Sankyo had forecast a Y65-billion profit for its full year, but on 5 January 2009, it warned that a major fall in value of the 64%

holding in Ranbaxy meant its full-year profits were to be lower than expected (Nakamoto and Lamont, 2009).

Daiichi Sankyo's write-down reflected the sharp drop in value of Ranbaxy's market capitalization, triggered by both the global financial crisis and the Indian company's problems with American regulators. In September 2008, the US Food and Drug Administration (USFDA) banned the import of Ranbaxy's medicines because of unresolved concerns over quality audits at two of its factories in India (Nakamoto and Lamont, 2009).

4. On 21 May 2010, **Abbott Laboratories** announced it would purchase the Indian drugmaker **Piramal Healthcare** for US$3.7 billion in order to expand in fast-growing emerging markets and its portfolio of low-priced drugs. In India, drug sales were expected to reach US$8 billion in 2010, double by 2015 and amount to US$50 billion in 2020 (Timmons, 2010).

Piramal Healthcare, based in Mumbai, makes generic and branded drugs in nine manufacturing plants in India, Canada and the United Kingdom, and had the largest sales force in India, with more than 6,000 representatives. The company's net profit increased 52% in the 2009 financial year to 4.8 billion rupees, or US$103 million. Abbott Laboratories stated it would pay US$2.12 billion in cash up front for Piramal Healthcare and US$400 million annually over the following four years. The deal was expected to add immediately to Abbott's earnings, executives said (Timmons, 2010).

Vertical integration, by adding research arms or divisions to conduct clinical trials, is another way to improve or increase generics manufacturers' competitivity. Actavis, an Icelandic generics firm, has acquired over two dozen rivals during the 1990s and early 2000s to become a global player, and was spending about US$236 million (€150 million) a year on research and development (*The Economist*, 2008 d).

There is another growth opportunity that will require more R&D and technical sophistication as well as more funding : "biosimilars" – the new wave of generic drugs that replicate biotechnology-derived medicines. About US$40-billion worth biotechnology drugs will come off patents in 2012, and therefore generics giants have to invest in such products. Within a decade, they hoped, the market of biosimilars could be as big as the entire generics trade nowadays. Teva Pharmaceutical Industries, for instance, had acquired Sicor in 2002 and CoGenesys in 2007 in order to become a leader in biosimilars. The confrontation with Sandoz which already markets biosimilars is very likely (Mamou, 2008 b).

India's generic drug production confronted with new intellectual property-right standards

These new standards should be applied by India, as the country became a member of the World Trade Organization (WHO) in 1995. In 2005, India had to adopt a law that recognized the relevance of patents in the pharmaceutical industry, while at the same time introducing measures to protect, to some extent, the interests of the national generic-drug industry. But in addition to the introduction into Indian legislation of the Trade Related Intellectual Property Rights (TRIPS), India is now encouraged to sign bilateral agreements that are more demanding. Pressure is coming from both Europe and America where multinational drug companies are lobbying actively; they always have denounced the "intellectual piracy" occurring in India on a large scale. The Indian drug market is dominated by generic drugs to the extent of 95% and by national companies in the order of 80%. Its annual growth rate (average) has been estimated at 12%-13% (Bobin, 2010).

Although the European Union has tried to reassure the Indian government that the new bilateral agreement did not aim to hinder the capacity of India to produce and export life-saving drugs, a leakage of the draft agreement raised a major concern. In particular, three clauses drew the attention of the defensors of Indian generic-drug industry. The first one, called "exclusivity of data" would make more difficult the approval by the Indian administrative authority of a generic drug on the basis of trials already carried out by the original producer, who is the owner of data considered as "confidential." A second clause would allow the lengthening of a patent duration by adding to its legal duration (generally 20 years) the period for studying the file by the Indian administrative authority (generally three years). Finally, the bilateral agreement would facilitate the upholding at the border of generic drug cargoes in transit towards third countries. Already, European customs had confiscated cargoes coming from India and in particular on their route to Africa, further to complaints by the original producers of the drugs (Bobin, 2010).

This agreement project made even more pessimistic some Indian professionals on their future. "In 2015, multinationals will control the whole market," stated Yusuf Hamied, the president of Cipla group – which produces generic formulae of retroviral drugs. However, Dilip Shah, secretary-general of the Indian Pharmaceutical Alliance, disagreed; he considered that the alliance Brazil-India-South Africa will strengthen India's resistance to the current multinationals' offensive (Bobin, 2010).

Another concern of Indian authorities relates to the threat over the supply and diversity of medicinal plants used in ayurvedic medicine. Of the 15,000 medicinal plants recorded in India, 7,000 were used in ayurvedic medicine, a traditional know-how which is more than 1,500 years old. More than 80% of Indian population is still using this kind of medicine. In 1988, the World Health Organization had reckoned the importance of these plants in India's health-care system and had adopted a resolution calling for "the safeguarding of these plants that save life" (Bouissou, 2010).

Over ten years, dozens of ayurvedic clinics had been opened in India; they welcome tourists from across the world. This market was estimated at €1.2 billion and its annual growth rate was about 8%. The plants needed for this ayurvedic medicine are often bought from the local communities, who collect them in the forests and woodlands without taking care of their conservation. According to the estimates of the Indian Agriculture and Rural Development Bank, this would be the case of 90% of these plants. Dr Kanjilal of the North-East Institute for Sciences and Technologies, stated that "despite important resources, the medicinal plant sector lacked investments in research and development and suffered from a market which became out of control; the way the plants are being collected leads to the destruction of biological diversity." India's health ministry has published a list of 359 plant species that are threatened with extinction because of their use in traditional pharmacopeia (Bouissou, 2010).

The Indian Council for Medicinal Plants, created in 2000 to safeguard those which are threatened with extinction, has begun to replant some of these plants, like *guggal*, used in the treatment of nervous disorders, and which India has now to import from Afghanistan. The council financially supports Indian States that decide to create conservation areas. About 30 of these areas existed in 2010. These initiatives also aimed at improving the quality of production of medicinal plants, and consequently the effectiveness of ayurvedic treatments. In 2007, during the opening session of a conference on the cultivation of traditional plants, Dr Siddhu, governor of the State of Manipal, indicated that the number of patients suffering from secondary effects further to a natural medicine treatment was increasing. "One of the main reasons is the use of medicinal herbs of poor quality," he stated (Bouissou, 2010).

Shift towards branded generics

It has been already mentioned that cost-conscious governments across the world are trying to decrease the overall cost of their health-care systems, and in particular to reduce the price of patented drugs, while at the same time promoting the sales of cheap generics. In addition,

regulators in the United States and the European Union have been fighting against anticompetitive practices whereby big drug companies pay generics firms to delay the launch of competitor medicines to drugs coming off patent. Governments are also liberalizing drug markets, thus eliminating barriers to the spread of generics (*The Economist*, 2009 g).

It is true that competition between patented drugs and generics is being exacerbated as a record number of drug patents are due to expire over the next few years. Evaluate Pharma, an industry consultancy, estimated that about half of the US$383 billion-worth of patented drugs to be sold in the world in 2009 were to lose patent protection within five years. In 2010 alone, the industry was to see nearly 15% of its revenue from patented drugs put at risk. For instance, in the United States, where competition from unbranded generics is harshest, the price of a given drug falls by more than 85% within a year of patent expiry (*The Economist*, 2009 g).

Consequently, in order to avoid calamitous chops in revenue, big drug companies are peddling "branded" (but not patented) versions of their original drugs for higher prices than unbranded generic equivalents. These branded generics often help the firm losing the patent retain half or more of the market, in value terms, even after generic competition is legally allowed (*The Economist*, 2009 g).

For instance, in the case of Pfizer's Lipitor, sales of this anticholesterol drug in 2008 amounted to US$12.7 billion, i.e. more than one-fourth of the company's annual turnover (US$48.4 billion or €30.9 billion). The best sold drug in the world was to lose its patent protection in 2011 in the United States and Europe. On 18 May 2008, Pfizer announced that the Indian generics manufacturer Ranbaxy had agreed to postpone to November 2011 (instead of 2010) the launch of the first generic of Lipitor on the American market; Ranbaxy also withdrew all the legal procedures against Pfizer's patents on Lipitor that were initiated in 2003. The amount of money agreed upon by both corporations for postponing the launch of Lipitor generic was not indicated (Mamou, 2008 a).

With Lipitor coming off patent, it was estimated that 41% of the company's annual sales would go to generics manufacturers as of 2010. Pfizer's losses started already in 2007 with the disappearance of the patents that protected : Zoloft (an antidepression drug), the sales of which only reached US$500 million in 2007, compared with US$2.1 billion in 2006; Norvasc (a hypotension drug), with US$3 billion sales in 2007, compared with US$5 billion in 2006, and only US$500 million for the first half of 2008; Zithromax (an anti-infection drug), with US$438 million sales in

2008, US$638 million in 2007 and US$2 billion in 2006. According to Timothy Anderson of Prudential Equity Group, Pfizer will lose, between 2010 and 2012, the patents on Aricept (drug used to treat Alzheimer's disease, US$401 million sales in 2007), Viagra (US$1.7 billion), Detrol (a drug used in urology, US$1.9 billion sales), Geodon or Zeldox (a drug used to treat psychiatric ailments, US$854 million) [Mamou, 2008 a].

In addition, Pfizer suffered setbacks in its endeavour to replace drugs losing their patent protection with new ones. Torcetrapib, a drug hat was expected to replace Lipitor, was abandoned after a study carried out on 15,000 patients showed that the death rate was higher among those patients taking the drug than among those taking a placebo. More than US$900 million had been wasted in the clinical trials that failed. Similarly, Exubera, an insulin codeveloped with the French corporation Sanofi-Aventis and thereafter acquired in 1986 by Pfizer for US$1 billion, was not commercialized; it was more expensive than other insulins present on the market and was not prescribed (it represented only 1% of the market). Chantix (used in weaning tobacco smokers), with US$880 million of sales in 2007, that initially was considered a profitable drug, was suspected for aggravating suicide behaviour; its prescription was therefore expected to decrease (Mamou, 2008 a).

However, Pfizer announced that 26 new drug candidates were being tested in clinical trials and that between 2010 and 2012 it should try to obtain the authorization for commercializing of 15 to 20 of them. Even though these 20 products were marketed, their potential sales would not compensate the losses due to generics replacing patented drugs (Mamou, 2008 a).

AstraZeneca was considering to acquire branded generic drugs in emerging countries, as it boosted its business outside the United States and Europe. David Brennan, AstraZeneca's chief executive, stated, however, that the company did not plan a large-scale move into generics. AstraZeneca's plans signalled a shift from its strategy of concentrating on selling high-price patented drugs in the key American and European markets, reflecting the growing importance of emerging markets for the industry as sales in developed countries' markets were slowing down (Berton, 2008).

Annual drug sales in emerging markets were expected to reach US$400 billion by 2020, according to the health-care information firm IMS Health. Through acquiring generic-making rivals, several big pharmaceutical companies could speed up a move into branded generics. And even more important, that helped them to gain better access to emerging markets. Sandoz reported, for instance, that its sales in the

six biggest emerging markets were 14% higher in the first half of 2009 than they were a year ago, while sales in Europe edged up by barely 3% (Berton, 2008; *The Economist*, 2009 g).

Branded generics command a premium in many emerging markets due in part to the fear that unknown products might be fake or of dubious quality. Satish Reddy, chief operating officer of Dr Reddy's Laboratories, an Indian generics firm, argued that by joining with local generics firms, multinationals could have a "cheap access to the middle class" in these markets. Thus, GlaxoSmithKline (GSK) had struck a deal in June 2009 to have Dr Reddy's Laboratories to make a portfolio of inexpensive drugs that GSK will sell as branded generics in the countries where it already has a sales forces. The deal with the Indian generics manufacturer was struck only the month before GSK had acquired 16% of South African maker of generics Aspen Pharmacare Holdings Ltd., gaining access to a number of low-cost branded generics to sell in emerging markets. GSK's Abbas Hussain stated that this new strategy aimed to "build new product portfolios of quality branded medicines which we can combine with GSK's existing extensive sales and marketing" (*The Economist*, 2009 g).

A similar move was made by Sanofi-Aventis in 2009, when it bought both Medley, Brazil's biggest generics firm, and Zentiva NV (Czech Republic), a big supplier of generics in Eastern Europe. In May 2009, Pfizer stated it was negotiating licensing deals with Claris Lifesciences and Aurobindo, both Indian generics firms (*The Economist*, 2009 g).

All these moves highlighted a clear trend for big pharmaceutical groups to embrace generics and to consider that "generics are not the enemy of innovative pharma any longer," as stated by Pfizer's David Simmons. But one may ask whether these alliances of convenience would really last. There is the issue of reconciliation of distinct economic and management cultures. Sigurdur Olafsson, chief executive of the Icelandic generics firm Actavis, remained sceptical. "Generics firms need to be nimble and aggressive," he observed. He reckoned : "Will one of these hybrid firms really be willing to challenge the patents of a rival pharmaceutical firm?" Jeff George, head of Sandoz, the generics branch of Novartis, insisted there was no such clash of cultures, only "a healthy debate of siblings" between his firm and its owner, Novartis. He stressed his company's aggressive entry into the market for "biosimilars." Atul Sobti, the executive officer of Ranbaxy, explained that his concern was not culture but rather the coming cash crunch. The generics business is facing "phenomenally high cost pressures," not least because "some government somewhere will always be ready to chop prices" (*The Economist*, 2009 g).

A "knowledge turn" for the health-care industry

A knowledge turn is a phrase coined by Andrew Grove, the former chief executive of Intel, that defines the time it takes for an experiment to proceed from hypothesis to results, and then into a new hypothesis – around 18 months in electronic chipmaking, but 10-20 years in medicine. A. Grove complained that the health-care industry seemed to innovate much too slowly, while he believed that it was ripe for disruption. The lack of proper electronic medical records and smart "clinical decision systems" concerned him, as did the slow-moving, bureaucratic nature of clinical trials. He therefore thought pharmaceutical firms should study the fast knowledge turns achieved by chipmakers, so that the cycles of learning and innovation are accelerated (*The Economist*, 2009 k).

A. Grove who was 73 in September 2009 and coping with Parkinson's disease, graduated at the top of his engineering class at New York's City College. He then went on to earn a doctorate at the University of California at Berkeley, and wrote a book on semiconductors that remains a reference text. He joined Fairchild Semiconductor, once a pioneering electronic firm, where he drew the attention of Robert Noyce and Gordon Moore. The former was a co-inventor of the integrated circuit, while the latter coined Moore's law (which roughly implies that the amount of computing power available at a given price doubles every 18 months). When the two scientists left Fairchild Semiconductor to found Intel in 1968 – initially to make memory chips, not microprocessors – they took A. Grove with them. He became the chief executive of the company in 1987 and kept that role until 1998, when he became chairman, holding that post until 2004 (*The Economist*, 2009 k).

A. Grove was called by Richard Tedlow, a historian at Harvard Business School, "one of the master managers in the history of American business." One reason for that was market success : under his tenure, Intel dominated the microprocessor industry and its market capitalization rocketed, making it at one point, the world's most valuable company. A more important reason lay in how he managed Intel to become such a spectacular success. In his bestselling book, *Only the Paranoid Survive*, A. Grove argued that every company will face a confluence of internal and external forces, often unanticipated, that will make an existing business strategy unviable. In the case of Intel, that occurred when its memory-chip business suffered the harsh competition from now Japanese rivals, willing to undercut any price Intel offered. A. Grove decided to bet the future of the company on microprocessors (a small niche at that time), a move that saved his firm and transformed the industry (*The Economist*, 2009 k).

The second major decision made was A. Grove's announcement that Intel would market its microchips directly to consumers, and he launched the "Intel Inside" campaign, which started in 1991. This incensed his rivals and his immediate customers, the computer-makers, but the strong demand for Intel's new Pentium chip showed that the strategy had worked. When a minor flaw was discovered in the Pentium chip, the firm was forced to offer a replacement for all affected chips, at a cost of nearly half a billion dollars. That was a painful event and a public relations disaster, but A. Grove believes that it actually benefited the company in two ways. Firstly, it proved to internal sceptics that Intel really had become a consumer brand. Secondly, it bolstered the company's efforts to improve the quality of manufacturing, to protect Intel from future failures. Retrospectively, the risky decision to turn Intel from a component-maker into a consumer brand was a very bright decision (*The Economist*, 2009 k).

In fact, when Intel introduced its widely anticipated 386 processor, A. Grove stunned the industry by declaring that his firm would not license any secondary manufacturer. This was a huge risk for computer-makers, but such was their appetite for the new chip that they bought it anyway. Intel's ability to deliver good enough chips in large numbers meant profits no longer had to be shared with secondary manufacturers (*The Economist*, 2009 k).

The lessons drawn from A. Grove's managerial skills are that the pace of innovation in the industry varies and that one should have a healthy attitude towards creative destruction. A. Grove believes fields such as energy and health care could be transformed if they were run more like the computer industry – and made greater use of its products. In the same way as A. Grove explained that oil extraction and transformation was a sunset industry, arguing that oil and cars were heading for a divorce, and regarding electricity as the most promising replacement fuel, Intel's former chairman strongly recommended that pharmaceutical firms accelerate cycles of learning and innovation, i.e. "knowledge turns" (*The Economist*, 2009 k).

Conclusions

There is no doubt that governments across the world will continue to struggle for reducing drug prices. They will also demand more innovative, targeted and effective medicines, and not just blockbuster drugs applicable to every patient. They want pharmaceutical groups to more actively participate in reducing the huge cost of health-care systems.

The pharmaceutical companies are aware of these demands and are also facing the losses that follow the disappearance of patent protection. They have to adapt and have responded in several ways : drastically reducing their functioning costs, laying off staff, reorganizing their R&D systems and management, making economies of scale as a follow-up to mergers and acquisitions, broadening and improving their pipeline of innovative products through the purchase of creative biotechnology firms (small and big), moving into branded generics, making new alliances with their erstwhile enemies, makers of generic drugs, and moving to promising emerging markets.

One cannot deny that the challenges are great and the design of new strategies in difficult economic times is inescapable. It is the appropriate period to strike deals between governments and health-care authorities, and all the actors of the pharmaceutical industry (including medical biotechnology), in order to increase the health-care effectiveness without hampering the research, development and innovation in the industry.

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PART TWO

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FOOD AND NUTRITION : A CONTEMPORARY CHALLENGING AREA FOR BIOLOGY, MEDICINE AND BIOTECHNOLOGY

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CONTENTS

FOOD, NUTRITION AND HEALTH : PERCEPTIONS AND COMPLEX RELATIONSHIPS

Brief historical perspective

In 1825, Jean Anthelme Brillat-Savarin, a renowned French gastronome, wrote: "Tell me what you eat and I will tell you who you are." Kate Colquhoun, in her book *Taste: The Story Of Britain Through Its Food* (2007), underlined that in the early 19th century it was easy to apply the dictum of J.A. Brillat-Savarin. She wrote that if, like Horace Walpole, at his country seat in Norfolk, you were "up to the chin in beef, venison, geese, turkeys, etc., and generally over the chin in claret, strong beer and punch," you were a member of the squirearchy. If, on the other hand, you were eating white bread, cheese, tea and sugar, you were one of the new urban poor whose diet had recently changed for the worst – from dark bread, souppy stew made with vegetables, grains and the occasional bit of fatty meat, and ale (*The Economist*, 2007 e).

For a long time, at least for the better-off, food has been more than a means of keeping body and soul together. It helped show power, wealth, taste and discrimination. It often reflected the mood of the times. That is why Mark Kurlansky, author of the books, *Cod: A Biography Of The Fish That Changed the World*, and *Salt: A World History*, is of the view that J.A. Brillat-Savarin's dictum should be expanded : "Tell me what you eat and I will tell you who you are, where you live, where you stand on political issues, who your neighbours are, how your economy functions, your country's history and foreign relations, and the state of the environment" (Kurlansky, 2007).

For instance, in England, whereas the court of Henry VIII used to dine in great splendour, the Cromwells did not go in for entertaining, and no one else did either. When England was Catholic, meat was not eaten every Friday, but also for the 40 days of Lent and other holy day as well. Fish was considered a poor substitute. With the dissolution of the monasteries in the 1530s, monastic fish ponds disappeared and fish consumption did not recover for centuries (*The Economist*, 2007 e).

Wars, invasions, immigration have played a key role in the supply of foodstuffs, the transfer and transformation of products and traditions. Potato, beans and tomatoes from the New World once arrived in Europe, became indispensable. When turkeys were introduced in the 16th century (at a great price), they soon appeared on the best tables, replacing roastswans and peacocks of medieval times. Sugar was rare until the Spaniards began to cultivate sugar-cane in the Canaries and Antilles in the mid-1400s, and even then only the rich people could afford it. Cane sugar was also brought by the Arabs when they conquered part of Europe. Only in the mid-17th century sugar started to arrive in quantity from Barbados and the Caribbean (*The Economist*, 2007 e).

The Chinese entered Japan and soybeans were added to the Japanese diet. Jews fleeing Portugal brought chocolate to southwestern France, and African slaves brought frying and okra (gombo, *Hibiscus esculentus*) to the Caribbean and South America. Modern historians questioned the influence of Catherine de Medici when she arrived in Paris in 1533 at the age of 14, with a large Italian cooking staff. That event probably did not upheave French cooking as it is sometimes suggested, but it did bring the artichoke north. And when Louis XVI married Marie Antoinette, sauerkraut became fashionable in Paris and remained popular (Kurlansky, 2007).

Technology also mattered: means of transportation of food and the kind of equipment available to process and cook it. It was not until the early 19th century that open fires were beginning to be replaced by more easily controlled closed ranges. When canned foods first became widely available in the second half of the 19th century (thanks to appertization process, developed by Nicolas Appert in France; see p. 442), they were considered a triumph of wholesomeness over the often adulterated and rotten "fresh" food of the day (*The Economist*, 2007 e).

The reciprocal relationship between food and history has been comprehensively described and seems nowadays obvious. Kate Colquhoun's *Story Of Britain Through Its Food* is among the best contributions to this field. Such relationship is now submitted to the acceleration of the globalization process. The fact that trade is fast and global, migrations are of an unprecedented magnitude, food no longer reflects its place or time. For instance, in the Caribbean, which acknowledged an endless succession of migrations and conquests, there is almost nothing indigenous in the "local" diets. Even China, renowned for its great cuisines, is full of imports in its traditional food, e.g. the hot peppers the Portuguese brought from the Americas. On the other hand, due to the relentless introduction and import of products, from all parts

of the world at any time or season of the year, we are offered endless choices in food more than ever in our history (Kurlansky, 2007).

Globalization, however, does not mean equality. Food remains a global issue with respect to discrimination in daily diets or food intake (quantitatively and qualitatively). The poor still eat mostly starchy products and fats, while the rich can afford animal protein – meat, fish and dairy products. How to improve human nutrition and how best to use locally available foodstuffs is a global challenge, where food biotechnology may play a significant role.

Most of the great cities of the world are increasingly enclaves of the wealthy where cooks and even farmers can experiment without considering cost. According to M. Kurlansky (2007), New York City has set up a system of neighbourhood "farmers' markets". The latter, serviced by local farmers trucking in their goods, are an example of collusion of small-scale farming and wealthy consumers. The prices of baby vegetables, such as tiny carrots, peanut-like squash and bean-sized Brussels sprouts – are very high. The well-off people who generally reject industrial farming, aimed at mass producing for the poor, pay higher prices for the products of small-scale (and often organic) farming (Kurlansky, 2007).

Another trend that characterizes contemporary food and nutrition is the focus of the media on the people who cook for the rich. These have become the luminary chefs and the luxury restaurants have proliferated across the world. But the creativity of great chefs contrasts with less and less cooking taking place in the average home; there is almost nothing in common between food eaten at home and that of restaurants. Although industrial food produced by intensive farming is rejected by a number of social actors and is under pressure to improve the nutritional value of its products, it still feeds the world to a large extent. It tries to respond to the challenge of quality and of keeping costs low. By contrast, there is a global growing market for organic agriculture, livestock and fish products, e.g. fruits and vegetables, free-range poultry, oysters from microbeds. But their high cost often makes these products out of the reach of less-privileged social classes.

Crop biotechnology already plays an important role in the production of food, via genetically modified crops such as maize, soybeans, canola and other species, particularly in the Americas and Asia, and much less in Europe. In 2010, Monsanto and Dow AgroSciences were expected to commercialize the first transgenic maize variety containing eight alien genes to make it tolerant to herbicides and resistant to several insect

pests and rootworm; currently, only three genes can be transferred to the plant. And in 2012, the first drought-tolerant maize variety will be commercialized by Monsanto. According to one assumption, transgenic crops will contribute to 60% of world agricultural production in 2050. Future transgenic crops will not only have useful agronomic traits such as pest resistance and herbicide tolerance, but also qualitative traits such as more nutritious content (vitamins, oligoelements, fatty acids and essential amino-acids, polyphenols and antioxidants) that meet consumers' needs and tastes. For instance, wheat varieties with better baking properties, fruits and vegetables containing omega-3 fatty acids and antioxidants, fresh tomatoes with extended shelf-life, low-fat meat are on the forecasts of the breeders and agroindustrial companies.

Ancient diets and new eating habits

According to Bryan Walsh (2007), "families across the globe are abandoning ancient diets and dining habits" due to accelerated urbanization and Westernization. For instance, in Japan, the characteristics of the country's cuisine are the essence of Japanese culture, i.e. simplicity of fresh and seasonal ingredients in small and well-balanced proportions, and artfully presented. What is true of Japan is also valid in other cultures, as food and diet are symbols of national identity. In Spain, during the very hot summer, a long lunch is followed by the afternoon siesta, a rhythm that is most suited to the prevailing climate. In China, meals of rice, vegetables and pork are usually served in big collective dishes to enable large families and groups of people to eat together. National and eating habits tend to reflect many societal aspects such as class structures, religious beliefs, economic constraints and geography. The act of eating together is a social act and when people share food they strengthen community ties and "are ordering the world around them", as stated by Martin Jones, a bioarchaeologist at Cambridge University, in his book *Feast: Why Humans Share Food* (Walsh, 2007).

Food and eating habits are borrowed from one culture and adapted to another. Thus, Japan received chopsticks from China and tempura from Portuguese sailors. Tomatoes, a basic ingredient of pasta and pizza, were brought to southern Europe by the Spaniards who discovered them in the Americas (the so-called Columbian Exchange). Global exchange has played a crucial role in the evolution and change of diets and eating habits. Nowadays, the rate of exchange has accelerated, trade and communication have grown considerably, and this has led to some kind of homogenization. A diet rich in fats and sugar is displacing grains and fresh vegetables and fruit, mealtimes are shrinking, fast-food restaurants

are spreading, while the risks of diabetes, obesity and heart diseases are increasing worldwide. There is also a countermovement in a number of countries to maintain traditional food culture, "slow food" replies to fast food, even though the trend of globalized eating habits is a reality. Another reality is that of 1 billion people who are starving around the world. Eating three full meals a day is out the reach of a large proportion of humankind, while in the remote history of agrarian societies the rhythms of the day and work shaped the rhythm of meals, and gave rise to the custom of the large midday meal eaten with the extended family and still observed in pockets of southern and western Europe. "There was a very important interconnection between eating together" and consolidating family ties, as stated by Carole Counihan, an anthropologist at Millersville University in Pennsylvania (Walsh, 2007).

Industrialization has led to the production and shipping of a wide range of foodstuffs, including huge increases in the amounts of animal protein and dairy products available. For instance, in China, where tens of millions were starving less than 50 years ago, meat and milk have become far more common, and Chinese youth are on average 6 cm taller than they were just three decades ago. On the other hand, traditional food and eating cultures are losing ground. For instance, the so-called Mediterranean diet – with its emphasis on olive oil, seafood and fresh produce – is not easy to maintain. Italians, for example, begin the day with a small meal called colazione, consisting of light baked goods and coffee; the big meal comes at around 1pm and includes a first course of pasta, rice and soup; a second of meat and vegetables; a third, fruit course, and wine; between the midday meal and a late smaller dinner, there is a small snack, the merenda. Today, there is little tolerance for interrupting economic activity for lunch and worsening traffic conditions in cities hamper workers to come back fast enough and return to their work. Consequently, the formerly small dinner becomes the main meal of the day, the only one which the family has a chance to share together (Walsh, 2007).

Not only do these changes in eating habits have a negative impact on family cohesion, but the nutritional quality of the diet also declines as well. People tend to eat whatever is cheap and quick. For instance, in Latin America, a large family meal at midday, containing much starchy grains like quiñoa and cassava, is preferred in rural areas, but migration to the cities has made such customary eating habits very difficult or even impossible, according to John Brett, a nutritional anthropologist at the University of Colorado at Denver and Health Sciences Center. Once again finding two hours of lunch becomes a luxury families cannot afford, and the pace of modern economy sets the tune. Furthermore, "parents

complain that when they make (traditional) dishes, the kids won't eat them. They want the things they see on television," stated Jeffrey Sobal, a professor of nutritional science at Cornell University (Walsh, 2007).

This is the paradoxical consequence of the less exposure younger generations have to the food their grandparents ate, and of the lesser development of the sensitive palates that allow them to appreciate it. This is due to the fact that women's numbers in the workforce are increasing. As they are working in order to improve the family income, they cannot shop, cook and prepare meals. Frozen and canned food is a quicker solution than preparing traditional meals (Walsh, 2007).

However, some societies have been successful in retaining food cultures and eating habits. For instance, in many Middle Eastern and North African countries, extended families still live together and women still prepare the kinds of traditional meals that women elsewhere no longer can. Diets in the Middle East also show the influence of religion. Besides widely observed taboos on pork and alcohol, the fasting month of Ramadan, when Muslims fast from sunrise to sunset, has become a "feasting month", stated Samy Zubaida, co-author of the book *Culinary Cultures Of The Middle East*; Ramadan nights are marked by heavy calorie ingestion, as the level of food consumption is much higher than during ordinary months; hence the tendency for Ramadan weight gain (Walsh, 2007).

The example of Japan

Among the many rice-growing nations of Asia, there is none so rich, innovative and modern as Japan. Its per capita gross domestic product is higher than that of Germany, its research-and-development budget is one of the highest in the world, its public transport system has no equivalent worldwide and many of its enterprises are global leaders in the manufacture of electronic components and in the car industry. But despite a rush to modernity, Japan is still very attached to its ancient rice culture. Rice-growing calendar has helped to shape Japan's identity since paddy fields were first created about 2,400 years ago. The Japanese are very proud of the quality, taste and stickiness of their rice (*The Economist*, 2009 c).

After each harvest, every farmer's crop is checked by inspectors with magnifying glasses; they shake 1,000 grains of rice into a saucer (the number that fits on the bottom) and count each imperfect one; anything below grade two is considered unfit for the table, and its price falls down accordingly. Yet, rice farmers are disappearing, almost half of them are over 65. Not only the physical effort but also the economics are discouraging.

For instance, in one of the richest rice-growing regions of Japan, Niigata, west of Tokyo beyond the Japanese Alps, to avoid overproduction, the government pays the households (which own about one hectare each) to leave about a third fallow, which means they produce, on average, 40 60 kg-sacks per hectare. A sack sold for about 20,000 yen (US$230) by the end of 2009. That amounted to a yearly income of only about 800,000 yen, which barely covered the cost of machinery (*The Economist*, 2009 c).

Yet this rice is among Japan's best – the snow, it is said, gives the local rice, known as *minami uonoma*, a particular purity. The melting of this snow in early spring in Niigata heralds the start of the planting season that has contributed so much to shaping the Japanese culture. According to mythology, rice was intimately associated with the creation of Japan : the Sun Goddess, Amaterasu, gave grains of rice to one of her descendants, the mythical first emperor Jinmu. This task was to transform Japan into a land of rice. Emperor Akihito, who reigns today (Heisei period), is Jinmu's 125th direct heir. According to Emiko Ohnuki-Tierney, a Japanese anthropologist authority on rice, commented : "It was about the transformation of a wilderness into a land of abundant rice at the command of the Sun Goddess, whose descendants, the emperors, ruled the country by officiating at rice rituals." As these rituals suggest, the planting of rice has an intimate bearing on Japan's religion, shintoism. The religion makes a virtue of the idea of subordination of self-interest to the well-being of the group. This may originate from the traditional labour-intensive rice cultivation, in which all members of the village were required to help sow, weed and harvest, and water had to be shared out with scrupulous fairness (even nowadays, two-thirds of Japan's water resources are devoted to its paddy fields) [*The Economist*, 2009 c].

Rice helped to shape customs and moral, and also history. Rice was a luxury good, served in the elegant bowls of the warlords and samurai at the cost of harsh work in the paddies. But the feudal lords made something else out of rice, that began to be reflected in art, aesthetics, even fashion. Rich women wore representations of rice woven into their 17th-century kimonos. It was during the Edo era, from about 1600 to 1870, a period of isolation in Japanese history, that the rice culture flourished most vividly. Edo, now Tokyo, and Osaka hosted rice-futures markets. The area around Tochikubo, now known as Niigata, was one of the most populated parts of Japan because of the quality of its rice crop. Over the centuries rice became so embedded in Japanese culture that it helped to strengthen a sense of national identity. In the 7th century, the emperor Tenmu commissioned the first myth histories of Japan, the Kojiki and Nihonshoki, to explain national origins. They are replete with rice and they served to

reaffirm Japanese identity just as China was influencing it with a writing system and new culture. But the myths did not recognize that rice came to Japan not from heaven, but from China via the Korean peninsula is about 400 BC, accompanied by Korean farmers who probably went on to populate Japan, outpacing the indigenous Jomon hunter-gatherers (*The Economist*, 2009 c).

As Japan entered the modern era, rice was again at the heart of Japanese history and culture. The Edo period ended with Commodore Perry's ships threatening in the early 1850s to blast eastern Japan with American cannons. Sumo wrestlers were made to carry heavy sacks of rice to show the Americans their strength. But the country was not strong enough to resist the American invasion, and it set to feeding up its people. Women were lured to factory work by the promise of three bowls of rice a day. Soldiers in the second world war were given bento boxes of rice with a plum in the middle to symbolize the rising sun. It was not until the 1960s, however, that everyone had as much rice as they wanted and at that stage, bulldozers made rice paddies squarer and flatter, which enabled farmers to use combine harvesters, pesticides and fertilizers, increasing productivity. The mission assigned to rice farmers was to return Japan to self-sufficiency. It coincided with Japan's industrial renaissance, and with a rising demand for labour in factories producing high-technology goods (*The Economist*, 2009 c).

But the fever of modernization tended to empty the villages. Even as rice harvests grew, Japanese people were eating less rice, and they turned increasingly to bread and meat, much of it imported. Nowadays, each Japanese consumes, on average, about 60 kg of rice a year, roughly half the amount of the early 1960s. Self-sufficiency in rice quickly turned to surplus, and from the 1970s onwards the government has paid farmers not to produce. Farmers felt that they were superfluous, although Tetsuhiro Yamaguchi, a young restaurateur in Tokyo, is doing his best to keep alive the "spirit of rice" – part of the Japanese DNA, he believes. His restaurant, Kokoromai (Heart of Rice) has more than ten types of rice on the bill of fare, and the farms from which each kind comes are listed. Like wine-tasting, the restaurateur makes rice-eating experience a little theatrical : "Rice is the backdrop, like the stage in a theatre; it needs stars and characters – that is where the sashimi comes in," he whispers. He is far from being the only Japanese to become lyrical about rice. The whiteness of the grain is like the soul, it should not even be stained with soy sauce. Its relationship with fish reflects a shared provenance: water. Rice is the only dish that is shared from a common bowl, it is a part of the concept of harmony and communality that is so dear to the Japanese

people. A proverb written about rice serves as a metaphor for humility, also a virtue that the Japanese hold dear : "The heavier the head of rice, the deeper it bows" (*The Economist*, 2009 c).

Many Japanese are also nationalistic over rice. Tetsuhiro Yamaguchi, for instance, would never serve foreign rice. "The Japanese people should only eat rice grown in Japan," he stated. That helps explain the extraordinary protectionism in Japanese agriculture. It is not just farmers who resist free trade, ordinary people in polls state that they are opposed to imports, even though prices would drop. But lower prices might encourage people to eat more rice. Farmers fear if market forces were unleashed, paddies would be forever lost, changing both the landscapes and the traditional orderliness of the Japanese mind. Despite the general trend of declining population of rice farmers, there are signs of entrepreneurship among the remaining old farmers, for instance in Niigata region, in order to keep alive all the traditions associated with rice farming and eating (*The Economist*, 2009 c).

To sum up, the pleasures of traditional eating, the attractiveness of ancient diets, considered as an important part of national culture and identity, the reaction against fast food and the excess fat and sugar in many current diets have inspired movements to advocate the merits of traditionally prepared meals as well as of a healthy approach to feeding oneself. For instance, in Europe, Asia and the United States, the Slow Food movement has been complaining against fast food while championing traditionally prepared meals. In Bolivia, foods fairs are regularly held to celebrate South American staples, even as ways are developed to speed up the time-intensive preparation of native meals. Yet while it is necessary to eradicate fast-food excesses and to preserve the diets that keep people both culturally and physically healthier, it is not realistic to think that can turn back the clock entirely. It should be recalled that, on the one hand, ancient diets or traditionally prepared meals are not necessarily healthier than some industrially manufactured foodstuffs, in terms of balanced nutrient contents and adaptation to modern life (e.g. ancient diets provided more fats to people who had a harsher life and spent more energy in physical tasks). On the other hand, we must adapt our diet to our current way of life, which is more hectic; time is becoming a limiting factor, and in fact fast-food restaurants which precisely tend to respond to the shorter time available for preparing meals and eating them, are increasingly updating their menus and respecting healthy nutritional requirements.

Family values, and flavours and meals of the past

Carlo Petrini, founder of the Slow Food organization, reports that in the 1960s and 1970s in Italy the creation of supermarkets and cheap foodstuffs was accompanied by higher spending on food but also on leisure. Women started to go out to work. Convenience foodstuffs were spreading: margarine tended to replace butter and refined oils displaced virgin oil, factory-produced cakes and pasta were increasingly marketed. In the late 1980s, food processing on industrial scale threatened the domestic and traditionally made produce. The Slow Food movement came into being at a time when uniformization looked set to prevail in a country that was renowned for its regional diversity in food, meals, flavours and eating habits. The founders of Slow Food organization strived to highlight this distinctive feature and the family values that are at the basis of food preparation and consumption. Since the mid-1990s, Slow Food founders and advocates have noticed some changes, which were also in relation with a number of food crises regarding safety and with the protection of the environment (Petrini, 2007).

The revolution in food production since the end of the second world war aimed at increasing this production markedly and also at decreasing the prices of foodstuffs. But, according to C. Petrini (2007), this had a high environmental cost and threatened cultural identity through uniformization. He considers that the example of Europe and North America is being followed by emergent countries; that the values of rural societies should be restored to current food preparation and consumption. Those values which have been lost or forgotten must come back to cooking foods and eating them, e.g. food is better when it is fresh and seasonal, when it is produced close to its place of consumption and when it is eaten by the whole family. Nevertheless, C. Petrini does not advocate a return to a past often characterized by poverty and social backwardness, or to bring back emancipated women to the kitchen. He believes that finding inspiration in the past could help applying good ideas to better produce and consume food (Petrini, 2007).

Spain offers many examples of preservation of old eating habits, which in several cases are not incompatible with modern life. For instance, in the northwestern part of the country, in the rural mountainous environment of Galicia, the midday meal or *cocido gallego* is made of ingredients produced in the immediate neighbourhood of the place where it is consumed. Chickpeas, potatoes, onions, and kale come from the family garden, the meat from the pig slaughtered at Christmas, the rooster was raised in the backyard, and only the veal was purchased for the occasion.

Timing is important in a good *cocido* : every ingredient must be added to the pot at the right moment, according to its relative cooking time. Once boiled, meats and vegetables are strained and served on separate plates, with a fillip of olive oil. *Cocido* is one of those seemingly simple dishes with a wide variety depending on the cook. Traditional cooking and eating in remote villages of Galicia may not necessarily disappear in front of modernity, although one can notice an ever-increasing proportion of purchased food items on the table nowadays. Farmers tend to live better under the European Union's regulations and they like to buy what they need. Another future is to invite visitors and tourists to share many traditionally prepared food during festivals. What used to be daily roborative meals consumed by hard rural workers becomes a gastronomic feast shared by many more people (Itoi, 2007).

Impact of globalization on preparing food and eating habits

Although it is not easy to accept the fact that many restaurants nowadays are an ever-evolving mixture of various types of cuisine and of meal preparation, we are in fact confronted with "a floating café of a whole new global order," as stated by Pico Iyer (2007).

We should acknowledge that, for instance, Thai food is being consumed a long way from Bangkok, just as claret and Burgundy wines can be found in Mexico City and Seoul; and it is not unusual to find *tacos*, *pizzas* and *moussaka* on the menu of some Indian restaurants, accustomed to receive tourists, or to find *wasabi* mousse on offer in London. This reflects the globalization trend as well as the unprecedented magnitude of human flows and migrations, and the subsequent blend of cultures.

Globalization has an impact on gastronomy and "fusion cuisine", but has given to fast food a new flavour. P. Iyer underlines that fast food is regarded by many as the very enemy of individualism and care, and he mentioned the antiMcDonald's movie *Super Seize Me* or the book *Fast Food Nation*. He does not deny the negative impact of fast or junk-food tradition on human physiology and morphology (it plays certainly a role in the obesity pandemic), as well as on family values. Yet, he considers that "it would be hard to deny that such food is not popular across the world just because it is everywhere; it is everywhere because it is so popular" (Iyer, 2007).

When the fast-food restaurants opened in Europe by the mid-1970s, many people had the possibility to eat something that was clean and whose quality was reliable and unpretentious. They could afford something that was not expensive, but not too cheap. For middle-class or even lower

middle-class people, there was an alternative to greasy eateries or to more expensive gastronomy, with a flavour of novelty and sometimes exotism. Furthermore, fast-food chains adapted their meals to local customs, while sticking to some basic items (e.g. hamburger and iceberg salad): they also increased their offer of vegetables and fruit. Finally, they are representative of the concept of "living globally by eating locally" (Iyer, 2007).

Another example of the globalization of food and culinary fusion is that of Mark Brownstein, a Californian food consultant who has specialized in supplying Western chefs with Asian food ingredients. M. Brownstein was originally a botanist and landscape designer. His interest in food began when he started cultivating organic vegetables in Los Angeles to meet the needs of Californian chefs who requested homegrown gourmet items. In 1998, on a trip to Vietnam, he was struck by the export potential of Asian food ingredients, and he set up a modest basement office in Hong Kong. He started supplying comestibles like Laotian *kaipen* (a Mekong seaweed cracker) and wild Philippine honey infused with *kalamansi* lime to renown Honk Kong's and Chicago's restaurants. Later on, he has been sending Vietnamese wild guava liquor, Indian pandan-flower sugar and coconut vinegar to top Los Angeles establishments, and he is now targeting specialty American grocers (Krich, 2007). M. Brownstein has been trying for over a decade, and quite successfully, to identify unusual foodstuffs, hand them over to daring chefs and devise the methods and combinations to make his findings be present on the world's most sophisticated menus. Thanks to an hour-long documentary aired in 2005, he has become known in Germany and France as the "food hunter". During months of filming, M. Brownstein has travelled to markets, villages and homes in India as well as in Vietnam's Mekong Delta and up-country hills. He established links between distillers in Rajasthan forests and cooperative fish-paste producers in Thai mangrove swamps with the most innovative restaurants of Honk Kong, Shangai and Los Angeles (Krich, 2007).

At the high-end of fusion gastronomy, there is also a wide range of examples and experiments across the world that underline the globalization of cuisine and eating habits. Jean-Georges Vongerichten, born in Alsatia, owns seven restaurants in New York City, where he never stopped testing culinary boundaries. Since 2003, he has opened three modern eateries in New York, Houston and Minneapolis; Asia-inspired spaces in lower Manhattan; a steakhouse; a couple of restaurants in London; a Caribbean-theme café in the Bahamas; and two other outposts in Shangai and Bora Bora. He is expanding his restaurant empire to Miami, Istanbul, St. Petersburg, Dubai and Mexico City, among others. In New York City alone, J.G. Vongerichten was employing 936 people and feeding roughly 20.000 persons a week in 2007 (Beech, 2007).

The Alsatia-born chef is not an exception. Although he is considered the most-globe-trotting of all chefs, Frenchman Joël Robuchon runs a string of international restaurants, Austria-born California cuisine purveyor Wolfgang Puck has opened six dining places just in Tokyo, Frenchman Alain Ducasse and Japan's Nobuyuki Matsuhisa are also expanding their business overseas. They all want to extend their culinary kingdom through the discovery of more exotic flavours and sophisticated preparation and presentation of meals, mixing ingredients to obtain different savours. But when does high-end cuisine give way to pure commerce? Celebrity cooks indeed must focus much time on brightening their fame and brand. They also have to take into account the rise of regional cuisine which can compete with their fusion gastronomy; for instance, American and European menus now often heighten the quality and taste of locally sourced ingredients (Beech, 2007).

The target clients of these high-end restaurants travel a lot and disseminate the reputation of culinary icons. In Shangai, J.G. Vongerichten's restaurant on the Bund has succeeded, in large part, because it is an expensive restaurant run by a world-famous chef. But as a gastronomic empire extends so rapidly, mistakes are inevitable: two Vongerichten's restaurants opened in London and Hong Kong have been closed, as well as a New York steakhouse along with the Chinese-inspired restaurant, which could not compete with the close-by Manhattan's Chinatown. Still, Vongerichten has more than a dozen eateries around the world serving hundreds of different dishes prepared by dozens of chefs (Beech, 2007).

J.G. Vongerichten appears convinced by the Asian concept, as he noted that 90% of his line cooks at Asian-themed spice market in New York were Mexican. Both cuisines share key ingredients like chillies, lime, cilantro and mangoes, and can therefore be combined into innovative preparations. The famous chef also summarized his approach to culinary fusion by stating that "we are a mixture of cultures, of races. The world is like that now. I'm cooking for this new world" (Beech, 2007).

Turning foodstuffs into global products : role of food research and biotechnology

Global products also suiting local tastes

Ever since a Swiss miller named Julius Maggi set up a factory in Singen, a southern German town, 150 km away from Stuttgart, in 1887, to produce his eponymous Maggi's Suppenwürze, a liquid condiment used in soups and sauces, Singen has been a kind of world capital of dessicated vegetables.

Singen's Maggi plant and food-research centres are part of a global network of research-and-development establishments run by Nestlé, the world's biggest food company headquartered in Switzerland and selling its products in 130 countries. It has owned Maggi since 1974 (Gumbel, 2007).

In a world where consumers tend to purchase the same kind of products – food as well as beverage or fashion items – Nestlé has not pushed towards standardization, because in food "you have to be very local", stated Thomas Hauser, who runs the Singen R&D centre. It is a policy drawn from 140 years of experience, which does not mean however that Neslté's efforts are also to make its products global. For instance, the company makes about 200 different types of Nescafé instant coffee, one of its biggest worldwide brands. These different types range from the "three-in-one" sachets sold in parts of Asia – which contain the supposedly perfect mix of coffee, milk and sugar for local taste – to the considerably more expensive jars of freeze-dried Colombian Nescafé targeted at French coffee amateurs. In addition to the brand variants, the 800-or-so components of Nescafé are subtly combined to suit national or local preferences and tastes. The same approach applies to chocolate bar that could look the same across the world, but it is in fact different. In the case of the Japanese variant, strawberry, banana and other fillings change seasonally; a Russian KitKat bar is slightly smaller than a Bulgarian one, and the chocolate is not as sweet as in a German one. It is the task of a Nestlé confectionery factory in York, England, to add or subtract key ingredients like sugar or cocoa during the production process, in order to obtain the right nuances in taste that suit the palates of the final consumers (Gumbel, 2007).

Once the right combination of ingredients is achieved, Nestlé's researchers have to come up with a cost-efficient way of manufacturing the product. Nestlé spends over US$1.4 billion in research and development, and employs around 4,000 scientists and technicians. For instance, researchers at Nestlé's Singen centre have developed and produced a "granulated seasoning" which has proved a huge commercial success in Poland and other East European countries and some parts of Asia; they also discovered how to preserve dried broccoli in such a way that it keeps its natural green colour and remains chunky. Maggi tomato soups (dehydrated) have been developed there for Turkish, Indian and European (one for Switzerland, one for Austria and two German offerings) consumers. Although the Maggi brand is the same, the differences concern the cooking time, that ranges from three minutes to ten, e.g. the Turkish one takes longer to prepare because Turkish flour is less refined and thus needs to be heated longer. Green basil is added to the Swiss type, while the German soup is

darker, creamier red than the pure, natural new version; the Indian one is almost orange. The Turkish variety looks pale and watery, because it contains the ingredients corresponding to low retail price in Turkey (it is said that Turkish people cut their own tomatoes into the soup anyway, which most likely compensates the limitations of what they have in their pockets) [Gumbel, 2007].

But how come that national or local taste preferences matter when so many people and different types of cuisine and meal habits are becoming international? Nestlé's answer is that "the taste profile is fixed very early in the human mind", and consequently one will be attracted by the kind of taste closest to the one developed in his/her childhood. But this may be an excuse for discarding or disliking any other food than the one eaten during the early ages. In fact, adaptation of a consumer product to national or local tastes is a trial-and-error process, and in the food industry eight out of ten new products are no longer to be found on shelves two or three years after they have been launched. Furthermore, it is not always possible for a food company to ignore economy of scale, and, for instance, producing four different types of the same consumer product (Gumbel, 2007).

One of Neslté's senior vice-presidents who heads the firm's ice-cream business at his headquarters in Vevey, on the shores of Lake Leman, explained that it was definitely in Nestlé's interest to make different vanilla ice creams for Germany and France: the French one is yellow and beany in taste, almost like a frozen crème anglaise, while the German one is much whiter and more buttery, leaving a warm feeling in the mouth, while the French type is more refreshing. However, a balance must be struck in order to avoid "hyper-complexity". In 2005, Nestlé introduced small squares of chocolate-coated vanilla ice cream in the United States, where they are called Dibs. As they were very successful, in 2007 the company introduced the same product in much of Europe under the brand Pops. The chocolate coating is different in France where people like dark chocolate, from that in Spain where consumers like it a little milkier, *but* the vanilla inside is the same. Nestlé also developed a brand of ice cream for Finland, named Aino. The Finns are among the biggest consumers of ice cream per capita in Europe, along with their Scandinavian neighbours. Appealing to local taste, the ice cream is sold in two flavours associated with Finnish childhood: blueberry pie and cranberry and caramel. The success has been outstanding (Gumbel, 2007).

There are, however, truly global products, which are the same everywhere. The super premium ice cream Häagen-Dazs, which is more expensive

than most Nestlé's ice creams, is not sold in different national types. It is therefore easier to market worldwide. MacDonald's hamburgers are also designed to taste the same everywhere, although the ingredients can differ slightly, and the food company varies its menus from country to country (Gumbel, 2007).

From the experience of Neslté, it appears that the key to success in the food industry is being local, but not too local, at the same time as being global, but not only global. The challenge is to find the appropriate balance from both the tastes and economic viewpoints (Gumbel, 2007).

Globalization of soy sauce

Another outstanding example of a successful global product is that of soy sauce. In 1959, at the International Trade Fair in Chicago, visitors were delighted by the salty-savoury taste of roast beef marinated in a novel condiment, called soy sauce. The young Japanese who were distributing these slices were not merely demonstration staff, but workers at the saucemaker's new American factory, who wanted to see how American consumers reacted to their product. Among them was Yuzaburo Mogi, 24-year-old student at Columbia Business School and the member of one of the founding families behind Kikkoman, a soy-sauce manufacturer which traces its origins to the early 17th century (*The Economist*, 2009 a).

In 1995, when he reached the top of the company, he was able to transform it into an international food business and to turn an Asian seasoning into a global product. Kikkoman indeed is now the world's biggest manufacturer of naturally brewed soy sauce. Foreign sales of the latter have increased by nearly 10% a year for 25 years. Its distinctive curvy bottle is found in restaurants and kitchens across the world. Interbrand, a brand consultancy, ranked Kikkoman among the most recognizable Japanese names in a list generally dominated by carmakers and high-technology firms (*The Economist*, 2009 a).

In 1973, Kikkoman, was the first Japanese food company to open a factory in the United States and Y. Mogi was running the American division by that time. Kikkoman bought American and Japanese companies during its expansion, and at the beginning of 2009, it adopted a holding-company structure which aimed at making acquisitions easier, among other things. Its corporate governance has been innovative, as since 2004 the firm's presidents have come from outside the founding families, and rather than being centrally run from Tokyo, Kikkoman is known for devolving power to the heads of its foreign subsidiaries (*The Economist*, 2009 a).

Kikkoman's annual sales have grown to more than US$4 billion, of which soy sauce accounted for 20%. Most of the company's revenue comes from selling other food products, in Japan and abroad. Kikkoman is the biggest wholesaler of Asian foodstuffs in America, with similar operations in Europe, China and Australia. It sells canned fruit and vegetables in Asia under the Del Monte brand. Foreign sales accounted for 30% of revenue but 55% of operating profit in 2008, three-quarters of which came from North America (*The Economist*, 2009 a).

According to Y. Mogi, America was the perfect place to venture abroad, because it is open to innovation and to incorporating novel ingredients into its cuisine. Y. Mogi understood that he should have to adapt its soy sauce to local cuisine if he was to be successful. In fact, the company promoted soy sauce in America by hiring cooks to develop recipes that incorporated the sauce into classic American dishes. The soy sauce was initially marketed not as a Japanese product, but as an "all-purpose seasoning". The phrase is still present on the bottles of sauce currently sold. In 1961, the company could recruit many new customers by introducing *teriyaki sauce* – a mixture of soy sauce and other ingredients devised specifically for the American market as a barbecue glaze. Kikkoman is also developing products for South American and European tastes, such as a soy sauce that can be poured on rice – which is not done in Japan. In Europe and Australia, Kikkoman's sauce is made without ingredients derived from genetically modified crops – mainly soybeans. In China, where soy sauce is part of Chinese cuisine and cheap products abound, Kikkoman wanted to introduce its soy sauce as a premium product to be bought by wealthy consumers (*The Economist*, 2009 a).

Traditional production of soy sauce

Globalization of soy sauce has not eliminated the traditional production of similar sauces, such as the one made by Kadocho, located in Yuasa, Wakayama prefecture, since 1841 (12th year of the Tenpo era). At the International Agrifood Fair, held in Paris during the third week of October 2010, the representative of this producer underlined that his products were the only soy sauces (*shoyu* in Japanese) made in a traditional way (Mesmer, 2010 a).

In Japan, 1,600 firms were producing *shoyu* in 2010, their overall production reaching more than 900 million liters per year and showing a slow but regular decrease. Each Japanese person consumed an average 7 liters a year in 2009. This sauce was discovered in the 13th century. In 1230, the Buddhist monk Hotto Kokushi travelled to China after

having made his initiation in the temples of Mount Koya, a holy area in Wakayama prefecture. He spent several years in China studying at Kinzanji, the temple of the gold mountain, one of the five most important Zen Buddhism centres during the Song dynasty (960-1279). He learnt the recipe of *miso* paste, also made from soybeans. After returning to Japan, he started producing Kinzanji *miso* in Yuasa, a village close to the sea and built along the Yamada river, whose water is very pure. Later on, Hotto Kokushi discovered that the liquid which settled during the preparation of the paste had a good taste and could be an excellent seasoning (Mesmer, 2010 a).

Soyu production started at that time, and the name given to the soy sauce was *tamari*, derived from the verb *tamaru* which means accumulate. *Tamari* was highly appreciated and was even christened *murasaki* (purple), the colour of Japanese aristocracy, and it still applies to the best soy sauces (Mesmer, 2010 a).

At the beginning of the Edo era (1603-1868), there were 400 *shoyu* manufacturers in Yuasa; the sauce was shipped from there to Kyushu and Shikoku islands. In Kyoto, the imperial court liked the sauce very much. After the discovery of a heating process with firewood, the sauce would be better preserved and exported farther. It was even stated by Kadocho that "King Louis XIV of France tasted Yuasa soy sauce." Nowadays, there are only a few soy sauce-producers left in Yuasa, including Kadocho, the most renowned. His workshop functions like in 1841. The small wooden buildings house big cedar-made vats where a mixture of Gifu wheat, soybeans from Okayama, salt and water ferments during months. Filtering and extraction of the sauce are still carried out with a hand-driven press. Consequently, production is small : 400 bottles of 1.8 liter per day; but there are different flavours. One of the most sought by Japanese consumers is *nigori*, which would taste as the sauce made during the Muromachi period (1333-1573) [Mesmer, 2010 a].

Globalization of kimchi, the emblematic condiment of Korean gastronomy

Cabbage is the basic ingredient of *kimchi*, along with turnip. *Kimchi* is the national condiment in Korea; cold and spiced, it accompanies any meal. It can also be added to some hot dishes. There is a wide variety of *kimchis*, but the one made from cabbage is the most popular. Consequently, the supply of cabbage is of paramount importance. By early October 2010, the price of cabbage soared, from €2 to €10 the piece of cabbage, due to the impact of weather vagaries in September on the overall production.

Some newspapers stated that it was a "national tragedy" for Korea, but the crisis was overcome thanks to imports from China. Cabbage therefore is not an ordinary vegetable for Koreans, and they consume about 2 million tons in the form of *kimchi* (Pons, 2010).

The condiment is prepared in the fall in order to be consumed during the winter. In a household, all women are busy, cutting, slicing, mixing and salting cabbage in a large recipient before letting it lose water, and then mixing it with a paste made of garlic, spices and red chili. The *kimchi* is then stored in brown earthen recipients in the house gardens, or on the balconies, and nowadays in specially designed refrigerators. Every family has its way of manufacturing *kimchi* that is transmitted from mother to daughter. The know-how consists of using the balanced proportions of ingredients, relying on the adequate processing and even "secret" recipes, which every housekeeper knows and keeps alive. One can find packaged *kimchi* in supermarkets, but for those who are faithful to tradition *kimchi* must be prepared at home (Pons, 2010).

It is true that originally *kimchi* was part of the fermentation processes aiming at preserving foodstuffs during winter. Initially, fresh cabbage was soaked in a salty solution – a method known in China and Mongolia, which has been probably adapted to suit local tastes during the Three Kingdoms period (first century BC to 7th century AC). Spices were added to the fermented food later on. Chili was brought into the Korean peninsula during the unsuccessful Japanese invasions at the end of the 16th century, after it reached Japan thanks to Portuguese sailors. Chili was added to *kimchi* during the following century in order to enhance the taste of food consumed by ordinary people. Although there exists a wide regional and seasonal variety of "white" *kimchi* (i.e. without red chili, and made from radish, stuffed cucumbers, small onions, eggplants, sesame leaves marinated in soy sauce, etc.), the "red" *kimchi*, made from cabbage and chili, is the most popular (Pons, 2010).

The emotional shock produced by the lack of cabbage on the Korean market in September-October 2010, testified to the fact that *kimchi* is not only a major condiment but the emblem of Korean gastronomy. There is a museum devoted to it, which was opened in 1986 in Seoul by Pulmuone, the leading manufacturer of fresh food products. There is also an annual festival in the south of the country, in Cholla province. Koreans believe that their national condiment has dietetic and therapeutic virtues : it is rich in fiber, it facilitates digestion, it contains minerals and vitamins which provide energy, while spices stimulate metabolism and garlic reduces the amount of cholesterol (Pons, 2010).

While Korea wants to promote the country's image, particularly through its gastronomy and dietetics, it has decided to open a world institute of *kimchi*, further to the personal initiative of President Lee Myung-bak. About 100 researchers are working in the institute in order to study the health benefits of *kimchi*, the fermentation process and microorganisms involved, with a view to making *kimchi* a global export product. The executive director in charge of new projects at Pulmuone, Kim Hyun-joong, stated that "globalization of *kimchi* implies we have a method which can determine its taste, improve its composition and adapt the product to foreign markets." This was "a great challenge" because, despite its virtues, *kimchi* has a taste and a smell that are not necessarily acceptable to non-Korean palates and noses (Pons, 2010).

The boom of seasonings in the United States

Nowadays Dijon mustard has become as American as French fries, and is the leading brand of all kinds of mustard according to Justin Parnell, director of Grey Poupon mustard, owned by Kraft Foods, which manufactures it from ingredients entirely produced in North America. SymphonyIRI Group, a research consultancy based in Chicago, has estimated that Grey Poupon mustard was the second most-purchased mustard in the United States. In July 2010, its sales reached US$76 million, a 7% increase compared with the figure recorded in July 2009. While in the early 1980s, buying mustard was considered snobbish, nowadays it has become part of the top food curiosity of Americans, observed David Kamp, author of *The United States of Aragula* (Sax, 2010).

There is now a global boost of seasonings or condiments. In addition to traditional brands such as Gulden's and Heinz, one can find on the shelves of supermarkets *wasabi* sauce with lemon, produced by Robert Rothschild farm, as well as Melinda's banana ketchup. Despite the fact that due to the economic downturn people make savings on their food purchases and eat at home more often, decades of tasting exotic food as well as television broadcasts on gastronomy have triggered the need for new flavours at a reasonable price. Consequently, sales of seasonings have increased by 9.4% between 2007 and 2009; they have become the second category of the specialty market, just behind cheese (Sax, 2010).

According to Mintel International Group, a research consultancy based in Chicago, the American market of seasonings has been estimated at US$5.6 billion. In 2010, 36 different kinds of mayonnaise have been marketed, a threefold increase in comparison with 2009. Consequently,

the market value in the United States would reach US$7 billion in 2015, according to Mintel, at a time when the main impetus will come from 18 to 34 years-old consumers. New mustards containing whole grains, spiced ketchup, fruit-seasoned sauces (*salsas*), chutneys and hot sauces will then reach groceries (Sax, 2010).

High-end seasonings in flasks, also sold in plastic bottles in supermarkets, are now being marketed, after being shown at the Fancy Food Show, held every six months and which awards the equivalent of an Oscar to the best pastes for sandwiches. In July 2010, the mildly hot curry ketchup of Dulcet cuisine – an enterprise of Oregon – was awarded the golden medal of seasonings, while its Moroccan mustard obtained the silver medal. "When a flavour becomes popular, other small enterprises manufacture it for their own products," explained Ron Tanner, spokesperson of the National Association for the Specialty Food Trade. "Large groups such as Kraft Foods keep an eye on sales. Once they realize that 10 to 20 small companies produce the new flavour, they start doing it. On average, it takes three to five years for a seasoning to move from a product purchased by few people to a mass product," he added (Sax, 2010).

For instance, Woeber's, based in Springfield, Ohio, which has been producing mustard for the last 105 years, is a forerunner. As a family enterprise renowned for spiced yellow and brown mustard, it now acknowledges a revival, and its annual turnover rose 50%. The recipe has been to produce new seasonings flavoured with *jalapeño* pepper, cranberries and *wasabi*. It also launched a new line of bio mustards, another of high-end mustards (in glass containers), and bought regional brands such as Mr. Mustard of New Jersey. Furthermore, in April 2010, Woeber's started to sell the fashionable flavoured mayonnaises, under the brands Mayo Gourmet (e.g. Kickin' Buffalo, Toasted Garlic and Cool Dill). Christopher Woeber, the grandson of the company's founder, who manages the firm, estimated that it had probably reached two or three times their forecast. In addition, Woeber's manufactures and packages seasonings for other brands or retailers, and this kind of business has jumped by 60%. Private brands, which make up about one-fourth of the seasonings market in the United States, register a growth rate which is faster than any other segment, according to Mintel. This is due to the fact that "people want quality products, less expensive and more specialized, like in the fashion area," stated C. Woeber (Sax, 2010).

Gastronomy hype : perfection on the plate; "molecular gastronomy"

Culinary tradition can lead to perfection in the hands of modern renowned cooks. For instance, Kyoto's Kikunoi Restaurant is a temple to authentic Japanese cuisine, where *kaiseki* – derived from the elaborate 16th century rituals of the Japanese tea ceremony – has evolved into a meal that celebrates the seasons by using only fresh, natural and local ingredients. Unlike other Japanese meal exports such as *teriyaki*, *tempura* and *teppanyaki*, *kaiseki* defies globalization. It is not exportable, it remains local and unique. Yoshihiro Murata is a third-generation *kaiseki* chef; after spending six months in Paris in 1973 to learn how to cook French food, he returned to Kyoto and became a Japanese food advocate. He has written 15 books (one of which, *Kaiseki, The Exquisite Cuisine Of Kyoto's Kikunoi Restaurant*) and he chairs the Japanese Culinary Academy. In addition to the flagship restaurant in Kyoto, he owns a Tokyo branch of Kikunoi (Baker, 2007).

Kaiseki emphasizes fresh vegetables, local and seasonal. Y. Murata qualifies that emphasis as "eating the seasons". The supplier of vegetables is a small farm whose owner Masataka Higuchi has been carrying out since the late 1980s a programme to preserve Kyoto's vegetables, with the help of Y. Murata: tiny eggplants, sweet carrots and spring onions. Taste is very important, as it helps to change cuisine. All ingredients must be of the best quality and in season, nothing is imported. While Western cuisine lays emphasis on contrasting flavours, Japanese cuisine seeks to temper extremes and bring them into a harmonious balance. What keeps those flavours in harmony is *dashi*, a broth made of dried fish and seaweed. Y. Murata uses this broth in every course, but dessert. *Dashi*, like salt, is a flavour enhancer; it helps remove bitterness from a radish, tenderize octopus, or make tomato soup much more tasteful (Baker, 2007).

Kikunoi's *dashi* is prepared in the following way: specially aged *kombu*, a cold-water kelp from northern Japan, is simmered at 60°C for an hour; the *kombu* is removed and the water is brought up to 80°C; translucent pink flakes of smoked bonito, a small tuna fish from southern Japan, are stirred in for 10 seconds (any more, and the broth becomes bitter), and the resulting liquid is strained through a colander. The water used for the preparation comes from a 450-year-old source located below the restaurant. The restaurant in Tokyo uses the same water, so that the *dashi* is as good as in Kyoto's Kikunoi (Baker, 2007).

According to Y. Murata, *kaiseki* is not just about tasteful food, "it is a dramatic art"… "with *kaiseki*, you need to tell a story." In addition, the presentation of the food on the plate is exquisite and mouth-watering; the aim is perfection in hand-crafted pottery, which is also essential to *kaiseki* (Baker, 2007).

By the end of 2007, Hervé This, an agronomy engineer and chemist, published the book titled *Kitchen Mysteries : Revealing The Science Of Cooking*. H. This is one of the founders – with the Hungarian-born physicist, Nicholas Kurti – of molecular gastronomy or cuisine science. The latter tries to explain the chemical basis of food tastes, flavours, and to eventually modify them and/or combine them. For instance, H. This tried "improving" the taste of cheap wine by adding paraethylphenol and paravinylphenol to it : both compounds are essential to the flavour of well-aged burgundy. He wrote that "it is time to discover the very substance of cooking."

He explained why microwaves cook fish well and meat poorly (they heat only water molecules, and those to just below boiling; in other words; in other words, they poach, producing succulent fish, but bland meat); why one should dress a salad just before serving (oil penetrates the thin waxy cuticle that coats vegetables, driving out the air that refracts light and gives vegetables their colour); why potatoes are the perfect food to deep-fry (because of the presence of much starch and sugar on the surface); and why the easiest way to rescue curdled mayonnaise is to wait until the oil separates completely, then just pour it back in, whisking constantly (This, 2007).

In 1984 was published the book *On Food And Cooking* by Harold McGee, who explained the chemical basis of food preparation, thereby debunking culinary myths. But, by so doing, both him and H. This are to a large extent retracing in the laboratory steps that cooks have taken in the kitchen for years.

Relying on molecular gastronomy to build up a reputation of a very high-end and costly gastronomy may be subject to criticism from both the cooks' community and the lucid consumers. Using sophisticated equipment and methods, such as lyophilization, thorough mixing and blending, microwaves and precisely regulated ovens, one could produce particular features of food ingredients, enhance some flavours and reduce others, and devise some odd combinations. Here there could be a relationship with food biotechnology and research, e.g. the addition or withdrawal of some of the numerous biochemical compounds that make

up the particular flavours of foodstuffs (fruits, vegetables, seafood, cured meats, condiments and spices), or the enhancement or repression of some enzymes to break down food ingredients and free their monomeric compounds (bread and cheese making, fruit juices, cocoa and coffee fermentation and roasting).

CURRENT FOODSTUFFS AND BEVERAGES : HISTORY, PRODUCTION AND NUTRITIONAL VALUE

The healthy menu : a global trend

In January 2008, New York became the first American city to pass a law requiring restaurant chains to state the number of calories in every item on their menus. Full enforcement of the new rules began in July 2008. Los Angeles was expected to vote on a similar law in September 2008, while California was considering a statewide bill (*The Economist*, 2008 f).

Those in favour of menu labelling stated it helped consumers, who tended to underestimate the calorie content of the foodstuffs they order. Companies explained that it was costly to reprint menus, and the National Restaurant Association underlined that restaurants would find it difficult to meet a rather wide range of county, city and State standards. The New York State Restaurant Association was so angry at the city regulations that it sued, claiming infringement of commercial freedom of speech under the First Amendment. The American restaurant industry, which had around US$558 billion in sales in 2008, had vigourously fought menu-labelling legislation. Some restaurants, already concerned about the slowing economy, worried they could lose customers if they drew attention to the calorie content of the food they served. But it may be premature to claim whether menu labelling would reduce sales or induce customers to order something different. One study, published in 2008, revealed that customers ordered foodstuffs containing an average of 52 fewer calories when the information was prominently displayed in fast-food chains in New York City. Another study found that diners ordered lower-calorie meals when the menu was labelled – but only on Mondays and Tuesdays (*The Economist*, 2008 f).

Many companies already started to introduce new low-calorie items and serve smaller rations. For instance, Starbucks changed its “default” milk from whole milk to reduced-fat milk, cutting the calorie content in beverages by 14% (reduced-fat milk is also cheaper). Dunkin’ Donuts has a new lower-calorie line called “DD Smart” that is designed to appeal to

the health-conscious clients with egg-white flatbreads and fruit smoothies. McDonald's has reduced the size of a serving of French fries, cutting the number of calories and costs. Le Pain Quotidien, a mid-range bakery chain with about US$165 million in annual worldwide sales and 17 outlets in New York, thought it had profited by adapting quickly to the new rules. A company's team was put together to overhaul the menu, cutting portions and eliminating high-calorie items. This proved a "strategic advantage", according to the company's vice-president of branding, and boosted sales. The company was providing calorie information voluntarily in Washington, D.C., and Los Angeles (*The Economist*, 2008 f).

Fast-food firms adapt their strategies

Fast food was thought to be recession-proof. When consumers need to cut spending, it is logical that cheap meals like those sold by fast-food companies become even more attractive. This was true during the latest economic recession (2009), when fast-food companies attracted customers who could no longer afford to eat at casual restaurants. This was particularly true for America, the home of fast food, with discounts and promotions, such as US$1 menus and cheap combination meals. As a result, in 2009, sales at full-service restaurants in the United States fell by more than 6%, but total sales remained about the same at fast-food chains. In some markets, such as Japan, France and the United Kingdom, total spending on fast food increased. Sales at McDonald's, the world's largest fast-food company, did not decline throughout the economic downturn. Panera Bread, an American fast-food chain known for its fresh ingredients, performed well too, because, according to its chief executive, it offers higher-quality food at lower prices than restaurants. But not all fast-food companies have been as fortunate. Many, such as Burger King, have seen sales fall. In a severe recession, while some people go to fast-food facilities, others eat at home more frequently to save money (*The Economist*, 2010 c).

Analysts expected the fast-food industry to grow modestly in 2010. But the downturn is making them rethink their strategies. Many companies are introducing higher-priced items to entice consumers away from US$1 specials. KFC (Kentucky Fried Chicken), a division of Yum! Brands, which also owns Taco Bell and Pizza Hut, has launched a chicken sandwich that cost around US$5 in 2010. And in May 2010 Burger King introduced barbecue pork ribs at a cheaper price (*The Economist*, 2010 c).

Companies were also trying to entice customers to buy new and more items, including drinks. McDonald's started selling better coffee as a challenge to Starbucks. Its "McCafé" line accounted for an estimated 6% of sales in

America in 2010. Starbucks has sold rights to its Seattle's Best Coffee brand to Burger King, which started selling it at the end of 2010. As fast-food companies shift from "super size" to "more buys" they need to keep customer traffic high throughout the day. Many see breakfast as a great opportunity: McDonald's was to start selling porridge in the United States in 2011 (breakfast has the potential to be very lucrative, because the margins can be high, according to Bernstein, a research firm). Fast-food companies are also adding midday and late-night snacks. The objective is that by having a greater range of items on the menu, "we can sell to consumers products they want all day," stated Rick Carucci, the chief financial officer of Yum! Brands (*The Economist*, 2010 c).

Yet growth opportunities in the United States seem to be limited because the market is considered to be "saturated" in outlets. China is the place where most fast-food chains, like so many industries, see big expansion. Yum! has the greatest presence in China of any Western fast-food company. Already around 30% of the company's profits were coming from China in 2010, and this was expected to grow to 40% between 2010 and 2014. India also looks as a good opportunity. Others planned to serve up more business in Russia and elsewhere in Europe. Given that around 75% of fast-food companies' revenue in Europe comes from people eating in the restaurants (compared with half in America), older European outlets are being refurbished to make them more attractive places (*The Economist*, 2010 c).

The economic downturn also highlighted the importance of size in competing for customers, which meant that more consolidation was likely. Wendy's and Arby's, two American food chains, merged in 2008. Smaller chains may attract private-equity firms, just as CKE Restaurants did at the beginning of 2010 when Apollo Management, a buy-out firm, purchased it (*The Economist*, 2010 c).

Regarding the compliance with government regulations meant to prevent or mitigate obesity, fast-food firms, by providing healthy options, like salads and low-calorie sandwiches, have at least given the impression of doing something about helping to fight obesity. These offerings broaden the appeal of outlets to groups of diners that include some people who do not want to eat a burger. In the future, simply offering a healthy option may not be good enough. The United States' health-care-reform bill requires restaurant chains with 20 or more outlets to indicate the calorie content of items they serve on the menu. A study by the National Bureau of Economic Research, which followed the effects on Starbucks of a similar calorie-posting law in New York City in 2008, found that the average calorie-count per

transaction fell 6% and revenue increased 3% at Starbucks stores where a Dunkin' Donuts outlet was nearby – a sign, it is stated, that menu-labelling could favour chains that have more nutritious offerings. In order to avoid other legislation in America and elsewhere, fast-food companies will have to continue innovating (*The Economist*, 2010 c).

A global trend

Over the past decade, the major trend in food marketing has been the shift towards organic, "natural" and even "whole" foods. Consumers in wealthier countries worldwide have demanded food with minimal processing, in a state as close as possible to their natural one, believing that these foodstuffs were healthier for them and environment-friendly. Such trend has led multinational food companies to increase investments in "functional" foods that are intentionally modified to make them healthier or more nutritious. Consequently, the global market for functional foods was expected to increase from US$78 billion in 2007 to US$128 billion in 2013, according to the consultancy PricewaterhouseCoopers (*The Economist*, 2009 b).

Examples abound, e.g. the enrichment of eggs with omega-3 fatty acids to control hypertension, the addition of phytosterols to margarines to impede the absorption of cholesterol, and the bacteria-enriched yogurts, such as Danone's blockbuster brand Activia, which are supposed to control constipation. Danone was selling yogurt is pharmacies in Barcelona as long ago as 1919. Vitamin B has been added to flour to fight pellagra and vitamin D to milk to combat rickets. Adding iodine to salt has also markedly reduced the occurrence of goitre.

This global trend started in Asia. Encouraged by a government keen to improve public health, Japanese manufacturers have long tinkered with packaged foods to allow them to make health claims. On average, the Japanese spend twice as much per person on functional foods as Americans and nearly three times as much as Europeans. When Coca-Cola wanted to experiment with a supposedly healthy-green-tea flavoured beverage, it targeted young Japanese women first (*The Economist*, 2009 b).

The trend is now spreading most rapidly in the United States. Firms selling energy drinks, breakfast foods or artificial sweeteners are adding ingredients to their items in the hope that the claimed benefits will attract customers. A number of rival food companies have even joined a coalition, called "Smart Choices" to agree on common labelling and advertising standards for functional foods (*The Economist*, 2009 b).

Vegetables and fruits

Dietary recommendations

One of the recommendations of the French National Programme on Nutrition and Health (PNNS), launched by the government is 2001, was "to eat five fruits and vegetables a day". The French health minister, Roselyne Bachelot, stated that this message had been fully received by French people. On 6 May 2010, the minister asserted that this slogan was quoted by 75% of interviewed people, compared with only 36% in 2005. While presenting the results of the national programme, the minister mentioned a study of the French Agency for the Sanitary Safety of Foodstuffs (AFSSA), which showed that the consumption of fruits and vegetables had increased by 10% between 1988 and 2006. However, the survey carried out by AFSSA did not differentiate fresh fruits and vegetables from prepared foods such as soups, preserved fruit salads or vegetables, marmelades, etc., that often contain sugar and preservatives. If one takes account of only fresh fruits and vegetables, consumption is decreasing; this trend started in the 1960s and it was amplified over the last two decades, with an average annual decrease of 0.5%. Another study made by the Research Centre for the Study and Observation of Life Conditions (CREDOC) between 1999 and 2003, also showed that the purchases of fresh fruits and vegetables decreased by 15% over that period. In 2007, an inquiry revealed that 25% of French people did not buy fruits and vegetables because they considered their cost too high (Santi, 2010 a).

French people between 20 and 30 years of age were eating eight times less fruits and vegetables than their parents at the same age. Why? Young people are less used to cook than their elder relatives; the constraints of modern life lead them to buy already prepared meals or food, which are less expensive than fresh products (since the 1960s, the consumption of preserved or frozen foodstuffs has grown considerably). According to the Fruit and Vegetable Interprofessional Association (INTERFEL), French people would consume about 300 g of fresh fruits and vegetables per day, i.e. far less than the 400 g recommended by the World Health Organization (WHO) and PNNS. About 60% of grown-ups are under the PNNS threshold and some 35% are considered small consumers (less than 3.5 servings a day) [Santi, 2010 a].

Households and young people with low income are the least consumers. According to the Abena study, concerning 1,500 persons that received food aid, 94.5% of them were under 3.5 servings a day. These figures were considered "alarming" by the director of the research unit on

nutritional epidemiology of the National Health and Medical Research Institute (INSERM) in charge of the PNNS implementation. The nutritional divide is increasing between those with low income or level of education (who consume less healthy foods such as fish, fruits and vegetables), and those with higher income and who are better informed about nutrition and health. Fruits and vegetables have a proven effect on reducing the incidence of cardiovascular diseases, diabetes, obesity and eventually cancers of the digestive tract. The World Health Organization considers that a low consumption of fruits and vegetables is at the sixth rank among the 20 risk factors of human mortality worldwide, just behind such a rife as smoking. That is why one of the key objectives of the PNNS is to convince "at least 25%" of "small" consumers to eat the five servings a day of fruits and vegetables. Since the beginning of 2010, fruits have been distributed in schools during the breaks between courses: 350,000 children in primary schools are benefiting from this initiative, which would be extended to high schools in 2011 (Santi, 2010 a).

On the other hand, one year after having been launched, the study called "NutriNet-Santé" and which concerned 130,000 volunteers, delivered its first results on 11 May 2010. The study was to be carried out over five years; it is coordinated by the research unit on nutritional epidemiology (INSERM/ National Agricultural Research Institute - INRA/University of Paris XIII) and its objective was to reach 500,000 persons. For the first time, thanks to this study, the intakes of food polyphenols have been measured. These natural substances of plant origin are found primarily in coffee (32.6%), fruits and vegetables (25%), chocolate (14%) and tea (13.4%). They exist in different forms and they are given several properties: antioxidants, antiinflammatory, anticarcinogenic, protection of the cardiovascular system. The study indicates that polyphenols could play a key role in the prevention of chronic diseases. Research is being pursued in order to better determine the recommended daily intakes (they are currently estimated at 835 mg per day for the French population) [Santi, 2010 a].

With regard to the consumption of fruits and vegetables, nutritionists are concerned by the presence of pesticides that could remain on them and which are harmful to the consumers' health. In France, a study carried out in 2007 on 3,742 samples by the General Directorate for Competition, Consumption and Repression of Fraud (DGCCRF), has shown that almost 50% of fruits and vegetables sold on the market contained herbicides, fungicides and insecticides. According to this study, 58.7% of vegetables did not contain pesticide residues, but 7.2% contained more than the maximum authorized concentration (LMR), which should forbid their commercialization. That concentration was found mainly in peppers, tomatoes, leeks, salads

and spinach. By contrast, carrots, potatoes and cucumbers did contain less pesticide residues than the LMR. Regarding fruits, 29.7% did not contain residues, while 8.5% did not comply with the regulation (this was the case primarily of strawberries, mandarines and grapes, while peaches, bananas and apples were less problematic) [Santi, 2010 a].

The recommended means to eliminate pesticide residues is to wash fruits and legumes, preferably with water and soap, and to rinse them abundantly. In the case of cabbage and salads, it is recommended to take away the external leaves. Consuming organic fruits and vegetables is obviously the solution, but it is expensive; the proportion of "bio" fruits and vegetables in the overall consumption is about 3%. It is also recommended to consume seasonal products, which generally contain less pesticide residues; by contrast, products that are transported over long distances in order to reach the markets at a different seasonal period, contain more biocides needed for their conservation (Santi, 2010 a).

It seems that fruits and vegetables contain less and less pesticide residues, according to Marie-Josèphe Amiot-Carlin, a senior researcher at the INRA/INSERM, but this specialist stated that we lack information on the combined effects of several molecules. This is a priority research area (Santi, 2010 a).

Chili peppers : spicing up world cuisine and nutritional value

Europeans had used black pepper (*Piper nigrum*) as a medicinal aid and to spice up their food since Greek and Roman times. The ingredient, imported from the Spice Islands of Asia, had triggered the economies of trading ports like Alexandria, Genova and Venice. By the Middle Ages, black pepper had become a luxury item, so expensive that it was used to pay rent and taxes. Later on, European traders looked for new ways to India and the lands beyond – not just for pepper but also for other lucrative spices, and for silks and opium (Robinson, 2007).

In 1492, when Christopher Columbus set off from Spain to find a westward route to Asia and the East Indies, he never arrived there, but in the islands of the New World he found a fiery pod that would, within years, not only spice up southern European cooking with new flavours, but also radically change cooking in India, China and Thailand, the very places C. Columbus had wanted to reach (Robinson, 2007).

Archaeologists have reported that they had found traces of domesticated chili peppers on 6,000-year-old cooking utensils used in Central and

South America. According to Linda Perry of the Smithsonian Institution's National Museum of Natural History, "it looks like people have liked spicy food for a very long time" (Robinson, 2007).

All hot peppers belong to the genus *Capsicum*, and contain a substance called capsaicin that causes the spicy hot flavour. Capsaicin comes from the Greek word *kapto* which means to sting, to burn. In the New World, seeds of chili peppers are widely scattered by birds, which apparently are not affected by the burning effect, while indigenous tribes used to scatter the seeds inside their canoes to drive away evil spirits.

The discovery made by the scientists of the Smithsonian Institution's National Museum of Natural History in Washington revealed that domesticated chilies were being eaten in southern Ecuador some 6,250 years ago. Because there are no wild chilies in southern Ecuador, domesticated plants must have been brought there perhaps from Peru or Bolivia, where chilies were probably first grown by humans. Only five of *Capsicum*'s 25 species have been cultivated, and in South America, where most of the world's wild chilies are still found, their shapes and colours are far more varied than the classic curved red or green ones of Mexican cooking or the small bullet-shaped "bird's-eye" chilies used in Thai cooking, or the sweet green and orange bell peppers or capsicums found in salads. There are pea-shaped chilies, heart-shaped chilies, and chilies that are flat and long like a bean. They come in purple, rusty red, yellow, black, bright orange and lime green. According to Paul Bosland, director of the Chili Pepper Institute at the New Mexico State University in Santa Fe, "there are thousands of types and we are still discovering new ones; the variations are incredible" (Robinson, 2007).

By the time Columbus sailed into the Caribbean in the late 15th century, chilies were a long-established part of most diets across the Americas. According to British author Lizzie Collingham (*Curry: A Tale Of Cooks And Conquerors*), Europeans initially were not so fond of the new spice that Columbus brought back from the New World. "On the Iberian peninsula," wrote Collingham, "chilies were grown more as curious ornamental plants than as sources of a fiery flavouring." However, Portuguese traders carried chilies to settlements and colonies in West Africa, in India and around East Asia. Within 30 years of Columbus' first journey, at least three different types of chili plants were being grown in the Portuguese enclave of Goa, on India's west coast. The chilies, which probably came from Brazil via Lisbon, quickly spread through the subcontinent, where they were used instead of black pepper (Robinson, 2007).

In Thailand, a short-lived Portuguese presence failed to convert the locals to Christianity, but succeeded in modifying Thai cuisine. European traders also introduced the spice to Japan. Chilies entered existing local trade routes and were taken to Indonesia, Tibet and China. The speed of their spread was extraordinary. Within half a century of chilies arriving in Spain, they were being used throughout most of Asia, along the coast of West Africa, through the Maghreb countries, in the Middle East, in Italy, in the Balkans and across Eastern Europe as far as present-day Georgia. Chilies probably spread so rapidly because they were easy to grow under a wide range of climates and conditions, and therefore cheap and always available. "It was probably the very first plant that was globalized," stated Paul Bosland (Robinson, 2007).

Few other foods have been taken up by so many people in so many places so quickly. In addition, chilies have become an integral part to the cooking of people who adopted them, and so deeply embedded in their culture. Comparatively, tomatoes and potatoes, also brought back by Columbus from his journeys, took much longer to spread through Europe and Asia. Another reason for the globalization of chilies was that they could be added to such staples as cereals (rice and maize) or even potatoes, and make them rich in flavour for hundreds of millions of poor who could spice up their basic food ingredients (Robinson, 2007).

In recent years, chilies have returned to Europe from Asia on the menus of Asian restaurants. For instance, Indian food is now the most popular cuisine in the United Kingdom. In the United States, where, of course, the chili had arrived thousands of years ago from further south, Mexican food is ever more popular; salsas and chili sauces have outsold tomato-based ketchup since the early 1990s (Robinson, 2007).

Each hot pepper has its own peculiar flavour, and not all are spicy hot. The heat in chilies comes from their capsaicinoids, concentrated in a chili internal ribs and seeds. These compounds turn on the pain receptors in our mouth and on our tongue. It is essentially a plant defence mechanism designed to repel animals from eating pod. At a very low concentration, the capsaicinoids induce the release by the brain of endorphins, a type of mild natural opiate, to ease the sting. It is therefore that blend of pleasure and pain that makes eating chilies such an exciting experience (Robinson, 2007).

Capsaicinoids can be diluted in alcohol, but not in cold water. That explains why it is preferable to drink a cold beer to relieve the burning caused by the pepper. In addition to enhancing flavours, capsaicinoids have several characteristics that may be beneficial to health: they have

low concentrations of sodium and calories, they store potassium (which prevents strokes) and contain beta-carotene, vitamins E and C (all antioxidants), as well as folic acid. They also act as vasodilators helping prevent heart attacks and as decongestants, alleviating sinusitis. According to researchers at the University of Sussex, England, consuming fresh hot peppers on a regular basis is an effective way to prevent headaches and toothaches. Adding hot peppers to food, in reasonable amounts, stimulates salivation and the release of digestive fluids, neutralizes acids, and cleans teeth.

In the past few years, chili lovers, e.g. in the United Kingdom and the United States, have become obsessed with eating the hottest chili, the hottest sauce. In September 2000, a military laboratory in the garrison town of Tezpur, in northeastern India, announced that it had identified the hottest chili in the world. Chili heat is measured in Scoville Heat Units (SHUs), from the American chemist Wilbur Scoville who invented the scale in 1912. Pure capsaicin measures 16 million SHUs. A bell pepper typically measures zero. An Italian *peperoncino*, used to spice up pasta dishes in southern Italy, measures about 500 SHUs, while the spiciest Thai chilies reach around 100,000 SHUs. Until the discovery made in Tezpur, the hottest chili ever measured was the Red Savina, a type of *habanero* grown in California by a commercial chili farmer, which measured 577,000 SHUs. According to the tests carried out by India's Defence Research Laboratory, pods from the *bhut jolokia*, a "ghost chili", grown across northeastern India, had measured 855,000 SHUs. In 2005, the Chili Pepper Institute in New Mexico finally grew enough *bhut jolokia* from seeds collected in India. The results were stunning: the *bhut jolokia*, also called the Naga chili after a local tribe that enjoys eating them, measured just over 1 million SHUs, the sort of heat found only in the hottest chili sauces made from pure pepper extract. Both the director of India's Defence Laboratory and the scientist in charge of cultivating the *bhut jolokia* explained that it was so popular in northeastern India that it was known as "the king of chilies" and celebrated in a festival that coincides with the beginning of the chili season in April (Robinson, 2007).

The Indian laboratory was contemplating applying for Geographical Indication Certification, which would result in only *bhut jolokias* from northeastern India could be sold as such. This chili could be used in medicines, or in antiriot weapons such as tear gas. By smearing it on strings encircling villages, it could keep elephants away from crops and humans (Robinson, 2007).

In 2005, Michael and Joy Michaud of Dorset, England, shattered the Scoville scale with the Dorset Naga, cultivated from a Bangladeshi pepper; it was potent enough for handlers to require gloves. The Dorset Naga measured 923,000 SHUs. It was later on outpaced by the *bhut jolokia* (Robinson, 2007).

There is therefore a "global warming" trend in the search, identification and cultivation of the hottest peppers. David Thompson, an Australian cook responsible for some of the most inventive Thai cooking of the past decade and owner of Nahm, London's Thai restaurant with a Michelin star, stated that "chili is not meant to swamp or overpower, but act as a counterpoint to something salty or sour or sweet, or to heighten the sensation of textures." Madhur Jaffrey, an Indian cook, added: "once we develop a taste for hot food, which provides a high, there is no going back. It turns into a craving… [The chili is] a conqueror, or, better still, a master seducer." Both opinions offer a rationale for the fact that in five centuries the chili has successfully spread across the entire planet (Robinson, 2007).

From the biotechnological viewpoint, there have been attempts to cultivate chili tissues *in vitro* in order to produce and extract higher quantities of capsaicinoids. The latter could be used in the food industry (spicy foodstuffs, snacks, frozen food, etc.). On the other hand, *Capsicum*, which is a member of the nightshade family that includes tomatoes, potatoes and eggplants, was among the first plants to be genetically modified, particularly by Mexican scientists working at the Irapuato Plant Biotechnology Laboratory of the Centre for Advanced Research and Studies (CINVESTAV). The genomics of the plant will help discover genes coding for not only the biosynthesis of capsaicinoids, but also for plant defence mechanisms, that may be transferred to other crops.

Fruit consumption : nutrition and culture

Consuming fruits is not just about the healthy intake of vitamins, antioxidants and other micronutrients, as well as meeting part of the energy needs, it has also to do with symbols, culture and mythology.

Fig

For instance, the fig, product of sunny countries, is the symbol of abundance. It is considered a present of God in the *Bible* and the fig-tree was part of the Garden of Eden. In the *Genesis*, it is reported that Adam

and Eve, after having discovered their nudity covered their sex with fig leaves and not grapevine leaves. Some specialists even consider that the prohibited fruit was not an apple but a fig. Cleopatra, the queen of Egypt, after the death of Marcus Antonious, committed suicide: she was bitten by a serpent hidden in a basket of figs – a fruit she liked very much. In Greek mythology, the fig-tree is the tree of Dionysos – the god of sap and juices, and protector of gardens – and it is often mentioned, for instance, the titan Syceus prevented his mother to fall into the arms of Zeus, by growing a fig-tree behind which his mother could hide from the almighty god. During the period of Homerus, as reported in the *Odyssey*, figs were planted in the orchards of the king of Phenicians, who always stored dried figs on their ships; in this form, they were easy to transport and were an important part of the sailors' diet (sugars, potassium, calcium and fibre) [Toula-Breysse, 2009 b].

Figs, eaten with cheese, milk and honey, or accompanying various meals, are synonymous of happiness. Apicius, the famous cook of the Roman emperor Tiberius, who was born in 25 BC, used to eat figs with cured ham. Considered "voracious and a rake" by Plinus, he also used to feed his pigs and geese with figs. The Romans indeed were very fond of this fruit brought from Mesopotamia; they planted in the orchards of Lutecia "the tree which gives fruit without making flowers." The fig was also used by Cato the Elder to convince the Roman Senate to engage in the third war against Cartago; he showed his colleagues a fresh fig that was picked three days before in the Phenician orchards, and argued that the enemy was not far from Rome. The Senate listened to him and Cartago was to be destroyed soon (Toula-Breysse, 2009 b).

In Eurasia, the fig-tree, associated to Vishnu and Shiva, is considered a sacred tree in the Vedic literature (Upanishad or Bhagavad Gita). The Buddha, Siddharta Gautama, is represented seated under a *Ficus religiosa* or the pagoda fig-tree (Toula-Breysse, 2009 b).

The fig (*Ficus carica*) is, according to Plato, "the food of excellence." Covered by a white, green or purple skin, the savoury pulp melts in the mouth. There are many recipes for cooking and eating figs. In France, the variety called *Violette de Solliès*, with a deep-blue skin and red pulp, is highly appreciated for its taste. It is harvested in the fall in the Var region (south of France and first producing region of the country); in 2006, the variety received a Geographical Indication Certification (AOC) and Solliès-Pont has become the French capital of the fig, with an annual harvest of about 2,000 tons of fresh figs (Toula-Breysse, 2009 b).

Citrus fruits

In Europe, citrus fruits are sold in shops and markets after the apples and pears of autumn have waned, bringing vitamin C to consumers during the shorter and darker days of mid-winter. Fruit expert Adam Leith Gollner wrote : "for me, they are simply a taste of sunshine at a bleak time of year." The author of *The Fruit Hunters*, published in 2009, added : "One of my most favourite things is to peel one whole and hold it up to the light; it is as though the sunshine is filtering through it." A. Leith Gollner was probably talking of a clementine, or a satsuma, or may be of a tangerine. The 5th-century Chinese poet Liu Hsun described the "fragrant mist" which clementines exude when they are peeled off to find the segments that pull apart easily and then comes the sweet, refreshing and aromatic taste (Kirby, 2009).

All the citrus fruits (satsuma, clementine, tangerine, tangelo, clemenule, kishu, minneola, etc.) are hybrids of the mandarin (*Citrus reticulata*), a type of orange whose name stems from a Portuguese derivative of the Malay word for counsellor (*matri*) – which is also used to refer to senior civil servants. It is now believed that mandarins may have been the original orange and along with citrons (related to lemons) and pummelo (a type of grapefruit) were the three primordial varieties from which all the other varieties of citrus fruits – lemons, limes, sweet and sour oranges, etc. – later derived. Mandarins were first cultivated from wild Indian varieties in ancient times in southern China. The kishu/tiny tangerine/clementiny may also be the same small mandarin variety that, according to the *Oxford Companion To Food*, "fashionable Chinese women would hold in their hands so that they were scented by it" (Kirby, 2009).

Compared with the other fruits and vegetables from the Far East, mandarins were late arrivals in the West. It was not until 1805 that the first ponkan cultivars – a paler, mild flavoured version which is still the most common mandarin variety globally – reached England. From there they spread rapidly to Italy and then the rest of the Mediterranean. They did not reach America until the 1840s, but soon became a staple fruit grown in Florida and California. It should be stressed that the citrus variety collection of the University of California has one of the biggest archives of its type in the world, and includes 197 varieties. While the name tangerine was common in the United Kingdom by the end of the 19th century, in the United States tangerine is the name used for other, darker varieties which are different from those sold in Europe and which used also to be referred to on the American market as "Christmas oranges". About that time, the richer-flavoured clementine is believed to have been

developed in Algeria. The mostly seedless satsuma was bred in Japan in the 16th century; some now regard it as a separate species, as it is more tolerant to cold than other citrus varieties (Kirby, 2009).

There are also the more obscure hybrids. The temple orange was a type of tangor, a cross between a mandarin and other oranges. The ortanique is the tangor most commonly sold in the United Kingdom. A tangelo is a mandarin-grapefruit hybrid, the best-known version being a minneola – a rich and juicy fruit with a distinctive knob on the stem end, which appears on European supermarkets in the early spring. The even more knobbly ugli fruit is a type of tangelo (Kirby, 2009).

In early December 2009, Tesco – the British retailer chain – launched the 50 p-sized clementiny, describing it as "small, rich tasting, easy to peel, great for kids, etc." The supermarket chain dubbed the clementiny the "world's smallest citrus fruit", but this is inaccurate. The citrus variety collection of the University of California has a citrus fruit in its collection that is no bigger than a pea. The clementiny is actually the same fruit as the "tiny tangerine" which Marks & Spencer has sold for several years in the United Kingdom, packaged in a plastic container as a premium product. Although giving it different names, the two chains agree that this under-sized fruit has been grown in southern China for about 1,300 years (allegedly it was once reserved for the imperial court). According to Marks & Spencer, they were first spotted by one of its own buyers in the region. In the Tesco version of events, it was a Swiss businessman who first imported it to Europe. The name tangerine did not originate in China but derives from the Moroccan port of Tangiers, from which large quantities of mandarins were first exported to the United Kingdom. Fruit expert Adam Leith Gollner thinks the tiny tangerine/clementiny is actually a kishu – which probably explains why the marketing people had to come up with a new name (Kirby, 2009).

Supermarkets and wholesalers lump clementines, tangerines and satsumas together as "easy peelers." This is an accurate description as the loosely attached skin is the main characteristic that distinguishes this group of citrus fruits from their relatives – the other oranges, lemons, limes, etc. One should add the ability to break up easily into bite-size segments, their small, convenient size, their sweetly seed-free child-friendliness, together with relative cheapness. All this makes a perfect consumer product. For "easy peeler" producers, one of the keys to a successful fruit is the amount of seeds – lack of seeds being a clear incentive to consumers. The seedless varieties of clementines, which the farmers find easier to grow, are therefore dominating the market (Kirby, 2009).

The winter months remain the peak season, when the "easy peelers" fill the gap on the supermarket shelves after the glut of late summer and autumn fruits, and, along with all the other oranges, are much better value than other imported fruits. A combination of hybrids designed to fruit both early and late, and the development of big, commercially minded exporting combines in Turkey, Morocco and southern Spain, have extended the season from October into early spring. And, although in much smaller numbers, southern hemisphere imports keep them on many supermarket shelves throughout the year. Consumption overall rose exponentially during the 1990s and the early part of this decade, rising by about 15% over the three-year period 2006-2008. According to market researchers TNS Worldpanel, during 2008 Britons spent around £324 million on "easy peelers", eating more than 180 million kg of them; they were included in the shopping baskets of almost 80% of households. The bulk of the sales – more than a third by weight – take place in the last three months of the year, when the amount spent reaches a total of £98 million. An overwhelming majority of the fruit sold – a total of about 157 million kg – are either satsumas or clementines, with clementines accounting for almost two- thirds of the total (Kirby, 2009).

Satsuma consumption has lost ground in recent years (from 67 million kg to 51 million kg between 2004 and 2007, although recovering somewhat to 56 million kg in 2008). Its sweeter rival, the clementine, has overtaken it (rising from 57 million kg in 2004 to 101 million kg in 2008). The drop in demand, mirrored in other countries, has resulted in a fewer satsuma trees being planted in the producing regions, threatening the levels of supply to the United Kingdom, which remains the biggest market for the slightly blander, paler satsumas (Kirby, 2009).

Strawberries

The fragile and flavoured berry found in the understorey of natural woodlands in the northern hemisphere is present in all continents. The Roman poet Virgilio, who was a faithful disciple of Epicurean philosophy, warned in his *Bucolics* younger people against serpents when they used to harvest the small red berries. In the Roman banquets, the fruit was often eaten, and women used to apply a paste of wild red berries on their face. Named *fraga* in Latin, as a remembrance of its fragrance, the wild berry was grown in the Middle Ages in French royal gardens. In the 14th century, King Charles V requested his gardeners to plant the berry-shrubs as an ornamental in the open spaces of the Louvre palace. During the Renaissance period, in France, women preferred to eat the wild berry with cream, while men preferred to eat it with wine. Louis XIV was very

fond of the berry; he used to consume large quantities until his stomach could not withstand it (Toula-Breysse, 2009 a).

Until the 18th century, all European strawberries were fingernail-sized. In July 1712, Louis XIV dispatched the spy, pyrotechnician, geographer and amateur botanist, Amédée-François Frézier, to Concepción, Chile, in order to collect information on the defences of Spanish colonial settlements. There, A. F. Frézier noticed local strawberries had the size of walnuts, collected as many as he could and shipped them home. Only five plants survived the six-months trip, despite A. F. Frézier using his own water ration to keep them alive. All were female plants, so unable to produce viable seed. But, by chance, A. F. Frézier found they could interbreed with the small and more intensely flavoured Virginian strawberries, which the first colonizers of America had sent back and were already popular in Europe. A. F. Frézier introduced the first cultures of strawberries in Plougastel, Brettany, and offered a few plants to the king's botanist, Jussieu.

As stated by Christopher Stocks, gardener and plant historian, author of *Forgotten Fruits*, a history of fruit and vegetables, "Frézier's story proves the desire to breed better varieties of strawberry is nothing new"... "the modern strawberry is a hybrid of species from the east coast of North America, the west coast of South America and Europe. Proof that horticultural development has depended on international trade for far longer than most people imagine."

The virtues of the fruit have been praised during modern history. For instance, the French philosopher and poet Fontenelle (1657-1757), the nephew of Corneille, was very fond of the berry and used to state that his long life was due to it. The world-known botanist, Carl Linnaeus (1707-1778), would have tried to cure his gout with a high consumption of strawberries. Mme Tallien (1773-1835) was nicknamed Notre-Dame de Thermidor, because she saved many prisonners from being beheaded during the Terror period. She had the reputation of being a bold free woman; she was very witty and shrewd, and she introduced the fashion of the Greek robe and of a new hairdressing. She took great care of her skin complexion, and according to Lamartine, she used to take baths of strawberry juice in order to keep the softness and glaze of her skin (Toula-Breysse, 2009 a).

Many of the varieties selected since the 18th century and which were hybrids between American and French varieties have disappeared. In addition to those contemporary varieties which are sold across

the world but which often lack flavour and savour, there are some interesting ones. For instance, a variety called *gariguette d'Aquitaine* had been selected in France in 1977: rich in vitamin C, it has a lemon flavour, and it is among the early strawberries during the season, often commercialized with the *pajaro* variety (sometimes called *fraise de Carpentras*). Thereafter comes, late in the season, the sweet variety called *mara des bois* (Toula-Breysse, 2009 a).

Strawberries are particularly vulnerable to the disease caused by the fungus *Botrytis cinerea* (grey rot), that can destroy a whole harvest from the Mediterranean to Asia, through Mexico and the United States. The global losses of strawberries due to this mould disease can reach 25% of the world's harvest of non-treated strawberries. The only way of controlling the disease is to spray fungicides, but unfortunately the fungus has become resistant to the chemicals. Henceforth, the interest of relying on a biological method of control that does not entail the risk of a genetic mutation of *B. cinerea*.

Researchers of the Faculty of Science of Tunis, Tunisia, have been working with their colleagues of the French Research for Development Institute (IRD), in collaboration with Tunisia's National Agricultural Research Institute and the Faculty of Agronomic Sciences of Gembloux, Belgium, in order to isolate halophilic bacteria that inhibit *B. cinerea*. These bacteria, *Bacillus subtilis* and *Bacillus pumilus*, were isolated from soil samples collected in *sebkhas* – depressions formerly occupied by the sea and whose soils are saline – or in *schotts* – lakes with very saline water due to a high evaporation. These kinds of hypersaline ecosystems are very common in arid regions, e.g. in North Africa and the Middle East. The effectiveness of these bacteria has been demonstrated on strawberry plants, but it was not tested during the storage of fruits. From 2006 to 2008, the Tunisian researchers with their colleagues have been testing during three seasons the application of a mixture of both bacteria to strawberries in warehouses of the Cape-Bon region, in the north-east of the country. The bacterial preparation could save a large part of the strawberry harvest: a significant reduction of the presence of grey rot was found, compared with non-treated strawberries and with those which received a chemical treatment. The assumption was that *B. subtilis* and *B. pumilus* produced toxic compounds and enzymes that could inhibit the growth of *B. cinerea* cells. Research is being carried out in order to optimize the process, with a view to obtaining its approval and commercialization (Essghaier et al., 2009).

Cocoa

When Christopher Columbus landed in 1502 on the island of Guanaja (Honduras), the Amerindians offered him, as a gift, beans of *tchocolatl*. One thousand years before the arrival of the Spanish navigator, Toltecs used to grow the cocoa tree, *cacahua quotchitl*, to comply with the demand of their legendary god Quetzalcoatl (the feather serpent) and because of the magic virtues of its seeds. The Indian beverage was made from cocoa ground beans; it was mixed with chili and maize; and consumed cold. It was considered a source of wealth and strength, quenching thirst and hunger, and it could cure many ailments. While Aztec nobles were the only ones to drink the *tchocolatl*, Spanish conquistadores mixed it with sugar, cinnamon, vanilla, and heated it before drinking it (Toula-Breysse, 2009 c).

The beverage became fashionable in New Spain (Mexico) and thereafter reached Europe via Spain in 1524. The Dutch and British people were seduced by what would become the "brown gold," not only because of the profits it generated but also because of its taste. Despite the criticism made by some religious orders, particularly the Dominicans, who considered the beverage dangerous because it could heat the human temper, cocoa became an important trade item, which conquered the palaces and royal families in Europe. For instance, Anne of Austria (1601-1666), the daughter of Philip III, king of Spain, and the spouse of Louis XIII, king of France, introduced the sweet beverage to the court. And Maria-Teresa, the spouse of Louis XIV, had a passion for chocolate. The marchioness (marquise) of Sévigné (1626-1696), renowned for the letters written to her daughter, Mme de Grignan, used to say : "Take chocolate so that the worst companies would look good." Giovanni Casanova (1725-1798) qualified the sweet beverage as aphrodisiac and bringing euphoria, and it might have triggered its sexual appetite. According to the French judge and gastronomist Anthelme Brillat-Savarin (1755-1826), chocolate had numerous virtues (Toula-Breysse, 2009 c).

Cocoa is mainly produced in Africa, Latin America and Asia by about 14 million farmers on an area of around 5 million hectares. More than 80% of global production comes from smallholders (less than 5 hectares per household). Global production reached 3.6 million tons in 2009, but almost 30% of the harvest is lost due to diseases and parasites. Cocoa consumption takes place mainly in industrialized countries : 80% in Europe and North America (Devailly, 2010).

Cocoa is extracted from the beans embedded in the capsule of the cocoa tree, which belongs to the Sterculiaceae family. The capsules are found on the trunk and branches of the tree. There are three main cocoa varieties: *criollo*, the most delicate and sought after (5% of world production); the *forastero*, more common, with acid aromas and bitter taste, more resistant to diseases (about 90% of world production); and the *trinitario*, a hybrid of the two former ones, rich in fats. The best beans come from Venezuela, Colombia, Trinidad, Ecuador, Madagascar, Indonesia and Sri Lanka. French chocolate makers, classic or innovators, who are considered the masters of *ganache* (fresh cream added to chocolate and sugar), have found the appropriate formula that reveals the delicate bitterness of the raw material, formerly dubbed "*the manna of Caracas*", i.e. reaching the balance between taste and texture. Chocolate making is a complex process. After selecting ripe beans, the latter are fermented and sun-dried; then, they are toasted, their skin is taken away and they are ground into a paste, from which cocoa butter is extracted; after another mixing of the paste the final product can be transformed into chocolate (Toula-Breysse, 2009 c).

On 24 December 2010, the price of a ton of cocoa reached £2,036 (€2,397) on the London market. The price fell down from the peak of £2,713 reached on 14 July 2010, but prices remained twofold higher than those of January 2008. The world cocoa market was to remain tense because the supply was lower than the demand. The political crisis affecting Côte d'Ivoire – the leading global producer (40% of world production) – was also harming cocoa plantations which were poorly maintained (Devailly, 2010). On the other hand, due to the increasing demand from China and India, world consumption has risen and this trend was to continue, despite a global harvest of about 3.8 million tons in 2010-2011 (+6%) forecast by the director of the International Cocoa Organization. There was also speculation from some buyers who were trying to withdraw the small available volumes on the market, so as to reach a maximum price of £3,000 per ton of beans.

Cocoa-tree genomics may help improve in the future the aromatic properties of cocoa as well as the resistance of cultivars to fungal diseases which cause the loss of one-third of global production every year. In the 26 December 2010 issue of *Nature Genetics*, the sequence of the cocoa-tree genome and the first analyses of this genome have been published by a consortium of about 60 partners from six countries, led by the French International Cooperation Centre on Agricultural Research for Development (CIRAD). The genome was that of the *criollo* cocoa cultivar (variety), collected in Belize and which may be the descendant of the first

cocoa trees grown by indigenous populations of Central America, more than 2,000 yeas ago (Argout et al., 2010).

The *criollo* cultivar is rich in aromas, but it is sensitive to fungal diseases. Consequently cocoa-producers have increasingly relied on hybrid varieties, more resistant to diseases, but less aromatic. The result is that the production of aromatic cocoas (high end for the consumers, including the *criollo*) amounts to only 5% of global production. Deciphering the genome of cocoa *criollo* cultivar has contributed to a more accurate analysis of two families of genes among the most important ones known in the plant kingdom for disease resistance. It is therefore possible to seek genetic markers and use them in the breeding of cultivated cocoa varieties. Also genes encoding the synthesis of aromas, e.g. those involved in the biosynthesis of polyphenols, in that of cocoa butter or terpenes (which give many flavours), would enable the breeders to screen more or less aromatic cultivars. In addition the analysis of the cocoa-tree genome has shown that, like that of grapevine, the genome has not evolved much since the first ancestors of the plant (Argout et al., 2010; Devailly, 2010).

Genome sequencing will help geneticists and breeders to analyze the diversity and qualities of cocoa trees still existing in equatorial Amazonia and threatened with extinction. The seeds of over 80 centenary trees have been collected by Claire Lanaud of CIRAD during the summer of 2010 and stored in the banks of genetic resources of Ecuador, while the French geneticist brought with her samples of leaves and beans in order to carry out genomics studies (Argout et al., 2010; Devailly, 2010).

The sequencing of the cocoa-tree genome started in August 2009 and the work lasted one year and half; it was mainly carried out at the Genoscope (Evry, Essonne, south of Paris) as well as at Pennsylvania State University and Cold Spring Harbor Laboratory, using sequencing techniques developed in 2008. Thereafter a team of bioinformatics specialists and biologists belonging to CIRAD and other institutions of the consortium took over and identified 28,800 genes. They also carried out the first analyses. Then the publication was submitted to *Nature Genetics*, which published it on 26 December 2010 (Devailly, 2010).

A parallel genomics work has been carried out by an American team and it was funded to a large extent by the agrifood group Mars Inc. (CIRAD had two chocolate-producing partners : Valrhona and Hershey Corp., the leading chocolate producer in the United States). It seems that this American team, aware of the fact their rivals were to publish their results quite soon, decided to put on line their own results without going through

the validation by a scientific peer-reviewed journal. They also claimed that they were the first to decipher the genome of a more common cocoa variety called Matina 1-6 and existing in Costa Rica. However, the results which were incomplete were not validated by the scientific community. Most probably the industrial group preferred to publish the results more rapidly and eventually patent the genes instead of making them available to anyone. By the end of September 2010, the journal *Science* made comments on both projects and the methods used by each group of researchers. Three months later, *Nature Genetics* validated the results of the consortium led by CIRAD, and these results were available to the whole international scientific community (Devailly, 2010).

Reducing the chance of suffering from cancer?

The recommendations from governments, charities and even the World Health Organization (WHO) to eat five portions (servings) of fruit and vegetables every day are meant to reduce the chance of suffering from cancer. But a group of researchers led by Paolo Boffetta, of the Mount Sinai School of Medicine in New York, have conducted a new study on the link between cancer and the consumption of fruit and vegetables, and found it to be far weaker than anyone had thought. It seems that some of the earlier investigations that concluded vegetables and fruit associated reductions of cancer rate were as high as 50%, may have been biased by the use of "case-control" studies. Such studies try to identify the factors contributing to cancer by comparing people who have the disease with those who do not, but are otherwise similar. The problem is that they can easily be biased if researchers do not adequately establish that the two groups being compared are, indeed, otherwise similar. P. Boffetta and his colleagues have therefore carried a different kind of study, known as prospective cohort study: they followed a group of individuals over time and looked at how different factors contribute to different outcomes – in this case, the development of cancer. The analysis of dietary data from almost 500,000 people in Europe led to a weak association between high fruit and vegetable intake and reduced overall cancer risk (Buckland et al., 2010).

According to Susan Jebb, of the British Medical Research Council Collaborative Centre for Human Nutrition Research in Cambridge, the new study suggested that if Europeans increased their consumption of fruit and vegetables by 150 g a day (about two servings, or 40% of the WHO's recommended daily allowance), it would result in a decrease of just 2.6% in the rate of cancers in men and 2.3% in women. Even those who ate virtually no fruit and vegetables, the paper suggested, were

only 9% more likely to develop cancer than those who applied WHO's recommendations (*The Economist*, 2010 a).

P. Boffetta's conclusions were played down, because critics pointed out there are a number of other factors that nutritionists would urge to be considered. One is that this kind of study has attempted to adjust for every possible factor that might contribute to the relationship, and isolate only the contribution that fruit and vegetables make. This means that if people who turn away from fruit and vegetables end up eating more processed meats and foods high in fat instead, they probably will increase their cancer risk, even though the direct cause is not the consumption of less fruit and vegetables. However, there is still good evidence that fruit and vegetables protect against heart disease and strokes by reducing blood pressure. A separate investigation of the people involved in P. Boffetta's study suggested that those who ate five servings a day of fruit and vegetables had a 30% lower incidence of heart disease and strokes than those who ate less than one and half servings. It is also possible that some specific foods, such as tomatoes, broccoli and other cruciferous vegetables, do offer protective effects against particular kinds of cancer. Consequently, the best advice is still to try to consume fruit and vegetables five times a day (*The Economist*, 2010 a).

The virtues of olive oil and "Mediterranean diets"

Brief historical record

The olive tree (*Olea europea*) has a wild variety (*Olea europea* var. *sylvestris*), which can be traced back to about 3.2 million years in the Mediterranean basin (traces of pollen). The cultivation of the tree was probably initiated in Syria, Lebanon, Israel and Turkey some 4,000 years BC. The common name of the tree and fruit is derived from the Greek *elaia* and Hebrew *zait*, which later on became *Olea* in Latin and *zaitun* in Arabic (Sánchez Muniz, 2007).

Mycenian tablets are the oldest written documents on olive oil and they highlight the importance this kind of oil had 4,500 years ago at the court of King Minos. In Egypt, the cultivation of the olive tree was probably initiated in the western part of the Nile delta; but the country imported oil from Palestine for food, the preparation of perfumes and for religious ceremonies. Leaves of the olive tree were part of the crown of justice which the pharaohs used to wear, like Tuthankamon (Sánchez Muniz, 2007).

In the *Bible*, olive oil is mentioned 200 times, with references to its culinary and religious uses. For many, Greece is considered the craddle of the olive tree, which is associated with the legend of the foundation of Athens in the 17th century BC. The discoveries made at Mount Testaccio, near the Tiber River, by Heinrich Dressell in 1878, revealed the economic importance of olive oil during the Roman Empire. It was estimated that the fragments of some 40 million amphoras were buried in the mount; they were used to transport olive oil from Spain (Betica), over 2 million tons of oil having been exported to Italy between 138 and 260 AD. Oil trade between Spain and Rome lasted until at least the fifth century of our era; Spanish olive oil was appreciated and it could compete with those produced in Apuglia and Campania (Sánchez Muniz, 2007).

According to written reports, the olive tree was being grown in the southern half of Spain. Many Roman authors highlighted the quality of Spain's olives. The fall of the Roman Empire was also followed by a decrease in olive-oil production, but much later the cultivation of olive trees was still again on the rise under the Wisigoths. When the Arabs settled in Spain, olives and olive oil were highly regarded and their quality has been praised by several authors. For instance, during the 12th century, the olive groves near Seville were very famous and their oil was highly appreciated (Aljarafe). Under the Catholic Kings, the broth (*gazpacho*) made with vinegar and olive oil was part of the basic diet in Andalucia and Extremadura (Sánchez Muniz, 2007).

Types and composition of olive oil

The olive tree is relatively easy to grow, it is not very demanding; it generally prefers sandy and well-drained soils; it can withstand extreme temperatures. The age of some individuals could reach several hundred years. The number of cultivated varieties is high, e.g. about 260 varieties in Spain, but around 20 varieties are mostly cultivated (for instance, Arbequina, Manzanilla Cacereña, Picual, Royal de Jaén, Hojiblanca, Gordal de Archidona, Empeltre, Picudo). There are also about 20 Geographical Indication Certifications of virgin oil in Spain, the world's leading producer of olives (30% of olive oil global production) [Sánchez Muniz, 2007].

When olives are crushed (without heating), virgin oil and olive paste are obtained. Virgin olive oil includes virgin olive oil extra, virgin olive oil and lampant virgin olive oil. The extra quality is the finest in terms of flavour and savour, and has an acidity equal or lower than 0.8°; the virgin olive oil has also excellent savour and flavour and its acidity if equal or lower than 2°; the lampant virgin olive oil is not to be consumed because of its high

acidity and must be refined; the refining process leads to what is labelled as olive oil, of which the acidity should be lower than 1.5°. Olive paste contains the olive skin and press pulp, fragments of the stone, and 5% to 10% of oil, which makes its extraction interesting economically; the result is an oil of olive paste or an oil of raw paste; once refined, it gives an oil with an acidity that should be lower than 0.3° (Sánchez Muniz, 2007).

The main fatty acids present in olive oil are: palmitic, stearic, oleic and linoleic acids. The content of trans fatty acids is very low. The fatty acid composition of olive oil is affected by climatic conditions, the variety and degree of maturation of olives. For instance, in Spain, the varieties Picual and Cornicabra give an oil very rich in oleic acid, while oils derived from Arbequina and Verdial varieties are less rich in oleic acid, but richer in linoleic acid. Olive oils have been classified into two types : one with a low content of linoleic and palmitic acids and high content of oleic acid; the other with a high content of linoleic and palmitic acids and low content of oleic acid. Olive oils from Spain, Italy and Greece belong to the first type, while those of Tunisia belong to the second one. Due to the composition of fatty acids, 70 or more triglycerides can be found in olive oil, but some of them are found in very small quantities; the content of triolein is significant: 40% to 59% (Sánchez Muniz, 2007).

Minor constituents of olive oil can be divided into two groups. The first one includes derivatives of fatty acids, such as mono and diglycerides, phosphatids, waxes and sterol esters. The second group includes compounds that are not related with fatty acids, such as: hydrocarbons, aliphatic alcohols, free sterols, tocopherols, carotenoids and phenolic compounds. These phenolic compounds are very rare and are not found in other oils; tyrosol and hydroxytyrosol are the main compounds, while others such as oleoeuropein, caffeic acid, vinylic, syringic and coumaric acids are rather frequent (Sánchez Muniz, 2007).

The oil extraction process has a profound impact on the content of biophenols. Oils that are obtained in two-phase containers contain more phenolic compounds and are therefore more stable with respect to autooxidation; they are also responsible for the olive oil flavour, as well as to a lesser extent, for some organoleptic properties (fruity, sweet or bitter) [Sánchez Muniz, 2007].

Nutritional value of olive oil

Olive oil is a source of essential fatty acids: linoleic and linolenic. A daily diet amounting to 2,000 kcal and containing 50 g of olive oil would

meet the needs of these two essential fatty acids. In addition, the balance between omega-3 and omega-6 fatty acids in olive oil is much better than in many other vegetable oils. Olive oil is also a source of liposoluble vitamins: 55% to 110% of the recommended daily needs of vitamin E would be met by the consumption of 50 ml of olive oil. The ratio vitamin E: linoleic acid in olive oil is 2 mg/g, higher than 0.6 mg/g, considered satisfactory and much higher than that in other vegetable oils. The latter are generally poor in vitamin A (retinol), but olive oil contains beta-carotene (0.6-1.3 mg/kg); a daily consumption of 50 ml of virgin olive oil would meet 7.5% to 15% of the recommended needs of retinol (Sánchez Muniz, 2007).

Other compounds such as carotenoids, squalene, phytosterols and polyphenols play a key role in the nutritive value of olive oil, but also in its gastronomic and organoleptic properties, mainly because they keep the original characteristics of the oil by hindering the processes of autooxidation and rancidity. Phenolic compounds are responsible for bitterness and, because of the consumers' tastes, their content is very low in the oils sold in supermarkets; this is considered detrimental to nutritional quality (Sánchez Muniz, 2007).

Olive oil and the "Mediterranean diets"

Olive oil has always been one the basic pillars of the Mediterranean diet. In this region, seed oils have appeared only in the 20th century. The concept of Mediterranean diet(s) was presented at a conference organized in 1993 in Boston by Harvard University School of Public Health and the Oldways Preservation and Exchange Trust; the diet was defined as a pyramidal structure, the basis of which consists of cereals (bread and pasta) and olive oil. The traditional Mediterranean diet is mainly composed of vegetables and fruit, it contains small quantities of foodstuffs from animal origin, fish, milk products and wine (in moderate quantities taken during meals), and virgin olive oil as the main source of fats. In addition, this diet implied the maintenance of a balance between food intake and moderate exercise; meals are considered a pleasant and convivial opportunity to share food among family members (Sánchez Muniz, 2007).

A basic constituent of the Mediterranean diet is therefore olive oil, an oil rich in monounsaturated fatty acids and with a moderate concentration of saturated and polyunsaturated fatty acids. In virgin and extra virgin olive oil, biophenols with antioxidant properties play an important role in the modulation of atherogenesis. According to the data published by some institutions and research teams, consumption of olive oil has drastically

decreased during the last decades and has been substituted by oils derived from seeds, up to 50%, e.g. sunflower oil. For instance, in Spain, the average consumption of sunflower oil is about 33 g per capita per day, i.e. higher than the quantity of olive oil (in a low saturated-fat diet) considered by the US Food and Drug Administration (FDA) as offering a significant protection against cardiovascular ailments (Sánchez Muniz, 2007).

In the Mediterranean diet, olive oil is used in the preparation of salads, but is also eaten with bread at breakfast or as a snack; it is an important ingredient of sauces, mayonnaise, aioli, as well as of *gazpachos*. In these cases, it is recommended to use an oil with a balanced composition of saturated and unsaturated fatty acids, rich in minor components such as antioxidants. Savoury sweet oils, with a very low degree of bitterness, are the most appropriate for seasoning salads, vegetables, boiled fish, or for preparing vegetable soups and other light meals. On the other hand, fruity oils somewhat bitter and with more or less flavour, are the most appropriate for enhancing fried meals. In Spain, frying cannot be separated from the Mediterranean diet. During the frying process, oil penetrates into food and modifies its texture, flavour and aspect. It should be mentioned that frying with olive oil does improve in many cases the balance between saturated, monounsaturated and polyunsaturated fatty acids in the food, and increases the content of liposoluble vitamins and antioxidants in the ingested fried meal. As frying takes place at 170-180°C, it is recommended to choose oils that are stable at high temperatures and remain nutritious. Olive oil should not be used for frying more than four or five times, when small quantities of oil are used, in order to avoid alterations that occur in the oil composition. When larger quantities of oil are used, and particularly to fry low-fat foodstuffs, olive oil can be reused up to 30 times, compared with 10 times for sunflower oil (Sánchez Muniz, 2007).

Effects on health of the consumption of olive oil

Many publications refer to the beneficial effects on health of the consumption of olive oil, for instance in the Mediterranean diet. For example, it is worth mentioning the book titled *Olive Oil And Health*, published in 2006 by J.L. Quiles and M.C. Ramírez-Tortosa of the University of Granada. Thus, the regular (daily) consumption of olive oil could prevent cognitive degeneration related with ageing, e.g. Parkinson's and Alzheimer's diseases. Human brain is very sensitive to variations in the content of fatty acids in the diet and also to oxidative stress. Olive oil could play a role in the incorporation of another fatty acid, docosahexaenoic acid, and in preventing the excess of arachidonic acid in the synaptosomes and other areas of the membranes of neurons

and neuroglia. Olive oil carotenoids can also contribute to reducing lipoperoxidation and DNA alteration in neurons (Sánchez Muniz, 2007).

It has been suggested that the Mediterranean diet, rich in olive oil and antioxidants, is associated with a significant increase in life expectancy. For instance, it has been shown through a study involving 2,600 persons of more than 70 years of age that old people of southern Europe had by far the longer life span, compared with those of northern and eastern Europe. Among the factors, that could explain this result, are the lifestyle (less stress, Mediterranean type diet) and some biological markers (e.g. a lower content of serum cholesterol) [longitudinal SENECA study, 2004, in Sánchez Muniz, 2007].

Keys et al. (1986) published the results of their study carried out in seven countries (Finland, Greece, Italy, Japan, Netherlands, United States and Yugoslavia) on 11,600 men of median age, with a view to understanding the relationship between the lifestyle of the 16 cohorts (Mediterranean and non-Mediterranean) participating in the study and the morbidity-mortality rate due to cardiovascular diseases. The Mediterranean cohorts had a diet rich in cereals, fresh vegetables, fruits and olive oil; wine was drunk during meals. Among the non-Mediterranean cohorts meat and milk were the main sources of fat, and beer and other spirits were taken between the meals. The members of the Mediterranean cohorts had a low intake of saturated fatty acids (percentage of total energy intake) and a ratio, monounsaturated: saturated acids, higher than 2. Mortality rate due to cardiovascular diseases, after 15 years, indicated that the Mediterranean diet was a protective factor against cardiovascular diseases – a primary cause of death in industrialized countries.

After the publication by Keys et al. (1986) of their results that suggested that olive oil and monounsaturated fatty acids play a key role in the protection of the cardiovascular system, a few studies did not lead to the same conclusion. Some of them have used small quantities of olive oil and sources of monounsaturated acids distinct from olive oil. Others did not follow a proper methodology. By contrast, the study CARDIO 2000, conducted by Pitsavos et al. (2002) on a sample of 661 patients of median age who had suffered from heart attack or coronary disease, and on a control group of 661 healthy persons from different regions of Greece showed that the adoption of a Mediterranean diet was associated with a statistically significant 16% reduction of the risk of a first coronary syndrome. In addition, the same researchers found that, after discarding several factors, the Mediterranean diet could reduce by 7% to 10% the risk of coronary disease among patients suffering from hypertension and being treated for

their illness. Finally, they concluded that the adoption of a Mediterranean diet was associated with a 35% reduction of the risk of coronary disease among a subgroup of persons suffering from the metabolic syndrome.

According to Trichopoulou et al. (2003), who studied a population of over 22,000 adults, there was an inverse correlation between cardiovascular mortality and the adoption of a Mediterranean diet. This correlation was found only in patients who were more than 55 years old; this result suggests that the correlation is due to the cumulative exposition to the Mediterranean diet over very long periods.

The "Lyon Diet Heart Study", the results of which were published by the De Lorgeril et al. (1999), also pointed out to a major reduction of the risk of coronary disease among 605 patients of both sexes, who had a serious heart attack. The control group of patients followed the prudent diet stage I of the American Heart Association, while those of the experimental group adopted the Mediterranean diet. The latter was similar to that of Crete of the 1950s and early 1960s, which was followed in the study of seven countries. After only 27 months, it was observed that coronary disease and death events had decreased drastically, and that these good results were confirmed after 46 months of study.

Several papers published in the Netherlands, Spain and the United States between the mid-1980s and 2002 have underlined that olive oil had a favourable lipidic profile, that it reduced inflammatory processes, reduced arterial pressure, increased the vasodilation of arteria, decreased thrombosis and improved the metabolism of carbohydrates in type-2 diabetes (Sánchez Muniz, 2007).

Sánchez Muniz and his colleagues of the Madrid Complutense University Faculty of Pharmacy have also studied the role of olive oil in the lipoproteic metabolism and atherosclerosis. They kept replacing the oil consumed in the diet of nuns living in cloisters, during four periods of one month; the intake of other foodstuffs was maintained constant. This kind of community followed classical Mediterranean eating habits. It was observed that the diet period that induced the lowest levels of total cholesterol and LDL-cholesterol was when the nuns consumed extra virgin olive oil, followed by the consumption of sunflower oil with high content of oleic acid, and by that of a mixture of olive and sunflower oils; palm oil rated much less.

The Spanish researchers also observed that a higher intake of monounsaturated fatty acids was followed by the lowest concentration

of apolipoprotein A-II in HDL (high-density lipoproteins), which suggests a more efficient reverse transport of cholesterol. In a pilot study of the same population (nuns living in cloisters), they found a lower aggregation of plaquettes, during the period of consumption of extra virgin olive oil, as the unique source of culinary fat. These effects were correlated with the content of polyphenols and other minor components of olive oil. This probably explains that sunflower oil, which is also rich in oleic acid, does not have the same effect, because it lacks polyphenols.

Other studies, such as "Diet and Reinfarction Trial" (DART), "Lyon Diet Heart Study" and "Gissi-Prevenzione Trial", have shown that the inclusion of olive oil in the daily diet (of a Mediterranean type) increased cardiovascular protection (20% to 30%) granted by a therapy with statins (Sánchez Muniz, 2007).

The group of Valentina Ruiz-Gutiérrez of the Seville Institute of Fat and Derived Products have published the results of their research on how olive oil and its components could hinder the formation of free radicals, the activation of the system cyclooxygenase and lipooxygenase, the production of prostaglandin E2, of interleukins, tumor necrosis factor (TNF) and adhesion molecules of monocytes (Perona et al., 2006).

The conclusions drawn at the end of a scientific meeting held in Madrid in March 2005 on the "State of knowledge concerning olive oil, nutrition and health" could summarize the virtues of olive oil (demonstrated or suspected) with respect to human nutrition and health.

1. When olive oil replaces a diet rich in saturated fats, it reduces the concentration of LDL-cholesterol in the plasma, and it improves the ratio LDL/HDL (i.e. reduces the risk of atheroma).
2. Olive oil intake reduces the concentration of plasmatic triglycerides and increases that of HDL-cholesterol, compared with a low-fat diet, but rich in carbohydrates.
3. Olive oil improves the postmeal lipoproteic metabolism.
4. Olive oil intake improves the endothelial vasodilatation and the inflammatory response. It reduces the aggregation of plaquettes, the postmeal activation of clotting factor VII and the plasmatic content of plasminogen inhibitor (PAI-I).
5. Olive oil consumption reduces the risk of hypertension.
6. It improves the metabolism of carbohydrates for those suffering from type-2 diabetes. It does not favour obesity and increases the lipolytic activity of adipous tissue (Sánchez Muniz, 2007).

Nutritionists who have studied the impact of olive oil on health, generally insisted on the fact that not only monounsaturated fatty acids of the vegetable oil (oleic acid) play a key role, but also several other components which exist in the virgin oil, but not in the refined oils or in virgin oil with a sweet savour.

On the other hand, the following nutritional recommendations are worth reminding:
- the dietary intake of saturated fat should not exceed 10% of total energy daily intake; it is even recommended to lower it down to 7%;
- monounsaturated fatty acids should remain the most abundant in our diet and represent 15% to 20% of total energy daily intake;
- polyunsaturated fatty acids should not represent more than 5% of total calorie intake, due to its higher sensitivity to oxidation;
- the ratio between omega-6 and omega-3 fatty acids should vary between 4:1 and 10:1.

The last three recommendations will not be met by consuming oils made from seeds (Sánchez Muniz, 2007).

Omega-3 versus omega-6 fatty acids

On 26 and 27 May 2010, at the Royal Society of Medicine in London, the numerous virtues of docosahexaenoic acid (DHA) were discussed. This is the most important of omega-3 fatty acids: it is a component of brains, particularly the synaptic junctions between nerve cells. It is found in fatty fish, e.g. in salmon, tuna (not the red tuna fish), anchovies, mackerels or sardines. Eating these fish species is supposed to have antidepression virtues, and even anticancer ones. However, David Khayat, head of the cancerology department at the hospital La Pitié-Salpêtrière (Paris), in his book titled *Le vrai régime anticancer* (The true anticancer diet), published in 2010, stated that the consumption of fish (all species included) decreased only by 3% or 4% the risk of colon cancer. The French specialist asserted that fatty fish consumption should be reduced or limited because of the high content of heavy metals and other toxic compounds: dioxine, arsenium, polychlorobiphenyls (or PoP, organic remanent pollutants), methylmercury, cadmium, lead. The higher the rank of a fish species in the food chain, the more contaminated it is. Both wild fish and farmed ones do not differ in this regard, because it is almost impossible to control their foodstuffs. It seems nevertheless that mackerels, sardines or anchovies, that are rich in omega-3 fatty acids, contain low amounts of mercury. David Khayat's advice is therefore to avoid salmon, red tuna and swordfish, and to consume much less fatty fish species such as the

flounder, sea bass and John Dory; it is also recommended to know the provenance of fish, because water contamination varies.

The displacement of DHA from modern diets by the omega-6 fatty acids in cooking oils such as soybean, maize and oilseed-rape (canola) worries nutritionists. Many researchers consider this shift – and the change in brain chemistry that it causes – may explain the growth in recent times of depression, maniac depression, memory loss, schizophrenia and attention deficit disorder. It may also be responsible for rising levels of obesity and heart disease which often accompanies overweight (*The Economist*, 2010 b).

Michael Crawford, researcher at the Institute of Brain Chemistry and Human Nutrition in London, has suggested that DHA had played a key role in the existence of nervous systems, and that access to large quantities of this fatty acid had permitted the evolution of big brains in humans' more recent ancestors. DHA has been the enabler of communication between nerve cells, particularly through electrical potential. Some 600 million years after animals became multicellular, more than half of the fatty-acid molecules in the light-sensitive cells of the human eye are still DHA, and the proportion of DHA in the synapses of the brain is not far short of that, despite the fact that similar molecules are far more readily available. M. Crawford has suggested that dolphins whose diet is based on fatty fish, have brains that weigh 1.8 kg, whereas zebra brains weigh only 350 g, even though the two species have similar body sizes. Furthermore, he argued that the dramatic increase in the size of the brains of humans' ancestors that occurred about 6 million years ago, was not because apes came out of the trees to hunt on the savannahs, but because they reached the coast and found a ready supply of DHA in fish. However, humans' ancestors were not exclusively living near the coasts and therefore M. Crawford's hypothesis is questionable. But most experts agree that DHA substitution by another fatty acid is not a good thing (Brand et al., 2010; *The Economist*, 2010 b).

Joseph R. Hibbeln, a researcher at the US National Institutes of Health, carried out a study in the early 1990s that showed that children who were breastfed had the same range of IQs, regardless of whether they had the ability to make their own DHA. In the case of those fed on formula milk low in DHA, though, children without the DHA-making ability had an average IQ 7.8 points lower than those with it (Steer et al., 2010).

It is not only intelligence that would be affected by a lack of DHA. Countries whose citizens eat more fish are less prone to depression, suicide and murder. J. R. Hibbeln's new research work showed that low levels of DHA were a risk factor for suicide among American servicemen

and women. Actual suicides had significantly lower levels of DHA in the most recent routine blood sample taken before they killed themselves than did comparable personnel who remained alive. More worryingly, 95% of American troops have DHA levels that these results suggest put them at risk of suicide. The US Department of Defence was to implement a programme which aims at supplementing the diets of soldiers with omega-3 fatty acids (Hibbeln, 2009; *The Economist*, 2010 b).

The US Food and Drug Administration may also change one of its policies. Thomas Brenna, a professor of nutrition at Cornell University, has written a letter (co-signed by many other scientists) urging the FDA to revise its advice to pregnant and fertile women that they limit their consumption of fish. This advice, promulgated in 2004, was intended to protect fetuses from the malign effects of methylmercury, which accumulates in fish such as tuna. The signatories argued that this effect was greatly outweighed by the DHA-related benefits of eating fatty fish (*The Economist*, 2010 b).

However, the popularity of omega-6-rich foodstuffs based on cheap vegetable oils would be difficult to reverse. In another experiment, Joseph Hibbeln fed rats diets that were identical except that in one case 8% of the calories came from linoleic acid (an omega-6 fatty acid), while in the other that value was 1%. These percentages reflect the shift in the proportion of omega-6 fatty acids in the American diet between 1909 and the early 21st century. In the 8% diet, levels of rat obesity doubled. In rats and also in humans, linoleic acid is converted into molecules called endocannabinoids that stimulate appetite. Those who consume omega-6 fatty acids want to eat more. And since in the case of humans, omega-6-rich food is much cheaper than omega-3-rich food, that is what they are likely to consume (Hibbeln et al., 2004; Hanbauer et al., 2009).

Eating fish remains a reasonable recommendation while at the same time bearing in mind than contamination with heavy metals, particularly mercury, is a serious threat to health. Crop biotechnology could be helpful in creating genetically modified soybeans with higher content of docosahexaenoic acid (DHA). Until then, a balanced diet containing adequate amounts of omega-3 fatty acids is the best approach.

Sugar alternatives, natural sweeteners

Artificial low-calorie sweeteners

Aspartame is a methyl ester of a dipeptide used as a synthetic non-nutritive sweetener that has been approved for use since 1981. Consumed in over

90 countries worldwide, it is used in over 6,000 low-calorie products (e.g. yogurt, cereals, chewing gum, etc.) and certain medicines. Sold under several brand names like Equal, NutraSweet, Canderel, aspartame is added to foods and beverages by manufacturers. On 5 May 2006, a study presented in Rome by the European Food Safety Authority (EFSA) concluded that aspartame was not harmful for human health. The executive director of EFSA, Hermann Koeter, highlighted that "the results of this new study on aspartame did not provide scientific evidence that would lead to question its use in food products." EFSA's conclusion was contrary to the conclusions of the work carried out by the European Foundation Ramazzini (Bologna), published in July 2005. The Foundation had advised pregnant women and children not to consume aspartame, after it was found that rats fed with aspartame at comparable levels per body weight to humans had a higher risk of developing lymphoma and leukaemia. Foundation Ramazzini's scientific director, Morando Soffritti, was worried by the insufficient evidence provided by the EFSA's study and announced the launching of a new three-year study on mice (Soffritti et al., 2005; Soffritti, 2007).

Since its discovery in 1965, aspartame has been the subject of controversial statements regarding its innocuity for human health. It is nevertheless widely consumed.

Sucralose is an artificial sweetener that is added to packaged foods and sold in packets or granulated form. It is marketed under the name Splenda for use in baking. Some studies have suggested sucralose may cause weight gain and have a harmful effect on beneficial gut bacteria.

Saccharin is the oldest artificial sweetener, discovered in the 1870s and used in products such as chewing gum and breath mints. It has been banned as a food additive in Canada since the 1970s, after studies linked the sweetener to cancer in rats. However, it can be sold to consumers as a sweetener, but only at pharmacies (Weeks, 2009).

Cyclamate is also an artificial sweetener, generally in the forms of sodium and calcium cyclamate. Sweet'N Low is an artificial sweetener that contains cyclamate. The latter was banned as a food additive after numerous studies linked it to cancer in animals, as well as possible male reproduction problems. However, in Canada, where it is banned for use as a food additive, it can be sold directly to consumers with a warning label (usually placed near the ingredient list) that it should only be used on advice of a physician (Weeks, 2009).

Stevia sweeteners

The leaves of *Stevia rebaudiana*, a plant species that grows in Paraguay and that is commonly used by the Guarani Indians to sweeten their meals and beverages, contain a sweetener (rebaudioside A) whose sweetening power is 300 times that of sucrose. *Stevia* has been used for about 40 years by Japanese consumers as a natural sweetener, in fact since the Japanese authorities had forbidden the use of sweetening additives such as aspartame. It has also been authorized in the United States by the end of 2008 by the US Food and Drug Administration (Mamou, 2010 b).

On 6 September 2009, the French Agency for the Sanitary Safety of Foodstuffs (AFSSA) authorized the use of *stevia* as an additive. The sweetener was commercialized in the form of a white powder, extracted from the plant leaves that have been macerated. It could be used in beverages, soups, confectionery, and its qualities could be highlighted through advertisement campaigns: high sweetening power, but zero calorie intake, no modification of glucose concentration in the bloodstream, recommendation for the food diets of diabetics and people suffering from hypertension and obesity (Ribaut, 2009). The authorization delivered by the French agency applied to a restricted category of food products, but it was extended to diet sweeteners in January 2010 (Mamou, 2010 b).

The European Food Safety Authority (EFSA), in charge of ensuring that *stevia* was innocuous for the health of Europeans, issued a favourable advice to the European Commission on 14 April 2010. In the meantime, at the beginning of 2010, Coca-Cola launched its FantaStill which, thanks to the addition of *stevia*, contains 30% less of sugar. In March 2010, Phare Ouest launched Breizh Cola Stevia, whose label bears a green leaf. In April 2010, Eckes Granini announced the marketing of Joker Vital Equilibre, a range of low-calorie (-30%) concentrated fruit juices, thanks to the addition of *stevia*. The Swiss Company Hermes Sweetener is commercializing sweet pills made from *stevia* in France, Australia and Switzerland. In June 2010, Danone launched the first yogurts containing *stevia* extract under its brand Taillefine (Mamou, 2010 b).

Health Canada did not allow *stevia* or its extracts to be used as a food additive in Canada because of insufficient evidence to support its safety. But in September 2009, the department released updated rules that allowed *stevia* and its extracts to be added as a non-medicinal ingredient to natural health products, which has opened the door to allowing food and beverage makers to expand the use of *stevia* in their products. For instance, PepsiCo Beverages Canada has launched the new vitamin-infused Aquafina water

beverage sweetened with Purevia. The company submitted an application to Health Canada's Natural Health Products Directorate to have the water approved as a natural health product. Coca-Cola Canada was planning to introduce beverages made with Truvia in the country (Weeks, 2009).

But not everyone is enthusiastic about *stevia* moving into the mainstream. Although it has a long history of use (in particular by Guarani Indians in Paraguay for centuries), there are fears that introducing *stevia* and its extracts in a wide variety of products could lead to potential health problems. For instance, Curtis Eckhert, professor in the environmental health sciences and molecular toxicology department at the University of California, Los Angeles, helped prepare a report in 2008 for the American Center for Science in the Public Interest that urged more testing on *stevia* extracts before it is widely introduced among consumers. Executive director of the Center for Science in the Public Interest Michael Jacobson stated he believed *stevia* was probably much safer than artificial sweeteners such as aspartame, but that more rigorous studies should be carried out. Stacy Reichert, president of PepsiCo Beverages Canada, stated the company believed there was "an extensive data-base" on the safety of rebaudioside A and that its long history of use provided strong evidence it would not cause any harm to consumers (Weeks, 2009).

The marketing push was working : consumer research firm Mintel estimated the market for products made with *stevia* could reach US$2 billion by the end of 2011 (Weeks, 2009). Euromonitor International forecast that the world market of *stevia* would increase from 52 tons in 2010 up to 421 tons in 2014 – an eightfold increase (Mamou, 2010 b), while Real Stevia, a Swedish distributor of the sweetener, estimated that *stevia* would make up 25% of the global market of sweeteners over a period of five years; the market value was estimated at US$50 billion (Ribaut, 2009).

Although these figures indicate a trend that is favourable to *stevia* (rebaudioside A), the low-calorie sweeteners available on the market – aspartame and saccharin – remain predominant. Thus, the global consumption of aspartame rose from 21,605.8 tons in 2004 to 24,323 tons in 2009; that of saccharin rose from 28,560 tons in 2004 to 32,479 tons in 2009. Could *stevia* replace sugar? Probably not. When *stevia* is used at high concentrations, it leaves an after-taste of liquorice, that changes the tastes sought by the consumers. That is why Coca-Cola uses *stevia* in its beverage FantaStill to lower the sugar content only by 30%; beyond such concentration, the taste of the beverage will be modified. Also Danone has added the equivalent of half a piece of sugar in its yogurts Taillefine Stevia (Mamou, 2010 b).

However, *stevia* offers new marketing opportunities to companies which, like Danone, are eager to foster the market of low-calorie fresh products. Due to its organic or "bio" quality, *stevia* could attract the consumers who, for instance in France, have been buying less low-calorie products since 2003. It is not just slenderness that matters, a more complex perception of the relationship between health and beauty is prevailing nowadays. It remains to be seen whether *stevia* may contribute to this trend (Mamou, 2010b).

Perception of sweetness

At the International Symposium on Olfaction and Taste, held in San Francisco during the third week of July 2008, two studies, one on mice and one on people, were presented and showed that two hormones seemed to fine-tune the perception of sweetness, and thus regulate the intake of sugar independently of the previously known mechanism of satiation located in the brain (McClintock et al., 2008).

The study on mice was carried out by Steven Munger, a neurobiologist at the University of Maryland, and his colleagues. They identified a hormone called glucagon-like peptide-1 (GLP-1) that is made by intestinal cells in response to sugar and fat. It was already known to act in the pancreas and the brain, where it helps respectively, to regulate blood-sugar levels and feeling of satiation that induces the cessation of eating. S. Munger, though, found that both GLP-1 and its receptor molecule that allows it to act are found in taste buds too (Scrocchi et al., 1998; *The Economist*, 2008 e).

To elucidate the role of GLP-1 in taste, the research team used a strain of mice, genetically engineered to lack GLP-1 receptors. They found that such animals were much less sensitive to sweetness than the unengineered controls. Indeed, the mutants were no more interested in a dilute sugar solution (or a solution of artificial sweetener) than they were in plain water. The control or "wild-type" mice, by contrast, drank significantly more of the sweet solution than they did of the water, even when the sweet solution was dilute. Moreover, the behaviour was limited to sweetness. The animals' responses to the other four fundamentals of taste – bitterness, sourness, saltiness and *umami* (the flavour of monosodium glutamate) – were unaffected. These results suggest, though they do not yet prove, that there is a feedback from the gut to regulate the desirability of eating sweet food (*The Economist*, 2008 e).

The study on humans was carried out by Yuzo Ninomiya, a neuroscientist at Kyushu University Section of Oral Neuroscience, Japan. He and his colleagues looked at leptin, another hormone known to regulate appetite and metabolism. Leptin concentrations are also known to fluctuate naturally over a 24-hour period, being lowest in the morning and highest at night, at least among people who eat three meals a day (Horio et al., 2010).

The Japanese researchers found that their volunteers were more sensitive to sweetness when their leptin concentrations were low. As the hormone concentration increased over the course of the day, the threshold for detecting sweetness rose. And when the researchers shifted the pattern of leptin production by changing the number of meals their volunteers ate, the volunteers' sensitivity to sweetness shifted as well, suggesting that it was the hormone rather than merely the time of day that was causing the effect. As in the case of mice, humans did not show any changes in their sensitivity to other tastes. However, individuals who had lower leptin concentrations, and thus more sweet-taste sensitivity before a meal, experienced sharper increases in blood-sugar levels when they had eaten (Horio et al., 2010).

The results of both studies may have an impact on the search for ways of reducing calorie intake of consumers that tend to eat much sugar and sweet products. See also *The Economist* (2006 c).

Cheese manufacturing : biotechnology and traditional know-how

Lactalis, a private group based in Normandy, is the largest cheese and milk producer in Europe, and also the world's biggest producer of unpasteurized cheeses. Globally, it is number 2 to Kraft Foods of the United States. Lactalis started using a mild form of pasteurization that heats the milk to a lower temperature than is the norm for pasteurization. "This so-called thermizing process removes potentially harmful bacteria," according to the company. In doing so, Lactalis sacrificed its appellation d'origine contrôlée, or AOC, status, a label supplied by a government body to verify that a product had achieved certain standards. But Lactalis subsequently wanted to win back the label, arguing that pasteurized cheeses should be included. In March 2008, the authorities stated they would protect small producers by reserving the AOC only for Normandy Camembert made in the traditional way. The small producers won the battle, but the broader war continues (Saltmarsh, 2008).

The debate is focused on the process of pasteurization and the effect that it is having on the product and the market. "Raw milk is the battlefield," stated Pierre Boisard, a sociologist who is author of *Camembert : A National Myth*. According to him, Lactalis has altered the landscape through its production of traditional products using industrial methods. Citing health concerns, and related import restrictions imposed by large markets like the United States, Lactalis moved away from making cheese from raw, or unpasteurized, milk, favouring pasteurization, which helps kill harmful bacteria. Small producers, as well as masters of the art of ageing cheese, are worried that industrial processes – from sourcing through production and distribution – are squeezing small farmers and threatening to deny consumers the choice, complexity and quality of a product, that is considered a luxury in many countries, but a staple on French tables. They also add that pasteurization eliminates useful bacteria as well. The giant producers reply that traditionalists are scared of losing market share to new techniques. Consumers, they state, are happy with the products available and prices charged (Saltmarsh, 2008).

Cheese indeed is a big business in France. There are an estimated 400 types of cheese in this country, and no other country offers the creativity and range in its cheese making. In addition to the world-famous AOC cheeses like Roquefort and Brie de Meaux, there are hundreds of cheeses with regional and local nuances. According to the Maison du Lait, which represents dairy producers, French cheese production rose 1.7% to 1.9 million tons in 2007 from a year earlier. Sales at large stores rose 2.2% and French exports were 4% higher. But production of AOC cheeses was down 1.2% in 2007 and raw milk cheese production fell 3.8% (Saltmarsh, 2008).

This may be explained by the scarcity of small producers and small cheese businesses, which are losing ground. Lactalis, meanwhile, employed 15,000 people in France in 74 locations, of which 19 were in mountainous regions (2008). While it mass-produces brands like Président Camembert and Bridel Emmenthal, it also makes a range of AOC cheeses. Dairy prices have risen alongside nearly all food prices in 2007, but small producers stated the price that farmers obtained for their milk had not risen in line, and hence only the distributors and the big players like Lactalis had benefited. While the big company claims that its objective is to develop a French corporation and to increase the consumption of cheeses and dairy products worldwide with good brands and consumer confidence due to quality, the founder of a regional cheese association complained about a "standardization" of the product as the mass market removes small producers by buying their land, pooling milk and hence deteriorating the final product (Saltmarsh, 2008).

In Italy, in a tightly defined area south of the Po River, mainly in the provinces of Parma and Reggio Emilia, the Conzorcio Produttori Latte in Baganzolino, in the flat farmland of the Po valley a few miles from Parma, makes the AOC Parmigiano-Reggiano cheese. An annual production of 120,000 tons of this kind of hard cheese is derived from the output of 500 dairies (*The Economist*, 2007 a).

Surplus whey from the dairy is used to feed 3,000 pigs, the meat from which is cured in hills south of Parma. About 170 local firms produce the ruby, sweet-tasting Parma ham; almost 10 million hams, each weighing about 8 kg, leave the curing cellars every year. Both parmesan cheese and Parma ham have behind them centuries of tradition, and their production is tightly regulated by certification agencies. So producers were mostly untouched by the collapse of Parmalat, a large milk and beverage group, based outside Parma that crashed in December 2003 with a hole of €13.2 billion in its accounts (*The Economist*, 2007 a).

Parmalat sold its foreign divisions, the number of brands has been reduced from 130 to 40, biscuits, snacks, bottled water and tomato sauce have been axed from the product range, the work force has been decreased, and Parmalat became profitable once again, with annual sales of about €4 billion. It now focuses on high-margin "functional" products, including milk, yogurt and fruit juices, which include health supplements. In January 2007, bottles of Zymil, a type of milk for people with digestive problems, were produced for the first time off a new €8 million production line (*The Economist*, 2007 a).

Parma's food-manufacturing industry is based on rich farming land and employed around 15,000 people in 2007 in businesses like preserved fruits and vegetables (Parma is a centre for tomato processing), sugar refining, ice cream and frozen foods. But as the food business grew strongly after the second world war, Parma developed an engineering industry specializing in food-processing equipment, employing about 8,000 people (2007). In 2005, the European Food Safety Authority (EFSA) settled in Parma (*The Economist*, 2007 a).

Salting and drying, or curing meat : the oldest method of food preservation

Cato the Elder served Rome as quaestor, praetor and consul, but he came from an ancient plebeian family in the Sabine region. His only complete surviving work, *De Agricultura*, is a farming manual where he gave the recipe for curing hams, after salting them for 12 days, hanging them in

the fresh air for two days, thereafter rubbing them with oil, hanging them in smoke for two days, rubbing them again all over with a mixture if oil and vinegar, and finally hanging them in the meat store. Neither moths nor worms will attack them. That recipe may have been an attempt to replicate the flavour of the hams smoked over juniperus and beech fires that Roman gourmets used to import from Germania (see the book *Salt*, written by Mark Kurlansky). In fact, the technique of salting and drying predated the Romans, who may have learnt it from Gauls and Celts, and also existed in the Far East who had no direct contact with Rome (*The Economist*, 2006 b).

Due to the simplicity of the ingredients – meat, salt, air and time, with smoke, seasonings and water as optional extracts – it is not surprising that the process of salting and curing was improved in many cultures, long before the chemical basis of it was discovered. Salting preserves animal proteins by inhibiting microbial growth (by dehydrating both the flesh itself and cells in the moulds and bacteria feeding on it, either ending or drastically slowing their growth). Salt can be rubbed into the meat, or the meat can be submerged in brine. Brined or pickled meat often has to be cooled or soaked, because wet cures permeate the flesh more thoroughly and the meat would otherwise be too salty to eat. The meat is then hung up to dry, either in fresh air or in smoke, which is itself a complex material made up of hundreds of components, including carcinogens, which inhibit microbial growth; phenolics, which slow down fat oxidation; and a range of sugars, acids and particulates that colour and flavour the meat. It should be stressed that excessive consumption of smoked and cured meats has been linked to several types of cancer (*The Economist*, 2006 b).

Western Europeans used to smoke meat over alderwood, though oak and beech are becoming more prevalent. North Americans tend to use hickory, mesquite, pecan, apple or cherry. Woods with heavy concentrations of resin, such as pine and fir, are unsuitable for smoking, because resin tends to flare and produce too much soot and tar. There is also a distinction between hot-smoking, in which meat is cooked in a smoky oven, and cold-smoking in which salted or brined meat is exposed to smoke but not heat. In hot-smoking, the muscle filament proteins uncoil and coagulate, while in cold-smoking they remain coiled but microbe-free. Cut thinly and with the grain, a salted, cold-smoked leg of pork – such as Western Europe's *prosciutto*, *jambon cru*, *jamón* or *schinken*, Appalachian America's country ham or China's Yunnan ham – retains the silky texture of the raw meat. While hot-smoking transforms the meat, salt, smoke and wind (cold-smoking) preserve it (*The Economist*, 2006 b).

History of cured meat, salted and smoked fish

The English word "jerky" for dried meat is derived from the Quechua word *charqui*, and archaeological evidence shows that the Incas sliced and salted meat surplus to requirements, and then left it to dry in the wind and sun. The cowboys of America's Far West did much the same. In the sun, fresh meat could take a full day to dry out; today the sliced, spiced, brined meat rolls on nylon screens through a drying oven with fans and exhaust pipes to draw out moisture, reducing the drying time to a few hours (*The Economist*, 2006 b).

Ships used to sail from European ports with casks full of salt pork. Cut from the fatty belly of a pig, like bacon, salt pork requires blanching to render it edible. Salted for a fortnight, it could last for two years in a cold climate. Nowadays, cooks use it mainly for flavouring (*The Economist*, 2006 b).

Salt cod fuelled the economies and the maritime explorations of much of northern Europe between the 15th and 19th centuries. Although Norwegian sailors caught cod off their coast as early as the 10th century, it was not until 1497, when John Cabot discovered Newfoundland and the abundant fish stocks around its banks, that salt cod became an important commodity. The Portuguese, who had been selling salt to Scandinavia since the 12th century, quickly set up cod-fishing outposts in Newfoundland, and salt cod became an important item of Iberian culinary culture (*bacalao* and *bacalhau* in Spanish and Portuguese respectively). Spain and Portugal, which remained Catholic as the Nordic countries became Lutheran, needed fish for Fridays and fasting days, and salt cod travelled even to Iberia's mountainous and inaccessible interior. Cod stocks in the northwest Atlantic have fallen by 96% in the past 150 years, causing the price to jump and consumption to drop. A famous Portuguese cookbook of the early 20th century contains 365 salt-cod recipes, one for every day of the year (*The Economist*, 2006 b).

To preserve cod, the fish is gutted, heavily salted and packed in barrels. As the salt leaches the water from the fish, they are soaked in self-created brine, and then they are hung up to dry. Smoked salted salmon, called *lox* in Yiddish, was as important to the Eastern European Jewish diet as cod was to the Portuguese. Its abundance in German waters made its cheap; salting made it transportable to inland regions; and fish neutral status in Jewish dietary rules meant the orthodox people did not have to worry about how and by whom it was killed and prepared. Its processing is nevertheless supervised by local rabbinic authorities (*The Economist*, 2006 b).

Except for some factory-produced smoked meats, most of which are also cooked for extra safety and durability, curing and smoking methods for meat and fish remain largely unchanged. Sailors who brought salt from the Adriatic Sea and up the Po River to Parma in the 8th century would be paid either in money or in ham. Parma remains the world's most famous producer of cured hams. The craftspeople themselves also continue to use the same methods they have always used. After the second world war, as food production across Europe became industrialized, making ham in the traditional labour-intensive manner ceased to be a necessary way of life (*The Economist*, 2006 b).

Curing meat : role of microorganisms

Cured meats, with almost identical ingredients from region to region, taste differently. For instance, Italy produces six *denominazione di origine controllata* varieties of *prosciutto*, all of which are made from the whole leg of a pig, salt and perhaps a bit of sugar or spice. Due to the airborne yeasts and moulds native to the particular environment, the variations in humidity, temperature and air quality, the diet and care of the pigs and the storage of the resulting hams, each of them tastes different from the rest. For instance, cured ham produced in Spain, in the hilly regions of Andalucia (near Granada) or around Salamanca (Gijuelo) under oak trees, is highly appreciated because the pigs (of the Iberian breed, with black feet) are fed with acorns, in a free-range system. A *prosciutto* from Parma is softer, pinker and milder than a *prosciutto* from Modena, and a Lyonnais *saucisson* has a tang that a *salame Piacentino* lacks (*The Economist*, 2006 b).

Dry-curing sausages, however, as opposed to whole hams, relies on fermentation in addition to desiccation. The interior of a raw ham, having never been exposed to air, remains relatively sterile; sausages which are made of ground (and therefore aerated) meat, along with added fat and seasonings, require an acid to kill bacteria from the inside out, and salt that dries microbes from the outside in. Dried air-cured sausages have a long history : the Romans learnt the craft from the Lucanians, a tribe in what today is Basilicate, in southern Italy. The tribe's name persists in dry-cured sausages across the Mediterranean : the Greek *loukanika*, Spanish *longaniza*, Italian *luganega* and Portuguese *linguica* (*The Economist*, 2006 b).

The traditional acid of choice is wine, although many sausage-makers nowadays use dried, powdered strains of lactic-acid-producing bacteria that kill *Listeria* and other harmful microbes. As they multiply, these

bacteria produce sodium nitrite, which protects against botulism, a lethal form of food poisoning caused by the anaerobic bacterium *Clostridium botulinum*, whose species name derives from the Latin word for sausage (*botulus*). They also inhibit mould growth inside sausage, but allow beneficial and tenderizing white mould to grow on the surface of the sausage. But both types of mould, as well as a range of bacteria, both harmful and helpful, are present in the air, and dealing with them, i.e. favouring the good ones and killing the harmful ones, is a matter of experience and know-how. For instance, in the 15th century, Bartolomeo Sacchi, a writer from the Po Valley town of Cremona, suggested testing the quality of a ham thus : "Stick a knife into the middle of a ham and smell it; if it smells good, the ham will be good; if bad, it should be thrown away." The smell-test remains a standard and reliable practice, both for whole muscle cuts such as *prosciutto* and for dry-cured sausages such as traditionally made *salami* or *saucisson*. A trained eye can also make the difference between the healthy white mould in the exterior of a sausage, which protects it from bacterial contamination and tenderizes the meat, and the furry blue moulds that can grow inside the casing and spoil the sausage (*The Economist*, 2006 b).

Industrialization and regulation of cured and dried products

Over the past century, as food production in the United States and Europe has become more industrialized and the population has become more urban, cured and dried products, manufactured according to old traditions, have become scarcer. Cooking or irradiating sausages, rather than curing them, is a global trend. Also the disappearance of the neighbourhood butcher and the decrease in meat consumption explain the decline in proper meat-curing. Stricter regulation of the process also plays an important role in the production and commercialization of cured-meat products. For instance, any producer wanting to export cured meat to the United States must have a representative from the US Department of Agriculture at the site of production and must follow the same stringent procedures as the American sausage-makers do. Producers must therefore keep voluminous records on everything from the temperature of the storage room – measured and registered every four hours – to the meat products pH, the proportion of water in dry-cured sausages and the cleanness of their employees' shoes and clothing. Spanish cured ham, *jamón ibérico*, has not been legally available in the United States until the end of 2006, but has become so from 2007 because the US Department of Agriculture has at long last decided that the methods used by Spanish craftspeople for centuries were unlikely to poison US citizens (*The Economist*, 2006 b).

Food industrialization, along with stringent regulation, offers safer products to the average consumer. The artisan-made products are considered more tasteful, but they have become a luxury item. Those who can afford them are targeted by some producers who can become very successful. For instance, Paul Bertolli, the grandson of an Italian butcher, used to run Chez Panisse, one the most celebrated restaurants of the United States, decided in March 2006 to abandon his job in order to start a business making hand-crafted, dry-cured sausages. The monthly turnover is over 35,000 pounds of pork, and his sausages rival those found in Italy. Having studied with the itinerant Tuscan sausage-makers, the *norcinos*, he realized he could replicate their know-how in California's East Bay, which has a similar climate (*The Economist*, 2006 b).

Canned food : a two-century-old technique of food preservation

Nicolas Appert (1749-1841) was the son of an inn's owner at Châlons-en-Champagne (centre-east of France). When he was 11 years old, he left his family and started to learn the profession of food supplier, first in champagne cellars and thereafter in a brewery. In 1784, he opened his first luxury shop in the centre of Paris and became a renowned confectioner. In 1789, he participated in the French Revolution and was jailed; after being freed, without having been judged, he abandoned politics and devoted his skills to the preservation of food. During that time, foodstuffs used to be preserved through drying, salting and smoking, or conserving them in vinegar and alcohol, or dipping them in fat or sugar for long periods. But all these processes resulted in the loss of many food qualities (Géné, 2010 a).

His obsession of not wasting food led him to discover a very ingenious conservation process, in a workshop located in the suburbs of Paris, at Ivry-sur-Seine. In 1795, he found that only intensive heat could preserve food. He chose glass recipients : champagne bottles with a larger neck, which he closed with a cork tap, maintained with iron thread; the bottles were wrapped in a cloth and put in heated water. The technique was called *appertization* and was developed 60 years before the research carried out by Louis Pasteur on food and beverage preservation, also using heat to kill microorganisms. N. Appert's technique could keep intact the natural qualities of fruits and vegetables (Gazsi, 2010 a).

While Peter Durand was granted a patent on this technique in 1810, N. Appert had been awarded before only a price, on the condition to offer his discovery to the public. His method was disseminated through a handbook published at his own cost and titled *L'art de conserver*

pendant plusieurs années toutes les substances animales et végétales ("The art of preserving for several years all animal and plant substances") [Gazsi, 2010].

The book was translated and republished many times, and it became the guide for numerous factories in Europe, and particularly in Great Britain, which were producing preserved foodstuffs (Géné, 2010 a). The technique was an immediate success and it was copied in the form of cans made of laminated steel or corrugated iron. It was acclaimed by the critics of that time – in particular by the famous gastronome Grimod de La Reynière – and it was adopted by the manufacturers of canned sardines in Brettany, the mining corporations and above all by sailors and soldiers across the world. For the latter, who often died from scorbut (vitamin-C deficiency), canned food was a blessing, even though it was not easy to open the cans (can openers were commercialized in 1850 only). People had to wait another 45 years to be able to use the "universal key", around which the lid of a sardine can is enrolled, making the opening of cans very easy (by the end of the 1960s, cans of soft drinks had a ring on their lid which one can pull to open the can) [Gazsi, 2010].

Nowadays, despite the predominance of frozen food (an annual global turnover of about €7 billion), canned foods still occupy an important place in supermarkets and 99.7% of households consume them. In 2009, due to the economic crisis, the production of cans rose to 3 billion units, the annual turnover reaching €4.4 billion. According to Laurence Silbert, of the Interprofessional Union for the Promotion of Appertized Can Industries (UPPIA), "the behaviours of French people have changed; they cook using fresh products, canned and frozen foods. In addition, the lower reputation of the metallic can is nowadays partly erased by the renewal of preserves in glass flasks". And in fact the biggest manufacturers of metallic cans have moved to glass recipients for preserving foods (natural vegetables, sauces or prepared meals) [Gazsi, 2010].

Currently, in France, 3.5 million tons of appertized products are consumed annually (i.e. 50 kg per inhabitant), of which 85% are canned products, far ahead of products preserved in glass flasks (Géné, 2010). Canned food is a success story. It has been largely supported by slogans and drawings in newspapers and the media. The can has even been adopted by many artists : Duchamp, Ernst and Pollock, and Andy Warhol, who, during the 1960s, has made Campbell soup an icon (Gazsi, 2010).

Coffee

Origin and cultivation

The coffee tree (*Coffea*) is a shrub or a tree belonging to the Rubiaceae family, which can reach 6-12 m, but is lopped to the height of 2-3 m in order to facilitate the harvest of coffee berries. At the age of three years, white and fragrant flowers blossom (henceforth the old name of the coffee tree, *Arabia's jasmine*), which wilt after fertilization. At the basis of flowers (30,000 per year per tree), the fruits or berries are formed. Each berry is filled with a sweet pulp and contains two seeds, rich in caffeine.

Coffee trees grow in a humid atmosphere, on soils that should be fertilized with nitrogen and potash. Dutchmen succeeded in creating optimum growth conditions in greenhouses built in Amsterdam for the plants received in the 17th century. But European countries that realized their weather conditions were not appropriate and that sufficient land was not available, developed coffee plantations in their colonies, where a warmer climate was much more favourable and labour, often from slavery, was abundant and cheap.

A coffee tree lives about 70 years; it shows its optimum development when it is 20 years old, and produces its first berries at the age of five. Of about 80 species of coffee, two are of economic importance, *arabica* and *robusta*. *Arabica* coffee was consumed by nomadic tribes living in Abyssinia and Arabia, where it was growing as a wild shrub in the region of Moka (cf. the story of the shepherd Kaldi who observed dizziness among his goats that consumed the berries of this plant). In these old times, the ripe fruit was ground, mixed with animal fat to form balls that were chewed by the nomads during their journeys. Later on, berries mixed with water gave a cold drink. Coffee became a hot drink only by 1000.

Researchers of the French Research for Development Institute (IRD) have demonstrated that coffee trees' origin is in Atlantic Central Africa and not in East Africa as was assumed during the 1980s. Due to the presence of coffee trees around the Indian Ocean, botanists thought that they came from East Africa before the dislocation of the Gondwana supercontinent, more than 100 million years ago. In order to establish the history of the coffee tree, François Anthony, a research director at IRD, and his team have sequenced the DNA of 26 species of the genus *Coffea*. They concluded that there were two lines that played a key role in the evolution of the coffee tree : one present in the overall distribution area from West Africa

to Madagascar, and the other found only in lower Guinea. This origin is known for the high diversity of its flora; it also shows the greatest diversity in the DNA sequences of coffee species. It is therefore their centre of species differentiation and most probably the craddle of the coffee tree. IRD researchers have tried to determine when the evolutionary process started. It was done with the help of a species close to the coffee tree and belonging to the genus *Rubia* (both belong to the Rubiaceae family). This close relative has been found in fossils and a rather precise datation has been made, in addition to the fact that genetic mutations accumulated in its genome since its appearance are known. The overall conclusion drawn by the French scientists was that coffee trees were born 400,000 years ago; they are therefore much younger that botanists initially thought.

Their diversification and dispersion have been therefore very rapid. Consequently, the genome shows little difference from one species to the other. But gene expression is, by contrast, very variable. This study on the origin of the coffee tree should result in the reorientation of the genetic research to be carried out on this species, with a view to improving this crop of high agronomic and socio-economic interest, that was discovered and domesticated in the 8th century.

Dutchmen propagated *arabica* coffee from Abyssinia in Java, while French people set up plantations in the Caribbean, and from there the tree expanded into America. *Arabica* trees grow in the intertropical zone, on the highlands. *Arabica* is a self-fertilized plant. Several subvarieties have been developed, such as the fruity moka, the bourbon in Mauritius, the maragogype in the region of Bahia-Brazil, and the highly appreciated Blue Mountain coffee of Jamaica. *Arabica* is appreciated for its fine taste and represents three-fourths of world production.

The rest of the production is contributed by *robusta* coffee. It is well adapted to equatorial climates and grows as a wild tree in tropical forests, in Africa, India, Indonesia, Sri Lanka and the Philippines, on lowlands. *Robusta* coffee plants are allogamous and therefore need pollinating insects. It grows faster than *arabica*, is more pest-resistant and has a higher output. It contains more caffeine, but it tastes stronger, “earthy”, and is less appreciated by coffee drinkers.

History

Less reluctant than the inhabitants of Venice, English people opened up coffee-houses very early in Oxford and London; these coffee-houses attracted men who spent a lot of time chatting and arguing,

while the spouses remained at home. Housekeepers therefore drafted the "Women's petition against coffee" and Charles II, in 1676, decided to close down coffee-houses considered as places of liberal unrest. However, the protests of husbands were so strong that the royal decision was cancelled. In 1700, there were over 2,000 coffee-houses in the United Kingdom.

Ship owners and sailors used to attend Edward Lloyd coffee-house, in Tower Street, where they collected their mail and could retrieve all kinds of maritime importations, sell their merchandise and ships, and even share between themselves wartime profits. In 1670, a coffee-house opened in Berlin, before the Procope in Paris.

With the ever-growing demand for coffee, Europeans moved to their colonies to plant coffee, especially when exports from Arabia tended to decrease. Dutchmen, both excellent navigators and agronomists, were the first to transfer coffee shrubs from greenhouses in Amsterdam, to Java in 1699, thereafter to Surinam. In 1714, Captain Gabriel Mathieu de Clieux robbed a cutting of coffee tree offered by Holland to Louis XIV and took it to the Antilles. In three years, millions of trees were planted in Martinique and Santo Domingo. In 1727, a Portuguese lieutenant, coming through Cayenne, robbed a few berries and bought them to Brazil. In Ceylon, British coffee plantations heavily suffered from a disease and were replaced by tea plantations; the British planted coffee in India only in 1840.

Coffee industry in the Antilles and South America was sustained by slavery and slave trade. After the abolition of slavery, the industry survived. Brazil, which was the last country to abolish it in 1888, is nowadays the world's leading producer of coffee.

During the 20th century, coffee suffered from several crises, mainly of a speculative nature. It is today the world's most consumed beverage, after water, and ranks second behind oil in terms of international trade.

Harvesting and processing

From 2.5 kg of coffee berries produced by a coffee tree, one can obtain one pound of green coffee. Berries become mature six to eight months after blossoming in the case of *arabica* and nine to 11 months in the case of *robusta*. Stripping, either manual or mechanized, consists of ripping off flowers, mature or green berries, from the branches. This process, indispensable in very large plantations, has the drawback of harming the

trees and of resulting in a heterogeneous harvest containing both mature fruits, and younger and acid ones. Stripping is poorly adapted to rugged landscapes where *arabica* grows.

Hand picking, by contrast, leads to high-quality coffees, but overloads production costs of small producers. In Brazil, 300,000 farmers make a living from coffee and harvesting mobilizes 3 million people. In this country or in Africa, farms with an acreage of several thousand hectares are suited to mechanical stripping (a machine replaces 100 persons and can harvest 95% of the fruits during one single trip), but 70% of world coffee production is from family farms with an average acreage below ten or even five hectares.

After having been harvested, berries are washed several times to take away their pulp (wet process) or sun dried (dry process); coffee beans are separated from their shell, selected and put in jute bags of 60 kg to be exported. Coffee beans can be stored for several years.

Roasting of coffee beans is an art. Before doing so, the roaster should know the properties of the beans, their origin, so as to make the appropriate blending. As a fine cook, each roaster has his recipes and secrets. Coffee roasting in a rotating cylinder can be made in different ways. The fastest one, called flash, consists of exposing the beans to a temperature of 880°C for 90 seconds. This technique is applied to current varieties and cannot lead to the expression of all aromas (there are more than 700 aromatic compounds in coffee). Another technique that lasts 10 minutes at 600°C, leads to the expression of a wide range of aromas. The best results are obtained by using the old process : beans are roasted for 10 minutes at 230°C, so as to become deep brown; aromas are expressed during the following 10 minutes.

While they become brown or black, beans double their volume, lose their oil and are cracked. Once roasting is over, beans are mixed with cold air so as to condense aromas, and thereafter they are packed (either ground or as beans) under vacuum. By contrast to wine, coffee does not bonify with age : beans are oxidized in 20 days; ground coffee is oxidized in five days. It can be stored for months in a hermetic box in the refrigerator (coffee should not be exposed to air, humidity, light or heat).

The domestic cubic grinding mill, made of wood, with its crank and drawer to collect ground coffee was first used at the beginning of the 19th century (in France, the model sold by Peugeot was on the market in 1832). It was thereafter replaced by an electric machine. This way of

preparing coffee in the household has encroached on one of the major social functions of coffee-houses; in those times, retailers had the almost exclusivity of the product and of the tools of preparing the beverage; thus, persons of a wide range of origins could find a reason or a pretext to leave the private sphere, to go out and mix with other coffee drinkers, to know each other, to chat and exchange ideas, a behaviour that was not agreeable to spouses and authorities.

Ground coffee, when prepared as a fine powder and exposed to a small volume of boiling water, is very aromatic and liberates little caffeine; this is the way it is prepared in Italy. There are five traditional ways to prepare coffee : Turkish decoction (very finely ground coffee, mixed with sugar, settles in the coffee-pot put on the fire); infusion (in a glass vessel, a piston presses down the residue); filtering (hot water is poured on ground coffee that is put on a paper filter); percolation (coffee powder is placed in a filter between two superposed compartments, and water heated in the lower compartment runs through bottom up); pressurized percolation (same principle as with percolation, but water pressure is increased by air compressed at 10 bars) is used to serve expressos rapidly and is suited for different tastes (small or larger volumes).

High-quality coffee is drunk immediately after having been prepared, generally very hot. Sugar can be added, or milk, cream, cocoa, or spices such as cardamom and nutmeg. In Rome, it can be mixed with ground ice, and called *granita di caffe*.

Trade

When about 1.5 billion cups of coffee are drunk every day in the world (2006), coffee cultivation is the mode of living of about 125 million people, including 25 million small farmers. In Colombia, in 2006, for instance, some 350,000 families made their living from coffee cultivation.

The demand for coffee originates from developed countries to the extent of 85% and coffee trade sums up to US$9 billion per year (2006). Annual production reached about 126 million sacks (60 kg each) in 2009 from 70 countries. The main coffee producers are : Brazil, Colombia, Mexico, Ethiopia, Guatemala and Costa Rica.

Robusta coffee is produced by six countries : Indonesia, Vietnam, India, Uganda, Côte d'Ivoire and Cameroon. Over the period 1997-2005, production rose 20%, i.e. twice the increase in demand, causing a glut of coffee and a drop in prices. In 1962, an international agreement fixed

export quotas in order to stabilize prices. As communist countries were used to barter coffee against machines, Vietnam started to cultivate coffee on a large scale and in ten years this country, supported by the World Bank, climbed up from 31st rank of producers to the fourth one. In 1993, the United States withdrew from the international agreement, followed by Canada. In addition, they tried to convince African countries to adopt a liberal policy, while ensuring the farmers that they will have a decent income. In fact, the poorest countries where coffee is a major part of their exports (e.g. 50% in Ethiopia and 80% in Burundi), lost half of their income. In Latin America, farmers (not owners of their farms) sold their coffee harvest at production cost. The consumers of developed countries were the beneficiaries of the elimination of the international agreement, not the producers.

In fact, the five multinational corporations [including Sarah Lee, Nestlé, Kraft Foods (Maxwell)], which purchase (in dollars) half of the world harvest, did not suffer at all from the crisis. In order to pay the fair price to the producers, a number of fair-trade operations were initiated; the oldest one (1988) bears the name of Max Havelaar, the hero of a novel published by the Dutch writer Eduard Douwes Dekker in 1866 (see p. 577).

On 16 June 2010, at the New York Stock Exchange, the pound of *arabica* coffee rose up to 183.45 cents (for a delivery in September 2010). This was a peak after 27 months and it was reached after a 20% increase in nine days. *Robusta* rose 18%. On 18 June 2010, the price fell back to 180.15 cents a pound. The reasons for such an increase have been known for a long time. On the demand side, coffee is drunk more every year (about +2%). For instance, according to the studies carried out by the American Coffee Association, coffee has become in the United States the second beverage after water. In the case of gourmet coffees (or terroir coffees) that are strictly selected, the increase in consumption was 5%, and there is a real fashion for "vintage" coffees promoted by Kraft, Starbucks and Nespresso. On the supply side, production is not sufficient, although *arabica* coffee is cultivated on 7 million hectares, compared with 4 million hectares for *robusta*, and its annual production amounts to two-thirds of the 126 million sacks produced globally. Colombia, the world's second biggest producer behind Brazil, has been faltering since 2008; it has uprooted its old coffee trees and replaced them with younger ones that will bear fruit only in 2013. Production fell down to 8 million sacks in 2008-2009 from 11 million sacks during the previous campaign. The International Coffee Organization announced in June 2010 that Colombian plantations will not increase their output before 2010-2011. Specialists of the Macquarie Group reported on 16 June 2010 that in 2010 the *arabica* global market will be again in deficit, even though

the Brazilian harvest was "exceptional" (Faujas, 2010). Consequently, investors thought it was a good deal to invest in *arabica* coffee, when oil and metal prices were hesitating. According to Benoît Bertrand, a researcher at the French International Cooperation Centre on Agricultural Research for Development (CIRAD), "despite the good prospects of harvest in Latin America, the market reacted to the fact that quality coffees were vulnerable to climatic vagaries, and speculation has bet on the deficit that would result" (Faujas, 2010). In fact the price of the pound of *arabica* coffee rose again up to 222.45 cents at the New York Stock Exchange on Friday 17 December 2010, a peak since June 1997; probably because of the fears about a harvest less abundant than forecast.

Trade of quality coffee

In 1999, Susie Spindler initiated in Brazil a competition, Cup of Excellence, that aimed to reward the country's best coffees. It is now conducted in seven Latin American countries. The competition is open to any grower in each country, tasting and scoring is systematic and blind, and the winning beans are sold worldwide in an online auction. By focusing on quality and transparency, S. Spindler has not just brought out excellent coffees from some unexpected sources, but has connected the best growers to buyers prepared to pay for quality (*The Economist*, 2007 b).

A trend led by Starbucks and other specialty roasters has introduced drinkers to coffee differentiated by origin and type. Small roasters such as Stumptown, based in Portland, Oregon, are taking this approach further, borrowing concepts such as terroir, vintage and appellation from the wine world, taking great care in roasting and preparation, and emphasizing quality. As a result, the coffee trade has bifurcated in the past decade into commodity coffee, sold in large quantities and at a low price, and specialty coffee where quality rules. Growers producing unexceptional coffee must either cut costs to compete with big mechanized farms – impossible for most – or improve quality. The benchmark "C" price is set at the New York Board of Trade, and varies depending on the weather, the level of demand, and other factors. The aim of the Cup of Excellence and other schemes is to enable high-quality coffees to differentiate themselves and command a premium over the C price. In Brazil, for instance, investments in quality can increase a farmer's profits by 50% (*The Economist*, 2007 b).

For poorer farmers in less developed countries, even modest investments that would greatly improve their coffee can be out of reach. In such places, targeted assistance can help. The tasting expertise and price-discovering transparency of Cup of Excellence can uncover remarkable global

competitive advantages in some regions. Starbucks' CAFE practices, Fair Trade and other schemes can have a similar effect in some countries by providing technical and management assistance, improved facilities and access to credit (*The Economist*, 2007 b).

With just under 800 sacks of coffee of 60 kg each, in its Brazil auction, Cup of Excellence is insignificant alongside worldwide production of around 126 million sacks a year. Yet, it is influential. The winner in Brazil's Cup of Excellence competition in 2001 went on to sell his coffee for US$700 a sack, doubling his farm's income. On 16 January 2007, 21 sacks of Fazenda Esperança – the top producer in 2007 contest in Brazil – fetched almost US$40,000 from Japanese and Taiwanese bidders, more than ten times the C price. The new diversity of buyers gives farmers a chance to maximize revenue by selling their coffee through many channels simultaneously : their best through internet auctions, a specialty grade through Fair Trade or other cooperatives, a commodity grade to big exporters and the rest to local markets (*The Economist*, 2007 b).

Wine and spirits

Wine consumption and its impact on health

In France, a global leader in wine production, tradition of drinking wine is very old. In the 19th century, Louis Pasteur – a founder of modern microbiology – used to recommend the consumption of one glass of wine per day, thus suggesting a positive impact on health of small quantities of the beverage, but at the same time warning against alcoholism. Since then, there have been recurrent controversies about the volume of wine that is reasonable to drink; any restriction was confronted to the powerful lobby of winemakers and other spirit manufacturers. Liver cirrhosis was a very common disease in France, but not all the forms of the illness were due to excessive consumption of alcohol. It has also been noted that the annual average consumption of alcohol by the French population has decreased, particularly among youth (although recent surveys tend to show a fashion for hard liquors among young people). In addition, wine, including champagne, is being preferred to spirits among middle classes.

On 4 May 2004, a study carried out by Onivins was made public and concluded that : "In 1980, wine was consumed with meals by one French person out of two; it was the preferred beverage of French people during meals, ahead of tap water. Twenty years later, wine was consumed with meals only by one French person out of four. It is bottled mineral water which they preferred during their meals," particularly at lunch time.

But at the beginning of the 21st century, French people are those who consume most wine worldwide, just after the Italians : an average of 58 liters per capita per year. And the French market, even though it is declining, remains the biggest one, ahead of Italy and the United States (Galinier, 2004).

According to Onivins, the percentage of those who declared they were not consuming wine had jumped from 24% to 37% between 1980 and 2000, which meant a loss of 3 million wine drinkers. This proportion has remained stable since the early 1990s, and, taking account of population growth, there were another 2 million wine drinkers between 1999 and 2000. Among those between 20 and 24 years of age, 57% declared they were not consuming wine in 2000, according to the study carried out by Onivins. It is not therefore surprising that France will be one of the few big countries where wine consumption is expected to decrease over the next years, while it is growing in the United States, the United Kingdom and even Italy (Galinier, 2004).

The proportion of those who recognised they were drinking wine every day fell from 47% to 24% between 1980 and 2000. During the same period, the proportion of those who used to drink wine once or twice a week rose from 30% to 40%. As a regular wine drinker consumes about six times more than an occasional one, the losses have been high for the wine industry – which employed 260,000 persons and had an annual turnover of €8 billion by the early 2000s (Galinier, 2004).

The overall conclusion was that wine consumption had decreased very significantly over the last decades, much more than other alcoholic beverages, and even though about 60% of alcohol consumption in France is in the form of wine. Those who are between 15 and 24 years old consume "only" 19 liters of wine per capita per year, according to the study of Onivins. However, they drink an average five glasses of strong alcoholic beverages during week-ends, according to the services in charge of road and traffic casualties (Galinier, 2004).

The wine lobby wanted to rehabilitate the "food qualities" of wine, but in fact what happened was the conversion of vineyards towards fine wines, with Certified Geographic Indications, more elaborated and also more expensive. This trend has been successful in several cases, for instance, sparkling wines and champagne, the consumption of which has risen fivefold since 1960. This trend has been detrimental to ordinary or table wine, that of regular drinkers and generally strongly criticized (Galinier, 2004).

In order to maintain production, wine producers have bet on the large distribution networks (supermarkets), which are selling 80% of the wine sold in France. But the producers and traders of fine wines did not adapt their prices and marketing to this new mode of distribution; on the contrary, prices rose considerably. And the Certified Geographic Indications have stuck to their old and complex rules, with labels that guarantee the geographic origin of the wine, but are not sufficiently clear with respect to the grapevine variety used and to quality (Galinier, 2004).

But again what should be the reasonable recommendation for wine consumption in relation with its impact on health, excluding of course children and teenagers? David Khayat, head of the oncology department at the hospital La Pitié-Salpêtrière (a very large medical compound in Paris), published a book titled *Le vrai régime anticancer* (The true anticancer diet) in 2010, where he presents a state-of-knowledge concerning the impact on cancer of a wide range of foodstuffs and beverages. In fact, he decided to write this book after the statement in 2009 by the National Cancer Institute that red wine was carcinogenic as of the first glass. D. Khayat had been appointed in 2002 by the French president Jacques Chirac to lead the new National Cancer Institute and the vast programme of research assigned to it. He left the Institute a few years later and returned to his faculty job.

While stating from the outset that "not a particular foodstuff can trigger a cancer, and that there is no foodstuff which prevents the disease surely" … "A type of cancer can take sometimes ten years to develop and drinking liters of pomegranate juice will not stop the differentiation and multiplication of cancer cells. Maybe it could delay the process," wrote D. Khayat. In the case of wine, he asserted that the first glass of wine is not carcinogenic, as stated in 2009 by the National Cancer Institute (INCA). This was also contradicted by the High Council of Public Health. D. Khayat indicates that 60% of mouth cancers are caused by a papillomavirus, of the same group as the virus that causes cervix cancer, now prevented thanks to vaccines (see p. 151). This risk is also low for other cancers, such as colon, breast and even liver cancer. Red wine, consumed in small quantities, can have a health benefit including against cancer, because it contains a strong antioxidant (resveratrol) that could inhibit the disease at several stages of its evolution. Consequently, the recommendation by D. Khayat is : two glasses of wine per day for women, and three glasses for men, i.e. a maximum 30 g of ethanol per day. If the wine is low in alcohol and sugar, it is even better, because excess consumption of sugar has negative effects on health.

On the other hand, Joseph Kanner and his colleagues of the Hebrew University of Jerusalem have discovered when meat and wine mix in the stomach, compounds of the wine thwart the formation of harmful chemicals, released when meat is digested. They hypothesized that if wine polyphenols arrive in the stomach at the moment when the fats are releasing malondialdehyde and its kin, then this might stop these toxic materials from moving any farther into the body. To test this hypothesis, J. Kanner and his colleagues fed a group of rats one of two meals – either red meat from a turkey (a foodstuff shown by previous research to raise malondialdehyde levels in humans) or such meat mixed with red-wine concentrate. An hour and half after the rats had eaten, they were killed. Their stomachs were removed and the contents were analyzed. The researchers discovered that the wine concentrate did indeed reduce the formation of malondialdehyde. It also cut the concentration of hydroperoxides, that also cause cell and tissue damage (Gorelick et al., 2008).

J. Kanner and his colleagues argued therefore that looking for antioxidants from wine in the bloodstream was a mistake; they do not need to be there to be useful. Their results also suggest that the habit of eating fruit at the end of the meal is a healthy one. Many fruits are rich in polyphenols (wine is made from fermented grape juice) and, eaten at the end of the meal, they arrive in the stomach at the point when meat digestion is producing harmful chemicals *(The Economist,* 2008 c).

The cult of fine wine

According to *The Economist* (2009 d), the birth of the cult of fine wine can be dated precisely : on 10 April 1663, Samuel Pepys, diarist in London, noted that he had enjoyed "a sort of French wine called the Bryan that hath a good and most particular taste that I never met with." He drank what is now called Château Haut Brion at the Royal Oak Tavern in the heart of London, one of many such places that spread after the return from exile of King Charles II three years earlier and which served tea, coffee and fine wines. Not only Château Haut Brion and the other great wines of Bordeaux were introduced, but also port from the Douro Valley in Portugal, the sparkling wines of Champagne and the brandy from Cognac, a small town north of Bordeaux (*The Economist*, 2009 d).

During the late 17th century, London was starting to replace Amsterdam as the hub of world trade. Its merchants were becoming more powerful, wealthier, and could appreciate luxury items, such as the claret, and by the 18th century Londoners were the biggest consumers of good claret in the world. By contrast to earlier periods when drinks became famous

and popular due to their connection with the royal family or the king's tastes (e.g. King Louis XIV used to drink Burgundy and the still wines of Champagne, and he was the arbiter of most alcoholic beverage taste), in England a wider social group including both commoners such as Samuel Pepys, and aristocrats, set the tone (*The Economist*, 2009 d).

The English had been drinking claret for five centuries before S. Pepys' time : it was of poor quality, shipped immediately after the harvest and consumed quickly before it turned to vinegar the following spring. The owners of Ho Bryan were the Pontacks, the top winemaking family of their time; they founded a fashionable restaurant, called Pontack's Head, in London, in 1663. John Locke, the philosopher, spotted the reasons for the superiority of the Bryan on a visit to the vineyard in 1667. J. Locke has captured the essential concept of terroir, the combination of soil, bedrock, drainage and microclimate which provide the conditions for the production of fine wine. Another connoisseur, the 18th-century economist Adam Smith, mentioned that "the vine is more affected by the difference of soils than any other fruit tree. For some it derives a flavour which no culture or management can equal" (*The Economist*, 2009 d).

The craft of claret-making had improved. By the early 18th century, the wine was designed to be kept for years not months, notably by being carefully stored in oak casks. Also better corks allowed wine to be stored longer and more safely. Bottles were produced that could be laid down on their sides to mature. By Adam Smith's time, the shape of the claret industry was established for centuries to come. Advertisements in the *London Gazette* at the time noted the sale of wines from four châteaux : Haut Brion, today a green area in Bordeaux's suburbs, and Latour Lafite and Margaux, all on the gravel banks above the Gironde estuary in the Medoc, the peninsula north of Bordeaux. The four estates remain the greatest brands in wine, and their main competitors, then as now, are a few tiny vineyards in Burgundy (*The Economist*, 2009 d).

British consumers of French wines had difficulties of supply. Britain, Portugal and their allies were at war with France and Spain. Port (from Portugal) was therefore considered the patriotic drink. In the 18th century drinking claret helped the rich to distinguish themselves from England's port-consuming aristocracy. Port was not only the more traditional drink, but also – because it attracted much lower duties – far cheaper. When Britain made peace with France in 1713, claret became more accessible and the wine trade boomed. Claret was not cheap, but rich Londoners, who were also by then big spenders on theatres, spas and music produced by fashionable immigrants, such as Friedrich Handel, consumed great quantities. Sir Robert Walpole,

Britain's first prime minister, used navy ships to smuggle his favourite wines from France. The most expensive one he bought was old burgundy, but that – as nowadays – was available in small quantities. Consequently, he relied largely on claret, and in a single year his wine bill amounted to over £1,200 (£100,000 today). By the time of the French Revolution (1789), the British were paying five times as much for their claret as the wine other main customers, the notoriously parsimonious Dutch, who preferred the cheaper, lower-grade beverage (*The Economist*, 2009 d).

The wines were no longer drunk, or even bought, when young : in 1714 R. Walpole was buying bottles of the 1706 vintage of the classier wines. Claret was still largely for the rich well into the 19th century. In *Every Man His Own Bullet*, published in 1839, Cyrus Redding, a wine merchant and author, wrote "claret for a bishop, port for a rector, currant for a curate and gin for the clerk." Three of the fine-wine merchant's contemporaries survived today : Corney & Barrow in the City of London, Justerini & Brooks and Berry Bros and Rudd in St James's Street (*The Economist*, 2009 d).

A free-trade treaty between Britain and France in 1860 drastically reduced the duty on French wines, thus encouraging the British middle classes to mimick their social superiors; and in that year the Chancellor of the Exchequer, William Ewart Gladstone, cut the duty on table wines to 40% of that on more intoxicating fortified wines such as port and sherry. The following year was promulgated the Single Bottle Act, allowing grocers to sell wine by the bottle. A very popular drink called "grocers' claret" was born, with the result that, between 1859 and 1878, sales of French wines, largely from Bordeaux, rose sixfold to 36 million bottles. The Gilbey family, one of the most remarkable commercial dynasties of Victorian England, franchised 2,000 grocers licensed to sell wine, largely claret. Their business grew so fast that by 1875 they were able to buy Château Loudenne in the Médoc to hold their enormous stocks of claret. As the middle classes turned to claret, so the upper classes abandoned it in favour of champagne (*The Economist*, 2009 d).

Thereafter, the fortunes of the claret business changed. In the late 1870s and 1880s, an attack of mildew ruined the reputation of wines : this was the case of Lafite, for instance, where the 1884 vintage turned mouldy after only a couple of years in bottle. At the same time, the phylloxera pest began to devastate Bordeaux's vineyards (*The Economist*, 2009 d).

In 1960, claret became again famous; the 1959 excellent vintage coincided with the arrival of big American buyers, and since then the wine popularity has been rising. London remains at the centre of the fine-

wine business – home of organizations such as the Institute of Masters of Wine, of *Decanter* and *World Of Fine Wine* magazines, and of most of the world's biggest wine auctions. Liv-ex, the world's first stockmarket of fine wine, is based in London; and its figures show that nine-tenths of the wine trade is still in leading clarets. Newcomers from vineyards in a dozen countries trying to launch their finest wines on the world market come to London first for validation. However, as consumers the British are not the top ones. In 2009, 57% of the fine wine that Sotheby's sold globally, by value, was bought by Asians, and four-fifths of those buyers were from China and Hong Kong (*The Economist*, 2009 d).

Global wine market

In 2005, global wine production has been estimated at between 274.6 million and 282 million hectoliters, while the global consumption reached 230.3 million to 240.9 million hectoliters. The estimates of production and consumption were the following with regard to : France (50.5 and 32.6 million hectoliters), Italy (50.6 and 27.6 million hectoliters), Spain (35.3 and 13.9 million hectoliters), United States (23.5 and 25.4 million hectoliters), Argentina (15.2 and 10.9 million hectoliters), Australia (14.0 and 4.5 million hectoliters), China (11.7 and 11.7 million hectoliters), Germany (9.1 and 19.6 million hectoliters), South Africa (8.3 and 3.4 million hectoliters), Chile (7.9 and 2.6 million hectoliters), Portugal (6.6 and 4.7 million hectoliters), Russia (5.1 and 9.0 million hectoliters), Romania (3.8 and 5.9 million hectoliters), Moldavia (3.0 million hectoliters), and New Zealand (1.0 and 0.8 million hectoliters) [Clavreul and Galinier, 2006].

In 2009, the annual turnover of the wine industry has been estimated at US$7 billion (€5.7 billion) in the world. Asia made up only 4% of world consumption. However, between 2004 and 2008, wine consumption in China has increased by 80% up to about 900 million bottles in 2008, of which 88% was red wine. Another study forecast a 31.6% increase in consumption between 2009 and 2013. Consumption of imported wines has been multiplied by four (+308%) between 2004 and 2008. France is the main supplier of China, and imported wines represented 12% of volume in 2008, but 40% of value (Changy, 2010).

Global wine exports grew from 55 million hectoliters in 1995 up to 78.7 million hectoliters in 2005, with a low figure in 2000 (60.4 million hectoliters), and then an upward trend. Still in 2005, Italy was the world's biggest exporter (15.8 million hectoliters), followed by Spain (14.1), France (13.9), Australia (7.0), Chile (4.2), United States (3.5), Germany (3.0), Portugal (2.8), South Africa (2.8), Moldavia (2.4),

Argentina (2.1), PECOs – Bulgaria, Hungary and Romania (2.1), New Zealand (0.6) and the Maghreb (0.5 million hectoliters) [Clavreul and Galinier, 2006].

Over the period 1986-1990, the first five European exporting countries made up 79% of the global market, compared with 3% for the New World countries, 9% for the PECOs (Bulgaria, Hungary and Romania) and 9% for other countries. Over the period 2001-2005, the average estimates were 65% of the global market for the top five European countries, 23% for the New World countries, 3% for PECOs and 9% for others. The top wine-exporting European countries are : France, Germany, Italy, Portugal and Spain; the New World countries include those of the Southern Hemisphere (Argentina, Chile, Australia, New Zealand, and South Africa) and the United States (Clavreul and Galinier, 2006).

The areas devoted to grapevine cultivation were the following :
- Spain, 1,180,000 hectares in 2005, compared with 1,196,000 hectares in 1995;
- France, 890,000 hectares in 2005, compared with 927,000 hectares in 1995;
- Italy, 847,000 hectares in 2005, compared with 927,000 hectares in 1995;
- China, 487,000 hectares in 2005, compared with 170,000 hectares in 1995;
- United States, 399,000 hectares in 2005, compared with 340,000 hectares in 1995;
- Chile, 191,000 hectares in 2005, compared with 122,000 hectares in 1995;
- Australia, 167,000 hectares in 2005, compared with 73,000 hectares in 1995;
- South Africa, 133,000 hectares in 2005, compared with 103,000 hectares in 1995 (Clavreul and Galinier, 2006).

Wine consumption per capita per annum was in 2005 as follows : France (55.4 liters), Italy (51.1), Spain (33.6), Argentina (32.1), Germany (24.4), Australia (21.3), United Kingdom (17.9), Chile (16.1), United States (8.1), South Africa (7.7) and Russia (6.1 liters) [Clavreul and Galinier, 2006].

The United States is set to overtake France as the world's largest wine market by 2012, according to a study commissioned by the organizers of the VinExpo trade fair in Bordeaux. The study forecast that global wine consumption over a five-year period would grow 5%, but the market value would increase 9% to US$117 billion from US$107 billion in 2005. The study predicted that US consumption of still wine (that is, not sparkling wine), would rise to 27.3 million hectoliters, or 721.2 million gallons, in 2010 from 23 million in 2005. That would exceed French consumption, which is predicted to fall to 24.9 million hectoliters from 27.4 million.

In terms of value, the still-wine market in the United States was expected to be worth US$22.8 million in 2010, up from US$19.2 million in 2005, according to the study, with fastest growth rates expected for bottles costing more than US$5 each – a trend also expected in other industrialized countries ("the world is drinking more and better, more expensive wines," stated Robert Beynat, the VinExpo secretary general).

Italy would remain the second-largest market in terms of volume with consumption in 2010 of around 27.2 million hectoliters, according to the study by the International Wine and Spirit Record, a London-based consultancy. In all, the global market for still wines with an alcohol content of less than 15% by volume was seen growing to 224.8 million hectoliters in 2010 from 211.9 million hectoliters in 2005.

Asia's wine consumption was expected to rise more than 21% from 2005 to 2010, with India and Taiwan leading the gains. China is Asia's biggest market and would grow about 36% in that period to 5.7 million hectoliters a year. China and Russia are emerging as growth areas for top-end, luxury French wines.

The VinExpo study also forecast that the global spirits market would be worth US$180.7 billion in 2010 compared with US$170 billion in 2005. Tequila, cognac and rum were set to replace vodka as the fastest growing spirit.

France seems to lose its dominant position to the winemakers of the United States and the Southern Hemisphere. Historically France has been the first global power in wine production, and it does conserve important assets : according to the figures published on 20 April 2006 by the International Grapevine and Wine Organization, it was at that time the world's biggest producer, on a par with Italy. It also remained the biggest consumer of wine, with about 33 million hectoliters per year. Its domestic market was on the downward trend over two decades, but the country has tried to compensate the loss of volume by gains in value, thanks to a quality-based policy that fostered the Certified Geographic Indications (AOC, or *appellations d'origine contrôlée*), i.e. 450 terroirs that produce 80% of French wine nowadays. In addition, the six grapevine varieties that are mostly cultivated and commercialized throughout the world are all from French provenance : merlot, cabernet sauvignon, syrah, pinot, sauvignon and chardonnay (Clavreul and Galinier, 2006).

France imports about 5 million hectoliters of wine annually (i.e. 15% of consumption approximately), mainly from Europe (85%), mostly table wine or cheap Spanish AOCs. According to a report on *Réussir l'avenir de la viticulture de France* ("Make successful the future of grapevine cultivation in France"), delivered to the French government in March 2006, "wines of the New World make up only 0.19% of wine sales by supermarkets, and their distribution through the networks of cellars and restaurants is insignificant." Among the New World countries that exported wines to France, Chile was in 2005 the most dynamic (250,000 hectoliters of wine had been imported), followed by the United States (110,000 hectoliters imported) [Clavreul and Galinier, 2006].

France's wine exports have dropped : their global share declined from 23% in 2001 to 18% in 2005, while global consumption has been growing over a decade by 1 million hectoliters per year. In 2005, 33% of wines were being consumed outside their country of production, compared with 18% in the early 1980s. While the exports of French wines have been dropping since 2003, those of Italy and Spain – France's historic rivals – are increasing. In 2005, and for the first time, Spain outpaced France as the world's biggest exporter, and France became the third-biggest exporter. With a vineyard area that has not changed, Spanish grapevine growers and traders have more than doubled their exports between 1995 and 2005. In addition, Spain has succeeded in protecting the image of wine as a cultural product, which has been cast in Spanish law in 2003 (Clavreul and Galinier, 2006).

Grapevine cultivation

Grape varieties

According to *The Wine Bible*, Karen MacNeil's massive compendium on wine, there are some 24,000 differently named vine varieties in the world. Of that number, only about 5,000 represent distinctly different grape varieties. Most of what is consumed, according to K. MacNeil's book, published in 2001, comes from 150 varieties, only nine of which are considered classic : chardonnay, chenin blanc, riesling, sauvignon blanc and sémillon among whites; cabernet sauvignon, merlot, pinot noir and syrah for reds. K. MacNeil indicated that varieties with the potential to cross over into the "classic" category must be planted in different places throughout the world and be increasingly demanded by consumers. Consequently, she singled out a number of varieties such as Spanish tempranillo and

garnacha (known as grenache in France, North America and Australia), Argentine malbec, nebbiolo, zinfandel and syrah (known as shiraz in Australia) [Hubbard Preston, 2005].

Malbec, tempranillo and garnacha are now widely planted varieties, although for centuries they have existed almost solely to be "blending grapes". Top-tier Argentine malbec, some of which sells for US$70 a bottle or more, is among the biggest imports in the American market. A centuries-old French variety used in making Bordeaux wines, malbec came into its own as a transplanted variety in the Mendoza region (pre-Andean area) of Argentina (Hubbard Preston, 2005).

Tempranillo, a long-respected blend made in Rioja, Spain, and port wines, is being produced for mainstream consumption by Spanish vintners, who, according to Paul Lukacs, author of *American Vintage*, published in 2000, "cannot be beat" for quality or value. Tempranillos are gaining momentum in the American import market. Garnacha, the favoured blending mate in rosé, is receiving positive critical attention. Robert Parker, the influential American wine critic, held a seminar on grenache in October 2005 at the Culinary Institute of America's West Coast campus in San Francisco. According to K. MacNeil, who directs the wine programme that recruited R. Parker, the US$500-a-ticket event sold out (Hubbard Preston, 2005).

Nebbiolo is an indigenous Italian grape variety that is now being grown in California, South America and Australia. Traditionally, it has been used for blending in barolos (in Piemonte). Australia has made the syrah grape one of its signature grape varieties and contributed to driving the grape into the mainstream consciousness. The annual case production of shiraz in Australia more than quintupled from 1998 to 2004, to 434,000 tons. Syrah is not only becoming more broadly available but also much more expensive. K. MacNeil stated that "syrah is moving into a level of familiarity with merlot", and noted that prices for French syrah continued to go up. "Even eight to ten years ago (1995), I could buy some of the best Côte Rôtie," the French appellation where syrah is grown, "for around US$50 to US$70. Not many of the great ones are now under US$200 and US$300" (Hubbard Preston, 2005).

Grape varieties with the largest increase in acreage planted between 1990 and 2002 were :

Red-wine grapes	Percentage change (1990-2002)	Hectares under cultivation (2002)
Syrah	257%	309,700
Pinot noir	103%	207,300
Cabernet sauvignon	95%	614,800
Tempranillo	84%	216,200
Merlot	56%	598,000
Malbec	50%	62,800
Cabernet franc	32%	129,200
Alicante bouschet	25%	60,700
Gamay	10%	94,900
White-wine grapes		
Welschriesling	192%	139,800
Chardonnay	140%	411,100
Moscato bianco	132%	101,900
Sauvignon blanc	61%	178,000
Isabella	48%	76,900
Feteasca alba	44%	65,200
Grüner Veltliner	18%	60,700
Malvasia blanca	16%	122,000
Macabeo	9%	117,500
Riesling	6%	136,900

Source: The Vineyard Handbook (Hubbard Preston, 2005).

There are dozens of unusual or lesser-known grape varieties that would come into vogue in the coming years. This has been the case with Chile's carmenere, originated from the carignan variety in France. It could be a passing fancy, as was the case with Sparkling Pink Catawba – a sparkling pink wine made from the catawba grapes grown on the slopes of the Miami River in Cincinnati in the 1850s. Poets from Henry Wadsworth Longfellow to Robert Browning sang the praises of the fizzy, sweet wine, putting it on a par with French champagne. According to Paul Lukacs, Sparkling Pink Catawba was "the first really successful commercially produced American wine."…"While nowadays wine connaisseurs "would

not like it", the wine "fit into the style and fashion of its day" (Hubbard Preston, 2005). Also wines made from the monastrell grape – a centuries-old variety native to Spain, best known as a blending grape – are still relatively obscure. However, Mark Pope, the creator of the Bounty Hunter Rare Wine Catalog in Napa, California, opted to feature a 2003 Antonio Candela Barahonda Monastrell-Yecla, which sold for US$13.95 a bottle. As the market for "classic" varieties is becoming saturated according to Liz Thach, co-author of *Wine : A Global Business*, one should be prepared to read and learn more about non-classic grape varieties, e.g. South African pinotage, Canadian vidal blanc, and even Japanese koshu and a Burmese version of German dornfelder (Hubbard Preston, 2005).

In Japan, major players including Château Mercian and Suntory have thrown their weight behind a few unique grape varieties. The most viable is koshu, which Gastin who travels throughout Asia regularly, has linked to a "more restrained" variety of sauvignon blanc. Also worth mentioning are: yamabudou, a mountain grape being grown for wine production in Japan, South Korea and China; and shokashi, a yambudou descendent, that produces rich flavours and colours naturally (Hubbard Preston, 2005).

Regarding rosé, the summer wine by excellence in France, also called the summer red, it is not a white wine mixed with red, as the European Union wanted to do in 2009 in order to reduce the wine glut in the European Union's main wine-producing countries. It is the real product of a fermentation process that is modified so as to obtain colours ranging from pale salmon to pink mixed with fuchsia or, sometimes orange yellow. According to Ribaut (2010), the origin of rosé is probably the old *vinum clarum*, produced in Gascogne, a French province owned by England from 1152 to 1453; the wine was exported to England till the end of the 18th century. Most of the wines made in Gascogne were red wines, called *bin vermeilh* or *bin clar*. The latter, named *vin cleret* in 1459, is the origin of the famous *claret* or *Bordeaux clairet*, highly appreciated by the English people. Jean-Bernard Marquette, an oenologist, has established that it was obtained after a shortened fermentation of red grapes juice. Nowadays, the clairet wines are produced by the Quinsac cooperative, in the region of Graves (Bordeaux area), and a few winemakers are striving to maintain this traditional production (Ribaut, 2010).

In Barsac, the Château Massereau has produced in 2007 some 5,000 bottles of clairet, that was the result of an assemblage of cabernet sauvignon and merlot grapes, the aromas of which were due to a maceration of grape skins, a fermentation in barrels, and thereafter a six-month aging, so as to avoid the excessive addition of sulfite. This process leads to a natural

wine, without addition of sugar (chaptalization) and acidification. It is an original wine, elegant and fruity, but also a wine that can be kept, which is drunk with grilled meat, spiced meals and desserts with red berries. In Bergerac, at Château du Jonc-Blanc, a couple of winemakers assemble cabernet sauvignon and merlot grapes to produce a wine called L'Heure osée, an appellation of Perigord wines 2007 (Ribaut, 2010).

DNA fingerprinting of grapevine varieties

Sine 1990, American and French researchers have been trying to establish the phylogenetic tree of the varieties of grapevine grown throughout the world, using the fingerprinting technique. The research consists of analyzing the structure of certain regions of the genome of some grapevine varieties and comparing it with that of other varieties, so as to establish possible phylogenetic relations. For this analysis, the DNA is extracted from young ground leaves, but also from fruits and branches. The results of this first research have been published in 1999 in the *Science* journal (Nau, 1999).

There are thousands of grapevine varieties cultivated worldwide. In order to establish the origin of a variety, specialists used to rely on phenotypic traits, such as the morphology of leaves, berries and grapes. In this way, varieties could be grouped in a few families. DNA fingerprinting enables the researchers to go further. It appears that the current grapevine varieties are the remote offspring of the grapevines grown during the Antiquity around the Mediterranean, or during the European Middle Ages. The current varieties are the result of lengthy breeding activities, identification and comparison work, and stabilization of the selected strains or lines (Nau, 1999).

The team led by C. Meredith and J. Bowers of the Department of Viticulture and Enology of the University of California, Davis, has confirmed that the cabernet sauvignon variety – which is dominant in the Médoc region of France and is at the origin of most red grapevine varieties in the New World – was in fact the offspring of the cabernet franc and sauvignon, two varieties deeply entrenched in the middle valley of the Loire river. The cooperation between the Californian team and the French specialists of the National Higher School of Agronomy, Montpellier, associated with the Genetic Research and Breeding - Viticulture Unit of the National Agricultural Research Institute (INRA), has led to undisputable results concerning the origins of various grapevine varieties : the chardonnay (the most famous and expressive variety in Burgundy), aligoté (also from Burgundy), gamay (red variety) and melon of Burgundy (which

has colonized the area of muscadet) are all cousins and almost siblings. Such a conclusion, derived from the analysis of DNA fingerprinting, was not a surprise for oenologists and tasters because the wines made from these four varieties share common structures and aromas. This kinship is particularly expressed in the ageing wines. Similarly, aged wines of the chenin variety resemble the Hungarian tokay (Bowers and Meredith, 1997; Bowers et al., 1999).

But more surprising than the discovery of that kinship among the four grapevine varieties, was the identification of one of the progenitors of the initial couple that gave rise to these varieties. Indeed, several historical elements were in favour of the creation of lines through the cross pollination between the pinot noir and the white gouais; these crosses have given birth – as proven by the fingerprinting analysis – to the three white and red varieties grown for a long time in various French provinces, and for some decades, in many regions of the globe. The surprising aspect of this discovery was that the white gouais is almost unknown, although the vine specialists in Montpellier continue to grow it and to make wine from it for their own pleasure. However, this variety has played a key role in the origin of French viticulture, according to R. Dion in his *Histoire de la vigne et du vin en France des origines au XIXe siècle* ("History of vine and wine in France from its origins to the 19th century"). A document dated from the 12th century mentioned this variety as a lower-grade one; in 1338, the white gouais was found in Metz under the name of goez; at that time, instructions were given to eliminate this variety from all the Metz territory and to privilege only the white and black fromental, considered as higher-grade varieties. The gouais was found in Paris during the 14th century and, owing to the expansion of the workers' population, it progressively replaced the pinot noir of Burgundy, which was a good variety of Parisian vineyards. The extension of the gouais was due to the wish of winemakers to produce a cheaper wine. However, the phenomenon was limited to Paris and its suburbs; in the vineyards located away from the capital, the gouais was rejected, more noble grapevine varieties were used and contributed to the reputation of French viticulture (Nau, 1999).

The white gouais was also formerly grown in the Jura and Franche-Comté. For the American and French researchers, this variety which has played a key role in the history of vine and wine, is the same as the heunisch variety of Central Europe, introduced in Gaul by a Roman emperor originating from Dalmatia. In Montpellier, the French researchers participating in the joint study with the American scientists from the University of California, Davis, have tried to reproduce the

breeding between the pinot noir and white gouais in order to seek confirmation of the genetic research. Other attempts were expected to widen even more the range of cultivated grapevine varieties, for both their fruits and wines derived from them. But this approach was hindered by a drastic regulation, which practically prohibits any venture of this kind, while non-French winemakers and vinegrowers could do it (Nau, 1999).

In Apulia, in the heel of the Italian boot, drawing on grapes grown by up to 1,600 small farmers in the area, a California wine consultant associated with another Italian wine consultant from Friuli (northeasten corner of the country) are producing and marketing wines that have scored a great success worldwide. The wines are called *A-Mano* – handmade – and by far the best known is a robust red made from a once-obscure grape named *primitivo* (Apple, 2004).

DNA testing by Carole Meredith at the University of California, Davis, established that *primitivo* is a descendant of a grape called *crljenak kastelanski*, widely known in the 18th and 19th centuries on the Dalmatian coast of Croatia (a *crljenak* cross with *dobrinic, plavac, mali*, is being grown on that area today). California's zinfandel, she showed, is genetically the same as *primitivo*, though how it crossed the ocean remains a subject of dispute (Apple, 2004).

Apulian *primitivo* and zin are not twins, of course; climate, soil and vinification all help to shape a wine's look, aroma and flavour, along with the grape variety. But the two share several characteristics : both are fruit-rich, chewy, sometimes lush wines, a deep violet-red in colour, often too high in alcoholic content for comfort, but much more subtle if carefully handled (Apple, 2004).

For years, *primitivo* was used to add unacknowledged heft to chianti, barbaresco and even burgundy. Nowadays, *primitivo* can stand on its own feet. In addition to *A-Mano primitivo*, other high-quality *primitivos* are grouped in an organization called the Academia dei Racemi, not a true cooperative but an association in which each member makes his own wine and joins the others for marketing support and technical advice. Based in Manduria, between the old cities of Taranto and Lecce, the group includes value-for-money labels like Masseria Pepe, Pervini and Felline (Apple, 2004).

Grapevine genomics

Among the forty species of grapevine existing nowadays, *Vitis vinifera* that originated in Eurasia, is the only one cultivated by humans in order to produce table and wine grapes. This species has been disseminated throughout the world during the neolithic period. *V. vinifera* is very sensitive to many fungal diseases and vineyards in Europe receive about 49% of the million tons of pesticides used annually in this region. One important line of research is therefore to select grape varieties that are more resistant to diseases and pathogens (Rached, 2007).

Sequencing the genome of the grapevine would enable geneticists to identify genetic markers related with plant physiology and wine quality. In 2005, the French-Italian Public Consortium for Grapevine Genome Characterization has been launched with the goal of developing new breeding tools to be used in a "more precise, more sustainable and higher-quality grapevine cultivation." The first results of the sequencing programme have been published on 26 August 2007 on the site of the journal *Nature*, and they were considered "a very good first draft" of the genome by Anne-Françoise Adam-Blondon of the National Agricultural Research Institute (INRA) which led the programme on the French side with Génoscope – the national sequencing laboratory (Jaillon et al., 2007).

On 18 December 2007, the first full genetic sequence of a grape variety (pinot noir) was published in the Public Library of Science. The Italian team was led by Riccardo Velasco of the Agricultural Institute of San Michele all'Adige, and they found a high degree of difference, 11.2% in all, between pinot noir two sets of chromosomes. Those sets come from the varieties originally crossbred to create the clone (grape varieties are normally propagated as cuttings; in other words, clones). These parental varieties must therefore have been very different from one another, because 11.2% is far more genetic variation than the one which exists between a chimpanzee and a human, for instance. R. Velasco and his team have also found hundreds of genes that encode enzymes which produce flavourings and aromatic compounds. That will help both those who want to make flavours more consistent and those who want to add new features. Anne-Françoise Adam-Blondon, on her side, stated that genes involved in the synthesis of resveratrol – often associated with the wine's benefits for health – have been identified; that "certain forgotten aromas might be rediscovered, but it was not easy to judge wine quality at the molecular level" (Pilati et al., 2007).

It should be recalled that wine qualities and character do not depend only on genetics, but also on a complex combination of soil conditions

and microclimate, i.e. the terroir, that gives *appellations contrôlées* or Certified Geographic Indications their peculiarity. Wine qualities are not just the result of nature (grapevine genetics), but also of nurture. However, some winemakers argue that where one grows his grapes matters, but not more than it does for any other crop. The true flavour is encoded in the genes and the consistency of a cloned crop species is an asset. Environmental diversity is something to be resisted, since it masks the character of the grape and makes it difficult to produce a consistent product. As in the debate over human character (nature versus culture), both sides have a point, but the naturist side receives support from structural and functional genomics, that will help clarify the role of many genes of grapevine varieties (*The Economist*, 2007 e).

Knowing the pinot noir genome should also help vintners who would like to grow grapes in places now off-limits to them – either for climatic reasons or because local diseases would kill them. Florida, for instance, produces a few grapes for the table, but these are muscadines, a different species from those usually used for wine. Its wine is one tenth of one percent of the American market. The reason is that the State is home to diseases that affect wine grapes : a bacterial infection called Pierce's disease, fungal infections such as mildews and rots, and viral diseases such as grape fanleaf virus and corky bark. Dennis Gray, a developmental biologist at the University of Florida, and his colleagues have been working for years on making vines resistant to these diseases by modifying their genes (Dutt et al., 2007). In 2007, he began field trials of grapes engineered to resist Pierce's disease and fungal infection (*The Economist*, 2007 e).

Similarly in France, at the Colmar centre (Haut-Rhin department) of the National Agricultural Research Institute (INRA), transgenic vines have been developed in order to make them resistant to the fanleaf virus, transmitted by soil nematodes and for which there is no natural resistance. After years of negotiation involving researchers, elected representatives, viticulturists, citizens, the director-general of INRA, Marion Guillou, was able to convince them about the interest to conduct field trials under very strict biosafety conditions. She was even able to make NGOs participate in a "follow-up committee." Indeed the threat to Alsatian vineyards was a major concern, as the only remedy to the viral disease is to uproot the diseased vines. The field trials aimed to assess the efficiency of the transgene, but also the eventual transfer of the gene to the rest of the vine (rootstocks were genetically engineered) and to the environment (Morin, 2009).

Unfortunately, despite the openness of INRA director-general and researchers, on 7 September 2009 70 transgenic vines were cut off by

an anti-GMO activist, who was sentenced on 19 November 2009 to a €2,000 fine by the court of Colmar. The sentence also included €1 for the moral prejudice caused to INRA. In fact, this destruction ruined several years of work carried out by the INRA researchers. The man sentenced in Colmar was known for his legal guerrilla against GMOs and he stated before the court that he decided to destroy the transgenic vines in order to "provoke a public debate on GMOs. "His action has received little support from those circles generally opposed to GMOs, and some, belonging to the French Green Party, even condemned it – one of them qualified the destruction as an "incommensurable human wastage." It should be recalled than on 18 November 2009, 58 antiGMO mowers had been trialed before the appeal court of Versailles, because they had destroyed a plot of transgenic maize of Monsanto in 2007 in the centre of France. They had declared during the first trial that they had acted "in a state of necessity," and they were relaxed; hence the appeal made by Monsanto (Morin, 2009).

The almost total destruction of the INRA trials, when these were not very far from delivering important results for the future of Alsatian viticulture, has shocked the researchers. Jean Masson, chair of the INRA-Colmar centre, stated : "it is our mission to carry out prospective research, and thereafter society would adopt or not the solutions we propose." Many researchers and executives voiced their concern and underlined that the destruction of greenhouse and field trials of transgenic crops will hinder French research in this area when other countries are making important progress; companies are also moving their trials outside France. For instance, the seed cooperative Limagrain closed two laboratories, in Cambridge and Evry (France). Since 2007, its trials are being carried out in Spain and Israel, and also in China and India. Limagrain was devoting only €10 million to transgenesis out of a research budget amounting to €140 million (Morin, 2009).

Regarding INRA, research on transgenic grapevine has been resumed in 2010, in addition to a field trial on transgenic poplars, carried out in the INRA centre located in Orléans with a view to producing bioenergy. INRA researchers hoped that the results expected from the grapevine trials could contribute to control the fanleaf virus disease, which seems on the rise in several French vineyards.

Most unfortunately and for the second time, on Sunday 15 August 2010, a group of about 60 activists invaded the INRA Colmar centre, uprooted the transgenic grapevine rootstocks that had regrown and cut them into pieces. These activists came from all over the country and operated before dawn, after having warned the press about their action. The INRA

researchers had obtained a four-year authorization in order to carry out their project, which had also received the support from the French High Biotechnology Committee. While the destruction of the transgenic grapevines was due to one single person in September 2009, that of August 2010 was the result of a collective action, at a time when there were no field trials of transgenic maize in France – the cereal species has been for several years the target of antiGMO activists (Morin, 2009).

The activists, who were brought to a police station and then released on the same day, stated they had "neutralized" the experimental plot, "because field trials of GM crops were the first stage of a commercial approach aiming at imposing, with the support of the European Commission, crops which are not currently authorized and which are not, above all, accepted neither by the people, nor by the professionals". The activists requested the French government "to devote public funds to finance research on other alternatives to control the fanleaf-virus disease than transgenic grapevines" (Morin, 2010).

INRA researchers and executives were appalled by this new destruction of their work and they stated that, by doing so, the activists "contribute, by attacking a public-research work, to expand fear about environmental risks which do not exist in this trial, while INRA is assessing, in complete independence, the relevance and possible risks of this kind of technology for the control of fanleaf-virus disease". The French ministers of ecology, research and agriculture have, in a joint declaration, strongly condemned the destruction of INRA's experimental plot, and said "they were shocked by this scandalous degradation action against a trial which had the merit to bring together scientists, professional agricultural organizations, local collectivities and environmental non-governmental organizations". The implementation of the field trial included a follow-up committee in charge of determining the precautions needed for the confinement of the trial, as well as of finding out alternatives to transgenesis in controlling the disease (Morin, 2010).

The director of the Association of Alsatian Viticulturists expressed his dismay regarding the repeated destruction of INRA's trial; he recalled that the trial had an objective of research and not of production and the "co-design" of the study had been "fully satisfactory" – INRA having made progress in parallel on alternative methods of controlling the virus (Morin, 2010).

However, many people hope that the diseases in grapevines may be overcome without genetic engineering that risks provoking consumer boycotts. An alternative to transferring genes from one line to another, is to create new varieties by cross pollination and then use genetic-sequencing

techniques to find out which offspring of such crosses have desirable combinations of genes in them. D. Gray is sceptical about this approach : when wild species of grapevine with good disease resistance are bred with cultivated varieties possessing great qualities of flavour, the result is very often a wine of lesser quality that has only some of the desired disease resistance. He thinks preferable to pick the genes precisely and engineer them into the target. Moreover, since the gene-products in question rarely turn up in the flesh of the grape (they are actually expressed in stems, roots, leaves and seeds), the wine from a genetically modified grapevine should not contain them (*The Economist*, 2007 e).

It remains to be seen if this genetic-engineering approach would be enough to convince European consumers, who are attached to the terroir, particularly as the benefits of disease resistance are reaped largely by producers. But research on genetically modified table and wine grapevines is going on in France, Italy, but also in Chile and the United States, spurred by the progress expected from genomics. Thus by comparing the genomes of cultivated grapevine with those of wild species, researchers can identify the gene sequences involved in disease or pathogen resistance; these sequences could then be transferred or used in advanced crop breeding programmes (Rached, 2007; *The Economist*, 2007 e).

Adaptation to climate change

At a symposium organized in Dijon by the University of Bourgogne from 28 to 30 March 2007, on "Climate warming, which likely impacts on vineyards?" (*Réchauffement climatique, quels impacts probables sur les vignobles?*), it was recalled by Emmanuel Le Roy Ladurie – a French historian of climate vagaries – that wine production has always adjusted to weather changes. However, the year 2003, according to the historian, has been exceptional in this regard. "The only year that could be considered comparable was 1523, when the grape harvest had started on 27 August in Burgundy". In 2003, it started on 19 August (Morin, 2007).

It is acknowledged that grape phenology, i.e. the different stages of the grapevine cycle from the appearance of buds to grape maturation, has been modified everywhere, starting in the early 1980s. Gregory Jones, a geographer from Southern Oregon University, has noted that these phenological stages occurred increasingly earlier and that the whole cycle was shortened by six to 17 days (average) over the period 1950-2000. He analyzed the data from 27 wine-producing regions throughout the world and found that temperature had risen by 1.3°C during the vegetative growth period. In the Napa Valley, California, wine rating improved at

the same time as sugar content increased. However, 75% of the whole production must nowadays be dealcoholized because the percentage of ethanol reaches 16% before this operation. G. Jones has estimated that since 1950 the geographical belt where grapevine can be grown (average temperature between 10°C and 20°C), has been displaced towards the poles by 80 km to 240 km. This also means that vineyards located in higher areas, where temperature is lower, became more promising (Morin, 2007).

Furthermore, G. Jones forecast, on the basis of the "pessimistic scenarios designed by the Intergovernmental Group on Climate Change (IGCC), that the displacement of the grapevine cultivation belt could reach another 280 km to 500 km by 2099. In this case, changes would have to be made among the vine varieties grown in the existing vineyards, some of which would have to be displaced because of the drought threat. This trend can be seen already in South Africa, where apple orchards located near the coasts are being replaced by vineyards (Morin, 2007).

In South Africa, in the region of Cape Town, average temperatures have risen 1°C in 25 years. According to Victoria Carey, a researcher at the department of vine cultivation and oenology of Stellenbosch University, "this phenomenon is being accelerated since 2000," and climate warming has already produced its first impacts, "as the new plantations of grapevine are displaced towards cooler areas, higher valleys or the coastal zone." The shortening of the winter period delays the fall of leaves and threatens the development of buds. The strong warming observed during the month of February, just before the harvest of grapes, accelerates the concentration of sugar in the grapes and modifies aromas. Alcohol content rises rapidly in wines which already contain 15% of ethanol (red wine). Victoria Carey underlined that the rise in temperatures is only one aspect of the environmental change. "Periodicity of rainfall is going to change, with heavy rains occurring at periods more distant from each other, and lower penetration of water into soils." Stellenbosch University is participating in workshops aimed at warning vine growers about climate change and at training them in the ways and means to mitigate the change while reducing CO_2 emissions (Allix, 2008; Carey et al., 2008).

The rise in average temperatures does not only mean the change in rainfall pattern or drought, but also the multiplication of new insect pests (such as eudemis in Champagne, France) or the spread of diseases like mildew and oidium. It also means more soil erosion due to heavy rains and storms. Consequently, Marie-Claude Pichery, an economist of the

University of Bourgogne, not to look for new areas for vineyards "would be an error for a wine producer or merchant" (Morin, 2007).

For the time being, winemakers are generally happy about the rise of temperatures, which has increased yields and improved wine quality. However, in the French southern vineyards, there is a fear about recurrent droughts. The National Institute of Origin and Quality has supported a decree on the regulation of irrigation of vineyards with Certified Geographic Indications (appellations contrôlées). The year 2003 that may be repeated during the second half of the 21st century, is considered a warning notice for winemakers. Indeed, many experts agree that early grape harvests, associated with a significant change in grape maturity and alcoholic content, could have an impact in the short term on the ageing of wines and their "typicity", i.e. their unique qualities (Morin, 2007).

In England, winemaking is benefiting from climate change, particularly the rise in temperatures, as well as from the progress in grapevine cultivation. A winemaker in Kent, south-east of England, stated in 2007 that they had almost the same weather as in Champagne 25 years back. And this situation has triggered the production of sparkling wines, which English consumers like very much. The best varieties are Denbies (Surrey), Camel Valley (Cornwall), Chapel Down, Ridgeview and Nyetimber (Sussex). While English wine represented only 1% of the British market during the late 2000s, its future seemed promising, as demonstrated by the interest shown by old French champagne-producing corporations in acquiring land in England for growing grapevine that is 100 times less expensive than in France (Langellier, 2007).

It is true that grapevine cultivation is expanding, mainly on the chalky and sunny hills of south-eastern counties – Kent, Essex, Surrey and Sussex. By the late 2000s, there were more than 400 independent wine producers, offering good-quality wines: pinot noir, pinot blanc or chardonnay. Despite the rapid expansion of vineyards, supply cannot meet demand, which is really booming. Initially to become a winemaker in England was an opportunity for the farmers, whose income from cereals and dairy products was stagnating, to diversify their resources. Heavy investments were compensated by the lesser cost of marketing networks. Many good-quality wines are directly sold through supermarkets (Langellier, 2007).

Vineyard extension and protection of natural ecosystems : the example of South Africa

Adaptation to, and mitigation of, climate change is associated in South Africa with the Biodiversity and Wine Initiative (BWI), in which 135 wine producers are participating, in collaboration with the World Wildlife Fund (WWF) and organizations of botanical conservation. The key objective of the initiative is to work with vine growers in order to make sure that they plant grapevines where biological diversity is less in danger, according to Inge Kotzé, the coordinator of BWI. The goal of this action is to protect areas (samples) of all existing types of vegetation and to create corridors which link them to national parks, so as to enhance migrations and pollination. It is true that under the Mediterranean climate of Western Cape more than 10,000 plant species are living and 70% of these plants are endemic. But in the valleys surrounded by ridges, the spreading of vineyards and urbanization have spared only 4% of the natural vegetation type, the *fynbos*, which resembles the Californian *chaparral* or the French *maquis*, composed of heather, proteas and succulent species. About 80% of the land belong to private owners and it is therefore of crucial importance to draw their attention on the need to protect plant diversity when they develop their vineyards (Allix, 2008).

Those who participate in the Biodiversity and Wine Initiative (BWI) make the commitment to conserve or to rehabilitate at least 2 hectares of natural ecosystems. The "leaders" – about 13 vine growers by the end of 2008 – have increased this area up to 10% of their land. In September 2008, 110,000 hectares had been put aside as protected areas, and in the Cape vine-growing area the acreage of protected natural ecosystems was equal to that of vineyards. While only 25% of wine producers participated in the programme of land conservation, 95% of them were applying the principles of BWI in order to mitigate the impact of grapevine cultivation on natural ecosystems, e.g. cleaning of water streams, bush fires management that help regenerate the *fynbos*, waste recycling, and limitation of use and dumping of chemicals. Consequently, after decades of excessive consumption of resources and massive spraying of biocides, an increasing number of vine growers are trying to adopt organic agriculture as well as renewable sources of energy. As a result, the professional meetings Cape Wine, that took place at the beginning of October 2008, evolved into a forum on "rational" agriculture (Allix, 2008).

For instance, Jonathan Grieve, the owner of Avondale vineyards, which can be reached after one-hour drive from Cape Town, considered that conventional agricultural practices should be modified. He decided

not to use any pesticide, herbicide or fungicide on its 300 hectares of vineyards, and he was of the opinion that soil microbial life must be restored instead of adding chemicals to the soil. It is better to rely on a great diversity of insects, plants and microorganisms, between the rows of vines, in order to restore soil fertility. There are many similar initiatives. At Spier vineyards, a nursery has been created in order to propagate plants of the *fynbos*, which are thereafter planted on 460 hectares; a waste-water treatment unit is also part of the domain and recycles all waste waters through phytoremediation. Another champion of the BWI, the Backsberg vineyards, has become in 2007 the first South African wine producer certified as "carbon neutral". Energy consumption and CO_2 emissions have been reduced drastically in the following sectors : lighting of the cellars, refrigeration of the fermentation vats, transport through the 230 hectares of farmland, and one thousand trees have been planted (CO_2 sequestration) [Allix, 2008].

Economic benefits are expected from these initiatives. The BWI logo is considered "a marketing tool that confers a competitive advantage to South Africa on the wine world market." The head of the department of vine cultivation and oenology of Stellenbosch University, located in the heart of the vineyards region, stated that the wine industry had become aware of the problem of soil fatigue, and that, beyond marketing objectives, the whole production system should be reviewed in order to adopt an integrated approach to an environment-friendly and "carbon neutral" grapevine cultivation and wine production in South Africa. This country has become in 2008 the world's eight-biggest wine producer, and its exports have been multiplied by 14 in 15 years : 360 million liters have been exported in 2008, i.e. one-third of the whole production. And its first five customers are European countries that are very keen on environment protection and nature conservation. Therefore, South Africa's Biodiversity and Wine Initiative (BWI) conveys a good image to the European customers (Allix, 2008).

Winemaking : from art to science and technology

In 2005, plummeting consumption of wine in Europe has led to a worldwide glut of 6.5 billion bottles a year. France was worst hit, but this country still led the world, with Italy and Spain, in making and selling wine. However, New World winemakers – Australia, New Zealand, South Africa, Argentina, Chile and California – have seen their share of global exports surge. Much of that growth is due to changing tastes, which favour the flavours of New World winemaking techniques. While in much of Europe, especially in France, heavy regulations and tradition prevent

the use of several newer techniques, e.g. roto-fermentation to soften tannins and quicken colour extraction, reverse osmosis to remove water, drip irrigation, mixing grape varieties, which New World winemakers have adopted. The openness to experiment and the commitment to produce wine adapted to the palate are in a way a revolution that has given a competitive edge to the New World winemakers. "We do not let wine judges or the producer define what wine should be; we make wine according to what the customer wants," stated Sakkie Pretorious, managing director of the Australian Wine Research Institute in Adelaide, South Australia (Margolis, 2005).

To that end, New World grape farmers and vintners are employing innovation to gain ground against traditional European producers, at every stage of the process : in the vineyards, while old rules ban watering, innovation includes surveillance, irrigation and genetic engineering with a view to boosting yields; infrared scans determine the best time to harvest; roto-fermentation, reverse osmosis, use of custom yeasts, tweak tastes with flavour chips; during storage, instead of long ageing in big wooden casks, a year in the barrel may be all the time wine needs; in the bottle, corks risk damaging flavours in the old way, but Australia has popularized the screw-tap bottle; computerized vineyard models. With as many as five different soil types found in a single vineyard, the South Africans have excelled in soil analysis, while Australians have mastered drip irrigation, which delivers precision jets of water to some of the world's most arid vineyards. A Chilean company, called Ingenium, using remote-sensing technology, developed in the United States, has designed an infrared ray gun that scans grapes for sugar and acidity without puncturing the fruit – data that ordinarily would take weeks to gather back in the laboratory (Margolis, 2005).

Nowadays, vintners can make the wine they want through manipulating numerous strains of yeast and acting on tiny doses of the nearly 800 key chemicals and compounds that exist in grapes, e.g. bringing on the isobutyl methoxypyrazines in shiraz wines, which are spice-bearing compounds that can be coaxed out of hibernation by reducing exposure of grapes to sunlight. At the Centro de Aromas, a small laboratory at the University of Santiago, Chile, researchers inject small samples of wine into a gas chromatograph, which breaks down and returns a digital footprint of the key chemical compounds of varying aromas. Eduardo Agosin, an agronomist and biochemist, predicted that once this digital library would be ready, winemakers would be able to develop grape varieties according to the aromas customers wanted (Margolis, 2005).

Pablo Morande, a production manager at Ventisquero Wines in Chile's Central Valley, is using aerial infrared images of vineyards generated by Ingenium to micromanage harvesting of Ventisquero's vineyards, down to the smallest plot, in order to harvest uniformly ripe grapes. P. Morande stated : "We harvest grapes within the same plot at different times, which makes an astounding difference in quality." Aridity is a major challenge to New World winemakers, and water management has become a key area of work and improvement. At the Graham Beck estate, near Stellenbosch, in south-western South Africa, viticulturists measure moisture and salinization levels in the soils with "neutron probes" in order to decide when to irrigate their vines and with how much water (Margolis, 2005).

Most New World wineries were also among the first to use techniques like reverse osmosis – which filters out excess water from the grape juice to concentrate flavour and ethanol – and micro-oxygenation, which blows tiny oxygen bubbles into the fermenting wine to soften tannins, enrich colours and round out the texture of wine on the palate (Margolis, 2005).

Traditional wine lovers do not appreciate all these technical tools. "We have used technology to destroy the formidable concept (good wine comes from God) that was a source of European heritage", states Nicolas Joly, an "organic" winemaker whose family produces fine chenin blanc in the Loire Valley on vineyards dating back to the 12th century. But New World viticulturists "use these inventions better than us," stated Michel Rolland, perhaps the world's most influential wine consultant, who boasted 100 clients on four continents. "They do not question themselves too much. They do not have heavy regulation. They simply make modern wine. Sometimes it is good, sometimes not. But sometimes it is very good" (Margolis, 2005).

A striking example of the quarrel between traditional winemaking and the reliance on unusual technologies in wine ageing is that of the use of wood chips. On 15 September 2005, an agreement was signed between the European Union and the United States that authorized the sales in Europe of wines made according to peculiar oenological practices in the New World; as a counterpart of this authorization, 17 names or brands were to be used in a more restrictive way in the United States, such as chablis, chianti, porto. In particular, sales were authorized in Europe of wines that have contained oak chips in order to give them a wood flavour. Later on, the European Commission authorized this process in the European Union's member states, while at the same time each

one of them was free to apply specific rules or modalities regarding the labelling, mandatory or not, as well as the partial prohibition of the technique (Ribaut, 2006).

In France, the Supreme Court (Cour de cassation) had stated on 6 February 2001 that the ageing of Bordeaux wine (with Certified Geographic Indication – AOC) in the presence of oak chips in the vats was a "falsification". Since then, there has been a debate over whether wine ageing must be done in new barrels of oak, or whether oak chips could be incorporated in wine; in other words, follow an old tradition or a new global fashion, a symbol of the new oenology (Ribaut, 2006).

The Spaniards seem to be the most reluctant to the generalized use of chips, while Italians are more willing to accept it. Bordeaux wine producers (AOC) are also favourable to the use of wood chips, one the arguments being the economic one : this use costs ten times less than a new barrel of oak. Oak chips can bring to the ageing wine wood and vanilla flavours, but not the natural micro-oxygenation which is associated with the porosity of wood. Oak chips act rapidly on wines to be consumed young and they are not useful for keeping wines over long periods. There is therefore a confrontation between winemaking techniques of the New World and the Old World. According to reliable figures, less than 10% of French wines and a maximum 3% of wines produced globally are matured in barrels (Ribaut, 2006).

Ribaut (2006) is of the opinion that viticulture would show two facets in the future : one will continue to produce wines that can be kept for long periods in oak barrels and whose marketing will be based primarily on their fame; another facet will be that of vintners who will make wines to be consumed rapidly, with a more or less marked wood flavour, but which have not benefited from natural oxygenation (those wines will be more in tune with the recipes of the agrifood industry). The use of wood chips, as a source of aromas, will accompany reverse osmosis or evaporation under vacuum in order to concentrate grape juices. Wines may even be mixed with some water, as it was specified in the United States-European Union agreement. Powdered tannins, Arabic gum and metatartrates are used along with ageing in oak barrels, or they can even replace it in order to obtain "toasted" or vanillin flavours. The use of gases is also frequent : nitrogen to blow out carbon monoxide (CO) from red wines, micro-oxygenation to speed up red wine ageing, carbon dioxide to give more body to white wines (Ribaut, 2006).

Global industry and international competition

France

France is the superpower of the wine world, the largest producer and, measured by per capita consumption, the biggest drinker. For more than a decade, globalization has transformed the business, bringing with it new markets, consumers and competitors. Producers from Australia, New Zealand, California, South Africa, Chile and Argentina have launched massive campaigns to promote their wines throughout the globe. They have risen production, introduced a new generation of consumers to inexpensive, fruity wines with labels easy to understand, and, in the process, created a big business. By the mid-1990s, France exported three times as much wine as all the so-called New World producers put together; by the mid-2000s, it sold about 15% less than they did. Europe imported almost as much wine as it exported, something that would have been unthinkable a decade ago. In the United Kingdom, for instance, one of the biggest and most competitive markets, Australia has become the undisputed market leader in a few years (Gumbel, 2006).

The French barely reacted to these changes, largely because global wine consumption has been growing, up about 10% in the decade 1995-2005 to 240 million hectoliters (hL). But overproduction occurred and in 2004 worldwide production hit its highest level in 20 years, almost 300 million hL, or 15% more than the previous year (2003). The glut hit producers everywhere, particularly in Australia : according to estimates by the Australian Wine and Brandy Corporation (AWBC), the government body that oversees the wine industry, the country had, in 2006, surplus wine stocks that exceeded a whole year of exports, and many grape growers simply left the 2006 crop on the vine rather than harvest it (Gumbel, 2006).

In France, a massive support system subsidizes producers who cannot sell their wine and therefore mitigates the impact of overproduction. Even though, revenues and incomes overall have been falling since 2002 – the first decline in decades – and the €11-billion French wine industry was in trouble in 2004, not the premium vintners of Bordeaux and Burgundy, but the lesser names that made up 80% of national production. The 2004 harvest was good and abundant, but selling it was difficult, at a time when many of the country's second-tier winemakers were struggling to survive (Pape and Mcnicoll, 2004; Gumbel, 2006).

On 3 November 2004, *Mondovino* – an exceptional documentary film on the world of wine – could be seen in the French movie houses. J.P. Géné (2004) of the French newspaper *Le Monde* seized that opportunity to enquire on the crisis in the French vineyards. *Mondovino* highlighted the opposition between a traditional approach to winemaking and a more modern and commercial one. This opposition is at the heart of the crisis striking the French vineyards. The latter was a serious one : in Bordeaux, hundreds of viticulturists, some on the border of bankruptcy, took to the streets in 2004; exports fell by 9% in value and 3.5% in volume during the first five months of 2004. In 2003, for the first time, wine exports from the New World (Australia, New Zealand, Chile, South Africa) overtook those of France. On the American market, French wines, at the top of sales earlier on, represented only 14%, compared with 26% for Australian wines which became the leaders. In the United Kingdom, a traditional customer of Bordeaux wines, French exports also plummeted : -27% in value; and the fall would have been even greater without the positive impact of champagne sales (Géné, 2004).

Some experts have tried to explain the crisis, like René Renou, a viticulturist at Bonnezeaux (Maine-et-Loire), member of the National Institute of Origin Appellations (*Institut national des appellations d'origine*, INAO) since 1984 and chair of INAO's National Committee of Wines and Spirits. He stated : "Until 1985 French vineyards were in a monopolistic situation with respect to top and middle-ranking wines. Italy was a leader for the cheaper wines, while Spain and Portugal almost did not exist. Supply was always lower than world demand and there was only a single code for reading and understanding wine : the French code – terroir, AOC, labelling, etc. When one Australian or American vintner wanted to grow grapevine and produce wine, he would buy a book on French wine and this was his unique reference." He added : "When you are in such a situation, you can do everything : produce more, in the way you like, increase prices, do a sloppy work, as everything is sold" (Géné, 2004).

But in 2004, the situation changed drastically. René Renou explained : "Since then, supply is higher than demand; 45 countries make wine and due to the irruption of New World vintners there is permanent overproduction. One is now in an environment of international competition, in a global economy and French viticulturists were not prepared. In addition a new code of understanding wine, inspired from the Anglo-American approach, has progressively replaced the French code. For these new consumers, wine is not "a link to God," but something simple, a colour, a grapevine variety, a pleasure and value for money. And we should not bother them

with our stories of classified vintages, historic traditions and complicated regulations. This is nowadays the mindset of 80% of the world market" (Géné, 2004).

So the issue is what to do for a very important sector of France's economy. In 2004, 876,200 hectares were devoted to viticulture, i.e. 2.8% of the whole agricultural acreage, and 95% of it was located in 30 departments. The number of viticultural farms was 144,000 and that of permanent viticulturists amounted to 240,000; also 36,000 persons were employed temporarily. There were 850 cooperative cellars, 1,400 businesses, and the annual production reached an average 53 million hectoliters. The annual turnover of the whole French industry amounted to €11 billion, including €5.8 billion in exports. Wine was the country's fifth export product, behind motor-cars, aeroplanes, pharmaceutical products and integrated circuits. Wine consumption has decreased over 20 years, down to an average 58 liters per capita per year in 2004 (Géné, 2004).

Advertising may be one answer to the question. Another is to change the system – and how French winemakers go about their business. But basically a painful exercise should be carried out about what French viticulture has done over the last decades of its land, its vines and wines, and end up into such a profound crisis by the mid-2000s.

French wine is complicated to understand. How Americans and British drinkers should select from 57 appellations solely in Bordeaux? Vintners realize that the solution entails simplifying. To reduce the range of choices facing consumers – and better compete against the ubiquitous Californian chardonnay and other New World brands – French vintners are showing a new enthusiasm for so-called varietals, wine sold by the type of grape (e.g. cabernet sauvignon or pinot noir) as New World wines often are. The Loire Valley producer Vinival grouped its wines under a new Boire & Manger (Drink and Eat) label aimed at young people, particularly women. They include drawings of fish, lambs and chickens to show which dish or meal the varietal best accompanies (Pape and Mcnicoll, 2004).

It is true that the worldwide-known *appellations d'origine contrôlée* (AOC) – Bordeaux, Burgundy, Côtes-du-Rhône – which make a very heavy imprint on the French viticultural landscape, have for a long time inhibited the emergence of strong wine brands, such as Jacob's Creek, Wyndham or Rosemount that are produced in the New World. The two main French commercial wine brands sold on foreign markets are Castel (of the same commercial group, the first industrial wine company in France) and JP Chenet (Grands Chais de France group). The first AOC Bordeaux

brand is Mouton-Cadet, an assembled wine produced by the owner of Mouton-Rothschild. Among these giants a new export wine brand has been launched by the company OVS : Chamarré. The ambitious objective of the company was to sell 4 million bottles in 2006 and 10 million in 2010. The first bottles were put on the market at the end of April 2006 and sold in the United Kingdom. Chamarré was initially conceived as an umbrella brand which includes AOCs, varietal wines and assembled ones (blended wines). Labels are simplified and legible. In addition, OVS has been able to convince large cooperatives in various vineyards to pool their financial means in order to become successful exporters. At least seven of these cooperatives have become shareholders of OVS which evolved into their common marketing structure. This brand is also sold in French supermarkets, while it was initially conceived only for export (Clavreul and Galinier, 2006).

Vintners in the Languedoc-Roussillon region are importing New World techniques and seeking advice to help ensure low cost and consistent taste for their wines. Taste-targeting appears increasingly necessary to succeed in the low-cost wine market, while French winemakers do not often realize they have to play to consumer tastes. New World vintners understand that perfectly, stated Gérard Seguin, an industry consultant. A report commissioned in 2002 by the French government called on wine producers to adopt international marketing techniques and abide by the consumers' wishes. The task is not easy and many are those who want to stick to tradition and the charms of "Frenchness", renowned for its nuances, subtlety and refinement (Pape and Mcnicoll, 2004).

Such a conflict can be perceived and examined in Bordeaux, the biggest French fine wine region and arguably the most prestigious. One can find there divisions between winemakers and the merchants who traditionally sold their vintages; between the handful of the top-name châteaux that enjoy worldwide fame and the 9,000 others, about 500 of whom are estimated to be in very difficult situation; between young and old, traditionalists and reformers; those who despise New World wine and those who think they should be copying some of its tricks; between the Bordeaux establishment and the French government in Paris, which wants to micromanage the business. All this has an impact on how the Bordeaux wine is made and sold. Winemakers are emphasizing a more direct appeal to consumers. There are moves to reduce the myriad of appellations and weed out some of the châteaux in order to make the wine more consistent and less of a puzzle. Some new brands are being created that raise some controversy, but they are far more New World in taste than traditional Bordeaux (Gumbel, 2006).

For the very top brands, prices are very high : for instance, the 2005 vintages of Château Cheval Blanc and Château Lafite Rothschild were retailing for about €500 per bottle by the end of 2006. That was about the same price as 1,000 bottles of the down-market wine sold in bulk. The ability of élite producers to raise their prices is fuelling Bordeaux's recovery. Overall, the region's wine exports dropped by about 20% over the five-year period 2000-2005, but during the first six months of 2006, exports once again rose, about 5% in volume and 24% in value to €1.2 billion. Whatever the sustainability of this recovery would be, Jean-François Bruère, who heads a 220-member cooperative in Landerrouat, on the edge of the Bordeaux region, towards Bergerac, stated that one thing was sure : "We are beginning to wake up, we understand that the consumer is what really matters. We can make the best wine in the world, but if nobody buys it, it is useless" (Gumbel, 2006).

Struggling to beat the Australians and Californians should not be confused with becoming pale imitators, or "losing the soul" in making such a high-quality product as a Bordeaux château vintage. But what to do when there is a glut of wine? The European Union's wine regime, when vineyards produce more than they are able to sell, is to hand out subsidies to transform the wine surplus into industrial ethanol. The money is supposed to be kept for exceptionally difficult years, but "crisis distillation subsidies" had been paid out three times between 2001 and 2006. At €500 million a time, it was considered a very expensive way to deal with a market imbalance. In June 2006, the European Commission published proposals for sweeping changes to the wine sector aimed at eliminating surpluses and making wine producers more competitive. Among the key measures it proposed to eliminate 400,000 hectares of vineyards over the five-year period 2006-2010 in order to stop overproduction; adopt a simpler labelling system to make European wine more attractive to consumers; and the end of all distillation subsidies (Gumbel, 2006).

In France, while the cooperative producers' association – whose members produce one out of every two bottles of French wine – have been reluctant to accept the European Commission's proposals, in Bordeaux many thought they made sense. In fact, Bordeaux has put in place a series of measures that are broadly similar to the ones proposed by the Commission. A new quality-control system is being implemented that, if enforced properly, could lead to underperforming wineries losing their right to call their wine Bordeaux. The 57 different Bordeaux appellations are encouraged to consolidate; for instance, five big areas – the Côtes de Blaye, Côtes de Bourg, Côtes de Castillon, Côtes de France and Côtes de Bordeaux – have negotiated an agreement to combine into one single

expanded Côtes de Bordeaux. There were still 17,000 different Bordeaux wine labels, and only a few hundred producers had signed up for the scheme to be paid to leave the business altogether (Gumbel, 2006).

A driving force behind these changes is a new attitude among Bordeaux's main customers : French supermarkets. They are pushing for good deals because per capita wine consumption in France has halved since the 1960s. Bordeaux made a big strategic mistake by stepping up plantings in the late 1990s – a move that increased production and aggravated the already growing pressure on prices (Gumbel, 2006).

The impact of the wine glut that occurred in 2004 was global. For instance, in California, big grape growers and some wineries have gone into bankruptcy, including the Legacy Estate Group that owned prestigious brands such as Arrowood, Byron and Freemark Abbey. The group was sold in September 2006 to a rival producer, Kendall-Jackson. In South Africa, grape prices have dropped about 30% in 2004 and 2005, while in the Friuli region of Northern Italy, which specializes in pinot grigio and other white wines, winemakers' cellars have been filling up with unsold bottles. In Australia, the situation was dramatic : unsold wine quantities were huge after doubling of output during a decade, and unlike France Australia has not subsidized distillation. According to the trade group Wine Grape Growers Australia, incomes for some growers have fallen by 60% in 2004-2005. The situation had become so bad that the trade group requested the government to pay severely affected farmers not to grow grapes. Australia's 20 biggest winemakers who accounted for 85% of the market had in fact reacted rapidly to the crisis, cutting prices and taking the financial hit early by writing down the value of their stock. Some grape growers decided to switch to citrus or almonds, and as a boon to consumers, many producers sold their surplus stock as bargain-priced bottles that showed neither the winemaker nor the winery. Australian vintners hoped that another two years would be necessary before supply and demand could reach a balance (Gumbel, 2006).

One of the reasons why the French winemakers lost out so badly in export markets was that they are bound by many strict rules. Unlike their Australian rivals, Bordeaux vintners are not free to grow as many grapevines or make as much wine as they want; quantities are strictly limited. Moreover, they cannot sell their wine as merlot, or any other single grape variety – one of the most popular New World innovations. And under a regulation passed in the 1990s, they are even forbidden from using their grapes to make table wine; the only production allowed in Bordeaux is of high-quality *appellation d'origine contrôlée* (AOC). By contrast, over the course of

15 years, the market share of Australian wines in the United Kingdom soared from about 1% to more than 21% in 2006 – five percentage points ahead of the French – as British drinking habits shifted. Wine has overtaken beer as the nation's most popular drink, driven in part by supermarket chains such as Tesco and Sainsbury that have made it affordable. Pubs are also contributing : one chain, J.D. Wetherspoon, is even serving wine on draught at its 650 pubs (Gumbel, 2006).

The Bordeaux region as a whole has to compete better. For instance, some winemakers seem willing to adopt techniques they long spurned, including, as mentioned earlier, sprinkling wood chips in maturing wine as a cheap alternative to keep it in oak barrels. They also decided to modify their regulations on AOC wine to enable producers to make a table wine, called Vin de Pays de l'Atlantique. Bordeaux merchants are already selling on the British market bottles with screw top rather than a traditional cork, and a modified label (on the front of the label, there is a drawing of a pretty château and the name : Bordeaux Classique; on the back, in English is the lure : "Steeped in heritage", it reads, "the winemaker's philosophy was to take classic Bordeaux but deliver it in a very modern way"). Other merchants have made exhaustive market research, targeting the American and British markets; some of their sales staff come from consumer-goods companies rather than the wine business; they try to go beyond the tasting of wines and talking about their qualities. A good example is that of Pascal Renaudat, an entrepreneur who aspired to create a French mega brand and who persuaded several cooperatives in Bordeaux and elsewhere in France to become shareholders of his firm; his brand, Chamarré, is itself a focus group-tested marketing creation (Gumbel, 2006).

While trying to compete, Bordeaux faces an image problem. The top wines of the region command very high prices because of their worldwide prestige, and their makers have no interest in being associated with the down-market wines some producers and merchants wish to sell. But producers in the middle are not happy, either; they worry that the massive price increases of the top wines will give Bordeaux a reputation among ordinary consumers for being unaffordable. Owners of châteaux that produce wines which were classified 150 years ago as *grands crus classés*, are convinced that they must continue to aspire to the best, to invest more and more, and to adopt at the same time a lot of savvy marketing. As one of them stated : "I am sure in the next 20 to 30 years people will be interested in products that are top. And I am doing all I can to be top" (Gumbel, 2006).

Medium- and long-term remedies for French viticulture

French wines, and not only the top ones, are produced in so-called *exceptional terroirs* and are derived from the best grapevine varietals. A major criticism made by Claude Bourguignon, a soil microbiologist, is that "during years soils have been neglected and the terroir has been destroyed." In Burgundy, this specialist considers that soil degradation started during the 1960s-1970s, with the use of wide-ranging herbicides and fertilizers (nitrogen, phosphorus ad potassium). Manual weeding in vineyards was abandoned and herbicides were sprayed, using tractors which dammed the soil (instead of aerating it). Viticulture is now consuming 30% of pesticides used in French agriculture, while its acreage is 2.8% of total cultivated land. While it is true that grapevine is susceptible to several diseases and is also sensitive to weather vagaries, the *terroir* has been submitted to a heavy chemical treatment and no vineyard has been spared. These excesses have spurred a reaction from vintners who adopted and are adopting organic or "bio" viticulture. But they are a minority. However, viticulture using too much pesticides has been denounced and many specialists recommend to till the soil better (C. Bourguignon asserted that in 80% of French vineyards vine roots are not found beyond a depth of 50 cm, because soil surface is dammed and thick; consequently, roots develop horizontally instead of growing deeper). During the hot wave that struck France in 2003, it was noticed that vines that were rooted very deeply had suffered less and gave excellent grapes, while young vines with a superficial root system were burnt by the very hot sunshine (Géné, 2004). It is therefore crucial to improve soil structure and to restore soil fertility, through allowing microflora and microfauna to multiply and play their role in improving soil physical and biological properties (Géné, 2004).

Another issue is the decrease in the diversity of cultivated grapevine varietals. There is a dominance of the five varietals cultivated internationally : merlot, syrah, cabernet sauvignon for red wines, and chardonnay and sauvignon for the white wines. Between 1979 and 2000 (the two dates of agricultural census), total vineyard acreage decreased in France, but the surfaces planted with syrah increased by 312%, that of merlot by 164%, that of cabernet sauvignon by 132%, that of chardonnay by 179% and that of sauvignon by 199%. The diversity in grapevine varietal cultivation is decreasing steadily. Several lesser-known varietals which have been abandoned because they used to produce a mediocre wine, but when they are blended with others they bring an originality that characterizes wine specificity. That is the case of the petit verdot in the Médoc area, of muscadelle in Montbazillac or Sauternes – worldwide-known sweet

wines –, or of the roussane, that has been replaced by the marsanne in the Rhône Valley. According to Guy Renvoisé, a former merchant of fine wines and author of two non-complacent books, it is "a decrease in quality and another step towards uniformization." In the Bordeaux region, another example is the coming back of merlot, a varietal that is easier to cultivate than cabernet sauvignon, which matures better and gives a less expensive wine : 17,000 hectares of merlot in 1970 compared with 62,000 hectares in 2000. As these varietals which are found across the world, are derived from clones, a vineyard can become totally homogenous quickly. It is true that clonal selection has improved the sanitary quality of vineyards as well as yields, but its generalization is a threat to biodiversity. That is why the vintners of the nine most prestigious Bordeaux wines have decided to go back to the traditional mass selection, which consists of observing and tagging the best grapevines for several years, and of taking scions of them, so as to obtain a more heterogenous plant material, but without sanitary guarantee. "Old" vintners continue to use this mode of reproduction and those who own old vines, which supply high-quality scions, possess a priceless heritage. But they are the exception, because these old vines have been superseded by the clones. Some experts conclude therefore that French vineyards have entered the industrial era to the detriment of their identity, and the logical implication is the increase in yields in all wine-producing regions (Géné, 2004).

Winemaking using modern technologies is part of that industrial era. J.P. Géné (2004) sees the flying winemakers as a symbol; these winemakers are always travelling in order to advise their colleagues across the world and have an imprint on their wines. Michel Rolland, who has his headquarters in Pomerol, Bordeaux region, is considered the most illustrious and expensive among them, and he is consultant to 400 Bordeaux winemakers and about 100 wineries in a dozen countries. In the *Mondovino* movie, M. Rolland is shown as always on the road and talking on the phone. He is a friend of Robert Parker, a worldwide known American wine specialist, whose opinions call the tune on the American market. Both are often accused – as shown in the movie – to team up in order to impose a new taste, one through his new winemaking techniques and the other through favourably classifying the wines made in the vineyards advised by M. Rolland. One can argue about the approach and the judgements, but J.P. Géné considers that thanks to Robert Parker and to the influence of his newsletter *The Wine Advocate* (40,000 subscribers at least) a new type of wine was born : that which is not drunk, but on which one speculates (Géné, 2004).

New winemaking techniques have already been mentioned, and they enable the vintners to produce a custom-made wine. It is worth

mentioning another innovation of the late 20 years : the use of industrially produced yeasts. The latter are indispensable to the fermentation of grape juice into alcohol; wild yeasts are present on the skin of grapes and winemakers do not like to rely on them, because their scarcity or quality could ruin a grape harvest. That is why more than 300 yeast varieties have been developed industrially and are maintained in collections. Among them, they are about 100 of so-called aromatic yeasts. For instance, at the Wine Cooperative Institute (ICV) in Montpellier, one can find in the yeast catalogue the strain ICV-GRE, which "can develop the mature and complex aromatic and taste characteristics of Mediterranean white wines." Enzymes are also available, e.g. the ICV Alpha white, for the 2004 harvest, which "develops the intensity of fruity aromas that are characteristic of white wines (fresh pineapple for chardonnay, exotic fruits for sauvignon) as well as the intensity of flavours in the mouth." J.P. Géné underlines that all these winemaking techniques, including biotechnology (yeasts and enzymes), contribute to the production of technological wines with a standard taste, that are recognized in the commercial classification (Géné, 2004).

Not only a standardized taste is considered a threat, but the bottling of wine as well. For instance, as mentioned above, the traditional cork or a screw-tap? The quarrel has been initiated in the United States by the *Wine Spectator*, an influential magazine in wine. Journalists considered that the screw-tap was a better guarantee than the cork for the protection of the organoleptic qualities of wine. Screw-tap has been used a long time ago for the 250-ml bottles of mediocre wine served with the meals in the cafeterias of Pechiney, the French aluminium giant (its Grand Stelvin is very successful in the New World vineyards). But in France using screw-tap instead of cork is almost a cultural war, but the Portuguese – the main producers of cork – were late in reacting and the screw-tap had already been preferred in the bottling of young wines to be consumed rapidly. In addition to Australian, New-Zealand or Californian wineries that are using the screw-tap, several French ones are following the same approach, as well as well known big brands, so as to meet the requirements of British and American supermarkets, which sell most of the wine available in their countries. For instance, Tesco in the United Kingdom is planning to sell 50% of its wines in bottles with a screw-tap (Géné, 2004).

To sum up, French viticulture has a long tradition of producing excellent wines, some of them the best in the world. This is the result of the combination of climate, soils and bedrocks, grapevine varietals, and winemaking techniques. But nowadays, French viticulture has to compete with New World winemakers, not only in terms of price, but

also in terms of simplicity of wine origins and organoleptic properties. There are those who advocate the keeping of tradition, e.g. rehabilitate the *terroirs* and particularly their soil fertility, improving winemaking techniques and selling good or very good wines at affordable prices. And there are those who, for the sake of survival, are forced to produce a "technological" wine with a standardized taste, under a mega brand, through the supermarkets at a competitive price. Maybe the medium- and long-term remedies for French viticulture lay in the middle as is shown by several initiatives, e.g. in the Bordeaux region (Vin de Pays de l'Atlantique and Chamarré).

Rebound of French wines abroad

Experts consider that the solution to the crisis affecting French viticulture will come from the world market. Between 2001 and 2010, national wine consumption was to decrease by 20%, while world consumption would increase by 9.15%, according to a study by ISWR/Vinexpo published on 30 January 2007. In 2010, the United States has become the world's biggest consumer, ahead of France, and commercial trade has grown by 15%. France should therefore reverse the trend : from 2001 to 2005, it was the only country whose exports decreased by 12.4%, while Spain's exports rose 40% and Australia's 64%. And the solution was to compete with the New World wines through copying the methods which have made them so successful. Not only it was decided to use wood chips in local wines (*vins du pays*) and in wines with AOC on an experimental basis, but also to create a new denomination *Vignobles de France*, which corresponds to blends of local wines of the same grapevine varietal and from different regions. This was meant to export a solid brand and a wine of constant quality. Merchants and wine distributors had requested these adaptations, but all producers were not of the same opinion. For instance those of the Languedoc-Roussillon region – the biggest exporter of local wines – opposed the creation of *Vignobles de France*, because they were afraid that this category would be an outlet for AOC surpluses. Others considered that the denomination would be an umbrella for other French products abroad and would therefore lower the prices. But Jérôme Despey, president of Viniflhor, the public office where *Vignobles de France* was created, was more optimistic : "Our *terroirs* are an asset which is difficult to copy," he stated, and France must conserve them, while at the same time it should compete with the New World on mass markets (Clavreul, 2007).

In fact, New World's viticulturists whose vineyards often belong to French companies, have been thinking for several years to copy the concepts

of *terroirs* and AOCs. "After having focused their quality approach on the regularity of their products, they are now working on their typicity," has asserted Claire Duchêne, marketing director of Interloire, while Jacques Gravegeal, chairman of the trade-union of producers of Oc wines (Languedoc region), stated that "the word California having become insufficient commercially, the vineyards of this American State had almost 100 local wine denominations" (Clavreul, 2007).

And there has been a rebound of French wine exports during the first half of 2006, according to the statistical office of the agriculture ministry: +16.8% up to €2.75 billion, compared with the first six months of 2005. The whole agrifood sector has increased its exports, with a surplus of €4.2 billion after the first half of 2006, i.e. a €1 billion increase compared with 2005. This was due, to a large extent, to beverages, the commercial surplus of which amounted to €3.9 billion, including €2.5 billion for wines and champagne (+18.6%) [Clavreul, 2006].

Despite this good start in 2006, professionals were awaiting long-term data before speaking of a real rebound. Good results were due above all to cognac, champagne, vodka and top Bordeaux wines. Exports do not concern all wines equally : while Bordeaux wine exports rose 34% during the first five months of 2006 and côtes-du-Rhône 33%, Burgundy wine exports rose only 9%; wines from Val de Loire were losing 6% and beaujolais 5% in their export value. The success of Bordeaux wine exports was mainly due to important deliveries of the 2003 and 2005 vintages, considered good ones. Results are more encouraging in terms of value rather than volume, while world viticulture is confronted with overproduction (Clavreul, 2006).

The case of champagne is worth mentioning because since mid-2005, despite the crisis affecting French viticulture, its production and business were again attractive for investors. Thus, on 4 July 2005 Pernod Ricard made official the purchase of the British company Allied Domecq, the world's fourth-biggest spirit company, which owned Mumm, the world's third brand of champagne. On 8 July 2005, the shareholders of Lanson International – the Mora family and the Caisse nationale des caisses d'épargne, holding respectively 56% and 44% of the capital – sold the group, the world's third-biggest producer of champagne behind the leader Moët-Hennessy (€1 billion annual turnover in 2004) and Vranken Pommery (€261 million). Finally, on 22 July 2005, the group Taittinger (€87 million of annual turnover in 2004), which includes a champagne business bearing the same name, was purchased by the American investment corporation Starwood Capital (Mallet, 2005).

During the 1990s, big groups such as Danone, Grand-Met, Seagram, had left the champagne sector. But the year 2004 has been an exceptional year of sales, but also of mergers and business concentration. The sector became attractive while more than 300 million bottles (75 cl) had been sold, including 40% outside France. These figures were close to those sold in 1999 : a record 327 million bottles. The countries of destination of French champagne in 2004 were : United Kingdom (29.3 million bottles), United States (18 million), Germany (12.3 million), Belgium (8.4 million), Italy (7.9 million), Switzerland (5.8 million), Japan (4.3 million), Netherlands (2.4 million) and Spain (2.0 million) [Mallet, 2005].

Champagne sales fell down in 2008 (-4.8%) and 2009 (-9.1%), but grew again in 2010 reaching 320 million bottles, i.e. +12% for the first ten months of the year. The rebound of sales reached 22% in the European Union and 33% outside the EU. The two main export markets, the United Kingdom and the United States, were quite reactive, while other promising markets like China showed a strong growth. Although the situation was very satisfactory and brought relief to all producers, there was no sign of triumphalism because the economic and financial conditions in Europe were still too uncertain. It should be underlined that in 2008 and 2009 winemakers had managed the crisis rather well because of the favourable French market. However those who earned most of their income from exports suffered. In fact the rate of export decrease was significant : -17.4% in the European Union and -25.4% in the rest of the world (Van Eeckhout, 2010).

But the price of champagne grapes did not decrease. The geographic area having a certified appellation is limited and the price of grapes is about €6 per kg for the best among them. The owners of champagne businesses do not own more than 10% of the vineyards with *appellation d'origine contrôlée* or AOC and they buy most of their grapes from winemakers, directly or through cooperatives. In addition, the process of making champagne has always been costly because it needs a second fermentation in bottles and at least three years of storage (seven to eight years for the renowned vintages) [Van Eeckhout, 2010].

Consequently, champagne businesses, such as Vranken Pommery or Lanson BCC, have been trying to develop a portfolio of complementary brands. This enables them to devote one or two of their brands to premium quality or great vintages, while selling large volumes of less elitist brands or even supplying distributor brands. But any kind of successful business tries to draw the largest part of its earnings from export sales.

Profitability is higher because champagne is perceived outside France as a luxury product. In France prices are lower, although in 2010 higher-end and more expensive vintages were increasingly consumed; this trend would be confirmed by a price increase forecast for 2011 by champagne businesses (Van Eeckhout, 2010).

In 2010, the expected wine production in France amounted to 47.3 million hectoliters, compared with 46.7 million hectoliters in 2009, but lower than the average production of the five-year period 2005-2009.

Germany

Germany produces well-known wines such as riesling, sylvaner and gewurztraminer. At the end of the 19th century, riesling wines, known as *hock*, were fetching higher prices than claret from Château Lafite and Veuve Clicquot champagne, according to a list from Berry Brothers & Rudd, a London wineseller. Today's Rheingau rieslings are again winning high reputation and prices. Their special quality relies on a mix of cool nights and warm days for slow ripening. But warmer average temperatures due to climate change may redraw the vine-growing map. Red-grape varieties such as cabernet sauvignon and merlot, generally grown in the south, will migrate northwards by 200-400 km and up hillsides by 100-150 m, according to Hans Schultz of the Research Institute at Geisenheim in the Rheingau. By 2040, cabernet sauvignon will flourish where riesling does now (*The Economist*, 2007 c).

The impact of the warming-up of temperatures is already felt. *Eiswein*, a dessert wine made from grapes that are picked frozen on the vine at a temperature of -7°C or below, is becoming even rarer. In 2006, the local growers had only two chances – on the mornings of 27 December and 26 January – to harvest grapes frozen enough. "That was our latest harvest ever for *Eswein,*" stated Arno Schales, whose family had grown riesling since 1783 and had made the sweet wine since the mid-1950s. In 2006, his *Eswein*, a production of 200 bottles instead of the usual 1,000-2,000, came from pinot noir grapes, which survived the late warm weather without rotting (*The Economist*, 2007 c).

In 2006, many Rheingau growers abandoned *Eswein* and instead picked the grapes for *Trockenbeerenauslese*, a wine made from grapes that have dried on the vine. German vintners are also more attracted by red grapes, although they favour the more traditional pinot noir and dornfelder varieties over merlot and cabernet sauvignon. For their part, German consumers were choosing more locally made red wines in 2006 than

they did in 2002 – 27% of their red wine intake in 2006, up from 17% in 2002. Nevertheless, the main focus is still on better and more expensive rieslings, for which there is a strong demand, particularly in the United States. Germany has 60% of world production, despite the progress made in Australia and America. Quality has improved as the new generation of growers has invested in better processing. Climate change is also forcing them to adapt : according to Hans Schultz, the Geisenheim vines are developing shoots seven days earlier, blossoming ten days earlier and starting to ripen 12 days earlier than the 40-day average; they are especially affected by the warmer nights (*The Economist*, 2007 c).

German viticulturists are adopting the same approach as the Bordeaux vintners in terms of quality improvement and competition on the world market, while at the same time adapting their viticulture to climate warming as other European growers and South Africans do.

Russia : a new start for viticulture

In the steppes of Kouban, in the pre-Caucasus area, wine production was upheld in 1986 when Mikhail Gorbatchev, then secretary-general of the USSR Communist Party, ordered the growers to uproot their vines, as a means to control the disastrous consequences of alcoholism. Some growers, however, continued to produce grape juices and even wine in a hidden way. Russian vineyards were later on affected by the collapse of the sovkhozes after the end of the USSR, the orders from the state having ceased (Vatel, 2007).

Nowadays, the region of Krasnodar has 31,000 hectares of vineyards, equivalent to almost half the total vineyard acreage of Russia, and it is expected to increase its production capacity. Despite the availability of land and good climate and soils, Russia is not internationally recognized for the quality of its wines. There are also difficulties at the internal market level : grape harvests only met 20% of the needs of wine-production units in 2007 (Vatel, 2007).

However, in the Kouban region, there are efforts aimed at restructuring the whole wine production and business, and at obtaining funding from local viticultural and wine-production entities, from import and bottling companies and from tycoons of the industry and raw-materials production who want to diversify their investments. For instance, the Moscow businessman Sergueï decided to invest in vineyards; he commercializes Château le Grand Vostock from his 160-hectare vineyards, and, like other producers, he has called on foreign know-how. Since 2004, his advisers

have been French graduates from the National School of Agricultural Work Engineers (ENITA), Bordeaux. Their main challenge was to upgrade former state farms the priority of which had been since 1937 to comply with plans and focus on quantity rather than quality (Vatel, 2007).

In 2006, total wine production in Russia amounted to 470 million liters, compared with 328 million liters in 2005, an increase of about 50%. But this production has to face competitors who are attracted by the importance of the national market. Among emerging countries, Russia is the first biggest importer of wines and spirits. Italy, Spain and the United States are encroaching on the share of older and closer suppliers. This trend has been stronger since 2006, when a total embargo was imposed on wines from Moldavia and Georgia, officially for sanitary reasons. In 2006, France has become the first supplier of Russia in value terms, while in terms of volumes it has been outpaced by Spain. On the consumers' side, they have to be educated so as to prefer wine to beer and vodka, and to select their wine not just on the basis of price, according to Franck Duseigneur, a French engineer who works for Château le Grand Vostock. Wine consumption indeed has increased markedly in Russia : from 5 liters to 7 liters per capita per year between 2000 and 2005. In addition, the share of AOC wines and top wines in this consumption is increasing, and Russians are having a preference for dry wines instead of sweet wines (Vatel, 2007).

United States

It has been mentioned that the United States is set to overtake France as the world's largest wine market by 2012. Still wine consumption in this country would also exceed that of France, with a trend in favour of the consumption of higher-quality and more expensive wines. The United States is obviously a threat to the Old World's traditional hegemony with respect to production and quality.

Californian wines have nowadays an established reputation and they often rank at the top of blind tastings organized worldwide. The story started on 24 May 1976 when an English wineseller, Steven Spurrier, of Caves de la Madeleine in Paris, organized a professional blind tasting of top red Bordeaux and white Burgundy wines. He also invited foreign wine producers to participate in the tasting. While French newspersons did not attend, George M. Taber, the *Time*'s correspondent, was present. Thirty years later, he wrote a book on the event: *Judgement Of Paris* (which probably will be the basis of an American movie). In 1976, at the InterContinental Hotel in Paris, professional wine tasters gave their

preference to cabernet and chardonnay wines made in California. The cabernet sauvignon-1973 vintage of Stag's Leap Wine Cellars, produced by Warren Winiarski, obtained a better ranking than the top 1970 vintages of Château Mouton-Rothschild Pauillac and of Château Haut-Brion Pessac. Other Californian wines drew the professionals' attention : the 1971 Ridge Monte Bello, produced by Paul Draper in the mountains of Santa Cruz, south of San Francisco, and young wineries of the Napa Valley, Mayacamas, Clos du Val and Heitz Martha's Vineyard. Regarding the whites, the 1973 chardonnay of Chateau Montelena was ranked above all the Burgundy whites (Mulard, 2007).

On 24 May 2006, thirty years later, the blind tasting now known as the *judgement of Paris*, has been repeated with the same wines competing, and 18 judges distributed between London and the City of Napa. The French vintners hoped that the young Californian ones that won in the past would not age as well as theirs. But American viticulturists did improve their performances and they won the first five ranks, leaving Château Mouton-Rothschild at the sixth rank. That was a real world recognition and award for the New World wines (Mulard, 2007).

By the 1880s, there were over a hundred wineries in the Napa Valley, but, after the phylloxera attacks and the prohibition law on alcoholic consumption, less than 20 of them survived. Nowadays of the 1,300 viticultural farms and vineyards existing in California, more than 400 are in the Napa Valley, from small family vineyards to big brands such as Mondavi, that is listed on the stock exchange. California, the Golden State, has become the world's fourth-biggest wine producer, behind France, Italy and Spain, with an annual turnover of US$27 billion in 2006. The renaissance of California's viticulture started in the 1960s when a few vintners, convinced that they enjoyed an exceptional *terroir* and climate, resumed growing vines, often on old abandoned vineyards. All the winners of the *judgement of Paris* are part of that renaissance. The first was Joe Heitz who grew vines near Santa Helena in 1961. Those vine growers were university people, diplomats, business brokers, none had grown in a farm, but all wanted to go back to farming. They learnt their vintner profession very quickly, sometimes at the oenology department of the University of Davis, California, but most often in the Bordeaux region. And thereafter, the students fared better than their masters (Mulard, 2007).

In 1983, official appellations such as Napa Valley American Viticultural Area or AVA are ushered, and Californian vintners have the advantage of carrying out their job more freely than their French counterparts, who must respect very strict regulations regarding irrigation, yield per hectare,

grapevine varietals grown. Napa Valley enjoyed the reputation of an environment where vintners exchanged their know-how generously. They have in common the dislike for an "international style" that some modern viticulturists wish to impose globally. The Napa Valley vintners do not irrigate much and harvest the grapes late in the day and preferably during the night. They know that each *terroir* will give a unique wine and that is feasible in a valley located in a 24-million-old volcanic fault (Mulard, 2007).

Warren Winiarski, a son of Polish immigrants whose name means "son of vintner", is the owner of Stag's Leap Wine Cellars and one of the most respected vintners of the Napa Valley. He arrived there in 1964 after having worked at Chicago University, and five years later he had a revelation when he tasted a cabernet wine produced by Nathan Fay, in a vineyard in the district of Stag's Leap. Since then, this hill has become one of the most prestigious appellations of the Valley; its 1973 cabernet sauvignon won the award of the best red wine at the first *Judgement Of Paris*. Warren Winiarski's *terroir* is unique : pebbles and alluviums under the volcanic rock of Stag's Leaf (Mulard, 2007).

Mike Grgich, who grew up in Croatia in a farm with vineyards, is another legend of the Napa Valley. He is considered as a top producer of white wines : its Montelena vintage won the *Judgement Of Paris* in 1976. Bob Travers, a finance specialist, became a vine grower in 1968 and bought a 19th century vineyard. The Mayacamas Vineyards are isolated on Mount Veeder, a volcanic crater with woodlands on its slopes in the western part of the Valley. Vineyards are located on the slopes so that water trickles rapidly, soils are rocky ones, grapevines are not highly productive, but grapes have a taste of great intensity. At the *Judgement Of Paris*, they won a third rank. Paul Draper, in partnership with Stanford University, produces the Ridge Monte Bello vintages, near the Silicon Valley. Trained as a philosopher, he learnt everything about vine-growing and winemaking in French handbooks. Its wine with "mineral quality," produced from vines growing on limestone soils, won the first award at the 2006 *Judgement Of Paris*. Paul Draper can also rely on a very sophisticated analysis laboratory at Monte Bello (Mulard, 2007).

California enjoys a very favourable climate, which is a big asset for grapevine cultivation. Rainfall season occurs between November and April, with warm and dry springs and summers (a typically Mediterranean climate). By contrast to their French counterparts, harvests are not threatened by storms or hails. Nevertheless the Napa Valley is permanently wetted by marine air coming from San Francisco Bay and going along the Napa

river. Cool and wet nights are very favourable to grape ripening and give them a special taste (Mulard, 2007).

The boom of Californian wine has had a clear-cut impact on the change of taste of the American people. In a country where beers and hard spirits are generally preferred, the new generations (baby-boomers) have learnt to drink wine. In 2004, American people drank about 3 billion bottles, a consumption that is increasing although inferior to beer. Wine consumption goes with the attraction for good food; the restaurants of the Napa Valley are renowned and have been distinguished by stars in the Guide Michelin, which published a special edition named San Francisco Bay Area and Wine Country. In the White House's cellars, top French wines are present (Thomas Jefferson, the third American president, had ordered 300 bottles of Château Yquem , a very renowned dessert white wine), but along with those from California which are highly appreciated by the president's guests (Mulard, 2007).

Not very far from California in the State of New Mexico, sparkling wines are being produced at the Gruet Winery by Nathalie and Laurent Gruet (sister and brother) who moved from the Champagne region to New Mexico in 1983. They took the risk of planting vines in this high desert climate and of producing what have become very successful domestic sparkling wines. The Gruet family had deep roots in France : Gruet & Fils was established in Champagne in 1952 by the family patriarch, Gilbert Gruet, in the village of Béthon. G. Gruet, who died in 1999, was an architect who agreed to help build a winery in Champagne in exchange for lessons in making the region's sparkling wine. He founded the first wine cooperative in Béthon in 1967, persuading the villagers to uproot sugar beets and plant grapevines instead (Kershaw, 2010).

In the early 1980s, like other winemakers in the region, G. Gruet decided that with a change in the French government and impending tax increases, it was time to start a winery in another country. With two of his children, Nathalie and Laurent, he toured California, Texas, New York and finally New Mexico. They wanted to try the Champagne method of making sparkling wines in New Mexico, so they sent samples of the sandy soil to France for testing, in order to make sure the vines could grow the deep roots they needed to thrive. Encouraged by the results, the siblings, Nathalie and Laurent Gruet, planted an experimental vineyard in 1983, in Lordsburg, New Mexico, and settled a year later on land near a small town about 150 miles south of Albuquerque, known among tourists and retirees for its natural hot springs and mild climate. The latter was an advantage, with cool evening temperatures that slow

the ripening of grapes to produce a pleasantly sharp acidity. The Gruet imported their first press and other machinery from France and rented a small production space in Albuquerque. Their operation grew in small increments over 20 years (Kershaw, 2010).

They went from producing 2,000 cases of wine in 1989 to 100,000 cases a year in 2010, doing everything but the growing in a 4,200-square-meter plant in Albuquerque. The French and New Mexican wineries (the other two siblings, twin sisters, stayed in Champagne and run the operation there) produce about the same amount of wine. The New Mexican members of the family stated that they had found the climate there even better for grapes than in Champagne; the days can be very hot, but the nights, as much as 17°C cooler, slow the ripening process in what could otherwise be a short growing season. The arid air that protects against rot also helps with the wine consistency, according to Laurent Gruet who stated that they used no pesticides on the vines (Kershaw, 2010).

While the economic recession has struck wine producers in Champagne, California (Napa Valley) and elsewhere, Gruet has held its own, and even made headway in some markets, with only some minor wholesale losses. This was due, to a large extent, to budget-minded pricing : most of Gruet's sparkling wines were selling for less than US$20 a bottle in 2010. Customers wanting champagne or sparkling wine have moved to taste less-expensive products, giving these French expatriates in New Mexico a timely edge. Their sparkling wines are especially popular in New York and California; they are in fact sold in 49 States – 60% of it at about 5,000 restaurants and the other 40% at retail stores, according to Laurent Gruet. They have even appeared in the past ten years on the wine lists of high-end restaurants, including Craft, Del Frisco's and Bar American in Manhattan, New York. The winery produced 10 sparkling wines in 2010 and a small amount of still chardonnay and pinot noir (Kershaw, 2010).

Some critics argued that despite their high quality-to-price ratio, Gruet's sparkling wines were inferior to top sparkling wines made in France and even in California. But many wine buyers and reviewers see Gruet as a source of affordable domestic good products that hold up – or can even outdo – more expensive items (Kershaw, 2010). It is by all means a good example of how New World wines can compete successfully with European ones, playing on the assets of climate, *terroir*, winemaking techniques and marketing policy.

South Africa

Peter Gumbel (2006) stated that "if French winemakers want to learn how to overhaul their business without losing their soul, they might want to visit South Africa."

During the apartheid era, South African viticulture and wine production were controlled by a government monopoly that set rules as strict as those prevailing in France. Since South Africa's independence, the old monopoly disappeared and producers have replaced over 40% of the country's vines – ripping out the white grapes long favoured for domestic consumption and planting a wide range of red grapes for export. They knew how to adapt to changing world demand and how to improve marketing. The consequence was an almost eightfold increase in wine exports since the early 1990s, a stronger performance even than Australia's (although from a lower base). Innovative marketing has helped : Charles Back, for instance, has made French authorities furious by making a successful côtes-du-Rhône-style range that he called Goats Do Roam. And Nick Dymoke-Marr, who created a new brand called Stormhock, added a date-code indicator on the back of bottles that highlights when they should be consumed. He then was trying to spread Stormhock's reputation through wine-loving bloggers in the United Kingdom to whom he had been sending out free bottles in the hope that they would post their tasting notes online (Gumbel, 2006).

But South Africa has suffered from the global wine glut, which caused a drop in grape prices, and producers had also to deal with a 50% appreciation in the rand between 2002 and 2005 that pushed them out of the sector of cheap and cheerful supermarket wines for the United Kingdom, where they initially made their reputation. But the impact of the global wine glut was nevertheless less harmful than in Australia or France. The South African solution was to continue to offer good quality and good value. South African producers are also pushing hard into new markets, including Germany, Russia, the United States and Sweden, which alone now takes 10% of South Africa's exports (Gumbel, 2006).

Producers have one problem in common with the French : even as their exports continue to grow, their domestic market is shrinking. Wine consumption in South Africa, already relatively low for a producer country, is falling, as some 1 million white people have left the country over the past decade or so and as tastes have changed. Reversing that trend could take years. For instance, the annual Soweto Wine and Brandy Festival is designed to encourage the nation's black middle class to consume

wine; this class still sticks to national beer and spirits. There are at least ten wine festivals per year in Johannesburg, but few people of colour attend them, according to Marilyn Cooper, who runs the local branch of the Cape Wine Academy. In 2004, the first wine shop was opened in Soweto, the Morara Wines & Spirit Emporium. The owner thought that the market opportunity was high and the wine market was largely untapped (Gumbel, 2006).

Most of the winemakers in South Africa are whites, who control the wine industry. According to Joshua Hammer (2005), there were about 20 black-owned vineyards in South Africa in 2005, and almost all of them have sprung up since 2002. While South Africa's wine industry has long been regarded as one of the last privileges of Afrikaners, more than a decade after the end of apartheid, a proliferation of black- and coloured-owned ventures has shown the changing face of South Africa – and also exposed hurdles to black empowerment. In 2004, the ministry of agriculture and land affairs announced that its goal was that 30% of all agricultural land to be owned by black and coloured people by 2014. As a result, many Afrikaners were breaking up their estates and selling a portion of the land to their employees. The Black Vintners' Alliance of South Africa counted more than 20 black-owned vineyards in 2005. But few, if any, of the new ventures have turned a profit : the employee-owned vineyards took on massive debt to start their business, and onerous repayments have made it impossible for them either to take money out of their ventures or to reinvest. For instance, African Terroir, outside Paarl, a Swiss-owned complex of vineyards and cellars, sold 173 acres to 28 of its grape pickers and their families. The workers purchased the land at below-market prices with government grants totalling US$68,000, or about US$2,500 per household. They are producing grapes and wines that are bottled under the Winds of Change label and sold abroad as evidence of South Africa's new ways. However, the pickers' lives have not changed much : they still toil in the fields and deliver the grapes they harvest to African Terroir's cellars for fermentation and bottling. They need training, but a four-year programme in winemaking at the prestigious University of Stellenbosch cost US$13,000 in 2005. Some cooperatives have set up a literacy programme and a once-a-week finance course on their premises in order to help the new owners to manage their property and sell their wines efficiently (Hammer, 2005).

Argentina and Chile

Argentina has been for a long time a big producer of low-quality table wines for local consumption. Since the devaluation of the peso, the local currency,

in 2002, foreign investment has poured in and wine exports have boomed : during the four-year period 2003-2006, Argentina's wine exports have more than doubled in volume and have tripled in value. Thanks partly to the arrival of foreign winemakers, quality has greatly improved. Argentines are drinking less but finer wine (*The Economist*, 2007 d).

Chile became an important wine exporter a dozen years before Argentina, and in 2006 it has exported almost 5 million hectoliters compared with less than 3 million hectoliters for Argentina (the respective value for these exports was US$1 billion and US$400 million approximately). Argentina was in 2006 the world's fifth-biggest wine producer, and it has a lot of suitable land for growth of the industry. Chilean winemakers may envy their neighbour's trademark malbec grape, a strong red wine that has become known worldwide. Also fresh, perfumed white wines made with torrontés grapes, mainly around Salta (northwest of the country), are distinctive. By contrast, Chile exports mostly standard varieties such as cabernet sauvignon and chardonnay, which are widely produced throughout the world. It nevertheless emphasizes the search for more typical varieties, such as carmenère (the old French grape variety called carignan), which can give good red wines (*The Economist*, 2007 d).

Argentina's exports to the United States are growing faster than those of Chile. Its wines probably please American taste. *Wine Spectator*, the highly appreciated magazine for American oenophiles, gave ratings of 90 points or over (out of 100) to 172 red wines from Argentina, compared with 138 from Chile. The US$120-a-bottle top wine of Achíval-Ferrer, a locally-owned small vineyard, was given a 95-points rating. But Chile's strengths should not be underestimated. It has many different *terroirs* nestled among secluded valleys (Maipo, Colchagua); cool Pacific breezes make it possible to produce fine white wine as well as red ones. In 2006, *Wine Spectator* gave 96 points to Don Melchior 2003, the top cabernet from Concha y Toro, the largest wine firm. Chile's greater economic stability has also helped the expansion of the wine industry. Since 1995, the area under vines in Chile has doubled to 100,000 hectares, while in Argentina the total area has remained static, but wine quality has been improved (*The Economist*, 2007 d).

Perhaps Chile's most important advantage is its web of free-trade agreements. Wines of Chile, a trade body, opened an office near London in 2002. Since then Chile's market-share in the United Kingdom (a huge wine importer) has risen from 5% to 7%, according to Ricardo Letelier, its general manager. It also has an office in the United States and hopes to boost worldwide exports to US$1.2 billion by 2010, with most of the

growth coming from higher prices. Although Chile has built a reputation for reliable, good-value wines, commanding higher prices depends on a clearer country image; Chile does not have the equivalent of the Argentine tango or Maradona, the famous soccer player, according to Patricio Tapia of Wine & Spirits, a New York-based magazine. While Argentina's winemakers have yet to unite in a single trade body, the industry opened a London outpost in 2006 (*The Economist*, 2007 d).

Both countries' wine exports – especially Argentina's – have the potential to keep growing. But Chile's producers are handicapped by a strong currency (the result of high copper prices, after a drop in 2007-2008), and Ricardo Letelier acknowledged in 2007 that most of the country's 150 exporters were barely managing to cover costs at that time. Their smaller local market does not offer a cushion, although some have sought to diversify by investing in Argentina. In the case of Argentine winemakers, record profits have attracted a lot of new investment, but double-digit inflation has caused wages and the cost of inputs to soar. A Spanish vineyard's owner in Mendoza – the pre-Andean region where many good Argentine wines are produced – has acknowledged that the cost of building a winery had quadrupled in dollar terms since 2002. A 5% tax on wine exports and the government's lack of interest in trade agreements were further obstacles. Mendoza's 800 separate producers (probably even more) may change the situation, but variety and innovation continue to be emphasized in Argentina and Chile (*The Economist*, 2007 d).

In this respect, the success story of Chile is worth being explained. The "South American Tiger", which is the world's fifth-biggest wine exporter behind France, Italy, Spain and Australia, commercializes about 70% of the wine produced abroad. In 2009, Chile exported 651 million bottles, mainly to the United States, United Kingdom, Japan, Brazil and Germany. The big wine corporations export up to 90% of their best wines, and top vintages such as Carmin de Peumo, produced by Concha y Toro, is almost exclusively sold on British and American markets (at an average retail price of US$100 a bottle) [Bernouin and Magnenet, 2010].

Analysts say that technical managers, agronomists and businessmen are progressively increasing the share of Chilean wines in a market historically dominated by French vintages. For instance, Ignacio Recabarren, a wine expert at Concha y Toro – the country's first wine producer with 276 million bottles a year in 2009 – is among the pioneers of innovation in winemaking. "Twenty years ago (in the late 1990s) Chilean wines were boring, many bottles were of low quality and there was no top-ranking vintage…" while

the country has remarkable assets for producing high-end wine, "such as an exceptionally healthy climate due to natural barriers against diseases, desert in the north, the Andes in the east and the Pacific Ocean in the west," reported Michel Bernouin and Jean-Christophe Magnenet, special envoys of the French daily newspaper *Le Monde*. It is in the immense central valley around the capital Santiago that grapes ripen today to make strong wines from three main varieties : cabernet sauvignon, cabernet franc and carmenère. The latter had been destroyed in the Old World by phylloxera during the 19th century, but has found a very favourable environment in the Andean hills (Bernouin and Magnenet, 2010).

The carmenère grape variety needs a warm and very humid environment in order to flourish and to be harvested late in the year. It has been confused with merlot for a long time, then it was left aside as vine growers preferred cabernet sauvignon, but now he attracts the favour of Chilean oenophiles, like Ignacio Recabarren. The number one of Concha y Toro has succeeded in making from carmenère his "dreamed wine," Carmin de Peumo. This top wine which sells at US$100 a bottle (2010) contains 90% of carmenère, a little of cabernet sauvignon and a touch of cabernet franc. The blending gives a strong wine, typical of the New World, with aromas of red fruits and spices. Robert Parker's *Wine Advocate* gave it a rank of 97, a record (Bernouin and Magnenet, 2010).

Alvaro Espinoza, son of a Chilean enologist, has its 8-hectares vineyards in the Maipo Valley, one hour-drive from Santiago. He spent two years in France (1987-1989) with his wife and discovered the winemakers of Saint-Emilion and Pomerol in the Bordeaux region. In 1996, they founded their winery where they invested 10 million Chilean pesos or the equivalent of €15,000. The first vintage Antiyal was marketed in 1998. It was again a carmenère and only 3,000 bottles were initially produced. Thereafter production doubled, but the bottles which sell at €65 a bottle, are still numbered and labelled manually. Antiyal means "son of the sun" in Mapuche language (Mapuches are the autochthonous people of Central Chile). A. Espinoza wanted to give the name *terroir* to the wine, not his own name. And the *terroir* is now on every Chilean winemaker's lips : a good wine needs not only optimal winemaking techniques, but above all the appropriate soils. Both J. Recabarren and A. Espinoza make the same discourse : in front of the trend towards some standardization of wines across the world, the *terroir* makes the difference (Bernouin and Magnenet, 2010).

Although all agronomic defects can be corrected in the laboratory, these imperfections give to a vintage its characteristics. A. Espinoza considers that the solution to making wines that seduce the customer is to respect

the *terroir*. It is a kind of agriculture that appeared at the beginning of the 20th century and was based above all on the respect of soils and plants. Called biodynamic agriculture, it is a mixture of organic farming and astral influence. It is supposed to lead to a healthier diet and it has become popular in Chile, a country very sensitive to "good waves" (*buenas ondas*). A. Espinoza who believes in this concept, imported from France the first fermentation vat, made of concrete and having the shape of an egg, because it is "a concentrator of energies," and energy is the basis of biodynamic agriculture (Bernouin and Magnenet, 2010).

At Montes wineries, another great name of Chilean wines, winemakers are trying to make a synthesis between science and mysticism. In the Colchagua Valley, Aurelio Montes founded its winery in the 1980s. It is a 100% owned Chilean company which sells 10 million bottles a year (2010) across the world. The son of the founder, Aurelio Montes Jr., is like his father an enologist and his new winery is a half-burried bunker, well designed according to the Chinese *feng shui* principles. But he does believe in the scientific understanding of each step of elaboration of his top vintages. Grapes of each plot are analyzed in a laboratory in order to follow the steps of ripening and then of harvesting and fermentation (Bernouin and Magnenet, 2010).

Without neglecting or underestimating the "mystical" approaches or concepts of winemaking in Chile, the overall success story of Chilean viticulture is largely due to good application of oenology, inspired from the Old World, but also keen to produce competitive wines including among the top ranking ones. The assets of an exceptional climate and good soils make also a key contribution to Chile's progress towards excellence in the world of wine.

Australia and New Zealand

Australian wines have conquered the world. In July 2007, their annual exports reached a record value of A$3 billion (€ 1.87 billion), according to national statistical figures published on 7 August 2007. This also represented a record volume of 805 million liters. Australia therefore was the world's fourth-biggest exporter of wine and the world's sixth-biggest producer. San Tolley, director of the Australian Society of Wines and Spirits, commented that these results "reflected the rise of price per liter and a range of better quality wines", and that was the goal for the future (Le Moël, 2007).

Wine production indeed has grown impressively over 15 years. While in the 1980s wine was consumed locally, the situation has changed and in 2008-2009 : 750 million liters have been exported. The vineyards area increased from 67,000 hectares in 1993 up to 170,000 hectares in 2009, with a population of 2,000 viticulturists. In the Barossa or Hunter Valley, two main viticultural regions of the country, a new kind of tourism has expanded : wine lovers can follow the route of vineyards and stop at tasting wineries (Le Moël, 2009).

Growing demand for Australian wines has caused a grape shortage and soaring grape prices, so that growers rushed to plant more vines in the late 1990s. In 1998, they put in a record 16,000 new hectares, double the new plantings two years earlier. In 2005, Australia produced almost 2 million tons of wine grapes, a quarter more than analysts said its markets could absorb (*The Economist*, 2008 a).

To sump up, over ten years Australia built up a powerful wine industry, more than doubling its vine acreage and concentrating its commercial strengths on a handful of groups and big brands, such as Jacob's Creek (property of the French conglomerate Pernod Ricard). Australian wine exports have quintupled since 1995 and in 2005 they amounted to half of the volumes exported by France. Thanks to Spain and Australia, 14 million hectoliters were added to the global market in ten years, i.e. more than half the total increase of 24 million hectoliters corresponding to global exports (Clavreul and Galinier, 2006).

The United Kingdom remains the main importer of Australian wines. It bought 285 million liters in 2006-2007, for a value of US$974 million. The United States is the second-biggest importer : US$962 million in 2006-2007. In 2008, each country imported volumes for a value of US$700 million (€412 million). Lawrie Stanford, in charge of information and analysis at the Australian Society of Wines and Spirits, stated that "the United States, like Canada, appreciate the characteristics of Australian wines, because the latter are affordable and easy to understand." Asian countries have increased their consumption : China has spent US$51 million, i.e. 125% more than in 2006. In Europe, Australia targets countries which have no or a very small production. France and Italy are not easy to penetrate; however the French have spent US$15.6 million to buy Australian wines (Le Moël, 2007).

In 2007, the boon turned to bust, due to the worst drought in the century. In north-west Victoria, where 150 years ago the gold rush enabled the most unlikely types to make a fortune and where the wine industry has

been booming, many farmers were forced to abandon their vineyards because they could not sell grapes or land. Fruit farms and vineyards could survive only with irrigation from the Murray River, the lifeblood of Australia's agriculture. Smaller firms, which supply the big winemakers with some of their grapes, faced falling grape prices and cuts to irrigation water. Stephen Strachan, chief executive of the Winemakers' Federation of Australia, an industry body, acknowledged the drought was a turning point, even if a tragic one in some cases, in forcing the wine industry back to "sustainable levels". The planting rush ended : the 3,600 hectares of new vines planted in 2006 almost equalled the 3,400 hectares of vines uprooted that year (*The Economist*, 2008 a).

Regarding the grape prices, it dropped from an average of US$710 per ton in 2005 to US$636 per ton in 2008. It was even expected to fall even more in 2009, warned Stephen Strachan. Also the price per bottle was falling. The major distributors tend to push downward; there was still too much low-quality wine on the market, which harms the producers of good-quality wines. The average selling price of a liter of wine was A$3.24, the lowest level in ten years (Le Moël, 2009).

Australia was a victim of not only an extreme and disastrous drought, but also of the global wine glut. The wine campaign 2005-2006 had started with a high global stock and a strong production. This situation was quite similar to that of the year 2000. In Australia, in 2005, 200,000 tons of grapes were left on the vines, which corresponded to 1.5 million hectoliters of wine, compared with 40,000 tons in 2004. Also in Chile, in April 2006 after the end of the grape harvest, grapes were not collected. The French vintners had also to uproot grapevines and to distil AOC wines (to produce alcohol) in order to survive (Clavreul and Galinier, 2006).

Consequently, in 2008, Australia's grapes harvest was not expected to be higher than 1.6 million tons. According to the Australian Wine and Brandy Association, the value of wine exports has fallen by 10% in 2008-2009 to reach US$2.42 billion, its lowest figure since 2003. In these conditions, Stephen Strachan considered that "20% of national production should be abandoned and priority given to publicize good Australian wine" (Le Moël, 2007, 2009).

The crisis indeed has led to much reflection among Australia's 2,000 wine producers about how the industry can recapture its reputation for quality wines. There is now stiff competition in the mid-market from other New World producers, notably New Zealand (see below). Much Australian wine during the grape glut found its way onto the world market as bulk

or "commodity" wine, sold at low prices or even at a loss. Australian producers now face the task of earning a reputation for quality rather than quantity. The appreciation of the Australian dollar, which makes Australian wines more expensive overseas, has made the job even more urgent (*The Economist*, 2008 a).

Historically, many Australian winemakers have derided the French approach to making wine, especially the concept that the finest wines come only from a *terroir* – the combination of climate and soil characteristics of each place. Australian vintners instead emphasize a simpler approach : blending grapes from different regions to achieve a consistent wine, and mastering the fermentation process. But some are now asking whether marketing an Australian wine locality, as much as its grape variety, might not work better. For instance, in Margaret River in Western Australia, small winemakers produce 3% of the country's production, mainly at the high end of the market, and independently of the big companies that predominate in eastern Australia; the wines, which pride their *terroir*, sold for as much as A$95 a bottle in 2008. Steve Webber, the winemaker at De Bortoli, a family winery in the Yarra Valley of Victoria, argued that Australia could no longer hope to compete on price alone (*The Economist*, 2008 a).

New Zealand had a bumper grape harvest in 2007. Wine overtook wool exports in value for the first time and it became the country's 12th most valuable export, worth US$610 million. New Zealand Wine Growers (NZWG), a national trade body, boasted that the industry sold about 1 billion glasses (approximately 100 million bottles) of wine in nearly 100 countries. Exports to Australia are booming and New Zealand accounted for over 10% of wines sold in the United Kingdom for more than £5 (US$10) in 2008. Exports to the United States are also increasing. Across the board, demand exceeded supply (*The Economist*, 2008 a).

The first cause of this success is that New Zealand's winemakers are working hard to improve quality and are exploiting the country's unique climate to produce distinctive wines, particularly the sauvignon blancs and rieslings, which benefit from unusually long, cool growing season. The increasing quality of its pinot noir is also attracting attention, while just a few years ago the country was known only for its sauvignon blanc. The second cause of success is that better wines have been promoted with more efficient marketing; for instance, NZWG has been putting on more overseas wine tastings, and many producers also travel to promote their wines to wholesale dealers and distributors. A third cause is that New Zealand's wines are benefiting from advertisements promoting

tourism in the country, that emphasize the freshness and purity of its landscapes (*The Economist*, 2008 a).

In order to attract consumers who are more conscious of environment protection, New Zealand's winemakers have decided to put on bottles a label indicating the quantity of CO_2 emissions produced during the winemaking process and transportation of wine to the site of consumption. Thus, in addition to the grapevine variety and the region of origin, consumers will be informed about the "ecological footprint" of the product (CO_2 emissions by the winemaking process, bottling and transportation), indicated on a small stamp sticked next to the main label. Since November 2010, the New Zealand Wine Growers has been commercializing bottles of sauvignon Mobius with this new indication : a 125-ml glass of this white wine drunk in New Zealand has an "ecological footprint" of 140 g of CO_2. This footprint increases when the wine is exported : in Australia, the same glass of wine will bear a sticker with 190 g of CO_2 (Le Moël, 2010).

New Zealand is a pioneer in this area. The first bottles bearing this kind of indication have been certified by Carbon Trust, a British organization which has set up a logo for carbon reduction to be adopted by companies that are striving to highlight their endeavours towards environment protection. This approach corresponds to the current trend aimed at supporting the environmental labelling of consumer products. In France, this kind of labelling was expected to be applied in 2011, but as an experiment (Le Moël, 2010).

The New Zealand wine making association wished to attract more consumers who want to reduce their ecological footprint. Craig Fowles, in charge of environmental affairs within the association stated : "Our company has been certified carbon neutral since 2006. And an increasing number of consumers are interested in reducing the carbon footprint, even though the price remains a very important factor in the purchase of a bottle of wine." New Zealand wanted to export its Mobius sauvignon to the United Kingdom – one of the key export markets for New Zealand's wines –, but at destination the quantity of CO_2 emissions per bottle will be much higher. Craig Fowles asserted that : "Due to our environmental practices, we are convinced that, even taking account of the impact of transportation, we should be able to compete on this ground with French and Italian wines." However, if European vintners decide to adopt the same environmental approach and the carbon labelling on their bottles, C. Fowles'assertion might be difficult to verify (Le Moël, 2010).

New Zealand's wine industry was expected to double exports between 2010 and 2015, but it faces a few hurdles. Firstly, the appreciation of the

NZ dollar means winemakers must lower prices to remain competitive. It is not easy on the other hand for small producers to find distributors in new markets such as the United States. Another problem is the shortage of labour. Stuart Smith, the owner of Fairhall Downs Estate, a Marlborough vineyard, stated that with 40,000 locals and 20,000 hectares of vineyards, grape growers and winery owners in Marlborough, the largest wine region, needed 3,000 people to prune their vines annually. They have therefore to rely on migrants from the Pacific Islands of Vanuatu and Tonga, but there were not always enough (*The Economist*, 2008 a).

China

Between 2004 and 2008, wine consumption in China increased by 80% and amounted to about 900 million bottles in 2008, 88% of which were red wines. Between 2009 and 2013, a study by Vinexpo-IWSR forecast a 31.6% increase in wine consumption. The consumption of imported wines has been multiplied by four (+308%) between 2004 and 2008. Imported wines represented 12% of the volume consumed and 40% of the value. France was the main supplier of China (Changy, 2010).

In 2005, the Asian giant became a member of the world's top-10-wine-consuming countries. And with annual consumption at a mere 0.7 liter per person – compared with 57 liters in France – there is much room for growth. It was not therefore surprising that analysts foresaw a 36% increase in Chinese wine imports by 2010. European wine producers as well as New World ones are keen to respond to China's demand. At the Vinexpo fair, held in Hong Kong from 25 to 27 May 2010, there was an overall feeling shared by the 1,000 attendants that the future of wine market depended on Asia and the Far East. Since the previous fair in 2008 the number of businesses present and coming from the Asia-Pacific region has increased by 30%. While wine producers and merchants are convinced that Asia will be a driving force of world viticulture, some express their perplexity : the Chinese market is new and nobody knows really what is going to occur; Chinese buyers order without really being aware of the qualities of the wines they are buying; some order a million of bottles of top Bordeaux or 2 million euros of wine just to diversify their investments; the more expensive a wine is, the better it is sold. However, beyond many anecdotes and since Vinexpo 2008, Chinese consumers are showing more expertise (Changy, 2010).

Enophilia may be a natural extension of China's rise. Dozens of luxury hotels and restaurants have been mushrooming in major Chinese cities ahead of the 2008 Summer Olympics. China's fast-growing upper-middle

class (with incomes of US$5,000 to US$13,000) was virtually non-existent two decades ago; by 2005, it made up 9% of the population. By 2025, the Chinese middle class was projected to include more than half-billion people. And the Chinese government is encouraging people to switch from hard rice and other grain spirits to wine, for health reasons as well as to spare crucial foodstuffs. It is not therefore surprising that taxes on many wine imports have plummeted from 120% to about 48% since 2001. Low-end imported wines sell in China for as little as €4 or €5 a bottle. But multimillionaires in the Chinese diaspora, and increasingly nouveau riche mainlanders, often buy wines at hundreds of dollars the bottle. "A bottle of Pétrus or a Château Yquem can sell at any price, because it is a symbol of wealth," asserted James Grégoire, owner of the Château de la Rivière, in Bordeaux (Baker, 2005). In 2010, a bottle of Château Lafite-Rotschild (*1er cru classé* of Bordeaux) has been sold at 10,000 yuans (more than €1,100), compared with 1,000 yuans in 2000. Earlier on, the renowned French winemaker Lafite-Rotschild put on the bottles of its 2008 vintage the number 8 in red colour and in Chinese – a symbol of luck and prosperity – that looked like a reversed "V" above the label. This marketing approach had a striking impact : at Sotheby's auctions in Hong Kong in October 2010,the 2008 Lafite vintage, estimated at 666 dollars, was in fact purchased at 2,860 dollars the bottle.

French wine merchants, facing a declining international market share, are reaching out to China as a potential saviour. Bordeaux is publishing a Chinese-language listing of its main châteaux. The Chinese "still believe French wines are the best," stated Shirley Tan of East Meets West Fine Wines, a Shanghai-based distributor. "It may not become the biggest market," stated Jean-François Bourrut Lacouture, who sells high-end wines to spot markets in Shangai, Hong Kong and Singapore, "but it will be near the top within ten years or so." Although Asia consumed half of the very high-end wines in 2010, it represented only 4% of global wine consumption. It is undoubtedly a promising market for both European and New World vintners (Baker, 2005).

But China is also trying to increase and improve its own national wine production. Serious winemaking was virtually unheard of in this country before the 1980s, when the government began to revive vineyards in the coastal provinces of Shandong and Hebei. Chinese had long preferred *baijiu*, a spirit distilled from grain. But in 1996, health concerns about hard spirits and worries about potential staple grain shortages led the government to renounce *baijiu*. Li Peng, China's Premier, called instead for wine to be used for toasts at Communist Party banquets. Wine was

also mixed with ice or soft drinks just about anywhere. But now one can see people in the main cities drinking with sophistication (Baker, 2005).

With domestic producers unable to meet the new demand, imports of wine poured in. But also investors looking for ripening profits considered that China could become a nation of wine enthusiasts. Dynasty Fine Wines Group, a vintner in Tianjin, that originated in 1980 from a co-enterprise between the French spirit group Rémy Martin and a state enterprise, offered stock to the public in January 2005; retail investors placed orders for 625 times the shares on offer, and the stock surged 43% in a few hours. Rémy Cointreau owned nearly 25% of Dynasty. The company planned to build a wine museum, the design of which was exposed at the Hong Kong Vinexpo fair in May 2010; it will be a mixture of the French châteaux of Versailles and Chambord (Loire Valley), with a replica of the Louvre Museum pyramid. Dynasty's director, Gao Feng, was optimistic about the future of his company, as he forecast a 20% to 30% increase in consumption of red wines in China for the years to come (Baker, 2005; Changy, 2010).

Foreign companies are interested in joint ventures. In addition to Rémy Cointreau, Illva Saronno announced in 2005 it would pay some US$58 million for 33% of a Shandong vintner, Yantai Changyu Pioneer Wine (Baker, 2005). In 2009, the Domaines Barons de Rotschild, which includes all the winemaking businesses of the Rotschild family, set up a partnership with a Chinese public group (Citic) with a view to creating a 25-hectare vineyard near the port of Penglaï (north-east of the country). This joint venture was expected to be very profitable, due to the high demand for the Lafite-Rotschild top wines in China.

China's three largest wine producers – Dynasty, Changyu and China Great Wall Wine – controlled about half of the country's wine market. They have specialized in cheap, mass-produced wines, but they are well aware of the need to produce better wines to rival foreign vintages in order to meet the demand of consumers. Foreign experts also visit China to advise local producers, e.g. an expert from Austria who helps Bodega Langes in Hebei to produce cabernet; the sprawling complex, which has been designed to evoke Vienna's Schönbrunn Palace, owned by Gernot Langes-Swarovski, a scion of the Austrian crystal family, boasts a four-star hotel, China's first wine spa, wooden tubs where visitors can stomp grapes, and a highly technically advanced winemaking facility (Baker, 2005).

Wineries like Grace and Bodega Langes are trying to achieve in one generation what it took most of the other wine-producing nations centuries to develop. "I want to show the world we can produce great

wines," proclaimed Hong Kong tycoon C.K. Chan, who stated he had already invested US$10 million in Grace. Daniel Schuster, an award-winner vintner and pioneer of the New Zealand wine industry, considered these aspirations were achievable. As a judge of Hong Kong's International Wine Challenge in 1999, he found two of the 15 Chinese entrants acceptable; in the 2004 contest, "no less than five or six were in the fine wine category. One of them was absolutely outstanding – Grace Vineyard Tasya's Reserve Merlot 2001…" "if this kind of progression continues, there is no doubt that in ten years China will be one of the major players among 'new wine' countries" (Baker, 2005).

However, a significant challenge is to gain consumers' confidence through strict labelling regulations, control of counterfeit (e.g. disguising cheap imported wines to make up for shortfalls in domestic production). In 2004, a Chinese well-known wine critic questioned if a 1992 cabernet sauvignon from Hebei-based vineyard was truly made from grapes harvested in 1992, whether it was genuinely made from grapes grown in the Changi region of Hebei and whether it was even made by the prestigious Great Wall Huaxia Vineyard. She argued that, in 1992, Huaxia's cabernet vines were too young to produce grapes and too few to account for the seemingly boundless supply of the '92 vintage in Chinese supermarkets. Before writing the article, the wine critic said she took a bottle of Huaxia '92 to a French laboratory, which found that its true vintage was between 2000 and 2002 (Baker, 2005).

A more basic issue is that most Chinese grapevines have not yet matured enough to produce wines worth laying down for more than three years. For a wine to age well, it needs well-developed tannins and a delicate balance of sugar and acid. The best wines come from rigorously pruned vines – a practice contrary to Chinese traditional agricultural methods which seek to maximize fruit growth (while a recommended practice is to have no more than six bunches of grapes per plant) [Baker, 2005].

He Wei, a distributor at Suntime International, a Xinjiang-based winemaker with the biggest vineyard in China, has summarized the future of the wine industry in this country in the following way : "For now, the priority is simply to get China to drink more wine…", "then, when the market is more mature, we will think about promoting the taste." For the time being, mainland China is home of some 300 wineries. Japanese, Italian and French investors own stakes in Chinese wine ventures, hoping they will become global players (Baker, 2005).

India

When an Indian decides to consume alcohol, he generally drinks whisky or beer, imported for some consumers, or locally produced for a majority of them. "Foreign alcoholic beverages made in India" are widely consumed, not without risk, as hundreds of people die every year after being intoxicated by counterfeit or smuggled beverages. Wine was ignored in India till the end of the 20th century, but nowadays there are many vineyards which not only try to meet the local demand, but also to export their wines (Géné, 2007 b).

For instance, Kanwal Grover who spent some time in Europe and learnt to appreciate wine, teamed with Georges Vesselle from the Champagne region in order to test several dozens of grapevine varieties in different locations. He selected a few ones which he planted by the end of 1980s in the Nandi Hills outside Bangalore, at Dod Ballapur. In 2006, Grover Vineyards produced 350,000 bottles of cabernet sauvignon, shiraz, sauvignon blanc, viognier and shiraz rosé. Its vintage Grover Vineyards La Réserve, made in collaboration with Michel Rolland, one of the best-known wine consultants in the world, is among India's most sought-after wines. The 2005 vintage is rich and smoky, with hints of roast peppers; its alcohol is listed at only 12% on a label that proudly highlights the collaboration with Michel Rolland (Géné, 2007 b; Fabricant, 2008).

Kanwal Grover, whose son Kapil is now in charge of the vineyard, will be remembered as the person who made "real" wine for the first time in India, without juices imported from anywhere and blended with table-grape juice as it was usually done. In 2005, Grover Vineyards exported about 70,000 liters of wine a year to France, including the exceptional cabernet sauvignon-shiraz (Géné, 2007 b).

In the State of Maharashtra, central and western India, the city of Nasik, 160 km northeast of Mumbai, has become the centre of India's expanding wine industry. More than 40 wineries are located in that region and were in 2008 at varying stages of development. Government officials stated that investment in wine increased by 74% in 2007. Indus Wines, which is the name the Terroir India company uses on its labels, are made in a boutique winery atop a hillside overlooking Lake Mukni, south of Nasik. The two-year-old winery (in 2008) started planting a vineyard, and bought its grapes from local farmers who, until recently, grew table grapes. The fruit and alcohol of Indus' fresh-tasting sauvignon blanc are well integrated, and the 2007 shiraz exhibits restrained richness. Sula Vineyards, established in 1996 on the outskirts of Nasik, is the brand most often on wine lists. Although

Nasik has a reputation as the Napa of India, Sula was in 2008 one of just a handful of wineries designed to receive visitors with a tasting room, tours and a guest house (Fabricant, 2008).

Another example is that of Rajeev Samant, who returned to India after being a financial director in the Silicon Valley, California; he decided to create vineyards on hundreds of hectares of not very fertile land which his father could not sell in the region of Nasik. He planted grapevines he was familiar with in California : zinfandel, sauvignon blanc, chenin blanc, cabernet sauvignon, shiraz, all imported from the New World. In 1999, he produced a total of 15,000 bottles and in 2006, 1 million. This is considered a success story in India and R. Samant's vintages are appreciated in the best restaurants of India, in particular a 2006 sauvignon blanc (Géné, 2007 b).

Chateau Indage, near Pune, another city of Maharashtra, was 25 years old in 2008 and, with production at 1 million cases, is said to be the biggest winery in the country. It was the first to make a sparkling wine (Fabricant, 2008).

Wine culture is developing quite rapidly in India. In 2006, 7 million bottles have been consumed, one-fourth being imported, and despite customs taxes between 150% and 265%. "In the next ten years there will be 300 million upwardly mobile Indians who can afford wine and for whom it will be a lifestyle choice. A lot of them will be drinking Indian wines," stated Ranjit Dhuru, the owner of the Château d'Ori winery, outside Nasik, who made his fortune in the software business and then decided to invest in the wine industry with the help of a consulting oenologist from Bordeaux. In 2009, R. Dhuru expected his production would reach a million bottles bearing the Château d'Ori label. The optimism of the owner of this winery was easy to understand, as wine consumption was increasing in India. Aman Sharma, the food and beverage director for the India-based Taj hotels, agreed with him: "There is already a large population eager for wine," he said. In 2006, the annual per-capita consumption of wine in India was estimated at about a tablespoon, but that represented a fourfold increase since 2000. Most wine made in India is consumed there. And as wine publications, websites, the first book on Indian vineyards, wine clubs, a wine academy, competitions and tasting dinners have taken hold, gradually, Indian wines with notable finesse are becoming available and appreciated (Géné, 2007 b; Fabricant, 2008).

South-East Asia

Indonesia

Fishermen from Java introduced grape farming to northern Bali in the early 1900s. Nowadays, vineyards protect their grapes from the tropical sun by using an Italian system of cultivating the fruit on small trees. Balinese wine is served on the island as an alternative to pricey imports (Baker, 2005).

When Hatten Wines started production at Sanur, Bali, with Alphonse Lavelle grapes in 1994, experts were incredulous and claimed that the table grape variety was unsuitable for winemaking. They were wrong. Hatten's rosé is medium dry, fresh and pleasantly fruity. Winemaker Vincent Desplat also produces an award-winning semi-sweet wine, Pino de Bali (Beale, 2006).

Thailand

Thailand's Chao Phraya delta was home of 6,500 hectares of wineries in 2005, including a floating vineyard where grapes are harvested by boat. About 80% of the grapes grown are Malaga blanc, a gift from Louis XIV to King Narai the Great of Siam in the 17th century (Baker, 2005).

Siam Winery began life producing blended wine and juice coolers, but lifted its wine quality with the help of French winemaker Laurent Metge-Toppin. Siam's Chatemp label won a bronze medal at the International Wine & Spirits competition in 2001 (Beale, 2006).

Japan

With Asia's most developed winemaking industry, Japan had some 230 producers in 2006. Almost 40% of Japanese wine hails from Yamanashi prefecture, where the nightly temperature drop gives rise to the perfect acidity. The area's oldest vineyard, Katsunuma, is the best known. Its award-winning Château Mercian label, comprises wines made from Japanese and European grapes. In the Katsunuma Valley is also produced the *koshu* – a purple table grape cultivated in Japan since the 12th century (Baker, 2005; Beale, 2006).

Yamanashi is the heart of Japan's viticulture. It is located in the centre of Honshu (Hondo) island and its landscapes are plateaux and deep valleys. This prefecture is well known for its historical figure : General Takeda Shingen (1521-1573) whose legend tells that none of his enemies

had been able to defeat him. The prefecture is also renowned for its production of fruits : persimons, grapes and apples. Since the 8th century, *koshu* has been cultivated in the prefecture's Katsunuma Valley. "These grapes, originating from the Caucasus region, were probably brought to Japan through the Silk Road, at the same time as Buddhism," according to Hideo Suzuki, in charge of Budo no oka, the municipal centre for wine promotion. During the Meiji era (1868-1912), Japan opened itself to Western influence and discovered wine. Katsunuma's authorities sent to France Ryuken Tsuchiya and Masanari Takano to learn French and the secrets of winemaking, in Champagne and Burgundy from 1877 to 1879, during 18 months. Despite this endeavour, wine remained in Japan and until the 1950s a beverage of the poor. This was the time of the sweet wine, that is a mixture of water, wine whose fermentation was stopped halfway, sugar and hard liquor, generally brandy (Mesmer, 2008).

During the 1960s, winemaking techniques improved and the image of wine changed. In 1975, Katsunuma decided to promote viticulture in order to attract tourists. A centre for the promotion of *koshu* production and consumption, Budo no oka, was built on a hilltop (Mount of the Grape); in addition to a winery, it houses a hotel, a museum, and it welcomes 600,000 visitors a year. From the centre's terrace, the visitor can look over the Katsunuma Valley and its vineyards which extend over 170 hectares (Mesmer, 2008).

In the Katsunuma Valley, 25% of Japanese wine is produced. In 2009, 2.5 million hectoliters of wine were commercialized in Japan, made from several varieties (delaware, cabernet, *koshu*) and of uneven quality. Winemakers are working hard to improve the quality of their wines and even to export some of them. For instance, Grace Winery as well as Katsunuma Winery are targeting a slice of the international market, thus trying to benefit from the success of Japanese gastronomy. Grace Winery is advocating a change in the system of grapevine cultivation, moving to the European system. On 25 January 2008, Katsunuma Winery succeeded in exporting its first batch of 480 bottles of a dry white wine with lemon aromas to the United Kingdom. This achievement was the result of a collaboration between the Japanese winery and Bernard Magrez, from the top Bordeaux vintage Pape-Clément, and it was in a way an outcome of the collaboration initiated during the Meiji era between Katsunuma and French winemakers (Mesmer, 2008).

Once of a better quality and recognized as such, Katsunuma wine can be served on European tables. Competition in Japan among wineries is growing, as "all prefectures want to have their grapes and wine; it is

considered a tool for revitalizing the rural areas; Nagano, for instance, has created an appellation that may threaten that of Katsunuma, because the climate is better," stated Naoko Matsuura, chief executive of Cinq-Sens, a company aimed at discovering wine (Mesmer, 2008).

When the leaders of the G8 countries met in Hokkaido in July 2008, they had a toast with a white wine, Cuvée Yoriko, made with the Champagne method in the Takeda vineyard of the northern prefecture of Yamagata. They also could taste white wines from the Yamanashi prefecture. That was a recognition of Japan's increasing know-how in the area of viticulture and winemaking. Japan could boast of having the "world's best sommelier," Shinya Tasaki, who won the award in 1995 (Mesmer, 2009).

Although the image of Japan, with respect to the consumption of alcoholic beverages, remains associated with a wide range of sake and tasteful beers, wine consumption is progressing rapidly : the annual per-capita consumption reached 3 liters in 2009, compared with 0.3 liter 25 years earlier. Of the 2.5 million hectoliters of wine commercialized in 2009, 60% were imported. A culture of wine is being disseminated in the Archipelago, for instance by the Wine Academy, in Tokyo (the equivalent of that of Paris), which was opened in 1987 and attracts some 4,000 students per year. The Cinq-Sens company organizes seminars on the discovery of wine and it has attracted 700 persons since its creation in 2008. The company's chief executive, Naoko Matsuura stated that : "Our students belong to well-off social categories; it is a pleasant leisure for them, and 70% are women" (Mesmer, 2009).

Wines from Takeda, Obuse or Sakaori vineyards are competing with those imported from some 30 countries. New appellations are created by Japanese authorities. New models of viticulture are being developed, e.g. a European system of viticulture where there are less grape bunches per vine (instead of up to 700 bunches per tree in the case of table grapes). As production is small, wineries like that of Haraoro, which produces 38,000 bottles per year, try to attract Japanese consumers to visit the winery, taste and buy the wines, instead of exporting them (because they would be too expensive). Labour is an issue : winemaking in Japan needs three times more labour than in France in order to produce a high-quality wine; but Japanese do not seem to be too much concerned about that, according to Naoko Matsuura (Mesmer, 2009).

And indeed Japanese wine has penetrated the American market and is now exported to Europe. In the latter case, the European Union's rules had to be respected : prove that 100% of the grapes used had

been harvested in the production area. In Japan, grapes from different regions could be mixed, including imported grapes; this is done by big companies like Mercian or Suntory, which flood the market with low-cost wines. The penetration of foreign markets has been facilitated by the international success of Japanese cuisine. For instance, in December 2008, the *shizen koshu* Cuvée Denis Dubourdieu started to be found in French cellars; since March 2009, it has been put on the wine list of Umu, a Japanese restaurant in London that was awarded a star in the Guide Michelin. This vintage, as well as that of Katsunuma Winery (*magrez aruga koshu*) exported to the United Kingdom on 25 January 2008, was made thanks to international collaboration. Denis Dubourdieu is both an enologist specialist of white wines and a passionate of Japan, and Bernard Magrez is the owner of several Bordeaux vintages (including the renowned Pape-Clément) and of vineyards in all continents. Both have built ties and invested in Katsunuma of the Yamanashi prefecture. The latter makes 90% of the 900,000 hectoliters of Japanese wine produced annually (2009). These wines are made from locally grown varieties but also from a high proportion of imported grapes (Mesmer, 2009).

Koshu wine, produced in Yamanashi, is highly appreciated in and outside Japan. There is a war between the vineyards and wine producers concerning prices. The former want to maintain the price of a bunch of *koshu* grapes at between 120 and 220 yens (€1 to €1.90), depending on sugar concentration. In this way, the price of wine will be kept at a reasonable level. A bottle generally cost between 1,500 and 2,000 yens (€13 to €17.50) at Tokyo cellars (2009). Many were of the opinion that a too high price would benefit foreign wines, when competition is already strong. But wine producers do not agree. They claim that *koshu* production has been decreasing, from 14,400 tons in 1990 to less than 7,000 tons in 2009. This decrease is due to the steady fall of acreage devoted to agricultural activities. Another cause is the fact that *koshu* is also a table grape that has medicinal properties (it would be beneficial for the skin). Consequently, many producers grow other vines, such as *kyosho* or *pione*, that are much more costly (Mesmer, 2009).

The crisis is worsened by the fact that 2008 was a bad year : cold and wet summer harmed the quality of the grape harvest. Consequently and despite an agreement concluded in September 2008 to increase the price of a grape bunch by 20 yens (17 cents of a Euro), some wineries had difficulties to find the quantities of grapes needed to keep their activity. For instance, the Marufuji Winery, created in 1890, had bought 52 tons of grapes in 2008, i.e. 30% less than it was customary. Some analysts therefore are afraid that *koshu* production may be stopped, and Mesmer

(2009) recalled that this would be catastrophic for those who honoured divinities at Daizenji, a temple built in 712, according to the legend, by a beggar-priest, Gyoki, who would have taught the inhabitants of the region the cultivation of *koshu* (Mesmer, 2009).

Israel

It is not just the *Bible* that testifies to the winemaking ability of the ancient Israelites. Archaeological evidence of amphoras, wine presses and other objects exists all over Israel. In the 7th century, Islamic conquerors put an end to wine production, and, for the most part, it did not resume until the beginning of the Zionist movement in the 19th century. But actually the Israel wine industry has come of age since the early 1980s or so, driven by advances in technology, improvements in viticulture and the global exchange of knowledge that has brought together winemakers from six continents (Asimov, 2008).

Paradoxically for perhaps the oldest winemaking region of the world, Israel offers no noteworthy indigenous grapes. It produces a wide range of wines from international grapevine varieties, particularly chardonnay and sauvignon blanc among the whites, and cabernet sauvignon, merlot and syrah among the reds. The pricing of the white wines makes them especially attractive : between US$10 and US$18, and generally under US$20 a bottle in 2008. The reds tend to be more expensive, generally more than US$20 a bottle in 2008. The 2003 Carmel shiraz from the Kayoumi vineyard in the Upper Galilee reached US$35 a bottle in 2008; it is a balanced and restrained wine, with pleasing flavours of berries, flowers and licorice (Asimov, 2008).

While grapes are grown all over Israel, many of the best wines tend to come from Galilee, in the northern part of the country encompassing the Golan Heights, occupied by Israel in 1967. Israel does not yet have a strict appellation system, so numerous variations on Galilee can be seen on labels (Asimov, 2008).

Kosher wines have improved considerably over the years, and so many are available nowadays that the issue of whether they are *kosher* is relevant only to observant Jews. There are basically two methods for making *kosher* wines. The first requires, among other things, that the wine be handled out by observant Jews throughout production and until it is poured. A second method consists of using flash pasteurization techniques that minimize the damage done through heating; these wines are labelled as *mevushal*, and they can be opened and poured by anybody. Most

of the good *kosher* and *mevushal* wines are made in Galilee, e.g. the top red 2003 Galil Mountain Yiron (a well-balanced Bordeaux blend with pronounced aroma of violets), the 2005 Dalton Upper Galilee cabernet sauvignon (*mevushal*), the 2006 Barkan classic chardonnay, the 2004 Segal's Special Reserve (with apple and pear flavours, an underlying minerality), and two sauvignon blancs, the 2006 Yarden and the 2005 Carmel from the Ramat Arad Vineyard (Asimov, 2008).

In modern Israel, winemakers had a captive market that demanded *kosher* wines and little in terms of quality. Traditionally, Jews drank mostly for Shabbat blessing and this wine was made sweet because a bottle had to last for several days and still be drinkable. A first wave of change has been brought by the Rothschild banking family who reintroduced grape cultivation at the beginning of the 20th century, mainly in the plains between Tel Aviv and Netanya, around Rishon Le Zion. Bordeaux grapevine varieties were thus introduced. Then, the country's economic boom since the early 1990s has created more of a market for fine food and wine. Israel's reduction of travel taxes prompted a wave of visits to Europe and exposure to good food and good wines. Import taxes were lowered and many of the most famous international wines finally became available in the country. Annual per-capita consumption of wine has doubled over the 1990 and 2000 decades to about seven litres (Echikson, 2008).

Before 1980, Israel counted only about 20 wineries. One company – Carmel – dominated the industry, vinifying about 70% of the country's total grape harvest. Nowadays, more than 200 small wineries are spread across the country, from the Golan Heights in the far north to the Negev desert in the far south. Sweet wines now represent a minority of the market.

By the late 1980s a small group of Israeli pioneers tried to create world-class white and red wines that progressively gained recognition from critics both at home and abroad. It is true that ever more-prosperous Israelis are demanding better drinking choices while connoisseurs in the United States and Europe are intrigued enough to taste these "new" world wines from an ancient land. For instance, Eli-Gilbert Ben-Zaken, a restaurateur and poultry farmer, planted a few grape vines on a hilltop next to his house in the Judean hills in 1988. He chose the name Domaine du Castel after a nearby crusader fortress, and, starting with a mere 600 bottles attempted to make high-quality, France-inspired wines (Echikson, 2008).

Ben-Zaken's Domaine du Castel was the first winery to plant vines in the Judean hills, in the centre of the country. Thereafter, more than 30 wineries

have been created there. The Judean's Mediterranean-style hillsides –where olive groves also flourished– benefit from relatively cool summers, which make them suited to quality winemaking. For his wines E.G. Ben-Zaken has given the hills a French name: Haute-Judée. This winemaker who grew up in Alexandria, Egypt, had initially no experience with wine. In the 1960s his family moved to Italy, where he acquired a taste for fine food and wine. Both himself and his wife, also from Egypt, attended the University of Geneva, which nurtured his Francophile feelings. In 1970, the family settled in Israel, on a small poultry farm about 17 kilometres from Jerusalem, where E.G. Ben-Zaken opened a restaurant called Mamma Mia. "It was the first restaurant serving fresh pasta in Israel," he stated. The restaurant required a wine list and he therefore began to travel to French and Italian regions, and he was inspired to plant his own vines. He adopted a French style in winemaking. His reds blend the five Bordeaux grape varietals led by cabernet sauvignon and merlot. His whites are 100% chardonnays and are made to resemble Burgundies. One of his two sons, Ariel, joined the winery after studying oenology in Burgundy. All Domaine du Castel wines are aged in French oak barrels (Echikson, 2008).

In 2008, the winery had 15 hectares under cultivation and produced 200,000 bottles each year. E.G. Ben-Zaken stopped raising poultry and turned the old chicken buildings into a modern cellar and winemaking facility. Being a secular Jew, at first he did not make *kosher* wines. But many of Domaine du Castel's customers began asking for wines that respect Jewish dietary laws, so he changed his production process to obtain rabbinic approval (rabbis or their assistants should supervise the wine production and animal byproducts such as gelatine, which is used to clarify wine, are prohibited). Despite criticism about the high prices of his wines (over US$35 in 2008), most of Ben-Zaken's vintages are sold out and the winemaker began mimicking Bordeaux growers in selling them en primeur – before bottling, while still aging in barrels. In fact, his wines' high prices reflect the low output and strong demand for top Israeli wines (Echikson, 2008).

Domaine du Castel regularly ranks among the best Israeli wines in international tastings. Its wines are exported to the United Kingdom, the Netherlands, Belgium, Luxembourg, France, Switzerland, Italy, Japan, the United States, Canada and Mexico. E.G. Ben-Zaken often shows off his wines in St. Emilion during the Bordeaux region's annual primeur tastings. Wine author and expert Hugh Johnson awarded Domaine du Castel his highest four-star rating and named the red Castel Grand Vin one of his 200 favourite wines in his 2008 pocket wine book. Robert Parker's *Wine Advocate* magazine gave the domain many of the best notes in its

December 2007 Israeli tasting. R. Parker grades on a 100 point scale, with anything over 90 considered superb. The red 2005 Grand Castel received 92 points, the Petit Castel received 90 points, and white 2005 C Blanc du Castel won 91 points (Echikson, 2008).

In addition to Castel, some of the best names of Israeli wines include Margalit, Tzora, Château Golan and Clos de Gat. Even Israel's largest and oldest wineries, led by Carmel, have invested in new production of high-quality, European-style wines. Carmel has launched the well-respected wineries Ramat Dalton, Zichron and Yatir (Echikson, 2008).

Morocco

In North Africa, Morocco is a good example of the revival of viticulture in a country of moslem faith. When Islam was born in the Arabian Peninsula, local populations used to consume alcoholic beverages made from fermented barley, wheat, maize, dates, grapes, honey. Table grapes have been grown for centuries in moslem countries. As mentionned by Fouad Rhouma in his book published in 1998 and titled *Statut de l'alcool dans l'imaginaire social des musulmans* ("Status of alcohol in the social imaginary of moslems"), the holy book – Coran – warned the believers against the dangers of alcohol, as well as those of money games, and that sin prevailed over usefulness (sourate II, versets 216-219). In sourate V, versets 90-91, wine consumption was prohibited explicitly, as the believers were requested to stand away from alcohol and money games, which are an invention of Satan. In addition to coranic prescriptions, the Prophet's lessons, reported by his companions or biographers, were clearcut : "Drinking wine is not compatible with faith", or "The oration of who drinks wine will not be accepted by Allah (God)" (Géné, 2010 b).

However, in 1955 (just before independence), there were about 100,000 hectares of vineyards in Morocco, whose wine production amounted to over 5 million hectoliters. In Algeria, in 1962 (before independence), there were 350,000 hectares of vineyards which produced 14 to 15 million hectoliters. When both countries became independent, the departure of French viticulturists, land nationalization and a stricter respect of Islamic rules caused a major decline of wine production in these countries and in the Maghreb (North Africa) in general. Vineyards were abandoned, grapevines were uprooted massively, the Rome treaty (European Union) forbade the blending of European wines with those imported from outside the Union, and consequently wine production and trade were ruined. A few years before the end of the 20th century only 50,000 hectares of vineyards devoted to wine production were left across the Maghreb (Géné, 2010 b).

Since the early 1990s there has been a gradual revival of viticulture in Morocco, as investors were encouraged to develop a high-quality wine industry. Some French groups such as Castel, William Pitters (Bernard Magrez) and Taillan responded positively. Even tough today only Castel is still present in Morocco, with more than 2,000 hectares and two production units exporting wine in bulk and bottled in France, the French companies brought their know-how, grapevine varieties and specialists, which all contributed to a revival of Morocco's viticulture. A key person was at the heart of the process : Brahim Zniber, who was almost 90 years old in 2010, owned 2,500 hectares of vineyards and produced 30 million bottles out of the total annual output of 50 million bottles of wine. He produces the only AOC of the country – les Coteaux de l'Atlas, of which Château Roslane is the most renowned vintage (Géné, 2010 b).

During 1960s, B. Zniber owned only a few vineyards; but being the only Moroccan active in this field, he became the chairperson of the winemakers. Half a century later, he was still chairing the Moroccan Association of Grape Producers (ASPRAM). He was able to acquire about 30 cellars left by French settlers. He created his first company, Samavin, the forerunner of the present Celliers de Meknès, headquartered near Meknès – the vineyard region of the country – in the centre-north of Morocco (Géné, 2010 b).

There are a few other winemakers, who also try to produce good-quality wines, drawing on the advantages offered by the climate, soils and grapevine varieties. For instance, a young French winemaker from the Bordeaux region, Jacques Poulain, who came to Morocco in 1977, worked for the company Thalvin – a property of B. Zniber – for more than ten years. He succeeded in making "expressive" white wines at Ouled Thaleb Domain, in Benslimane, 60 km from Casablanca. In December 2009, Jacques Poulain left Thalvin and in partnership with a Moroccan family, who owned the land, made his first grape harvest and wine at the Ferme Rouge Domain, located 60 km from Rabat inland; 2010 was its second vintage in a region located at an altitude of 600-700 meters, where grapevines are growing on clay-limestone soils, under drip irrigation, and where summer temperatures reach more than 40°C. As labour is cheap, weeding is done manually and herbicides are not used. In general, phytosanitary products are not necessary. Grapes are harvested manually at the beginning of August, preferably at night, the main concern of the winemaker being to control temperature and to guarantee a minimal acidity for the wine. After fermentation of the juices, the degree of alcohol often reaches 16°C or even more (Géné, 2010 b).

In another region, north-east of Meknès, a 70-hectare vineyard was created in 2002 at la Zouina Domain by Christophe Gribelin, a young winemaker also from the Bordeaux region. He adopted the grapevine varieties cabernet sauvignon, syrah and tempranillo for the reds, and chardonnay, viognier and vermentino for the whites. His 2007 Volubilia Classico, a blend of cabernet sauvignon (60%), syrah (20%) and tempranillo (20%), is considered a perfect example of French know-how applied to the Moroccan *terroir* (Géné, 2010 b).

Another example of a winemaker who is cultivating small acreages for market niches, with low yields, but who prefers quality over quantity is Charles Mélia. Raised in Morocco, he is the scion of a French family settled in Algeria since 1836. He was a winemaker in the famous region of Châteauneuf-du-Pape, when he decided to come back to Morocco and to settle in the region of Essaouira, which has never been a wine-producing region. In his vineyard located at more than 200 km south of Casablanca, he grew grapevine varieties that are from the French Rhodanian region : grenache, syrah, mourvèdre for the reds; ugni, clairette and bourboulenc for the whites. He therefore created the vineyard that is closest to the equator : "One has to go to South Africa to find vines," he stated. He owns 35 hectares and produces 35,000 bottles a year, under a trade mark, El Mogador – the name given to Essaouira during the French Protectorate. His red, white and rosé wines are certified as "bio" (Géné, 2010 b).

Morocco's annual wine production amounts to about 400,000 hectoliters; 100,000 hectoliters are exported as bulk by Castel to France where they are bottled. A small quantity is exported after having been bottled locally. The rest is consumed in the country, sold in supermarkets, and drunk by tourists whose number is increasing steadily. The annual turnover of the wine industry is around €900 million; the industry employs more than 10,000 persons and procures an annual revenue to the state of about €20 million of taxes (Géné, 2010 b).

In the rest of the Maghreb, viticulture is catching again. Algeria produces 500,000 hectoliters a year and has about a dozen appellations. Tunisia produces 300,000 hectoliters on 15,000 hectares, under the leadership of the Central Union of Viticultural Cooperatives. The annual output of the three countries of the Maghreb amounts to 1.3 million hectoliters, and the quality of their wines is undoubtedly increasing (Géné, 2010 b).

Cider and perry

After the successful rebranding of red wine as a health-enhancing beverage, at least, if consumed in moderation, in 2006, Britain's cider makers were hoping that the same thing might come true for their own drink. Phenolic compounds help give cider its taste, but they also have antioxidant properties. Britain's National Association of Cider Makers has thus sponsored Serena Marks, a researcher at the University of Glasgow, to look into this matter. She first measured the phenolic content of 24 types of British apple cider and found that all of them contained more phenolic compounds than Golden Delicious, a bland variety of eating apple. Some contained ten times as much (Marks et al., 2007).

To check whether the drinking of cider has a beneficial health effect, plasma and urine were collected by the end of August 2006 from 12 volunteers who went away US$95 richer for having had a cider breakfast after 36 hours without consuming any antioxidant-containing food or drink. These samples should reveal how many of the phenolics found in cider were excreted. They should also give clues as to how the rest were metabolized, in other words whether they might give any health benefits. In particular, S. Marks wanted to know if the cidrous molecules that were transferred into the blood were those associated with a reduced likelihood of developing heart diseases, etc. If this likelihood existed, it might mean that there was some truth to the old proverb that "an apple a day keeps the doctor away" (*The Economist*, 2006 a).

Perry (or *pirie*, as it was once called, a derivation from *pirige*, the Anglo-Saxon word for pear) is a delicate drink that had its heyday between the late 17th and early 19th centuries. Because the perry pears (as opposed to table pears) used to produce it are difficult to grow, with orchards taking at least 25 years to yield fruit, production of perry began to decline with the introduction of mechanized farming methods from the late 19th century. There are more than 100 kinds of perry-pear tree (many are rare, with just a handful of trees confined to individual orchards). Colourfully named varietals such as Mumblehead and Merrylegs are used to make either a single-variety or blended perry. While a little perry is made in Wales, the English counties of Herefordshire, Gloucestershire and Worcestershire are its heart land. Since the late 1990s, the dedication of several artisanal producers in these areas has made the drink back on the map. The beverage indeed is enjoying a strong revival, thanks in part to a renewed interest in cider, its close cousin, and a desire by some farmers to return to traditional agriculture (Horsford, 2010).

Perry is not easy to make, not least because pear juice is more difficult to ferment than the apple juice used to make cider. The skill, according to James Marsden of Gregg's Pit Cider & Perry, is judging and balancing ripeness, maceration, pressing, sugar levels and the time that perry is left to ferment in the barrels – usually between three and six months. Some producers add another element to the fermentation by maturing the perry in old whisky, brandy or rum casks. This kind of handmade quality only adds to perry's appeal and places it far from the so-called "pear cider", a mass-produced beverage from large commercial brewers. Tom Oliver, a perry and cider maker in Herefordshire for over a decade, makes nearly 38,000 litres of perry a year, a tenth of which he supplies to the United States, and he stated that "perry is completely different to any other drink; it is more demanding than cider and, less alcoholic than wine, has the same nuances of nose and flavour – notably citrus and hedgerow fruits." In fact, many producers argue that perry should be treated as an equivalent to wine because it goes very well with food (Horsford, 2010).

"Pear cider is an oxymoron since cider is made with apple," stated J. Marsden, who considered there was a relationship between the perry revival and the United Kingdom public's growing appetite for seasonal and authentic produce of known provenance. Bottlings of perry range from sweet to dry, and can be still or slightly sparkling. The 2009 vintage was a particularly good one because of favourable weather conditions (Horsford, 2010).

Whiskies

To make whisky, one needs spring water, peat from the heather land, malted barley, yeasts, and a know-how that has been transmitted over generations. The production of a single-malt whisky, i.e. a scotch whisky produced from a single distillery, comprises several steps. The first one consists of dipping barley seeds in pure water; when the seeds have been soaked for sufficient time, they are spread on the ground by the malting workers; germination starts and thereafter malted barley is dried on fire set and fed with peat – the traditional fuel in Scotland's Highlands – and then thanks to burning coal. Once dried and smoked, malted seeds are ground and mixed with boiling water and lactose in order to start the brewing phase. Yeasts are added in order to ferment the sugars of the whole mixture into ethanol. Fermentation vats with a volume of 50,000 liters (an average size) are made of Douglas fir in the traditional distilleries, while in modern ones they are made of steel. An artichoke

smell comes out from the fermentation vats. The last stage of single-malt production is the distillation made in copper kilns. Finally, the ageing of whisky takes place in old casks of sherry, bourbon whisky, rhum, cognac or port. Generally, these casks are maintained and repaired by a team of craftsmen, attached to every distillery, in a vast workshop. Ageing in these conditions confers over years a flavour and a "character" to every whisky; it also gives to the beverage an amber colour. According to David Mair, who works for The Balvenie, a small distillery of Speyside, set up in Dufftown, in the Grampians, "70% of the quality of a whisky is due to the cask where it matures" (Toula-Breysse, 2010).

On The Malt Whisky Trail, in Scotland's Highlands, there are many renowned whisky distilleries, including The Balvenie, set up close to the ruins of a castle bearing the same name and dating back to the 13th century. The Balvenie is considered a small traditional distillery, which is the only one in Scotland that grows part of the barley it consumes on plots near by. When one travels along The Malt Whisky Trail, the scent of whisky is smelled throughout the heather land, because whisky loses about 2% of its volume every year through evaporation (Toula-Breysse, 2010).

Single-malt whiskies are different from each other, even within the same distillery. One reason is the quality of the casks where they age, another is the number of years of maturation. For instance, The Macallan Fine Oak, 15 years old, is matured successively in three kinds of oak casks (where bourbon whisky and sherry have been "raised" previously); Glenfiddich, 21 years old, terminates its maturation in rhum casks from the Caribbean (Toula-Breysse, 2010).

While a single-malt Scotch whisky is exclusively made from malted barley in a single distillery and is not blended, pure malt whisky is made from the blending of single-malt whiskies produced by several distilleries. Blended Scotch whiskies are made from blending malt whiskies with whiskies produced from maize, wheat, rye and oats grains, and by different distilleries. Blending relies on the know-how of every distillery and gives each whisky its aromatic and colour touch. David Mair considers that "the ideal temperature for drinking a whisky is around 15°C." After having tasted a single-malt whisky dry, whisky lovers add in their glass a small volume of water, so as to discover other aromas (Toula-Breysse, 2010).

Whisky producers like winemakers are privileging quality over quantity and target high purchasing-power consumers, the emphasis being laid on single-malt whiskies and on Certified Geographic Indications.

In October 2008, Paul Kennedy, a history professor at Yale University, wrote in an article on the "unintended consequences" of the financial crisis in the *International Herald Tribune* : "This is not a time to be in Silicon Valley, better by far to be producing single-malt Scotch whisky. At least you can sip it." Even as they cut back on expensive items, such as cars, boats and homes, they can still find a little spare cash for a few bottles of *uisge beatha*, the Celtic drink today known as whisky. Martin Riley, international marketing director of Chivas Brothers, which includes the Glenlivet among its brands, claimed "the world will still continue to want Scotch whisky, it is a very versatile drink." He stated that in 2007 Chivas had been "inundated with orders" after releasing a 25-year-old version of its Chivas Regal blended Scotch whisky, which was selling for US$300 in the United States and US$600 in Russia, a fast-growing market (Wiggins, 2008).

Over the last 20 years (1985-2005), whisky volumes were almost flat, while vodka production rose by 3.5% a year. The explanation is that Scotch whisky, even basic blends, has to mature in a cask for at least three years, while vodka can be distilled at the beginning of the week and bottled, shipped and drunk by the end of the week. As demand went down, many small distilleries closed or were bought by international beverages groups. That led to a change in strategy : a move upmarket which has been paying off since 2008 (*The Economist*, 2010 d).

Although exports of Scotch whisky fell by sales volume in 2008 (down 3%), they were up in terms of sales value. Total shipments rose 13% to £1.86 billion compared to shipments of £1.65 billion in 2007, according to the Scotch Whisky Association. Campbell Evans, the association's director of government and consumer affairs, commented that the drop in sales volumes reflected a decline in shipments of so-called "bulk" whisky (sold to buyers who bottle the whisky under their own labels). But exports of more expensive single malt whiskies have been increasing, up more than 18% in 2008 to £293.7 million, as have exports of blended Scotch whiskies, up 14% to £1.48 billion (Wiggins, 2008).

With the "premiumisation" of both blended and malt whiskies (some of which are aged for 12 years or more), Scotch whisky exports have risen by over 40% in value since 2000. In 2009, records were set : the volume of exports rose by 4% to 1.1 billion bottles worth US$4.9 billion (£3.1 billion) [*The Economist*, 2010 d].

Investment is pouring into Scotch whisky production: about £600 million over the three-year period 2007-2009. New distilleries are set up and

old ones are expanding. In 2009, Diageo, the world's biggest beverages group, which dominates whisky sales with blended brands such as Johnnie Walker and J&B, and Lagavulin, a malt whisky, opened a £40 million distillery near Elgin, the first new one in Scotland for 30 years. Its whisky will come to market in 2012. France's Pernod Ricard, the world's second group with brands such as Chivas Regal, Ballantines and Glenlivet, is investing £10 million in a modernization plan aimed at boosting production of Glenlivet by 75%. It hoped to rival the world's best-selling malt whisky, Glenfiddich, made by William Grant, still an independent firm (*The Economist*, 2010 d).

In the United Kingdom, whisky sales have been stagnating. Charles Allen, global malt whisky director at Diageo, stated that Europe remained a "tough and difficult market" for spirits due to weakening economics and smoking bans, which have kept some people out of bars and pubs, but sales are booming in Asia (Wiggins, 2008).

Diageo, the world's biggest single producer of Scotch whisky, is trying to make single malt whisky more accessible by giving people information on how whiskies taste. It encourages shopkeepers to show customers a "flavour map" which describes what different brands of whiskies taste like, with some tending towards "dry smoke pepperyness" and others to a "floral, herbal, grassy freshness". Diageo is also trying to attract people to single malts with its "Singleton" brand. First developed for sale in Asia, the company links the brand with specific distilleries, such as Dufftown in Speyside, and then sells a whisky called "The Singleton of Dufftown" with information about where it is made. Diageo is selling Singleton in the United Kingdom, as well as in the United States (Wiggins, 2008).

France has become a consumer of Scotch whisky since the end of the second world war, and is now the world's biggest importer of single-malt whiskies ahead of the United States (Toula-Breysse, 2010).

Also a big source of demand for expensive single-malt whiskies is emerging markets. The Macallan distillery, a single malt owned by the Edrington Group, was investing £40 million bringing a disused stillhouse back into production and building two new warehouses to cater for overseas demand. The whisky group Glenmorangie was investing £45 million developing its core single malt brands Glenmorangie and Ardbeg, and pulling back from bulk whisky production (Wiggins, 2008).

An important growth stimulus for the whisky market is coming from South American countries, such as Brazil and Venezuela, where premium

blends are appealing to increasingly affluent consumers. Much of the investment in boosting volume is to build up reserves in the expectation that something similar will occur in India and China. Nowadays, international spirits such as whisky and brandy account for less than 1% of the Chinese market. The Chinese are developing a taste for whisky, although the challenge for Scotland's producers will be to ensure that they choose Scotch whisky. Kavalan is being made by a new Taiwanese whisky distillery which aims at selling its brand on the mainland Chinese market. Kavalan may be fruitier than a smoky single-malt Scotch whisky, in order to appeal to Chinese palates, but the local brand is praised by some connoisseurs (*The Economist*, 2010 d).

Mezcal : a harder spirit than tequila

Santiago Matatlan is the "world's capital" of *mezcal*, a harder spirit than tequila. This quiet city can be reached after a half-hour drive from Oaxaca, the capital of the State of the same name, that is located south-east of Mexico City (one-hour flight). Matatlan is close to the archaeological site of Mitla, where lived Zapotecs, the ancestors of local indigenous people who still speak the *zapotec* language. While tequila is a distillate made from the fermentation of the sap of *Agave tequilera* in the State of Jalisco, north-west of the City of Mexico, *mezcal*'s original plant is an agave, but a different species: *magueys espadinas*, with long, sword-like leaves. Those who produce and drink *mezcal* dream that this "worker's brandy", scoffed by the middle classes, would become as fashionable as tequila. Already some knowledgeable people are requesting this spirit in the bars of elegant areas of Mexico City. Some connoisseurs have even founded clubs with a view to "defending the diversity of local productions, as well as the different varieties of *maguey* against an excessive genetic uniformization" in the plantations that would benefit only a few big producers. Matatlan's small *mezcaleros* are afraid of having the same fate of former *tequileros*, who were eliminated by five millionaire families. While there were more than 300 *mezcaleros* in Matatlan 20 years ago, nowadays there are only about 40 *palenques*, i.e. open-air plots where the agave distillate is produced (Stolz, 2007).

The first stage of the production process consists of pruning at their base all the leaves of *magueys*; the remaining core is called *piña*, because it resembles a big pineapple. Then the *mezcaleros* dig a five-meter-diameter hole, which is internally surrounded by layers of stones and wood; the *maguey piñas* are piled up in the hole like walls, and the oven thus made is covered by a layer of earth. During three days, the *piñas* are cooked slowly. On the fourth day, the cooked pulp is crushed with a horse-drawn stone press; from the crushed and very sweet orange pulp

a brownish liquid is recovered and is fermented in wood barrels. On the seventh day, the mixture of fermented juice and pulp is distilled (this is the major difference with the production of tequila, where the pulp is taken away and is not incorporated to the distillation process) in brass cauldrons put on stoves fuelled with wood. The craft process' output is not more than 200 liters per day, with a concentration of alcohol of 40 degrees Gay-Lussac (Stolz, 2007).

A French connoisseur, François-Nicolas d'Epoisse, has been attracted by this *mezcal*, "100% agave", with a smoky taste, which he commercializes in France under the label Ultramarine. This is the result of a triple distillation and the quantities available are therefore limited (Stolz, 2007).

After a very bad year 2006 for the local economy and the 70% downfall of *mezcal* sales, the craftsmen who survived (and were able to clean their *maguey* fields three times a year) had set up marketing networks in Mexico and in the United States, not only for *mezcal* but also for weaved fabrics. Indeed, weavers living at Teotitlan del Valle perpetuate their craft by using natural dyes : pomegranate for obtaining yellow-green colours, indigo for deep blue, cochineal – extracted from mealybugs living on cactuses – for crimson-red hues (Stolz, 2007).

NUTRITIONAL AND HEALTH CLAIMS FOR FOODSTUFFS : ALLEGATIONS AND DEEDS

Using a healthy food or a drug?

Over a century ago, Elie Metchnikoff, a renowned Russian biologist, started drinking sour milk every day, having concluded that it could promote longevity. His work at the Institut Pasteur in Paris was later developed in the 1930s by Minoru Shirota, a Japanese microbiologist who isolated the bacterium used in the drinkable yogurt developed by Yakult, the Japanese food company, in the middle of the 20th century (Birchall, 2007).

In the United States, when Dannon, the American subsidiary of Groupe Danone, first attended conventions of gastroenterologists in 2000, it found a sceptical audience that was in favour of prescribing a drug than a yogurt to persons having gastro-intestinal ailments.

Dannon has tried to launch Activia yogurt since 2003 among the American medical community. The probiotic yogurt contains patent-protected live bacteria that have been shown in clinical tests to ease digestive problems or constipation. While Europeans and Japanese have been consuming probiotics in increasing numbers, the idea of bacteria delivering functional health benefits was relatively new to the United States, which also consumes less yogurt overall. According to the figures from Euromonitor International, Americans were expected to consume about US$5.4 billion worth of yogurt in 2007 – a third of the US$17.4 billion in Western Europe. While the sales of probiotic yogurts reached US$3.2 billion, that was about four times the American consumption (Birchall, 2007).

In the first year of its launch, Dannon's Activia, with its patent-protected *Bifidus regularis* culture, delivered retailed sales of about US$200 million. In 2007, the company was following up with the launch of DanActive, the American version of the yogurt sold as Actimel in Europe, which it says is "clinically proven to naturally strengthen the body's defence system." The company came to the conclusion that broad trends such as an increased

focus on wellness would support the launch, even if initial consumer tests showed a less than enthusiastic response (Birchall, 2007).

In 2006, in order to promote academic research, Dannon worked with rival Yakult to fund a symposium on the health impact of probiotics at Harvard Medical School's Division of Nutrition. At the same time, its consumer marketing is based on a strong functional message that brings it closer to pharmaceutical marketing. The company is now facing competition, a sign of the American consumers' changing tastes, hopes Dannon. General Mills' Yoplait launched Yoplus, marketed with a "unique Optibalance" mix of special cultures and fibres. In 2006, Kashi, an American natural food company, launched a probiotic cereal, also aimed at "digestive wellness." Dannon has reacted by promoting its clinical credentials, highlighting the results of research trials proving two bacteria's effectiveness – setting the stage for a likely battle over the medical benefit of patent-protected individual microbial strains (Birchall, 2007).

Probiotics and the difficult assessment of health benefits of microflora

Probiotics can be defined as live microorganisms that contribute to restoring the balance of intestinal bacteria and raising resistance to harmful microbes. Taken in sufficient amounts and over long enough periods, they can promote digestive health and comfort. However, only a few probiotics have been proved effective in clinical trials. For instance, *Lactobacillus* is a probiotic that can be supplied in the form of a number of bacterial strains : *Lactobacillus* GG (often called LGG), which can be found in the diet supplement Culturelle as well as several milk products in Finland; *L. casei* DN 114001, included in Dannon products; and *L. casei* Shirota, found in Yakult, a popular probiotic beverage from Japan. Studies have shown that all these strains are associated with reducing diarrhoea; LGG has also shown a benefit in treating atopic eczema and milk allergy in infants and children. Both LGG and Dannon's *L. casei* strain have been shown in studies of children attending day care to reduce illness (Parker-Pope, 2009).

After agreeing to reimburse dissatisfied consumers with a US$35-million settlement and to make labelling changes (among them adding the scientific names of probiotic strains it uses), Dannon stated that it settled the suit to avoid litigation and that it stood by all of its product claims. The company's website lists numerous scientific studies of its patented probiotic strains. It is, however, difficult to exactly specify the health benefits of probiotics. A panel of 12 experts who gathered at Yale University concluded that there was strong evidence that several probiotic

strains could reduce diarrhoea, including that associated with antibiotic use. Several studies have also suggested that certain probiotics may be useful for alleviating the irritable bowel syndrome, with the strongest recommendation for *Bifidobacterium infantis* 35624, the probiotic in the Procter & Gamble supplement Align (two members of the panel had ties to Procter & Gamble; three others had ties to other companies selling probiotics) [Parker-Pope, 2009].

Scientists continue to debate whether probiotics offer a meaningful benefit to the immune system. Barry R. Goldin, professor at Tufts University who helped discover LGG, but no longer receives royalties from the patent, stated : "The evidence for the general immune strengthening is just not there." Consequently, health-conscious consumers interested in probiotics should look for products that list the specific strain on the label and grant readers easy access to scientific studies supporting the claims (Parker-Pope, 2009).

Intestinal microflora contains about 100,000 billion bacteria, corresponding to 500 species. Their knowledge is still limited, but it is progressing through major international cooperative research projects. Their physiological role is important, as they eliminate wastes, degrade part of the cholesterol, transform some drugs into bioactive compounds, make more digestible some milk proteins, increase the acidity of the intestinal environment (which inhibits the proliferation of pathogens), and enhance the assimilation of vitamins and minerals. Intestinal microflora is therefore essential to the good functioning of the human body; it is formed at the age of two years, and it could deteriorate when nutrition is unbalanced, or further to stress and fatigue, and following recurrent treatments with antibiotics. Consequently, the consumption of probiotics (i.e. bacteria in foodstuffs or in food supplements) could improve the reconstitution of intestinal microflora (Amalou, 2006 a).

Recent research work has shown that these ingested bacteria could reduce the risk of infection, decrease digestion illnesses (e.g. inflammation of the intestinal mucosa, bloating, uncomfort), stimulate the child's immune system and have a positive effect on some allergies. But more research is needed in order to clarify the exact role of intestinal microflora (Amalou, 2006 a).

However, commercialization of some products by agrifood corporations has been successful. For instance, Danone's Actimel has been well received by the public : in 2005, in Europe, the sales of small bottles of this probiotic amounted to €1 billion. This product contains billions of live

bacteria belonging to the species *Lactobacillus casei*. Danone claimed that its product could be effective against diarrhoea. But it should be recalled that the health benefits of such product (sold at a higher price than a yogurt) would be observed only if three to five bottles are consumed daily, bearing in mind that each bottle of this very sweet product brings in 80 kcalories. Danone has been able to obtain the approval of the French Agency of Food Sanitary Safety (AFSSA) for Actimel (Amalou, 2006 a).

To provide a health benefit, a probiotic must comply with the following criteria : it should contain the highest number possible of bacteria; these bacteria should be alive when the product is consumed (this property cannot be checked when the product is bought); and the bacterial species ingested should be adapted to the illness or dysfunctioning to be treated. For instance, the good results obtained with *Lactobacillus rhamnosus* GG in the treatment of eczema, cannot be extrapolated to another strain. It seems that many probiotics do not contain enough bacteria, or bioactive compounds that are sufficiently resistant to stomach acidity, to be really effective. That is why the consumption of probiotics should follow a physician's advice (Amalou, 2006 a).

On 15 April 2010, while its shares fell by 1.56% on the Paris stock exchange, Danone announced that it was withdrawing its requests for a health-benefit labelling for its Activia yogurts and its fermented milk brand Actimel; these requests were filed at the European Food Safety Authority (EFSA). Both products made up almost 25% of the annual turnover of the French food company : €2.6 billion in 2009 for Activia and €1.2 billion for Actimel (Mamou, 2010 a).

Regarding Actimel, the request to EFSA was to authorize or not the mention that the product improved the defensive reaction of the intestine and could help in the mitigation of severe diarrhoea. With respect to Activia, the product could be advertised as a facilitator of digestion and intestinal transit. Following the withdrawal, Danone announced it was changing its communication policy and will not mention any health benefit for its dairy products, Activia and Actimel. Danone nevertheless indicated that the request for withdrawal was temporary and that the company was waiting for a meeting with EFSA in order to clarify the evaluation criteria. On its side, the communication office of EFSA asserted that the evaluation rules have been clear since 2006, that the scientific work is complex, and it also recognized that the selection criteria could be improved. It is, however, clear that Danone did not want to repeat the misfortune about Immunofortis, a food ingredient whose supposed medical benefits (improvement of newborns' immune defences) were not approved by

EFSA. Nestlé and Unilever, like Danone, have been confronted with the strict regulation enacted by EFSA (Mamou, 2010 a).

It is therefore obvious that food companies which have adopted the consumer's health as a strategic approach, should invest more important funds into clinical trials in order to substantiate their future requests for health-benefit claims. In the case of Danone, its strategy focused on products that are good for the body. In 2007, Danone sold its biscuits LU, and purchased Numico, a firm specialized in medical and child nutrition. In addition, the market of dairy products, where Danone is the world leader, is characterized by the transition from conventional products to nutraceuticals. If therefore Danone cannot enjoy the positive impact of health-benefit claims for its widely distributed products, it runs the risk of being less competitive versus similar products sold under distribution brands. The challenge is great, because the European market of "healthy" foodstuffs has grown by 25.6% over five years, from €100.1 billion to €125.7 billion (2009). In France, the market value rose from €15 billion to €17.3 billion, a 15.6% increase (Mamou, 2010 a).

Dietary recommendations and disease prevention

Food safety : a top priority

In the United States, the Centers for Disease Control and Prevention estimated that 325,000 people a year were hospitalized and that 5,000 died as a result of food-caused illnesses (2009). For instance, during the summer of 2010, salmonella contamination at one egg producer sickened nearly 1,500 people and led to the recall of more than half a billion eggs. In 2009, a disease outbreak that sickened 700 people and killed nine was traced to a processing plant in Georgia where state inspectors repeatedly had found unsanitary conditions but did little or nothing to force a cleanup. In the case of schoolchildren, a *USA TODAY* investigation found that for six years, federal inspectors discovered unsanitary conditions at a tortilla (a maize crepe) plant in Chicago that supplied the school lunch programme and was repeatedly associated with outbreaks of vomiting and diarrhoea in schoolchildren. Officials finally moved to shut the plant down in 2009. The investigation also found that the government's standards for the hamburger meat it bought for the schoolchildren in the lunch programme were lower than those routinely enforced by fast-food restaurants (*USA TODAY*, 23 November 2010, p. 8A).

Regulators often lack sufficient authority to do what is needed to protect the public. When internal testing at one peanut plant found salmonella,

for instance, the owners kept testing until the findings were negative, then shipped the product. The government had to use a bioterrorism statute to force the company to publish the positive tests (*USA TODAY*, 23 November 2010, p. 8A).

A measure pending in the Senate to improve the food-safety system with bipartisan backing (12 Democrat co-sponsors and eight Republicans), was scheduled for vote after 25 November 2010. The bill was so common-sensical that it even had support from the staunchly antiregulatory US Chamber of Commerce, the Grocery Manufacturers Association and cereal maker General Mills. Among other things, it would give regulators the power to order recalls of hazardous food, ending companies' ability to delay or refuse action. The bill and its House of Representatives-passed counterpart would also require regulators to inspect food facilities at least twice as often as they did before the approval of the bill (*USA TODAY*, 23 November 2010, p. 8A).

An opposing view expressed by Tom Coburn (2010), a medical doctor and a Republican senator of Oklahoma, was that the Senate should reject the false assumption that growing government meant safer food and instead market forces that worked should be promoted. He considered that the United States had the safest food supply in the world, and it had never been safer. The rates of food-born illnesses have been declining for more than a decade. The so-called FDA Food Safety Modernization Act of 2010, according to him, would expand duplicative and ineffective food-safety bureaucracy. The Government Accountability Office had called this bureaucracy "high risk due to (its) greater vulnerabilities to fraud, waste, abuse and mismanagement."

T. Coburn opposed the "250 pages of new bureaucracy and regulations." He was of the opinion that expanding the Food and Drug Administration will harm small businesses and raise prices at the grocery store – all without having a meaningful impact on food safety. He added that for the past 100 years, the free market, not the government, had been the primary driver of innovation and improved safety; consumer choice was a far more effective accountability mechanism than government bureaucracies. T. Coburn was offering an alternative bill to require better coordination among agencies and improve outdated information technology systems and other processes agencies use to protect food supply. His plan leveraged the free market by utilizing private inspections and allowing FDA to focus on bad actors (Coburn, 2010).

Whatever the viewpoint one may have on the need to regulate more strictly the innocuity of foodstuffs, it should be stressed that nowadays globalization of food trade as well as the increasing range of possible routes of food contamination make necessary the systematic control of food safety from farm (and factory) to fork. See also Sasson (2009).

Consumption of healthy foodstuffs and change in lifestyle for reducing risk of illness

Francis Larra, professor of medicine and president of the French League Against Cancer, has warned about such kinds of statements : "Eating broccolis will prevent cancer," or "If I had eaten curcuma, I should not have been affected by cancer." Instead of recommending anticancer foodstuffs, F. Larra prefers to talk about foods that are "protection factors" or "risk factors". We do not know actually if a foodstuff can act on the defence and repair mechanisms of cells. By contrast, there seems to be a consensus about the fact that Mediterranean diet (Crete's diet) has beneficial effects with regard to the prevention of cancer. Fruit and vegetables play a key role because they inhibit the accumulation of free radicals in cells. Similarly, food fibres, e.g. those contained in whole bread, reduce the contact between oncogenic compounds and the intestinal mucosa, which lowers the risk of digestive tract cancers. On the contrary, the repeated consumption of saturated fatty acids is a risk factor for the development of colorectal, uterus, prostatic, pancreas and kidney cancers. Also obesity increases the risk of colorectal, kidney and prostatic cancers (Larra, 2010).

F. Larra supports that healthy food consumption should accompany chemotherapy or radiotherapy. However, diet is not the only factor to be taken account of. Exercise has an important impact, because it decreases the amount of estrogens, which are directly associated with hormone-dependent cancers : breast, uterus and prostate. F. Larra recalled that France was one of the best performing countries with respect to cancer treatment (55% of cancers are cured), but it was not faring well in terms of disease prevention. Finally, regarding the relationship between cancer and stress, F. Larra is very careful, as such relationship has not been proved. Cancer is a multifactorial disease and duration is an important factor : for instance, a breast cancer would appear after ten years. It is also true that the state of knowledge evolves and that what we know currently about cancer and foodstuffs will change with more research being carried out on this complex relationship (Larra, 2010).

In an updated version (2010) of his best-seller, *Anticancer* (initially published in September 2007 by Robert Laffont Publishers in Paris, translated into 35 languages, and of which 1 million copies have been sold), David Servan-Schreiber (also called "DSS"), a professor of clinical psychiatry at the University of Pittsburgh, seems to be more assertive than F. Larra concerning the virtues of some foodstuffs to fight cancer. In 1993, D. Servan-Schreiber had been informed, while working in his own laboratory, that he had a brain cancer. In the first version of his book*, Anticancer* : *les gestes quotidiens pour la santé du corps et de l'esprit* ("Anticancer . Daily gestures for the body and mind health"), he narrates how he survived and reached a health status that he had never acknowledged before. He gave a long interview to Christophe Labbé and Olivia Recasens (2010) of the French weekly magazine *Le Point*.

He stated that nowadays at least 40% of cancers could be avoided by changing eating habits and the level of physical exercise. In France, this would represent 108,000 cases of cancer that could be avoided annually. Such statement, according to DSS, would apply to a large extent to cardiovascular diseases, diabetes and Alzheimer's disease. DSS mentioned in the first edition of his book (2007) the results of a study of women who suffered from breast cancer. Part of these women had followed a programme comparable to that proposed by DSS in his book, including nutrition, exercise and stress management. Eleven years later, these women had a mortality rate that was 56% lower than that of women who did not change their behaviour. DSS considered that such a result was comparable to those obtained with the most effective treatments administered to prevent the relapse of cancer. For instance, the drug Herceptin reduces the mortality rate by 50% and is effective among only 20% of women. DSS raises therefore the question why this approach that seems more effective and has no secondary effects, is not systematically proposed in anticancer centres (Labbé and Recasens, 2010).

DSS considers that big American corporations which spend about 60% of their net profits (after taxation) in health-insurance funds for their employees, are increasingly trying to make their staff more aware of uptaking healthy food, exercising and managing their stress. The French physician reports that every dollar invested in this approach delivers a profit of six dollars in terms of health-care expenses. He even suggests that the French social security could devote 0.5% of its annual budget, i.e. €1.5 billion, to this preventive approach [i.e. the equivalent of what was spent in 2009 to control the epidemic of A(H1N1) influenza]. DSS goes on to state that nobody questions the key role of food and eating habits in health. It is not just the question to have a balanced

and diversified diet, but DSS emphasizes that some foodstuffs are more protective than others. For instance, curcuma is as effective as ibuprofene, the most prescribed antiinflammatory drug worldwide, or green tea (a few cups a day) halves the risk of breast or prostatic cancer. Also a good balance between omega-3 and omega-6 fatty acids can protect against cardiovascular diseases. The concept of "food synergy" is important : tomato and broccoli, which combat cancer through distinct mechanisms, have a synergetic effect when they are consumed together. In other words, according to DSS, the beneficial impact on health of the Mediterranean diet is not due to olive oil alone, but to the mixture of foodstuffs that are part of that diet, fish, aromatic herbs and vegetables (Labbé and Recasens, 2010).

If some foodstuffs are protective, others are risk factors. For instance, according to DSS, sugar is involved in cardiovascular diseases, it plays a key role in the development of breast cancer, even more than substitutive hormone treatments. It is also an important factor in the extension of diabetes, as well as a risk factor in Alzheimer's disease. In France, the average annual consumption of sugar is 37.5 kg per person. DSS strongly recommends the reduction of this addiction to sugar, and to try other sweeteners that are less harmful, e.g. fruit, acacia honey, agave syrup or coconut sugar (Labbé and Recasens, 2010).

DSS also points out that inorganic phosphates used as food preservatives could stimulate cancer cells, particularly in the lung, as shown in mice. Calcium phosphate or phosphoric acid are found in soft cheeses (used for spreading on bread), meat and meat products, industrial ice creams, sodas and fruit syrups, pastries. In the 1990s, an industrial food diet would bring about 470 mg of inorganic phosphates per day; nowadays, this intake could reach 1,000 mg per day. DSS considers that a person treated for a lung cancer should be informed about this risk in order to reduce the intake of inorganic phosphates (Labbé and Recasens, 2010).

DSS mentions two studies the results of which were known in 2009. The first study concerned women where the risk to develop breast cancer was 80%, because they had both genes BRCA1 and BRCA2. With a diet rich in vegetables and fruit, the risk could be lowered to 25%. The same observation was made in the case of men genetically predisposed to inflammatory prostatic cancer. If they ate fish twice a week, they could almost eliminate the risks of cancer associated with their genes (Labbé and Recasens, 2010).

DSS agrees with F. Larra when he emphasizes the role of physical exercise. When the latter is associated with a healthy diet, the synergy is very

effective in decreasing mortality due to cancer. DSS has quoted the results of the work carried out by researchers of the universities of San Diego and Stanford : among women treated for breast cancer for nine years, and adopting both a healthy diet and exercise, mortality was divided by four, while the beneficial effect was very limited among women who adopted only one of the recommendations (Labbé and Recasens, 2010).

To sum up, the recommendations of D. Servan-Schreiber regarding the change of lifestyle (diet, exercise, hygiene) would enable people to avoid 40% of cancers in France. In this country, one man out of four, a woman out of three will develop a cancer during their lifetime. Cancer is the first cause of mortality among men, and the second cause of overall mortality after the cardiovascular diseases. It is still the first cause of a premature death. About 150,000 deaths due to cancer are recorded in France annually, and more than 70% of cancers are associated with eating habits and behaviour (Labbé and Recasens, 2010).

Nutrition and food-intake recommendations are therefore :
- consumption of fruit and vegetables (organically produced; otherwise, peel and wash them thoroughly);
- replacement of red meat by fish, vegetables, legumes, tofu (soybean curd), poultry (without the skin) and eggs (bio);
- consumption of olive, flax or canola oils, instead of sunflower, maize or soybean oils;
- consumption of milk products (bio) rather than industrially produced ones, yogurts with lactobacilli, soya milks or yogurts;
- replacement of white bread, flours, pastries, rice biscuits and industrial cereals, by whole bread, flours containing several cereals;
- consumption of whole rice, from Thailand or India (basmati), quiñoa, pasta made from a mixture of cereals;
- substitution of potatoes by sweet potato and yams; discard industrially made potato purées;
- reduction of salt consumption and seasoning with aromatic herbs or spices;
- avoidance of fried, smoked and preserved foodstuffs;
- careful consumption of foods that are too grilled, because oncogenic compounds could be synthesized during the process;
- replacement of refined sugar by honey or by coconut flower sugar, and of fruit preserves or fruits in syrup by fresh fruit;
- avoidance of industrial sodas and industrially made fruit juices;
- drink filtered tap water (in intensive agriculture regions), using a carbon filter or reverse osmosis, or bottled water;
- a maximum of one glass of red wine per day (Labbé and Recasens, 2010).

D. Servan-Schreiber has underlined the high risk of cancer associated with the consumption of red meat. In 2006, Harvard University Department of Epidemiology has concluded, on the basis of a longitudinal study of 91,000 nurses carried out for 12 years, that the risk of breast cancer among premenopause women was twice higher among those who consumed red meat more than once a day, compared with the risk among those who consumed red meat less than three times a week. DSS considers therefore that the risk of breast cancer could be halved just through the reduction of red meat consumption. In Europe, a large survey called EPIC, of more than 400,000 persons living in ten different countries, has led to the same conclusion with respect to the colon cancer : the risk is twice higher among those who are big consumers of red meat, compared with those who eat less than 20 g a day (the consumption of fish, rich in omega-3 fatty acids, halves the risk of cancer). It is not clear whether the risk associated with the consumption of meat is due to organochloride contaminants present in the fat of slaughtered animals, or to the way of cooking meat (e.g. heterocyclic amines that are formed when meats are grilled too much, or nitroso compounds used in the preservation of cured meats and which are well known oncogenic substances); it may also be due to xenoestrogens in plastic recipients where meat products are stored and transported. Finally, another explanation may be related to the fact that big meat eaters generally consume less anticancer foodstuffs, e.g. vegetables and fruit (Labbé and Recasens, 2010).

By contrast, it is well known that meat and milk products, as well as carnivorous fish, which are at the top of the food chain, expose human beings to oncogenic contaminants, such as dioxin, PCBs (polychlorobiphenyls) and some pesticides, which remain in the environment for very long periods. This kind of exposure to oncogenic pollutants could reach 90% of total exposure of humankind to these substances. In the case of France, vegetables contain 100 times less oncogenic contaminants than animal products, and, according to DSS, organic milk is less contaminated than conventionally produced milk (Labbé and Recasens, 2010).

D. Servan-Schreiber raises more controversial issues when he tries to establish a relationship between cancer and stress. In his opinion, reducing stress, e.g. through reasonable physical exercise, building strong family and friendship relationships, could be as effective or even more effective than an anticancer medical treatment. He cites his own experience and the ways and means he used to recover from his cancer. Consequently, in addition to adopting a healthy Mediterranean, or Asian diet, and to exercising at least 30 minutes a day, he recommends to master one's stress (meditation, sharing of emotions with friends and relatives, sufficient sleep, no smoking) [Labbé and Recasens, 2010].

In his crusade in favour of healthy nutrition and health-conscious eating habits, D. Servan-Schreiber has also highlighted the key role of vitamin D. In addition to helping the fixation of calcium and potassium in the bones, vitamin D may have a protective effect against some cancers, cardiovascular diseases, and stimulate the immune system. Sun exposure for 20 minutes per day of the face, arms and hands, is considered sufficient to the metabolic production of vitamin D at the level of the skin; thereafter, it can be stored in muscles, liver and fat tissue. Storage made during summer can supply the body during the winter. But another source of vitamin D is in the food intake : it is mainly found in fish, seafood and cod liver oil. It is not therefore necessary to consume food additives or foods artificially enriched with vitamin D; eating fish and/or seafood with reasonable skin exposure to sunshine, is sufficient to meet the body's needs in vitamin D. However, DSS considers that in France the daily intake of vitamin D, officially recommended, is too low, and he strongly suggests to increase it considerably, particularly for old persons and for people with black complexion (because their skin synthesizes four times less vitamin D than people with light complexion). His suggestion also applies to populations living in regions that receive less sunlight. With the help of some thirty renowned researchers, DSS has launched a campaign aimed at raising the awareness of physicians and the large public, on the dangers of vitamin D deficiency. By so doing, he reiterates the advocacy role he played for the consumption of omega-3 and omega-6 fatty acids (Labbé and Recasens, 2010).

In order to meet the body's needs in vitamin D, it is recommended to :
- expose the face, arms and hands to sunlight for 20 minutes per day (this brings in 10,000 to 20,000 international units -IU- of vitamin D);
- consume cod liver oil (440 IU), or raw herring (900 IU), or salmon (500 IU) or canned sardines (300 IU), canned tuna fish (230 IU); eggs (40 IU); calf liver (14 IU). These quantities of vitamin D (IU) are calculated per 100 g of the food item ingested (Labbé and Recasens, 2010).

Anticancer food diet

David Khayat, head of the oncology department at the hospital La Pitié-Salpêtrière, Paris, published a book titled *Le vrai régime anticancer* ("The True Anticancer Diet") in 2010, where he tries to elucidate the relationship between nutrition and cancer, questioning a number of ideas about supposedly carcinogenic or non-carcinogenic foodstuffs. D. Khayat was the first director of the French National Cancer Institute, which was created by the former president Jacques Chirac in order to implement the

national plan for combating cancer (2002). He explains that writing this book is primarily motivated by the concern about cancer prevention. He considers that prevention is not sufficient in France, a country where one man out of two and one woman out of three suffer from cancer during their lifetime.

D. Khayat asserts that "no foodstuff is sufficient to trigger a cancer, and no one can prevent cancer for sure... " "A cancer sometimes develops over 15 years, and drinking liters of pomegranate juice will not stop a cancer cell that is developing; maybe it could slow down the process." It is very difficult to determine the specific role of each of the 25,000 biocompounds existing in current diets, and it even becomes impossible to do so when these compounds are mixed in our meals, because their combinations are numerous. In addition, we are not all equal versus the cancer risk and versus the impact of food. Each individual has his/her own genetic make-up, which influences the susceptibility to the wide range of cancers, as well as the interaction between his/her diet and the protection against disease (or not).

Eating five servings of fruit and vegetables per day, as it is hammered by health authorities since the early 2000s, is not considered a mantra by D. Khayat. He mentions a study published in the United States, at the beginning of April 2010, in the *Journal of National Cancer Institute*, the conclusions of which are that leeks, pears and cabbage would decrease the risk of cancer by only 2.6% among men and by 2.3% among women. In other words, the impact is almost nil if one takes into account the methodological uncertainties. This vast study has involved some 500,000 Europeans and its results contradict the slogan of the French National Programme on Nutrition and Health (PNSS). However, pesticides that are sprayed on fruits and vegetables, if not eliminated, can be very harmful, and they are certainly carcinogenic. In 2007, a French study indicated that 7.2% of vegetables and 8.5% of fruits sold on the markets contained amounts of pesticides that were higher than the accepted standards. Eating organic fruit and vegetables will be a solution, but it is an expensive one. Peeling them or washing them out is cheaper, but not all pesticides are water-soluble. D. Khayat suggests to clean them with soap and then rinse them abundantly. He also considers that consumers must keep eating fruit and vegetables, because their action against cardiovascular diseases and diabetes is not questioned. Being hypocaloric, they are indirectly anticancer, because obesity increases the risk of cancer.

D. Khayat (2010) suggests to eat fruit and vegetables according to their respective pigments which have specific virtues : consume those of orange and yellow colour in the morning (e.g. orange, mango, grapefruit,

pear); red and white ones during the whole day (tomato, red cabbage, garlic, onion, soybeans); and the green ones in the evening (broccoli, cabbage). Better avoid the purple and blue ones at night because of their frequent acidity (eggplant, blackberries).

Among the fruits, pomegranate (*Punica granatum*) seems to have all qualities. Both the skin and juice are very rich in antioxidants, and in this respect "its action would be three or four times that of red wine or green tea;" and D. Khayat (2010) considers that pomegranate is "one of the most powerful foodstuffs for the prevention of cancer." Consumed as juice produced industrially it is very efficient : it would slow down the multiplication of carcinogenic prostatic cells, and would have some favourable effects on some breast cancers.

Regarding orange juice, the consumption of which is very high in France, as well as across the world, D. Khayat (2010) expresses some concern with respect to a probable relationship with malign melanoma. The number of new cases of melanomas diagnosed annually is steadily increasing. In fact it doubles every ten years in developed countries. In the United States, for instance, there was one case of melanoma per 1,500 inhabitants in 1935, while in 2000 it rose to 1 per 75 inhabitants; and this trend continues. For a long time, it was thought that this skin cancer was due to a prolonged exposition to sunlight of young children. And it is true that very young children should never be exposed directly to sunlight, particularly between 11 am and 4 pm. But apparently this is not the only cause. In 2009, dermatologists of the University of Tennessee published a study that raises the possible role of orange-juice consumption in the steady increase of the number of melanoma cases. Orange juice is rich in various furocoumarins and particularly in psoralenes, which are highly carcinogenic for the skin in the presence of sunlight. According to the American researchers, there is a very good correlation between the curve of orange-juice consumption and that of melanoma incidence. D. Khayat does not conclude that the consumption of orange juice should be forbidden. He thinks, however, that persons who may develop a melanoma (e.g. with white complexion, blond or reddish hair, blue or green eyes, who quickly become red under the sun instead of acquiring a sun tan; or those who have many patches (naevi) on their skin or whose a close relative suffered from a melanoma) should refrain from drinking orange juice until more research clarifies the issue (Khayat, 2010).

D. Khayat (2010) also questions the relationship between the consumption of red meat and colon cancer. It used to be asserted that eating 100 g of red meat per day would increase by 30% this type of cancer; and

that probably explains the decrease in red meat consumption by French people, down to 50 g per capita per day, compared with 140g per capita per day in the United States. D. Khayat quoted two studies that demonstrated this recommendation was not well founded : on the one hand, California's adventists of the seventh day, who are vegetarians, run the same risk of developing a colorectal cancer as the non-vegetarians; on the other hand, the study published in 2007 in the *Journal of the National Cancer Institute* and concerning the survey of 500,000 Europeans between 1992 and 1993, led to the same conclusion. D. Khayat (2010) underlines that it is not so much the consumption of red meat that would be the cause, but the way it is cooked. The French oncologist considers that grilled meat or fish is unhealthy : at high temperatures, meat or fish give rise to heterocyclic amines and polycyclic aromatic hydrocarbons, which are highly carcinogenic. The French Agency of Food Sanitary Safety (AFSSA) recommends to avoid the direct contact between meat and fire, because such contact induces at 500°C the synthesis of these carcinogenic compounds. Grilled meat should therefore be consumed with moderation and barbecues should not become too frequent. It seems that eating raw meat (28% of French people do so) or fish (1 French person out of 8 eats raw fish) or eating meat that is not well done (60% of French people do so) is a good habit (Khayat, 2010).

Some food items, whose health benefits have been known a long time ago, are now recommended on the basis of scientific studies. Thus, curcuma, the main spice of curry, is highly recommended by D. Khayat (2010), because it can detoxify carcinogenic compounds and inhibit the multiplication on many cancer cell types, and induce their apoptosis (suicide). It is recommended in association with soybean sprouts or green tea, and as a seasoning of vegetables, rice or fish with olive oil and black pepper. The French oncologist also praises garlic and onion, which contain allicine, a strong antioxidant, antiviral, anticarcinogenic and detoxifying compound. Garlic should be sliced into small fragments in order to exert its therapeutic effect, e.g. in the prevention of colon and stomach cancers (40% reduction of the risk of cancer among big consumers). Quercetin, which is found in hot yellow peppers, cocoa and particularly in capers, is a flavonoid that has the proven capacity of reducing the carcinogenic effect of tobacco and smoking. Finally, green tea is known for its antioxidant properties; a polyphenol, known as EGCG, can induce the apoptosis of cancer cells, inhibit the formation of blood vessels (angiogenesis) in tumours and reduce the dissemination of metastases. For instance, the regular consumption of green tea for at least three months has enabled persons having precancerous lesions in the mouth to halve the risk of developing a real cancer. It seems, according to D. Khayat, that Japanese green tea is

even more effective than Chinese green tea, and that its therapeutic effect is enhanced when it is associated with dried papaya leaves.

Regarding food supplements, selenium is a micronutrient quite effective against prostatic cancer. It can also decrease the risk of lung and colon cancer. It should be nevertheless taken on medical prescription. D. Khayat (2010) is very sceptical regarding the dietary intake of vitamin E. He quotes a study carried out by the American National Cancer Institute that has been interrupted, when the researchers realized that patients taking this vitamin had a higher proportion of prostatic cancer than those receiving a placebo. A similar study on beta-carotene had been interrupted in the 1990s in the United States, when it was discovered that this compound (provitamin A) increased by 30% the risk of lung cancer among smokers or former smokers. There had been also some controversy about the role of beta-carotene in the prevention of liver cancer; various studies have shown that it was not the case. It is not advisable to take beta-carotene pills, or in vitamin cocktails, as some people do, but to consume vegetables or fruit which contain beta-carotene, as well as xanthophylls (yellow carotenoid pigments) and flavonoids.

D. Khayat's recommendations, beyond the consensual ones concerning exercise, balanced and diversified diet, focus on men and women and the cancers they can develop. In the case of men, he recalled that 71,000 cases of prostatic cancer had been recorded in France in 2009, i.e. far ahead of any other male cancer. He highly recommends eating tomato, rich in lycopene that may reduce by 30% the risk of prostatic cancer, as well as that of mouth, oesophagus, stomach and lung cancers. Industrially made tomato products, like tomato juice, tomato sauce or ketchup, are even more effective, because they contain more lycopene. Associated with olive oil, tomato is healthy for men, as are most of red vegetables and fruit, or white ones (garlic, onion, shallots). After 50 years, dairy product consumption should be reduced; milk, yogurt and cheese, consumed in high amounts, would significantly increase prostatic cancer. The reasonable recommendation is to eat cheese with wholesome bread and vegetables in order to accelerate intestinal transit (thanks to the fibers contained in cereals and vegetables) and prevent the inflammation of intestinal mucosa. Overweight must be avoided, because fatty tissue cells (adipocytes) produce male hormones that may stimulate prostatic cancer cells (Khayat, 2010).

Breast and uterine cancers (linked to female hormones) are generally diagnosed after the menopause; but they start developing much earlier, between 18 and 50 years. Consequently, preventive measures must be taken, including eating habits and a good nutritional behaviour.

Green and white vegetables, such as cabbage, broccoli, cauliflower and soybeans, are highly recommended. Tofu, derived from fermented soybeans, is rich in phytoestrogens, which may reduce the risk of breast cancer. Also women must consume more fibers, not only to keep off overweight, but also because colon cancer is becoming more frequent among them (in France, 18,500 cases in 2009, compared with 21,000 cases among men). Consequently, they must eat more of green vegetables, garlic, banana, chicory, onion, asparagus, and wholesome bread. All these fiber-rich foodstuffs speed up the intestinal transit and reduce the contact between the intestinal mucosa and potentially carcinogenic compounds. The consumption of dairy products, three servings a day (milk, yogurt or cheese, with reduced proportion of fat), is highly recommended, in order to meet calcium needs. Overweight should be avoided : adipocytes produce female hormones that may stimulate breast cancers, henceforth the need for exercising. Menopaused women ought to avoid overweight and too fatty foodstuffs, while consuming dairy products not only meets calcium needs, but also helps preventing osteoporosis (Khayat, 2010).

Cognitive effects of foodstuffs; role of antioxidants

Antioxidants and brain function

Fernando Gómez-Pinilla, professor of neurosurgery and physiological science at the University of California, Los Angeles, has been studying the effects of food on the brain for years and completed a review in July 2008 published in *Nature Reviews Neuroscience* that has analyzed more than 160 studies of food effects on the brain (Gómez-Pinilla, 2008).

On 20 January 2007, *The Lancet* published research results showing that folic-acid supplements – sometimes taken by pregnant women – can help those between 50 and 70 years old ward off the cognitive decline that accompanies ageing. In a study lasting three years, Jane Durga, of Wageningen University in the Netherlands, and her colleagues found that people taking such supplements performed better on measures of memory, information-processing speed and verbal fluency. That, plus evidence that folate deficiency is associated with clinical depression, suggest eating spinach and orange juice, which are rich in folic acid (Durga et al., 2007).

Another suggestion from F. Gómez-Pinilla's review is that people should eat more antioxidants-containing food or drink beverages also rich in these substances. Vitamin E, for instance, a powerful antioxidant found in vegetable oils, nuts and green leafy vegetables, has been associated

(in mice) with the retention of memory into old age, and also with longer life. F. Gómez-Pinilla underlined that brain is peculiarly susceptible to oxidative damage. It consumes a lot of energy, and the reactions that provide that energy also generate oxidizing chemicals. Moreover, brain tissue contains a great deal of oxidizable material, particularly in the fatty membranes surrounding nerve cells. That suggests the value of a diet rich in berries, which have strong antioxidant effects, although only a small number of their constituents have been evaluated in detail. Polyphenols have been shown in rodents to reduce oxidative damage and to boost the ability to learn and retain memories. In particular, these chemicals affect changes in response to different types of stimulation in the hippocampus – a part of the brain that plays a crucial role in the formation of long-term memories, and which is the region most affected by Alzheimer's disease. Another polyphenol, curcumin, reduces memory deficits in animals with brain damage. It may be no coincidence that in India, where a lot of curcumin in consumed (it is the pigment that makes turmeric yellow), Alzheimer's disease is rarer than elsewhere. Although the precise action of antioxidants is not well known, it is likely, according to F. Gómez-Pinilla, they protect the synaptic membranes. And many of the nutrients associated with brain function are known to affect transmission at the synapses (*The Economist*, 2008 d).

Docosahexaenoic acid (DHA), an omega-3 fatty acid, for instance, provides membranes at synaptic regions with "fluidity," i.e. the capacity to transport signals. It also provides "plasticity" – a synapse's capacity to change. Such changes are the basis of memory. Since 30% of the fatty constituents of nerve-cell membranes are DHA molecules, keeping the DHA concentrations topped up contributes to having a healthy brain. Indeed, according to the studies reviewed by F. Gómez-Pinilla, the benefits of omega-3 fatty acids include improved memory and learning, and resistance to depression and bipolar disorder, schizophrenia, dementia, attention-deficit disorder and dyslexia. These fatty acids are found in oily fish such as salmon, as well as in walnuts and kiwi fruit, and there is a strong negative correlation between the extent of which a country consumes fish and its levels of clinical depression. On the Japanese island of Okinawa, for instance, people have a strikingly low rate of mental disorder – and Okinawans are notable fish eaters, even by the standards of a big fish-eating country like Japan. By contrast, many studies suggest that diets which are rich in trans- and saturated fatty acids, such as those containing a lot of deep-fried foods and butter, have bad effects on cognition. Rodents put on such diets show declines in cognitive performance within weeks (*The Economist*, 2008 d).

In the past few years, several studies have examined the effects of adding omega-3 fatty acids to people's diets – particularly those of children. One such, carried out in the British city of Durham, was controversial in that it was funded by a maker of children's omega-3 supplements and did not include a control group being given a placebo. Work by other researchers, however, has suggested such supplements do improve the performance and behaviour of school-age children with specific diagnoses such as dyslexia, attention-deficit disorder and developmental coordination disorder. Moreover, although more work is needed to elucidate the effects of omega-3 fatty acids on healthy school-age children, F. Gómez-Pinilla stated that younger children whose mothers took fish-oil supplements when pregnant and while they were breast-feeding, do show better cognitive performance than their unsupplemented contemporaries (*The Economist*, 2008 d).

To sum up, eating well is one key to a healthy brain. But one should not overeat, as this puts oxidative stress on the brain and risks undoing all the good work antioxidants can do.

Phase-2 proteins and their inducers

A number of proteins tend to either directly or indirectly scavenge strong oxidants or to decrease the probability of production of strong oxidants; these proteins are referred to as phase-2 proteins and, since most of them are enzymes, they are commonly referred to as phase-2 enzymes. The latter include the classical ones such as NAD(P)H: quinone oxireductase 1, glutathione S-transferases A, M and P families, UDP-glucuronosyl transferases, as well as the more recently defined phase-2 proteins : ferritin H and L chains; cystine/glutamate antiporter, peroxiredoxin I, heme oxygenase 1; L-gamma-glutamyl-L-cysteine ligase, metallothioneins, etc. All these proteins directly or indirectly inhibit strong oxidant formation, e.g. ferritin through sequestering iron, or promote strong oxidant scavenging, e.g. NAD(P)H: quinone oxireductase. Phase-2 protein genes are coordinately upregulated through activation in their promoter regions. Phase-2 protein inducers can be found in our diet : kaempferol, a flavonoid fraction found in blueberries/cranberries; enterolactone, a metabolite of the principle lignan secoisolariciresinol diglucoside found in the flax seeds; ellagic acid found in strawberries and raspberries/blackberries; the flavolignan silibinin obtained from milk thistle (*Silybum marianum*) fruit; sulforaphane, the isothiocyanate metabolite of the glucosinolate glucoraphanin (Juurlink, 2003).

There is much evidence that dietary intake of such phase-2 protein inducers can increase phase-2 gene expression in a number of tissues and that such induction can decrease the incidence of chemically induced tumours. At the Plant Biotechnology Institute (PBI, National Research Council of Canada, Saskatoon, Saskatchewan), Juurlink and colleagues have been working on the phase-2 protein-inducing isothiocyanate derivatives of certain glucosinolates, 4-methylsulfinylbutyl glucosinolate, commonly known as glucoraphanin. The Canadian researchers have shown that dietary intake of glucosinolates that give rise to phase-2 protein-inducing isothiocyanates can reduce hypertension in the spontaneously hypertensive stroke-prone rats; in addition, oxidative stress and inflammatory changes in various tissues in the ageing rats was downregulated (Juurlink, 2003).

The NRC-PBI's researchers also examined the effects of administration of the flavonoid quercetin in a neurotrauma model. They found that quercetin administration after spinal cord injury promotes retention of function, correlated with decreased inflammation. Not only is quercetin a very selective kinase inhibitor, but it is also known to be a phase-2 protein inducer (although more than an order of magnitude higher concentration of quercetin is required for this activity than is the case for sulforaphane) [Juurlink, 2003].

In collaboration with Shawn Ritchie and Dayan Goodenowe, Juurlink has studied the effect of broccoli sprouts containing glucosinolates that are converted into phase-2 protein gene-inducing isothiocyanates on the metabolic profile and they have seen pronounced effects in liver and other organs (Juurlink, 2003).

In summary, we are beginning to understand how phytochemicals influence metabolism and gene expression. Since many phytochemicals can have multiple actions such as activating signal transduction pathways that directly or indirectly alter gene expression or influence protein function that result in adverse metabolic reactions, one must use multiple approaches to understand how phytochemicals either individually or in combination affect us. Henceforth, a combined metabolomic/proteomic/genomic approach is required (Juurlink, 2003).

Consumers' scepticism and the need for regulation

The trend in favour of functional foods may be thwarted by consumers' scepticism and regulatory disapproval. For instance, in 2009, Coca-Cola was sued by a consumer group over health claims made for its Vitamin

Water brand. Danone also faced a similar lawsuit over its yogurts. On 18 September 2009, the firm settled that case, admitting no wrong-doing, but agreeing to set up a US$35-million fund to reimburse dissatisfied yogurt-eaters. When Cheerios, a popular cereal brand owned by General Mills, tried in 2009 to claim on its box that it was "clinically proven to reduce cholesterol," the US Food and Drug Administration (FDA) decided that the company had gone too far. Also the decision made by the "Smart Choices" coalition to endorse sugary cereals had attracted criticism. On 21 September 2009, an American Congresswoman, Rosa DeLauro, sent a letter to the FDA demanding an investigation to see whether unhealthy products were being "misbranded" (*The Economist*, 2009 b).

In France, it was estimated that the number of food supplements available on the market amounted to 28,000. Before commercialization, these products must receive an agreement by the minister of industry. However, despite this strict regulation, the results of an enquiry carried out by the General Directorate of Consumption, Competition and Fraud Repression (DGC-CRF) in 2008 showed that 35% of the companies that had been inspected, were not applying the regulation correctly. Five hundred enterprises were inspected, labelling of food supplements was checked in order to verify if it reflected their quality and chemical analysis. Of the 1,760 verifications made, 220 were considered unsatisfactory. The enquiry revealed a significant proportion of defects in quality control and traceability. The DGC-CRF advised consumers to remain "vigilant" and requested them not to associate food supplements claiming similar effects in order to avoid risks of overconsumption. It also warned consumers about products sold outside the European Union, and to refrain from buying products with labels not written in French.

The French consumers' association UFC-Que choisir ? also warned consumers about some food supplements which it considered irrelevant and sometimes dangerous. The association studied 33 products belonging to three groups of mostly sold food supplements and containing vitamin C (nine products), omega-3 fatty acids (12 products) and "fitness" products (12).

Scientific studies on the real nutritional benefit of food supplements generally lead to carefully formulated conclusions. According to experts, persons in good health and having a balanced diet do not need to consume additional vitamins or antioxidants. It may be an issue for patients who could suffer from deficiencies due to nutritional problems (e.g. anorexia, alcoholism) or from particular diseases such as the ageing-associated macula degenerescence (for which some food supplements

may be recommended). In 2008, the large-scale analysis of the data from 67 studies led to the conclusion that uptaking vitamin C for long periods was not beneficial, nor was that of selenium (present in tuna or calf liver). Another conclusion was the uptake of vitamins A or E, or of beta-carotene, would result in a very slight increase in the risk of premature death.

There is nevertheless an interest in functional foodstuffs, i.e. foods that bring a real benefit for the health of consumers, in addition to meeting nutritional needs, quantitatively and qualitatively. The interest is in conformity with the old precept of Hippocrates of Cos (ca.460- ca.377 BC, author of the *Corpus Hippocratum*), considered as the father of "Western" medicine : "*Your food should be your first medicine.*" And it is true that trying to confer a health benefit or healthy characteristics to a foodstuff has been a prominent feature of the history of food search and supply.

Food quality is also closely associated with food safety, e.g. after European consumers have been confronted with the very negative implications of the bovine spongiform encephalitis (BSE) or "mad cow disease," or of the presence of dioxine in chicken meat. Henceforth, the emphasis on traceability of food "*from fork to farm,*" a wide-ranging programme of the European Commission since the early years of the 21st century. Food should be safe, i.e. innocuous, and also healthy. It should be therefore submitted to very strict control measures, made by independent authorities.

Correlation of genetic markers with food quality

In 2002, the Maryland (Savage)-based biotechnology company, MetaMorphix, acquired the livestock genotyping business of Celera Genomics, a company founded in 1998 by Craig J. Venter to sequence the human genome; it then joined up with Cargill, Inc., to commercialize a genetic test that will help to reveal, prior to slaughter, a cow's propensity to produce desirable meat. That task is being carried out by analyzing thousands of so-called single-nucleotide polymorphisms (SNPs) in the bovine genome. A SNP is a place where the genomes of individual animals vary by a single nucleotide. SNPs are therefore convenient marker versions of particular genes, and different versions of genes result in differences between animals (*The Economist*, 2004).

MetaMorphix and Cargill tried to find out which SNPs were associated with variations in meat quality, such as flesh colour, amount of marbling, wetness and tenderness, so that these could be identified before slaughtering an animal, and suitable animals will thus be reserved

for breeding. In 2002-2003, Cargill studied 4,000 cattle, trying to correlate MetaMorphix's genetic markers with meat quality – and with other important traits, such as growth rate. Almost 100 useful SNPs have been identified from this study. As a result, a prototype testing kit was to be used by the firm as of August 2004. The first 'designer meat' produced this way was expected to be marketed in 2005 (*The Economist*, 2004).

Species-specific DNA markers can be used to identify animal species such as commercial molluscs and crustaceans, which represent a high proportion of aquacultural species. For instance, in Thailand, at the National Centre for Genetic Engineering and Biotechnology (BIOTEC) Marine Biotechnology Unit, species-specific markers based on 16S ribosomal DNA (rDNA) polymorphism have been developed for penaied shrimps, tropical abalone and oysters.

The black tiger shrimp (*Penaeus monodon*) is the most commercially important cultured species in Thailand. Because of outbreaks of diseases, the white shrimp (*P. vannamei*) has been introduced into Thailand and cultured commercially. On the other hand, external characteristics of *P. monodon* and *P. semisulcatus* are similar, but the growth rate of the latter is approximately three times slower than that of the former. In addition, *P. merguiensis* larvae, which could not yet be successfully cultured, were sold as those of *P. vannamei*. Species-specific markers were therefore developed for identifying the afore-mentioned species and *P. japonicus* as well. These markers can be applied to ensure quality control by properly labelling traded shrimp larvae.

These species of tropical abalone are found in Thailand's waters : *Haliotis asinina*, *H. ovina* and *H. varia*. However, *H. asinina* is the most productive one, as it provides the highest ratio (85 per cent) between meat weight and total weight. *H. asinina* specific markers based on 16S rDNA polymorphism have been developed in order to prevent supplying the wrong abalone larvae for the industry as well as foster quality control of abalone products from Thailand.

Oyster farming has shown rapid growth in Thailand. Taxonomic difficulties relating to Thai oysters have had a limiting effect on the culture efficiency and development of their closed life cycle. Molecular genetic markers have therefore been developed to identify the three commercially cultured oysters, *Crassostrea belcheri*, *C. iredalei* and *Saccostrea cucullata*.

European Union's regulation

In order to streamline health allegations or claims for a wide range of manufactured foodstuffs, and to help the European consumer make his/her choice among those which are really healthy, in order also to harmonize national regulations, the new European set of rules concerning "nutritional and heath claims of foodstuffs" have been issued on 1 July 2007 in all Member States of the European Union (Blanchard and Amalou, 2007).

This new regulation adopted in December 2006 after long and difficult negotiations with the representatives of the agrifood industry, highlights the need to develop, between July 2007 and January 2010, a European Union registry of "authorized" claims for foodstuffs. Until January 2010, any product already on the market will continue to be commercialized. Even before the official list is issued, some claims that are often used are on the annex to the regulation with the conditions for their use. For instance, a product whose manufacturer claims it has "a low content of fats" should contain less than 3 g of fat per 100 g of product. Saturated fatty acids and trans fatty acids should not be present at a concentration of more than 1.5 g per 100 g an should not contribute more than 10% of the energy content of the product. The label "low content of sugars" should entail no more than 5 g of sugar per 100 g (2.5g if it is a liquid). "Low energetic value" should entail a maximum of 40 kcal per 100 g (20 kcal if it is a liquid). "Rich in fibers" should contain at least 6 g of fibers per 100 g. A "Light" label is acceptable only if there is at least a 30% reduction compared with a similar non-light product. Finally, the label "Source of proteins" will apply only if at least 12% of the energy provided by the foodstuff is supplied by proteins (Blanchard and Amalou, 2007).

But most importantly, a *new* product that claims to be "good for health" should bring the proof scientifically. This quality will be prohibited as of January 2009, for too fatty, salty and sweet products. The evaluation of health-benefit claims will be carried out *a priori*, i.e. before the commercialization of products, by the European Food Safety Agency (EFSA, Parma, Italy). This new measure is considered of major importance, because many nutritional claims are not substantiated by solid scientific data and consequently cheat the consumers. In France, for instance, the French Food Sanitary Safety Agency (AFSSA) is regularly requested to evaluate the validity of such claims *a posteriori* by the ministry of finance's General Directorate for Competition, Consumption and Fraud Repression (DGC-CRF). An example of such an evaluation was that of a soybean-based beverage enriched with vitamin D and calcium, to be consumed by menopausal women, or that of a wheat cookie, containing chocolate enriched with vitamin B9 and iron,

which claimed a health benefit for procreating women. It is also the case of products that can help losing weight or claim their benefit for child's development. The DGC-CRF was in charge of recapitulating a list of 400 mentions that did not seem reliable, to be delivered to the European Commission in January 2008 (Blanchard and Amalou, 2007).

All these measures lead to the fact that a foodstuff should be evaluated like a medicine, and a nutrivigilance should be put in place like pharmacovigilance. And it is expected that the new regulation based on the scientific proof of a health benefit will lead to the authorization of claims relating to the reduction of a pathological risk. For instance, "calcium promotes bone density" could become "calcium reduces the risk of osteoporosis." Finally, more products that induce body slenderness, as claimed in their commercial labels, will have up to January 2022 to prove that they really do what they promise (Blanchard and Amalou, 2007).

Clinical tests or trials are therefore indispensable; they will be imposed by law in the European Union. They are costly and they will increase the price of nutraceuticals or really functional foodstuffs. For instance, the company BioPolis, in Valencia, Spain, is trying to cut costs and time of these clinical tests, by working on *Caenorhabditis elegans*, a tiny worm, whose genome had been sequenced and that shares many genes with the human genome. The same approach is valid for a zebra fish. Another advantage is the high number of generations in a relatively short time as well as the detailed knowledge of the development process of the worm. One could therefore assume that the study of the impact of a specific food compound can be facilitated and pave the ground to the tests to be carried out subsequently on mammals and humans.

To sum up, the current situation shows that consumers are very much interested in the improvement of the quality of their foodstuffs and particularly their potential benefit for health. Functional foods providing scientifically proven health benefits have a promising future and could represent large markets. But the regulations are becoming more stringent in order to control abuses, false labelling or promises, and to respond to the consumers' need for unbiased and scientifically based information. In the case of the European Union, the increasing stringent regulation of food quality and safety (*lato sensu*, i.e. not only innocuous, but if possible providing a benefit for the good health of the consumer) applies not only to foodstuffs produced and processed in the European Union, but also imported from outside the Union. Biovigilance will be applied to the safety of foods as it is done for drugs.

That is why and for the first time in Europe, the French National Agency for the Sanitary Safety of Environment and Work (ANSES, *Agence nationale pour la sécurité sanitaire de l'environnement et du travail*) has launched on 9 December 2010 a nutrivigilance programme, aimed at identifying the hazards to which consumers may be exposed, when an increasing number of "enriched" foodstuffs are being sold in the supermarkets or on the Internet. The pilot stage of this programme was initiated in September 2009 and concerned food additives (about 30,000 of them were commercialized); it will be extended thereafter to foodstuffs and beverages enriched with nutrients such as vitamins, minerals, amino-acids, plant extracts, etc., and to novel foods. Consumption of these foodstuffs or ingredients was inexistent in the European Union countries before 1997. About 237 novel foods had been authorized in 2007 in Europe in conformity with the new regulation on novel foods, e.g. guar gum, noni juice, dehydrated pulp of baobab fruits (Santi, 2010).

Foodstuffs that targeted specific populations such as infants, persons suffering from some food intolerance, people practising sports, will also be scrutinized. According to Marc Mortureux, director-general of ANSES, "the evolution of food supply and dietary habits requires nutrivigilance, e.g. the behaviour of certain teenagers who consume large volumes of energetic beverages." The system that is set up by ANSES aimed at better protecting the consumer and could be compared with that of pharmacovigilance which concerns the consumption of pharmaceuticals. It is considered as a novel part of the current sanitary safety system existing in Europe (Santi, 2010).

ANSES recalled that for the vast majority of the French population a balanced diet brings in all the nutrients needed for the healthy functioning of the organism, and that there was no need to consume food additives. However, an adult out of five (19.7%) and one child out of ten (11.5%) consumed food additives at least once a year. Women used to consume them twice more often than men (23.6% compared with 12.6%). ANSES has signalled a number of undesirable effects further to the ingestion of food additives : for instance, products containing alcoholic extracts of yam drew the attention of ANSES' experts, and it was recommended to study the chemical profile, composition and toxicity of these products as soon as possible (by those who manufactured and commercialized them). In addition, the General Directorate for Competition, Consumption and Fraud Repression (DGC-CRF) was expected to take appropriate measures (Santi, 2010).

Since the nutrivigilance programme had started, ANSES received 123 reports concerning undesirable effects due to the consumption of food additives. More than 50% of these effects were associated with food additives that are supposed to reduce weight and to improve vision (23% of purchases). Marie-Christine Favrot, head of ANSES directorate of evaluation of nutritional and sanitary risks, explained that the aim of the nutrivigilance programme was "to increase awareness of health professionals (physicians, dieteticians, ophtalmologists, etc.) so as to send back the relevant information when they suspect a relationship between the consumption of food additives and effects on health." Also ANSES wishes to increase awareness of manufacturers, when the Syndicate of dietetics and food additives (80% of the French market) asserted that the effectiveness of these products had been demonstrated (Santi, 2010).

Food industry between pleasure and health care

For food industrialists, who have to compete with large distributors or retailers (e.g. hypermarkets and big retailer stores), innovation is costly, but crucial to let them increase their sales. In France, for instance, in 2007, the big "brands" have emphasized the concept of "pleasure," but creativity, based on the concept of "health" has been the most dynamic. At the International Food Fair (SIAL), held at Paris-Villepinte until 23 October 2008, the consultancy XTC, specialized in the analysis of the launching of new products, reported that 25.8% of innovative products worldwide had put emphasis on "health", compared with 21.7% in 2006 and 16.6% in the early 2001. Innovation emphasizing "pleasure" made up 42% of marketed products. But in the United States, for the first time, "health" innovations (37.7% of marketed products) were more numerous than those emphasizing "pleasure" (Clavreul, 2008).

Companies' commitment to producing healthier foodstuffs

Kraft Foods' hostile bid for British confectioner Cadbury has been making headlines since September 2009. Kraft Foods' chief executive officer, Irene Rosenfeld, explained to *Time*'s Barbara Kiviat in March 2010 that the maker of Jell-O, Maxwell House and Tang, based in Northfield, Illinois, bought Cadbury because gum and chocolate tended to have faster growth rates. The second benefit was that it greatly expanded Kraft Foods' geographic footprint, particularly in developing countries, e.g. Mexico, India, and enabled the company to have an infrastructure through which other snacking products could be sold. Thirdly, where Kraft was very strong in traditional grocery channels, Cadbury was very strong in impulse channels like convenience stores (Kiviat, 2010).

Kraft Foods' CEO saw the greatest growth in the company's snacking, confectionery and quick-meal products. Cookies, crackers, brands like Oreo continued to grow at a double-digit rate. This was indicative of the fact that people were looking for quick meals, because of their mobility and lack of time to cook. It was also found that the company's lower-sugar, lower-fat product versions were experiencing very attractive growth rates (Kiviat, 2010).

Irene Rosenfeld stated that the role of the company was to help the consumer to make informed choices. Kraft Foods launched a 25%-reduced-sugar CapriSun in 2009. It also continued to make sure that its lower-fat salad dressings and cheeses taste good for the consumers. It announced a 10% reduction in salt across its North American portfolio over the two-year period 2011-2012. At the same time, appropriate technologies are available to add whole grain to a number of the company's products without compromising taste (Kiviat, 2010).

One of the best ways for Kraft Foods to know what people will want to buy next was by studying restaurant menus. That is typically a leading indicator of what consumers are eating. For instance, the company has a whole programme around using Philadelphia cream cheese to make Alfredo and other white sauces, since that is a growing trend in the restaurant world. The company spends much time talking about who the target is for a particular product and what the lifestyle of the targeted people looks like. For products where the company thinks it has more teenage audiences, it focuses on digital media to advertise the relevant products. When other peer companies were reducing their investment in advertisement, Kraft Foods made some significant investments in aggregate in its advertising spending in 2008-2009. In fact the company continues to see growth for a number of products that offer very good value, despite the recession, particularly in the Asia-Pacific region and in Latin America (Kiviat, 2010).

In France, it seems that food industrialists are still emphasizing the "pleasure" concept (49.5% of innovative marketed products compared with only 15% for "health" innovative products). However, according to the consultancy Precepta, healthy foodstuffs (bifidus yogurts, mineral or vitamin-enriched foods) offer a real growth opportunity in a rather gloomy market (Clavreul, 2008).

By mid-July 2010, Fleury-Michon – the French leading corporation in the production and sales of cured and salted meats – signed with the health ministry a charter through which it committed itself to reduce the salt

content of its products (precooked meals, ham, surimi) by 6% to 10% by 2012. At the same time, Herta, that belongs to the group Nestlé and is a direct competitor of Fleury-Michon, boasted on the television screens its voluntary decision to reduce the salt and fat contents of its products. Herta had signed a similar charter as that signed by Fleury-Michon at the beginning of January 2010. In addition, 19 big corporations which supply foodstuffs very familiar to the French consumers, had made the voluntary decision to reduce salt, sugar and fat contents in their products, e.g. Maggi (soups), Davigel, Findus (frozen food), McCain (potato fries), Lesieur (oil, vinaigrette sauce), P'tit Louis (cheese). By so doing they show their health consciousness, as bad food-eating habits are considered by nutritionists a serious risk factor of hypertension, diabetes and cardiovascular diseases. Excessive consumption of salt would cause about 25,000 premature deaths per year (Mamou, 2010 c).

These charters for nutritional commitment are a key component of the French National Programme on Health and Nutrition (PNNS) and they are spearheading government action aimed at progressively and profoundly changing the quality of food supply by supermarkets. In 2002, French people consumed "9.5 grams of salt per day, and the average was about 8 grams per day in 2010, the objective being to fall to 6 grams per day as soon as possible," stated Michel Chaulieu, in charge of PNNS at the health ministry. It is not an easy task : Finland, a very good example to follow, took 20 years to reduce its salt consumption by 25% (Mamou, 2010 c).

Herta's overall commitment concerned 74% of its sales. In ten years Nestlé asserted that it had to change the recipe of more than 80% of its products in order to comply with the new nutritional requirements. Such compliance implies more investment in research and development, because salt reduction makes food less tasty. Consequently the challenge is to reduce salt without eliminating the taste and adding chemical additives. According to the statistics of Euromonitor International, the French market of low-salt products was estimated at €64.7 million in 2009; this was considered a low figure, but it increased by 62.5% between 2004 and 2009. The European consumption (including France) of these low-salt products was estimated at €83.4 million in 2009. While the first 15 charters signed by the food corporations with the French health ministry led to forecast an average 1% decrease of salt consumption, one should wait until 2012 – the yardstick year for the PNNS – in order to assess the real impact of these charters (Mamou, 2010 c).

Modification of food tastes and healthier food production

A breakthrough in the food industry would be to offer healthier versions of popular foodstuffs without affecting the taste. If it succeeds to do so, grapefruit juice could be sweet without added sugar and potato chips flavourful with half the current content of salt. This kind of research could have applications in medicine manufacture. In April 2003, Linguagen Corp., a biotechnology company in Cranbury, New Jersey, conducting taste research, was granted a patent for the first molecule that will block bitter tastes in food, beverages and pharmaceuticals. The compound, adenosine 5'-monophosphate or AMP, occurs naturally (in human breast milk, among other sources) and, when added to certain foodstuffs, including coffee and canned or bottled citrus juice, the company stated, it blocked some of the acidic tastes from being felt by the tongue (Day, 2003).

The finding of a bitter suppressor attracts all food companies, e.g. Coca-Cola Co., Kraft Foods and Solae, a soya-foods firm owned by E.I. Dupont de Nemours and Co., Inc., and Bunge have each expressed interest in flavour and taste biotechnology. Kraft Foods and Solae are Linguagen's clients while Coca-Cola Co. has signed a research deal with Senomyx, another biotechnology company (Day, 2003).

Some research has focused on finding compounds that would trick the receptors on the tongue by accentuating or blocking certain elements in the food, allowing people to taste a cup of coffee without adding cream or sugar, or the sensation of full fat in low-fat products. Processed foods such as canned soups, sauces and snacks like potato chips contain high amounts of salt to mask the bitter tastes that result from the very hot cooking process. Soft drinks are sweetened to tone down the bitter taste of caffeine. Food and beverage companies are interested in finding compounds that keep food tasty, minus salt, sugar and fat (Day, 2003).

Scientists at Linguagen Corp. have discovered about 20 compounds that blocked bitter tastes and have been granted patents to use four of the compounds as bitter blockers. Because humans have more than 30 separate bitter taste receptors, finding a universal bitter blocker is nearly impossible. Linguagen Corp. is also trying to discover and market a natural sweetener to replace artificial ones like aspartame or saccharine, which often leave a bitter after-taste. The company planned to license bitter blockers to food, beverage and medicine manufacturers in the United States (Day, 2003).

Senomyx, based in La Jolla, California, is also developing bitter blockers, as well as molecules that block unpleasant smells and others that increase the salty taste in low sodium snacks while decreasing the product salt content. The Coca-Cola Co. – the world's first-biggest soft-drink company, commercializing 400 beverage brands in 200 countries – is one of the company's clients. PepsiCo, Inc., is also interested in taste biotechnology and in anything that can impact food or beverages on a large scale.

Since AMP is not bioengineered and regarded as safe, it will be accepted by people and not shunned by consumers like previous additives which were supposed to revolutionize low-fat foodstuffs but later performed far below expectations. Much of current taste research is the result of radical rethinking of the mechanisms of perception of tastes by humans that has taken place since 1993. Researchers have shown that the human brain had the ability to recognize a variety of flavours including bitter, sour, savoury and sweet all over the tongue rather than in specific areas of the tongue, as it was thought before. The tongue papillae contain the taste buds; when food mixes with saliva, molecules dissolve on the papillae and, through the taste buds, send a signal to the brain, which interprets the flavour of what is being eaten. When a bitter blocker hits the tongue, it prevents the bitter taste receptors from being activated. The brain is thus unable to recognize the bitter flavour, while the latter is still embedded in the food or beverage (Day, 2003).

Developing an industry between food and pharmaceuticals

The annual turnover of "bio" and dietetic products (in the wider sense) had reached €8 billion in 2007, i.e. 6% of the whole food market, and would amount to €10 billion in 2012. A reduction in the purchasing power of consumers could have a negative impact. Also due to the global economic crisis, companies may reduce their innovation capacity, and consequently only the big corporations could maintain the pace of innovation (Clavreul, 2008).

For instance, in December 2006, Nestlé purchased Novartis' division of medical nutrition (nutraceutics) for US$2.52 billion (€1.91 billion). Nestlé, which had only 7% of the global nutraceutical market, became the world's second-biggest producer behind Abbott Laboratories (30%) after buying Norvatis' division that employed 2,000 persons and had clients in more than 40 countries. The purchase was part of Nestlé's strategy to be present in new ventures and areas where profits are higher than in conventional agrifood industry. Peter Brabeck, Nestlé's chief executive

officer at that time, stated it was an important step towards the strategic transformation of its group into a company focused on nutrition, health and well being.

On 27 September 2010 Nestlé announced that it planned to invest SFr500 million (€377 million) over the next decade to support the creation of standalone health science business to tackle obesity and chronic disease. To that end the Swiss food group appointed Luis Cantarell, one of its most experienced executives, to "pioneer a new industry between food and pharma" that will develop products to combat diabetes, heart diseases and Alzheimer's. This decision reflected a trend among food and pharmaceutical groups that are converging around high-margin non-prescription health products, for both humans and animals. Pharmaceutical groups including Pfizer, GlaxoSmithKline and Sanofi-Aventis, have laid increasing emphasis on consumer health products sold over the counter, in an endeavour to diversify away from high-risk conventional drug development (Simonian and Lucas, 2010).

Nestlé completed the divestment of Alcon, its specialist eye-care business, to Novartis. The new health science division, to open in January 2011, will include the group's existing health-care nutrition business, which had sales of SFr1.6 billion in 2009. By setting up a standalone subsidiary, the group may avoid perceived conflicts between core products, such as chocolate, and obesity – which in 2010 affected a sixth of the world's population (Simonian and Lucas, 2010).

Some analysts were sceptical about a food company's ability to break into this area, quoting the time it had taken its rival Danone of France, which had invested heavily to see a significant payback. But Nestlé retorked it was no newcomer and already held back Alzheimer's disease in dogs. The group stated its Pro Plan Senior was "the first and only dog food to contain ANTI AGE – a nutrient blend proved to improve cognitive function and mental alertness in senior dogs" (Simonian and Lucas, 2010).

BIOPRODUCTS DERIVED FROM ORGANIC AGRICULTURE

Benefits of organic agricultural products versus premium prices

In the United Kingdom, the Soil Association (headquartered in Bristol) was created by the end of the second world war, when the move towards intensive farming began. In 1943, as the British government ordered the intensive use of artificial fertilizers, Lady Eve Balfour wrote the organic movement's seminal book, *The Living Soil*; but it was not until 1946 that she and a group of scientists and growers started the Soil Association at her farm in Haughley, Suffolk (Wale, 2009).

Patrick Holden, the Soil Association's director, has been very influenced by a book written by Charles Rice called *The Greening of America*. He did some gardening in outside San Francisco in 1970, when he was 20 years old, and "became full of green idealism and interest in the community." He returned to England in 1971 and took a job at a conventional dairy farm in Hampshire. That was followed by a year at Emerson College in Forest Row, East Sussex, where the teaching was based on the theories of Rudolf Steiner. Although R. Steiner had no farming experience, he gave a series of controversial lectures, towards the end of his life, arguing that high-intensity, industrialized farming technologies made for less-nourishing crops. R. Steiner laid the foundations for organic farming's more testing cousin – biodynamic agriculture (Wale, 2009).

P. Holden helped to create the organic movement, while working on a farm in deepest West Wales (Bwlchwernen Fawr). Pre-eminent among P. Holden's contacts is Prince Charles, who gained his Soil Association certification as an organic producer as far back as 1987. The Prince of Wales visited P. Holden's farm in 2009; "he is the global leader of the sustainable agriculture movement. No one has done more to promote the cause," stated P. Holden (Wale, 2009).

Due to the economic downturn, sales of organic produce have fallen fast in 2008-2009. Vegetable sales were down nearly 20% in a year,

according to the Organic Trade Board, while organic bread sales have been halved. Underlying these steep declines in a market estimated at £2.1 billion in the United Kingdom in 2009, is the fact that organic produce cost more than conventionally farmed equivalents. In times of crisis, many customers have reduced their purchases. This situation has triggered the critics towards organic farming. For instance, Lord Taverne, a former supporter of both Greenpeace and Friends of the Earth, is now firmly against the organic movement. He stated : "The fundamental illogicality is that people have to pay more for organic food, not because the producers are crooks, but the yield is less. In world terms, how can you say something is sustainable, when it produces a less efficient use of land?" P. Holden replied that organic farming did not necessarily mean low-intensity farming. Still, Lord Taverne's point about cost stands. If, by eschewing man-made products such as fertilizers and pesticides, organic farmers produce less output per unit of land than conventional farmers, then their products have to cost more. Why should the consumers continue to pay that premium, particularly in times of economic crisis? The answer has been a general faith in the idea that organic food is healthier. But in the summer of 2009, the British Food Standards Agency published a report concluding that organic food was no more nutritious than conventionally produced food (Wale, 2009).

P. Holden countered that "the FSA report was entirely focused on whether there are more beneficial nutrients in organic food than in conventional food," and "there are major environmental benefits as far as producing organically is concerned – pesticides, herbicides, animal welfare benefits. There is a wide range of public benefits, which on their own justify eating organic food." The challenge, then, is to articulate the benefits of buying organic to a market that is motivated more by price than by provenance. It is true that sales of eggs laid by free-range hens have gone up 30% over the past 15 years (mainly as a result of growing unease about battery farming). But free-range eggs still accounted for only 40% of the total egg market. Certainly, some shoppers are rejecting organic produce for cheaper alternatives. Finding a simple message that will persuade them to buy organics once more is the immediate challenge. The Organic Trade Board, an industry body, has been planning a campaign to promote the benefits of organic food (Wale, 2009).

If economic development forces up the prices of oil and basic chemicals, then the cost of farming with chemical fertilizers, herbicides and pesticides will rise too. P. Holden believed there would be a food crunch as big as the credit crunch and that the cost of food would rise, pricing organic producers into the general market. In his vision, a food crunch would

turn a niche, middle-class market into a mass market in which organic farming would rise to the rescue. Yet, if organic farming has lower yields, more land under cultivation would be needed to feed the world. And could organic farming feed the world? P. Holden replied : "It's not only conceivable. I think it's inevitable." But many dispute this view and believe that genetic modification of crop species and varieties are a more promising answer than a return to pre-industrial farming methods (Wale, 2009).

The following figures concerning the cereals produced and consumed worldwide illustrate the major challenge for agriculture in feeding humankind : in 2050, the developing world would produce 1,799 million tons of cereals and consume 2,096 million tons, while the developed world would produce 1,212 million tons and consume 914 million tons (in 1999-2001, the respective figures were 1,026 and 1,125, and 859 and 741 million tons) [Wale, 2009].

Bioproducts and organic agriculture in France

France, with 580,000 hectares devoted to organic farming in 2010, ranked at the fifth position among the European Union's member states. In the Union, according to a report published on 16 July 2010 by the European Commission, 7.6 million hectares or 4.3% of total agricultural acreage, were devoted to organic farming in 2008. Organic farming acreage has doubled between 2000 and 2008. In France, the annual increase of the acreage devoted to organic farming was 4.8% (+11.6% for cereals and +25% for grapevine) during that period. According to the Agence Bio (Bio Agency), the increase rate was about 10% in 2009. To support the extension of organic farming, the French agriculture ministry had allocated to the farmers an additional annual subsidy of €12 million on 8 October 2008 (*Le Monde*, 14 October 2008, p.5).

On 30 September 2010, the ecology ministry announced that by the end of July 2010 there were 3,000 "bio" farms more than in 2009, the total number thus reaching 20,000. The French agriculture ministry emphasized the multiple benefits of organic (bio) farming in terms of job creation, soil-quality preservation and conservation of biological diversity, water and air quality. At the end of 2009, biocertified farms represented 3.14% of the total number of farms, i.e. 2.46% of useful agricultural surface in France (excluding overseas departments and territories). This percentage was expected to rise to 6% in 2012, and 20% in 2020, according to the objectives set up by the national negotiations on environment and development (*Grenelle de l'environnement*) [Dupont, 2010].

Although the largest acreages converted to organic farming existed in Spain, Italy and Germany, the annual increase of farm conversions to "bio" farming reached 23.4% in France in 2009. The growth rate was even higher in 2010 and the number of biofarmers has gone beyond 20,000. Conversion to biofarming is subsidized during the first five years. According to the National Federation of Biological Agriculture (FNAB), funding needs have been estimated at €96 million for 2010 (over five years) in 18 regions, while only €81 million had been budgeted for the whole country (half of this amount was supplied by European funds) [Dupont, 2010].

In 2008, the market of bioproducts had a growth rate of 25%, with a total of €2.6 billion. Between 2000 and 2007, the annual growth rate had reached 10%. The sales of bioproducts in 2008 (€2.6 billion) were distributed as follows : 18% (€567 million for grocery and beverages); 17% (€451 million for fruit and vegetables); 16% (€578 million for milk and milk products); 13% (€330 million for bread and flours); 10% (€254 million for wine); 7% (eggs); 6% (€303 million for beef, lamb and pork and aquaculture products); 4% (other beverages); 3% (poultry); 3% (prepared food sold in special shops or groceries); 1% (€108 million for frozen and prepared food); 1% (cured meats) and 1% (smoked and cured fish and seafood) [Clavreul, 2009].

The overall share of bioproducts within the food market had reached 1.7% in 2008. Most of these products (42%) were sold in supermarkets and department stores, where the annual growth of sales was the highest (+39%). In three years, the sales of biomilk had doubled, while those of eggs had been almost duplicated. Also in 2008, 46% of school restaurants had put bioproducts on their menus, and the forecast for 2012 was that 77% of these restaurants will do so (Clavreul, 2009).

In 2010, France was the second-largest consumer of bioproducts in Europe, behind Germany. But it was only the fourth-biggest producer, which meant that about 38% of these products (in value) were imported. Consequently, the pace of conversion of farms to organic or biological farming should be accelerated. FNAB executives are optimistic, because they consider that the state's commitment, as well as that of collectivities and the crisis of conventional intensive agriculture are pushing organic farming and consumption of bioproducts. However, the trend may change due to price evolution in the conventional market. The price of bioproducts should be decreased in order to make them more affordable by a larger proportion of population. A 15% difference in price between "bio" and conventional products was considered reasonable, but no more. One solution is to sell a large proportion of bioproducts directly to the consumers or through specialized shops (Dupont, 2010).

Marketing, certification, cost and health benefits

The bioproducts sold in supermarkets, which made up 45% of total sales in 2009 compared with 40% in 2005, are the same as those that can be purchased directly (12%) or in specialized shops like Biocoop and Naturalia (38%). The explanation is simple : the constraints are the same, i.e. the prohibition in crops and livestock of genetically modified organisms (GMOs), of pesticides or nitrates as fertilizers, and of pigments (biosalmon is pale). Nevertheless, the concentration of prohibited compounds or GMOs is not 0%, because , despite "the existence of protection zones and the three years imposed for the conversion of agricultural land into organic farming, organic farms are not completely isolated from the rest of agricultural space, i.e. 98% of total farmland. In the latter fertilizers and pesticides are still used and in very significant volumes" (Malaurie, 2010 a). However, the risks in organic farming have nothing to do with those in conventional agriculture, asserted William Vidal, director of Ecocert, the main French body responsible for the certification of bioproducts AB. He also said that the safety of the biolabel was even more credible in supermarkets, where quality control is very strict (Malaurie, 2010 a).

But the consumers of bioproducts are more demanding : they want higher amounts of nutrients, savours and flavours of the old times and a certified local origin. It should be admitted that the label bio cannot fulfil all these requests. It is nevertheless obvious that the production of bioproducts is booming : there are about ten new conversions per day of farmers, sometimes opportunistic. Biobusiness is obviously pushed by the growing demand; and this is a matter of concern for the National Federation of Biological Agriculture, which has launched its own brand "Bio Coherence", in order to focus on a bioproduction that is very strict in its regulations, fair and cooperative, and that can compete successfully with the large food distribution network (Malaurie, 2010 a).

Malaurie et al. (2010) have tried to bring the appropriate responses to a number of questions or issues raised by the consumers of bioproducts in France.

1. Why are they more costly?

More labour is needed to produce bioproducts. For instance, in viticulture or in the cultivation of vegetables and fruits, 20% to 30% more work is needed, and even more humanpower, in order to obtain yields that are generally lower than those achieved with fertilizers and chemical weed-killers. Bioproducts are therefore 20% to 30% more costly, even though their average prices have remained stable in 2009, according to the Bio Agency (Malaurie et al., 2010).

2. Are they healthier?

About 45% of fruit and vegetables produced conventionally contain pesticide residues and 8% contain more than the authorized concentrations. These residues could cause disease or increase the risks of certain illnesses. Scientific and medical independent expertise is certainly needed to ensure that bioproducts or conventional products are healthy. The French Agency of Food Sanitary Safety (AFSSA) is very aware of the need for this kind of independent expertise, because too many researchers have links with the agrifood industry, and it is therefore crucial to avoid conflicts of interest. Some experts consider therefore that public authorities should take the responsibility to advise pregnant women, for instance, to consume bioproducts. This aims to avoid that chemical products absorbed by the mother during pregnancy reach the vulnerable embryo (Malaurie et al., 2010). See also Kluger (2010).

3. Do bioproducts taste better?

It may happen, but the AB label does not offer any taste guarantee. A bio apple produced in Chile and sent by air to France in a refrigerated cargo does not generally have a particular taste. The shorter the distance between the producer and the consumer, the greater chance for aromas to be kept. In addition, plant growth is not accelerated in organic farming; this is particularly true for salads, tomatoes or carrots, which may therefore keep their flavour (Malaurie et al., 2010). See also Kluger (2010).

4. Is the European label reliable?

Since 1 July 2010, a new pictogram has appeared on the packagings of bioproducts : a green leaf surrounded by a crown of stars. This is the new indication of the European biolabel that will coexist in France with the very popular AB. Despite some rumours, it is absolutely wrong to oppose both labels. The constraints of awarding the European biolabel are very strict, even though some experts consider that one could go further. The origin of the suspicion was the authorization of 0.09% of GMO residues. But this threshold did not really exist before, the use of GMOs is strictly forbidden and this tolerance is possible only if “contamination” has been technically unavoidable or accidental. Ecocert carries an inquiry when traces of 0.01% of GMO are detected (Malaurie et al., 2010). In the case of other examples, regulation has been made tougher : yeast used in bakery should become bio as of 1 January 2013; regarding spices and algae that are not available as bioproducts and the list of which is updated, the amount of conventional products cannot be higher than 5%. A yogurt made with biomilk, but whose

strawberries are not 95% bio, will not be labelled as bio. One questionable case is the authorization without restriction of antiparasite treatments in livestock. These were forbidden before. This was the result of pressure from the Nordic countries and the United Kingdom, who are very sensitive to preserving the "well-being of animals" and to avoiding pain for domestic livestock (Malaurie et al., 2010).

5. Are controls of the bioquality serious?

Biocertifiers are reviewing bioproducts at least once a year, or even four to five times a year in case of doubt. And without previous warning. They look through the bills of purchase very meticulously, in particular the purchase of seeds, they check any traces left on agricultural machinery, pharmaceutical stocks, cleaning products, they examine satellite imagery and check if the furrows in the fields are dammed or not. At Ecocert, headquartered in Toulouse (southwest of France), about 17,000 French producers of bioproducts are checked annually. The withdrawals of license – the heaviest sanction – amount to no more than 50% per year, and the farmers have to convert again their farms as if they had become conventional farmers; this will take three or four years – a serious handicap. Biocertification is also a business : the growth rate of Ecocert has been 23% in 2009 and probably 30% in 2010. Ecocert is a private body, but it is accountable to COFRAC, a public body which renews its authorization every four years and controls Ecocert every 12 months (Malaurie et al., 2010).

6. Are the seeds bio?

In principle, the seed of biowheat or vegetable should be bio. Except if the farmer cannot find it on the market, he could then buy conventional seeds. It is authorized and obviously less expensive : up to less than 40% for vegetable seeds, and even less than 100% for lettuce seeds. The number of authorizations to buy conventional seeds rose from 17,735 in 2004 to 30,782 in 2010. These authorizations have been generally granted to large-scale farmers who are dependent on the big distribution (hyper- and supermarkets) and who are requested to decrease their production costs drastically (that is particularly true for carrots, cucumbers or zucchini). The three largest companies of bioseeds are Swiss, German and Dutch. Agrosemens is one of the few French 100% organic seed producers (Malaurie et al., 2010).

7. What is the value of bioproducts in supermarkets?

French hyper- and supermarkets are selling about 45% of bioproducts. After Carrefour and Monoprix which were the first to launch their

specialized brands in the 1990s, all the other companies have followed suit, even discounters such as Leader Price (Casino group) and ED/Dia (Carrefour group). Their strategy consists of selling a smaller range of products – 500 compared with more than 5,000 in specialized shops –, products that contain less costly ingredients like hazelnuts, chocolate, quiñoa, and offering lower prices – these are generally 25% lower than those found in specialized distribution networks. Supermarkets have induced traditional producers to mechanize their production and to hire humanpower in order to deliver higher volumes of bioproducts; this will reduce production costs and retail prices (Malaurie et al., 2010).

The main weakness of supermarkets is about the provenance of the products. About one-fourth of bioproducts come from outside France : Central Europe for wheat, Poland and Germany for dairy products, and even during the French season of production carrots from the Netherlands or eggplants from Spain or Italy, which are purchased at a lower price than in France (but the carbon footprint of these biovegetables should take into account their transportation in lorries and the corresponding CO_2 emissions) [Malaurie et al., 2010].

Another issue is that of sustainable development. The constraints of the production of bioproducts do not necessarily mean that this production is sustainable. For instance, Anecoop – a cooperative of producers of Almeria, in the south of Spain – has been producing cherry tomatoes since 2007-2008, using certain techniques of conventional agriculture (greenhouses, hydroponic culture), but without chemicals. Henri de Pazzis, chief executive of ProNatura, a wholesale dealer of biovegetables, stated it was “impossible to grow only melons or tomatoes without exhausting the soil rapidly,” a warning addressed to French producers who may be tempted to adopt questionable practices under the demand pressure; for instance, by not applying crop rotations or leaving the land fallow, which are indispensable to maintaining soil fertility (Malaurie et al., 2010).

8. How about biowine?

Biowines have no official existence; only exist wines made from grapes produced by organic farming. The European agriculture ministers have tried to draft a list of compliances that would entail the regulation of winemaking. Up to now the AB label concerned just the grapevine and did not care about what occurred in the winery. If biowines do not exist, 100% biowinemakers do exist; they do not use any inputs that may modify the natural aromas of the fruit. However they do not prohibit the use of sulphur, they use it moderately. There is nevertheless a majority

of winemakers who want to authorize techniques of conventional viti-viniculture, such as flash pasteurization. This technique consists of heating the wine up to 73°C for two or three minutes, in order to kill all the germs that may produce unusual tastes. But its drawback is to also kill natural yeasts existing on the grapes; henceforth the need to use exogenous yeasts to start the alcoholic fermentation and to eventually rectify aromas. As an agreement could not be reached, the Northern European countries being opposed to those of the South, particularly with respect to the amount of sulphite, the European Commission has renounced for the time being to the concept of "biological wine." In the meantime, the consumer could rely on labels like Demeter in order to identify wines derived from biodynamic farming (Malaurie et al., 2010).

9. Can organic agriculture feed the planet?

It obviously cannot. But what is the level of pesticides used in conventional agriculture that can be acceptable? The European Union decided to halve their use over ten years, in order to preserve underground water tables, which are often polluted by both pesticides and fertilizers (nitrates), to prevent professional diseases among farmers and to protect consumers' health.

A study published in January 2010, called *Ecophyto R&D*, coordinated by the National Agricultural Research Centre (INRA) and requested by the agriculture and environment ministries, led to the following conclusions : if the use of phytosanitary products was reduced by 40% in the large-scale crops, this would bring a great benefit; yields would decrease by only 7% compared with those obtained in 2006; the farmers' profits would remain stable due to the savings made in the use of pesticides. This study was the result of a huge work carried out by 80 experts and was mainly based on the observation of test farms that had adopted the "integrated system of production" in the Eure department (west of France) or in Picardie (north of France). This system is inspired from organic agriculture : crop rotation, use of machinery that preserves soil structure. This system is considered a "third way" between the chimaeric all organic agriculture and the all chemical agriculture. This was the subject of a book written by the agronomist Philippe Viaux and titled *Une 3e voie en grande culture* (Editions Agridécisions) [Malaurie et al., 2010].

In an interview reported by G. Malaurie (2010 b) in the French weekly magazine *Le Nouvel Observateur*, the agriculture minister, Bruno Le Maire, has insisted on the need to reduce by 50% the use of biocides in 2018. Attending this interview, Marc Dufumier, professor at the National

Agronomic Institute, expressed his concern about an extensive organic agriculture on thousands of hectares, as it is practised in Ukraine. He stated that an intensive organic agriculture should be intensive in work and labour in order to create jobs, as well as through the intensive use of renewable natural resources. He also questioned the fertilization with costly chemical nitrogen fertilizers, made from natural gas imported from Russia or Norway (Malaurie, 2010 b).

The minister was against the idea that organic agriculture would remain a niche the products of which are to be consumed by only an elite. He mentioned that in 2010 the number of farmers who converted their farms to organic agriculture amounted to 300 per month, and that the acreage devoted to this type of agriculture will increase. In addition to making bioproducts more available to all, it was forecast that 20% of food consumed in collective restauration supported by the state will be bioproducts in 2012 (Malaurie, 2010 b).

The financial support for the conversion to organic agriculture has increased from €90 million in 2009 to €130 million in 2010, and this support will be maintained according to the minister, because in his view French people will not accept that agricultural development be harmful to people's health and the environment. He also raised the issue of ensuring a stable income for the farmers who accept to reduce the use of fertilizers and biocides; this entails the regulation of agricultural markets. M. Dufumier highlighted the need for reviewing the whole European agricultural system. For instance, to avoid the proliferation of green algae on the beaches of Brettany, livestock raising (particularly of pigs) should become more environment-friendly in order to drastically reduce the volumes of nitrates which pollute underground water-tables. Raising of sheep and goats should be reeastablished in the Parisian Basin, so as to fertilize soils with the manure produced and not with nitrates. He insisted on the particular care for soil fertility, which is advocated by organic farmers and which in his view is common sense. The farmers can find the appropriate solutions by themselves provided that the prices reward their work correctly as well as their care for the environment (incentive economic measures) [Malaurie, 2010 b].

Problems of supply

The significant growth of sales of bioproducts could not be entirely met by local production : in 2008, imports of these products reached 30%, compared with less than 20% in 2005, according to the French Bio Agency, a public body in charge of the promotion of organic farming and production.

The deficit was important with respect to biomilk, cereals and particularly to fruit and vegetables. On the other hand, the supply of bioproducts is slow because at least two years are needed to enable a dairy farm to become an organic one, and three years is the time necessary for a cereal farm to produce organic grain; during those periods, chemical products that have been used before slowly disappear from the soils. On the other hand, demand has grown strongly, and, according to analysts, this increase was particularly due to two events which heightened the awareness of the risks of a non-sustainable consumption : the Ecological Pact published by Nicolas Hulot, and thereafter the national negotiations on environment and development, called *Grenelle de l'environnement*. Consequently, agrifood industrialists and brand distributors which had invested in the sector more recently, have increased their supply, but without being able to find all the needed products locally. Cécile Frissur, general delegate of SYNABIO, the trade-union of bioproduct processors – more than 5,600 enterprises in 2008 – stated that newcomers, as did the older in the profession, should adapt their growth objectives to the rate of supply of raw materials (Clavreul, 2009).

For instance, Biolait, which collects biomilk from 500 dairy farms, had to import produce from the United Kingdom in 2009 to supply its customers. Biolait's director-general Loïc Dété stated that "they could supply 40 million liters of biomilk, but the demand was almost 50 million liters." But in 2011, production was to increase by 50%, and therefore imports would not be necessary. In the case of cereals, most processors used to import organic wheat from Italy. Even though the agriculture ministry announced a 19% increase in production, imports will still be needed. At the Bio Agency, it was considered that the gap between supply and demand was temporary (Clavreul, 2009).

In addition to financial support for organic farming by the agriculture ministry, processors of organic raw materials are striving to convince conventional farmers to move to organic production. For instance, they have added to the price of milk a premium of €30 per 1,000 liters during the period of conversion of dairy farms. Some companies would even propose a higher premium in order to ensure a steady production. Another kind of incentive is technical assistance. That is the case of Danone which assists its dairy farmers settled around its plant located at Molay-Littry (department of Calvados, north-west of France), which manufactures its bioproducts (brand *Les 2 Vaches*). The group has launched a range of dairy bioproducts for children under the same brand in October 2009. Danone's supply of biomilk only comes from Biolait and is therefore locally produced. Also Terrena, the main French agrifood cooperative, provides technical assistance to the producers. In 2008, it suffered from

a bad image caused by the imports of soybeans from China (to feed its poultry) that were contaminated with melamine (Clavreul, 2009).

In France, therefore, demand has grown too rapidly and local supply did not follow suit. Some producers have claimed that imports of organic raw materials were not the appropriate solution, and that low-cost imports will not be easy to substitute in the future. Another issue concerns the impact of the economic downturn on the consumption of bioproducts. In Germany and the United Kingdom, it has decreased, but in France it does not seem to be the case, probably because the French market is not yet mature (Clavreul, 2009).

A global trend

Worldwide the acreage of biocrops has trebled between 1999 and 2004. In 2006, the principles of organic agriculture were applied to only 0.65% of cultivated lands throughout the world. In Uruguay, 6% to 7% of total agricultural land was devoted to organic farming; 2% to 3% in Australia; 1% to 2% in Argentina, Dominican Republic and Tunisia, respectively; 0.5% to 1% in Peru, the United States and Uganda, respectively (*Le Monde*, 14 October 2008, p.5).

But in Europe, 7 million hectares and more than 200,000 farms were bio labelled, i.e. were not using chemical pesticides, synthetic fertilizers and genetically modified seeds. Yields were generally 50% lower than those of conventional agriculture, but organic agriculture generates less greenhouse-effect gases. According to a report by the International Federation of the Movement for Organic Agriculture, organic farming uses 30%-70% less energy per area unit than conventional agriculture (use of less machinery).

With respect to the acreages devoted to biocrops, Italy was leading in 2005 with 1.148 million hectares, followed by Spain (926,000 ha), Germany (826,000 ha), United Kingdom (605,000 ha), France (553,000 ha), Austria (361,000 ha), Greece (302,000 ha), Ukraine (260,000 ha), Czech Republic (281,000 ha), Sweden (225,000 ha), Finland (145,000 ha) and Switzerland (126,000 ha). As a percentage of total agricultural land, Austria and Switzerland were the leaders with more than 10% devoted to biocrops, compared with 5.1% to 10% in Italy, Greece, Sweden and Finland, and with 0.5% to 5% in France, Spain and the United Kingdom (*Le Monde*, 14 October 2008, p.5).

FAIR TRADE AND BIOTRADE

Definition

Fair-trade consists of partnerships aimed at providing "fair wages" to family farmers, and its goal is therefore to establish more equity in international trade. These partnerships propose better buying conditions to small and poor farmers, and ensure that their rights are respected. The largest proportion of fair-trade products is labelled Max Havelaar. This label was developed in 1988 by Frans van der Hoff – a Dutch worker-priest and ex-member of the non-governmental organization Solidaridad – and Nico Roozen. Both have been struggling to ensure that small and isolated poor producers receive decent prices of their products and also enjoy a sustainable and autonomous development. Their approach was not charity, but that producers become the actors of their own development. In fair trade, the producer has the right to say what is the real price for its product, and ideally producers and consumers should agree democratically on their trade deals. The producer should obtain a price enabling him/her to live decently and the consumer should buy a good quality and healthy product, that in addition could be traceable (Breuillac, 2005; Amalou, 2005, 2006 b).

In fact, the idea of a fairer trade followed the appeal made in 1964 at the United Nations Conference on Trade and Development (UNCTAD) : "Trade, not charity." In 1969, the first shop selling fair-trade products was opened in the Netherlands, while in 1974 a shop of Craftspeople of the World was inaugurated in France. In Mexico, at Ixtepec, farmers of Uciri – Zapotec Indians who live in the mountains of the south of the State of Oaxaca, a few ten kilometers from the Chiapas – have developed, with the help of Frans van der Hoff, an agriculture cooperative that spearheaded the Max Havelaar international initiative. Nowadays, the Uciri cooperative is selling an increasing part of its harvest on the conventional market (Amalou, 2008).

When fair-trade products are derived from organic agriculture or are harvested in natural ecosystems (e.g. baobab fruit, açai) and mixed with other foodstuffs and beverages, their trade is qualified as biotrade. Thus, fair trade and biotrade are often associated, especially when the producers are poor smallholders who cannot afford buying chemical fertilizers, pesticides and other agricultural inputs, and depend almost exclusively on human labour. They can also be labourers or villagers who harvest natural products and sell them to retailers or wholesale dealers. But sometimes a product can be a conventional one and be traded as a fair-trade product (e.g. coffee or tea).

It should be underlined that Max Havelaar is neither a brand, nor a private company : its black and blue logo is just an international guarantee, a tool that makes sure that the product sold under this label has been manufactured and checked according to the rules of fair-trade. FLO International (Fairtrade Labelling Organization), in the Netherlands, manages the international certification system. The same label, granted to producers, importers and industrialists, is called Max Havelaar, Fairtrade or Transfair, depending on countries. Since 15 September 2003, certification, i.e. the series of checks that establish the conformity to the rules and criteria of fair-trade, have been carried out by a distinct entity, FLO-Cert, established in Bonn, Germany (Amalou, 2006 b).

Since 1988, the rules have been the same : the product is purchased at a price higher than world market prices (in order to cover production costs); a subsidy to development, in order to finance collective equipments; an emphasis on the grouping of producers in cooperatives in order to foster the establishment of a democratic system; prefunding of harvests in order to decrease or even cancel producers' debt; establishment of commercial ties in the long term, as to avoid speculation (Amalou, 2008).

Importance of fair trade

Even though fair-trade represents a very small share of international trade (0.02% or little more), its success is outstanding. Sales of foodstuffs or crafts labelled as "fair" have increased up to 20% per year in Europe, where are concentrated 70% of the outlets. In 2005, these products have been sold in 79,000 shops or sales places for an estimated value of €600 million, compared with €200 million in 2000. In Switzerland, of two bananas sold, one is from the fair-trade circuit. In the United Kingdom, 5% of tea and 20% coffee sold have a fair-trade origin (Amalou, 2006 b).

In France, 60% of consumers stated that they had heard about fair-trade and one French consumer out of three has bought a fair product (banana, coffee, tea or chocolate). In 2004, the annual turnover of fair-trade products was estimated at €72 million (compared with €37 million in 2003) and they represented 0.5% to 1.5% of the market (essentially banana, coffee, cocoa and chocolate, tea, fruit juices and fresh fruits, sugar, honey and rice). France was still behind European countries : on average, in 2004, the French consumer spent €0.60 per year to buy this kind of products, compared with €2.26 in the United Kingdom and €14 in Switzerland. But consumption is progressing : some 10,000 supermarkets are selling Max Havelaar-labelled products; big brands like Carrefour and Leclerc (€15 million of annual turnover for fair-trade products in 2005, with 180 products being sold) have joined pioneers like Monoprix, Cora and Auchan. In 2005, French consumption of fair-trade products reached €120 million (Amalou, 2006 b).

According to Alexandre Pasche, president of the company Eco&Co, those who buy fair-trade products belong to urban middle class; they tend to be younger, they give a meaning to their purchases and they privilege the common interest rather than a selfish behaviour. To meet their needs, the marketing of fair-trade products has been modernized and emphasis has been placed on tasteful foodstuffs often produced by organic agriculture. For these consumers, price is not an insuperable obstacle. The difference of price between conventional products and fair-trade ones has been reduced from 30% to 8%-10%; i.e. 15 cents for an average price of €2, according to Alter Eco – the first brand in French supermarkets – which gives back 27.6% of the price paid by the consumers to the small producers (Amalou, 2006 b).

In 2005, out of 3 billion producers, mainly in the southern hemisphere, excluded from conventional markets, 1 million persons in 50 countries, gathered in 548 cooperatives certified by Max Havelaar, have directly benefited from fair-trade, and another 5 million indirectly. Some operators like Alter Eco (whose annual turnover has doubled between 2005 and 2006) develop and manage the whole production chain (34 cooperatives of food producers in 30 countries in the case of Alter Eco), but other young operators have never seen the producers who supply them (Amalou, 2006 b). The role of European and American operators, such as the enterprises Alter Eco, Malongo, Fruidor, Méo, Lobodis and Ethiquable, is to find new outlets for the producers, e.g. supermarkets, hotels and catering groups (Accor, Best Western and Starbucks Coffee). The results are advantageous for the producers: for instance, Kilimandjaro's Union Cooperatives is selling up to 60% of its production (3,000 tons of coffee

per year) on Western markets; Kibena tea plantation in Tanzania has been able to fund 25 schools and to create a network of social assistance in 17 villages thanks to the benefits drawn from fair-trade; in Mali, villages involved in a new production chain called "fair cotton" (used in France by Kindy, La Redoute and Monoprix, among others) could have access to drinking water after selling their first cotton container; in Bolivia, in 15 years, the El Ceibo cooperative has been able to produce 70% of organic cocoa of the country and could defend its interests at the government level (Amalou, 2006 b).

In addition to Mexico's Uciri cooperative, another Brazilian cooperative, Coagrosol, specialized in fruit juices is considered a fair-trade project among the 632 projects supported in 2008 by Fairtrade Labelling Organization (FLO) [Amalou, 2008].

Multinational corporations (Nestlé, McDonald's, etc.) are also interested in fair-trade, but, just like Kraft Foods (which commercializes Jacques Vabre coffee in France, for instance), they consider the Max Havelaar certification system too constraining. They prefer therefore to adopt the American ecological label Rain Forest Alliance, which for instance, mentions a coffee that is "responsible, ethical, sustainable," but not necessarily fair (Amalou, 2006 b). There is in fact a need for more consistency about the norms of fair-trade products; there is more confusion among consumers who wonder whether their purchases really benefit a poor producer, and who do not always understand the criteria adopted by an increasing number of operators that sell fair-trade products. The European Commission has been requested by the French government to devise a European norm.

Pricing of fair-trade products and benefits for the producers

While critics state too much so-called fair-trade money winds up in the pockets of retailers and middlemen, including not-for-profit organizations, Paul Rice, chief executive of Transfair USA, which controls fair-trade certification in the United States, stated the programmes sometimes eliminated as many as five middlemen – a local buyer, miller, exporter, shipper and importer – and instead allowed farmers to deal directly with an American wholesaler. Transfair USA and 19 similar not-for-profit agencies in other countries collect licensing fees on each product that uses the Fair-trade label. In 2005, $1.9 million in licensing fees from companies had been generated. It also spent $1.7 million on salaries, travel, conferences and publications for the 40-employee organization. Some critics found such expenses excessive (Alsever, 2006).

The fact is that fair-trade programmes which promise a "fair wage" to family farmers have grown rapidly. In 2006-2007, more than 35,000 American retailers carried products bearing the fair-trade label, a 60% increase in three years. Since 1999, more than 45,000 tons of certified fair-trade coffee, tea, rice, sugar, bananas, mangoes, pineapples and grapes have been imported to the United States. Sales of fair-trade coffee trebled in that time, making it the fastest-growing part of the specialty coffee business (Alsever, 2006).

An analysis using information from Transfair USA showed that cocoa farmers received 3 cents of the $3.49 spent on 3.5-ounce chocolate bar labelled "organic fair-trade" and sold at Target stores. Farmers received 24 cents for one-pound bag of fair-trade sugar sold at Whole Foods markets for $3.79. Fair-trade coffee producers received $1.26, higher than the commodity rate of $1.10, whether the consumer paid $10 or $6 for a one-pound bag (Alsever, 2006). Amalou (2006 b) has compared the prices paid by French consumers for a conventional and a Max Haveelar-labelled bag of coffee, and the amounts received by the producers :

	Conventional	Max Havelaar
Price	€1.83 - 3	€2.30 - 3.3
Import cost and roasting	€1.41 - 2.45	€1.41 - 2.65
Export cost	€0.14	€0.14
Max Havelaar fee		€0.05
Local middlemen	€0.06	€0.08
Amount received by the small producer	€0.15	€0.60 (i.e. four times more)

Each fair-trade commodity has its own fair-trade price, or the lowest price that farmers will receive even if conventional commodity prices fall. That price is meant to allow them to cover their production costs and improve their lives. This is a means to struggle against rural poverty. Yet a price that is fair in one country may not be in another. In Brazil, "$1.26 per pound of coffee is a fortune," stated Kevin Knox, a coffee consultant in Boulder, Colorado. "In the forest, in the mountains of Mexico, the money barely is enough to justify doing it," he underlined (Alsever, 2006).

Consumption of fair-trade products

Fair-trade has become a real business, amounting to a €1.6 billion turnover in 2007 (+42% than the 2006 figure), according to FLO International. In France, the 2007 turnover reached €200 million. According to TNS

Worldpanel, which evaluates the real purchases made by a representative panel of 13,000 households, 22.5% of French households bought Max Havelaar-labelled products in 2007, the average annual budget amounting to €16 for these purchases. Between 2005 and 2007, the total number of French households consuming fair-trade products had increased by 16 million, according to TNS. A study published in February 2008 by Datamonitor in London has concluded that "the increasing sensitivity of consumers to ethical purchases" will have a decisive impact on the market (Amalou, 2008).

Coffee is the first fair-trade product to be commercialized. Its success is undeniable, but tea, banana and cotton (a fair-trade production chain has been set up in West Africa in 2005) are also successful products. In the United Kingdom, Marks & Spencer, in 2007, decided to sell all its coffee and tea under the fair-trade label; it has been followed by Sainsbury's for bananas and tea (Amalou, 2008).

Only 3.3% of coffee sold in the United States in 2006 was certified fair trade, but that was more than eight times the level in 2001, according to Transfair USA. Specialty coffee sold in 2006 represented 7.1% of the American market. For farmers, certification means better prices and higher standards aimed at improving working conditions, enhancing the local environment and limiting the use of chemicals. This fair-trade coffee was imported from Peru (25%), Mexico (12.2%), Nicaragua (11.2%), Indonesia (9.6%) and Ethiopia (8.8%) [Downie, 2007].

Coffee usually passes from farmers through roasters, packers, traders, shippers and warehouses before being displayed in stores. But Sam's Club, the warehouse chain of Walmart Stores, buys shelf-ready merchandise directly from Café Bom Dia, a roaster in Varginha, Brazil. Fair-trade coffee farmers in Brazil are paid at least $2.58 a kilogram, compared with the market rate of roughly $2.11 a kilogram (2007), according to Sydney Marques de Paiva, president of Café Bom Dia (Downie, 2007).

A farmer, member of a 3,000-members cooperative, usually cultivates less than 10 hectares of land, and produces around 200 60-kg sacks for the cooperative, with 70% of that sold as fair-trade to Café Bom Dia. The fair-trade crop brought about 258 reais ($139) a sack in 2007, compared with about 230 reais for the sacks that were not fair-trade. For the 2007 crop, that meant an additional 3,920 reais ($2,116) for the farmer, a large sum in the impoverished mountainous areas of Minas Gerais State (Downie, 2007).

Simpler labelling to help poor nations

The Overseas Development Institute, a London-based think-tank, has stated that poor farmers would benefit from a simpler "good for development" label as well as existing standards like the Fair-trade mark. Karen Ellis and Jodie Keane have written in the Institute's report : "It is likely that there are many products which are of significant benefit for developing-country producers but which are not explicitly organized as such, as they may not qualify for any of the existing ethical labelling schemes." They referred to a study of green beans purchased from Guatemalan growers by the American retail giant Costco, which benefited poor farming families but were not covered by any of the ethical trading schemes. A "good for development" label on such produce would show consumers that incomes were being raised and poverty relieved, simply by buying from developing-country farmers (Beattie, 2008).

A proliferation of ethical consumer labels, such as Fair-trade, Rainforest Alliance and the Forest Stewardship Council, bestow marks in return for products meeting certain criteria, often including environmental and social standards. Fair trade also guarantees minimum prices to farmers. But the ODI report argued that such labels benefited too few producers. Even in the United Kingdom, one of the Fair-trade's most developed markets, Fair-trade- labelled produce made up less than 0.5% of food and non-alcoholic drink sales in 2007. The certification fees charged to producers who sign up for Fair-trade and the cost to retailers of establishing new supply chains and trading relationships, appeared to have restrained the growth of the market, according to the report (Beattie, 2008).

Barbara Crowther, a spokeswoman for Fair-trade in the United Kingdom stated : "We think that Fair-trade is already a "good for development mark." Simply labelling all developing-country exports as being good for poverty reduction could reduce pressure on buyers to raise standards, she explained. Gareth Thomas, United Kingdom's minister for trade and development, stated : "We need to look at ways of expanding beyond labels such as the successful Fair-trade mark to look at environmental and workers' rights issues" (Beattie, 2008).

Another illustration of the positive impact of bio and fair-trade is the creation of an agricultural cooperative in the Peruvian Amazon region by former producers of coca, who are now cultivating cocoa and participating in a reforestation project in that area. In northeastern Peru, along the river Huayabamba, in the very small village of Santa Rosa, that can be reached only by boat and which has no electricity nor running water, three hours away from Juanjui, the

closest human settlement of San Martin region, coca has been grown and sold to Colombian narco-traffickers since the 1970s. Nowadays, the Peruvian farmers are producing cocoa for bio and fair-trade, and many of them are also involved in a large reforestation programme, in partnership with Nestlé – the world's leading agrifood company (Cousin, 2010).

The "cocaleros" used to transform coca leaves into a paste in rudimentary laboratories disseminated in the rainforest, and to sell it at about US$900 per kg, and up to US$1,500 if the paste was washed and cleaned. By the mid-1990s, government's pressure and that of the international community, the detention of traffickers, but also the fall of coca prices resulted in the departure of Colombian traffickers who preferred to rely on coca grown in Colombia. But due to the merciless policy of Colombia's government towards the narco-traffickers, drug production was again displaced, and Peru could even become the world's biggest producer of coca in 2011, according to the last report of the United Nations Office against Drugs and Crime. It is now in the southern part of the country, in the valley of Rio Apurimac Ene, that the narco-traffickers are doing their lucrative business and challenging the Peruvian authorities (Cousin, 2010).

But in Alto Huyabamba, cocoa cultivation seems to have won the battle. After the United Nations' Alternative Development Programme had proposed to the farmers to start growing cocoa, in 1997, 26 farmers decided to form an agricultural cooperative; they appointed a manager and they explored the markets by themselves; as soon as an outlet was found, new members were requested to join the cooperative. This initiative has been a success : more and more farmers have joined Acopagro, production has been increasing, fostered by the rise in cocoa prices. From US$800 the ton in 2000, it increased to an average US$2,855 in 2008. In 2001, the cooperative decided to grow organic cocoa, and in 2005 it was certified fair-trade by FLO. Consequently, the cooperative guarantees a minimum price of US$1,950 per ton to the farmers, and strives to attract new buyers, such as TCHO in the United States, Pronatec (Switzerland), Chocomundo (Netherlands) and French Alter Eco (Cousin, 2010).

By the end of 2009, the cooperative with more than 1,500 members has become the leading cocoa exporter of Peru. This cocoa is a high-quality one : it belongs to the *criollo* variety, which is from Central America and makes up 5% of cocoa traded worldwide; it has exceptional aromatic qualities. In October 2009, the French magazine *Que choisir?* tested 24 black chocolate tablets and awarded the first price with a mark of 18/20 to Alter Eco's Intense Black chocolate, made from the cocoa beans produced by Acopagro (Cousin, 2010).

When asked about their current revenues, compared with the former ones drawn from coca, most farmers stated that their revenues did not increase significantly. Cocoa cultivation needs more work and maintenance, and farmers have to work more to gain as much. But they said they are now living well, while before they just survived. In addition, another opportunity will improve their economic situation : a reforestation project was proposed to the farmers by Tristan Lecomte, the founder of Alter Eco, with a view to planting trees in the areas devastated by coca cultivation and also to increasing the yields in coca plantations through agroforestry. Cocoa planters have realized that timber could also be a source of income within a 20-year period, depending on the native forest-tree species that are introduced. Timber could even become more interesting than cocoa. Among the native forest-tree species, there are many that can supply timber for construction : from the most rustic such as *pino chuncho*, *palipero* or *caipirona*, to the most precious such as teak, cedar or mahogany. All volunteers were rewarded for every tree planted and were trained in sustainable forest management (Cousin, 2010).

The Peruvian initiative is part of a large reforestation programme called Pur Project, launched in 2008 by Tristan Lecomte, who is planning to plant 2 million trees in the area. To fund the project, he called on Western companies which want to mitigate their CO_2 emissions. T. Lecomte was able to convince GDF (Gaz de France), Hugo Boss or Cogedim (a public-work enterprise), and then he called on Vittel, that belongs to Nestlé Waters. After one year of negotiations, Nestlé made a commitment for only three years, because in the Amazon region it is the minimal period for the establishment of forest cover. The goal is to plant 350,000 trees in Peru and Bolivia in order to capture 115,000 tons of CO_2, i.e. the equivalent of the emissions of Vittel in France and Belgium for the year 2010. This initiative was announced on 25 March 2010, when the public at large perceived the bottle of mineral water as not environment-friendly, and also when the main competitor of Nestlé Waters, Evian (a subsidiary of Danone), was claiming that it would be "CO_2 neutral" in 2011 (Cousin, 2010).

So, in northeastern Peru, bio and fair-trade of cocoa, combined with reforestation and agroforestry projects, are transforming the life of small and poor formers through the provision of a sustainable income.

In Guatemala's western highlands, near Quetzaltenango, poor farmers growing coffee, like the 25 million other small coffee-growers, become more indebted each year. What they earn is not enough to buy food for their families. Fair Trade, that for 25 years has sought to bring struggling

Third World farmers out of poverty, does not seem to enable coffee growers in Central America and Mexico to meet their food needs. In a private-industry survey in 2008 of 179 fair-trade coffee farmers in these regions, more than half of them stated their families had been still going hungry for several months a year (Fieser, 2009).

In the case of a small coffee grower in Quetzaltenango, Fair Trade paid US $1.55 per pound of organic coffee in 2009, almost 10% more than the market price. But the farmer was left with only 50 cents per pound, after paying Fair Trade cooperative fees, government taxes and farming expenses. By the end of the year, from the few thousand pounds produced, the small farmer would pocket about US$1,000 – around half the meagre minimum wage in Guatemala – or US$2.75 a day, not enough for Starbucks' cheapest cup of coffee. Without Fair Trade, these small farmers would not be growing coffee anymore, even though Fair Trade prices have not kept up with the costs small farmers face (Fieser, 2009).

In 2007, Fairtrade Labelling Organization (FLO), which sets worldwide prices and standards, raised the minimum per-pound price of organic coffee 9 cents, to US$1.35 (10 cents of which go to social programmes like scholarships for growers' children). That was 15 cents higher than the current market price. And yet, according to Fair Trade researcher Christopher Bacon of the University of California, Berkeley, the per-pound price needed for farmers to rise above subsistence was really more than US$2. Farmers'advocates are urging FLO to consider raising the price that much. But such a jump would probably mean that Fair Trade could help fewer farmers: "What good is it to have US$-2-per-lb coffee if you can only serve tens of thousands of farmers instead of millions?" asked Paul Rice, president and chief executive officer of Transfair USA, the California-based non-profit organization that oversees Fair Trade in the United States. Instead, FLO's main growth strategy is to keep recruiting retailers like Starbucks, in order to increase the market for farmers (Fieser, 2009).

When by the late 1990s, coffee prices dropped to as low as 45 cents per pound, Fair Trade was the small farmers' saviour, paying twice the current rate. Starbucks joined the cause and in 2009 has pledged to double the amount of Fair Trade coffee it used to buy, to 40 million pounds, 40% of the Fair Trade beans the United States was importing. Instead of imposing a major price increase, FLO is reviewing other ways to help farmers. It is making cheaper loans more widely available, providing more technical assistance to help farmers grow better-quality beans and may begin automatically adjusting its minimum price for inflation (Fieser, 2009).

With US$1.75 billion in worldwide sales in 2008, Fair Trade was still a small player in the US$70 billion global coffee industry, dominated by big corporations such as Nestlé and Kraft Foods. Because producer countries reaped only US$5 billion of those US$70 billion, Fair Trade could help growers receive more of their share (Fieser, 2009).

FOODSTUFFS, BEVERAGES AND PRODUCTS DERIVED FROM BIOLOGICAL DIVERSITY : POTENTIAL HEALTH BENEFITS AND IMPACT ON BIOTRADE

Both the Andean countries and those who share part of the huge Amazonian ecosystem are home of numerous plant species, wild or domesticated, that could be exploited as functional foodstuffs or botanical drugs (and even medicines) if clinical tests are carried out on their relevant organs (roots, fruits, seeds, leaves) and if they can be cultivated on a commercial scale, once their nutraceutical or medicinal properties have been proved scientifically. For instance, William Roca, a former plant biotechnologist and breeder at the International Potato Center (CIP) in Lima, has highlighted the potential of several Andean crop species with respect to their nutritional and health properties (Roca, 2006).

Indigenous Andean potatoes

The United Nations proclaimed 2008 as the potato year, and the official inauguration took place at the United Nations headquarters in New York on 18 October 2007 (Ribaut, 2008). Potato originated in the Andes, it was transported and disseminated by the Spanish colonizers in Europe at the beginning of the 17th century. In France, Augustin Antoine Parmentier (1737-1813) made it a widely consumed vegetable. It saved from famine poor rural areas of northern and eastern Europe. Until the early 1990s, potato has been cultivated and consumed mainly in North America, Europe and the USSR. But in 2005, for the first time, the production of developing countries (about 161.5 million tons) was higher than that of developed ones (155.9 million tons). In 2006, the main producers were : China (70.3 million tons), Russia (38.6), India (23.9), United States (19.7), Ukraine (19.5), Germany (10), Poland (9), Belorussia (8.3), Netherlands (6.5) and France (6.4). As the fourth food crop in the world, potato is cultivated on 195,000 km^2, producing over 315 million tons (Géné, 2008).

The annual per capita consumption (2006) was : Belarus (835.6 kg), Netherlands (415.1 kg), Ukraine (414.8 kg), Denmark (291.1 kg), Latvia (286 kg), Poland (271.5 kg), Belgium (267.4 kg), Lithuania (261.2 kg), Russia (259 kg) and Kyrgyzstan (219.4 kg).

In Lima, Peru, considered as the world capital of potato, the day of papa is celebrated on 30 May. Potato was born on the banks of Lake Titicaca, when the first Inca, Manco Capac, requested his spouse Mama Ocllo to "grow maize in the lowlands and potatoes in the highlands." Nowadays, more than 4,000 varieties are stored in the world's largest potato bank at the International Potato Center (Lima, CIP). For the last 25 years, CIP's scientists have been travelling through the *Altiplano* (highlands between 3,000 and 4,000 metres) and Latin America to collect more than 15,000 samples of wild or cultivated potato. They have been analyzed and compared with existing samples and have been classified as 4,383 unique morphological types. They are stored *in vitro* as tubers, seeds in test tubes in liquid nitrogen in the CIP's division of genetic resources (Géné, 2008).

These varieties or types have all kinds of shapes, colours and sizes. Their local names are also quite picturesque: *illa pilpinta* (radiant butterfly), *puma chaqui* (puma's paw), *munya tuta* (midnight passion), *paq'ariyt'ika* (morning flower), *kusi song'o* (cheerful heart). In addition to the difficulty of peeling them because of the number of "eyes" deeply encroached in their skin, these Andean potatoes are floury, generally have a uniform taste and do not well withstand cooking in water; they can be fried as chips but cannot be sautéed. Jacques Benoît, director of Lima's school of Cordon Bleu, has selected 52 varieties that are interesting for cooking, out of over 2,000. After many attempts, he remained sceptical about the prospects of *papa andina* export. Cultivated and consumed for centuries by the Quechuas and Aymaras, it is considered a staple food of poor people, that is shunned by coastal populations who prefer to eat rice and pasta (Géné, 2008).

Peruvians consume an average of 80 kg of potato per capita and per year, the highest figure in Latin America. In the *Altiplano*, potato is consumed as *chuño blanco*, which the Quechuas call *moraya* and Aymaras *tunta*. This preparation was mentioned by the conquistadores in the 16th century. They noted that Andean potatoes could be divided into two categories : the sweet ones, the majority, are consumed after being harvested in the fields; and the bitter ones which should be processed into *chuño blanco* to be edible. The process lasts 45 days; after being harvested, bitter potatoes are frozen in the open air during 45 nights (in June-July, temperature can go down to -10°C at more than 3,000 m). Thereafter, they are dipped in the river to wash away the bitter compounds for 30 days. After being taken out of the water, they are frozen again for a night, and then treaded in order to lose water and their skin. For about 10-15 days, they are spread over pebbles along the river and sundried. Once dehydrated, the percentage of humidity is 14% and they have lost three quarters of their weight. They are

rubbed for the last time to take away the last impurities and they acquire the typical chalky white colour of *chuño blanco*. They could be stored for years in this form. Before consumption, they are soaked for one or two hours before cooking, like dry mushrooms. One ton of Andean potatoes yields 140-150 kg of *chuño blanco* (Géné, 2008).

Carlos M. Ochoa, an agronomist and a world authority on potato, has been roaming the *Altiplano* for 40 years, from Venezuela to Chile, and has discovered 85 new papa varieties, some of them bearing his name (*Solanum ochoanum, S. ochoae, S. cochoae*). He has confirmed that the south of Peru and the north of Bolivia around Lake Titicaca are the areas where one can find the greatest variety of wild and cultivated papas. He stated that "forty years ago, in the caves along the river Chilca, south of Lima, traces of cultivated potato had been discovered, dating back to 7,000 years on the basis of ^{14}C tests." This discovery underlines that potato has been grown in Peru long before the Inca civilization (Géné, 2008).

In France, Olivier de Serres grew potatoes in 1600 in the region of Ardèche (south-east of France), and it was used as feed for pigs. Antoine Augustin Parmentier (1737-1813), a young military chemist who was imprisoned by Prussians during the Seven Year War, discovered *Hannover roots* during his captivity. After being freed he showed a great interest for the tuber and delivered to the Academy of Besançon a publication titled *Examen chimique de la pomme de terre ("*Chemical Analysis of Potato*")*, because the academy made a call for studies on "foods that could lessen the catastrophic implications of famine." Potato was already common in northern Europe and in eastern France, but the court of Louis XVI had to be convinced. Parmentier grew the tuber in the Sablons plain (Neuilly, north-west of Paris) and the plots were protected by the army. But Parmentier took the initiative to cancel the night surveillance by the army, thinking that "every robbery would make a new proselyte of the crop." He was right and he offered a few flowered twigs of the plants to the king, who pinned the flower on his hat as well as on the upper part of the queen's dress. The overall result was that the nobles grew potatoes on their lands and clergymen in their gardens. In 1793, 35,000 hectares of potatoes existed in France; in 1815, ten times more (Géné, 2008; Ribaut, 2008).

The International Potato Center (CIP, Lima) Programme of Andean Potato (Programa Papa Andina-INCOPA) aims at advocating the qualities of indigenous Andean potatoes (*Solanum tuberosum*) and increasing the added value of these crops. These potatoes contain vitamin C (20 mg/100g), cartenoids (lutein, zeaxathin and violaxanthin 50-2,000 µg/100 g), phenols (10-60 mg/100 g), including chlorogenic acid (80-90% of all

the phenolic compounds), flavonoids (anthocyanins, including peonidin, petunidin and malvidin) and flavonols (kaempferol and quercetin). At the CIP, about 400 clones of indigenous potatoes have been analyzed and their contents of micronutrients have been compared with those of commercial varieties and published data; there were generally higher, e.g. 12-37 mg of iron per kg, compared with 14 mg/kg in commercial varieties; 22-140 mg of vitamin C per 100 g, compared with 50 mg/100 g; and 115-2,601 mg of phenolic compounds per 100 g compared with 40-1,108 mg/100 g.

Quiñoa

Quiñoa (*Chenopodium quinoa*) belongs to the Chenopodiaceae family, like spinach and beetroot. It is characterized by a great genetic diversity, from one variety to another, and within the same variety. Thus, the colours of the stem, leaves, ears and grains vary considerably : green, orange, pink, red or purple, spotted or not. The height of the plants ranges from 50 cm to 1.50 m, depending on the variety and the growth conditions. Edible grains are either conical, cylindrical or ellipsoid. Quiñoa is a traditional crop in the Andean *Altiplano*. It has been domesticated 7,000 years ago in the border regions of Peru and Bolivia, and is the staple food of people living in the Andean highlands. The seeds, dubbed as "pseudocereal", are consumed in the form of flour, flakes or popped grains. Quiñoa seeds are rich in proteins, essential minerals, lipids, antioxidants and vitamins, they have a balanced composition of amino-acids and do not contain gluten. These nutritional characteristics made quiñoa a successful product for dietetic food chains and for bio fair trade (Del Castillo et al., 2008).

In Bolivia, 50,000 small farmers in the *Altiplano* grow and sell quiñoa. National production amounted to 23,000 tons in 2008, 20% of which was exported. In order to ensure the sustainability of quiñoa production, Bolivia's ministry of rural development has launched a national research programme on this subject. The work to be carried out will include the ways and means to preserve a wide diversity of the crop varieties in order to sustain quiñoa production in the long term (Del Castillo et al., 2008).

Quiñoa is well adapted to arid environments and to poor, and even saline, soils; it is also very resistant to cold, frost and wind. At the beginning of the growth cycle, the plant can withstand up to three months of drought; it therefore stops to grow, its stem becomes fibrous and its roots become stronger. As soon as water becomes available, the plant rapidly recovers its physiological activity. Quiñoa can also survive at lower temperatures (-4°C to -6°C) during its young growth stage. The low water content of leaves contributes to delaying their freezing (Bois et al., 2006).

Night freezing temperatures are a major stress for plants grown on the *Altiplano*. They are very frequent during the cold and dry season of the southern hemisphere winter, from May to October, but also during the period of farming, from October to April, that corresponds to summer. In order to overcome this climate risk, farmers have developed during centuries appropriate agricultural techniques as well as dozens of local crop varieties with a high genetic diversity. This is shown in the great heterogeneity of plant growth and development in the same field (Bois et al., 2006).

In particular the great heterogeneity of the plant size limits frost damage; in other words, the tallest plants, that are mostly exposed to frost, protect the smaller ones. Researchers of the French Research for Development Institute (IRD) and their colleagues of the University Mayor de San Andrés, La Paz, Bolivia, have shown that this protective effect depended on the height of stems, leaf surface and cloudy status of the sky; air temperature and humidity played a minor role. The difference in height results from the selection of varieties carried out by the farmers, the variation in terrain conditions and farming practices. The researchers have shown that the tallest plants limited heat loss by the smaller plants underneath, because of their role as a screen against cold during the night. The smaller plants could therefore benefit from a thermic gain up to 2°C; they have therefore better chance to survive frost. However, the most promising ears are lost, and the researchers try to evaluate the share of the smaller plants in the final production of the crop. Knowing better the survival strategy of quiñoa would lead to the most appropriate agricultural practices aimed at stabilizing yields in harsh climatic conditions (Winkel et al., 2009).

Tuber or root crop species

Sweet potato

Sweet potato or camote (*Ipomoea batatas*) white, orange and purple flesh varieties have been analyzed at the CIP (about 51 clones of CIP's germplasm). The orange varieties contain an average 12.4 mg of beta-carotene per 100 g of dry weight, while the purple varieties contain an average 191.0 mg of cyanidin per 100 g of dry weight and have an antioxidant capacity (µg Trolox equivalent /100 g of dry weight) of 8,657. These were considered interesting properties for human nutrition (Roca, 2006).

Sweet potato is indeed the staple food of hundreds of millions of people in the tropics, and like potato, is the produce of small family agriculture.

There are many local varieties and this germplasm can help breed more nutritious varieties, for instance enriched in beta-carotene (provitamin A). Sweet potato is one of the target crop species of the HarvestPlus programme – an interdisciplinary alliance of institutions and scientists working to improve the nutritional status of the undernourished people through biofortifying staple food crops with micronutrients. HarvestPlus coordinates more than 60 institutions across the world. The magnitude of global malnutrition is illustrated by the following figures : more than 4 million preschool age children suffer from eye damage due to vitamin A deficiency and many of them will become blind because of xerophtalmia; more than 2 billion people suffer from iron deficiency that causes mental impairment in children and compromises working ability among adults; billions are at risk from zinc deficiency – children are stunned and are more vulnerable to infections and disease.

HarvestPlus is a Challenge Program of the Consultative Group on International Agricultural Research (CGIAR), and is coordinated by the International Centre for Tropical Agriculture (CIAT, Cali, Colombia) and the International Food Policy Research Institute (IFPRI, Washington, D.C.). It is supported by the Asian Development Bank, the Bill and Melinda Gates Foundation, Department of International Development (DFID, United Kingdom), Danish International Development Agency (DANIDA), Swedish International Development Agency (SIDA), the United States Agency for International Development (USAID) and the World Bank. In addition to sweet potato, the target crops of the programme are cassava, beans, cowpeas, lentils, groundnuts, pigeon peas, yams, banana/plantains, potatoes, sorghum, millet, maize, rice and wheat.

The HarvestPlus overall strategy consists of the following stages :

1. identify malnourished populations that can benefit from biofortification; determine appropriate nutrient target levels for selected populations; screen crop varieties and germplasm for breeding;
2. breed new biofortified varieties of staple food crops with higher micronutrient concentrations; test the performance of these varieties in the field; measure retention of micronutrients in crops and foods; evaluate the capacity of human body to absorb and use micronutrients from biofortified crops;
3. develop strategies to distribute biofortified crop seeds to producers; promote marketing and consumption of biofortified crop-derived foodstuffs in order to improve nutritional status.

In most cases, family and subsistence agriculture is primarily targeted by biofortification projects, as it is very often the main supplier of foods to developing countries' populations. For instance, smallholders provide 60% of foodstuffs.

In the case of sweet potato, the International Potato Centre (CIP) had launched by the laste 1990s a project in Mozambique with a view to producing orange sweet potato varieties among the crops grown in this country of southern Africa. In 2008, about 1 million farmers were involved in this €2.5 million project. Another dozen African countries are following the example of Mozambique within a programme titled *Vitamin A for Africa*.

In Mozambique, around 70% of children between six months and five year of age have a vitamin-A deficiency and many of them become blind. Orange sweet potato contains much more beta-carotene than the usual yellow varieties. The government of Mozambique agreed with CIP to expand the cultivation of the orange sweet potato, rather than distribute vitamin-A capsules to children every six months.

In 2002, following catastrophic floods in Mozambique, one thousand small farmers were offered 250 sweet potato plants each, and after three seasons this crop species became widely cultivated. Mothers had also to be convinced to include this non-traditional food in the daily diet. CIP's communication strategy in the villages had a key role in publicizing the crop through posters and advertisements on trucks, caps and even women's traditional attire. Radio and theatre shows emphasized the slogan : "Sweetness that brings health," with a view to convincing the population to eat this tuber that is sweeter than potato and to improve thereby their nutritional status. CIP's representative in sub-Saharan Africa claimed that over 18 months there has been a 15% decrease in the number of persons suffering from vitamin-A deficiency. The tuber has been processed into biscuits, cookies, donuts, fruit juices and even bread. Some bakeries are producing bread that contains sweet potato; it has a golden colour and a heavier texture than white bread. Homemade jams are also appreciated by children (*Le Monde*, 24 December 2008, p.4).

Sweet potato is vulnerable to drought that strikes Mozambique during three to six months every year. At the agricultural research centre of Umbelizi, 30 km north-west of Maputo – Mozambique's capital – drought-tolerant varieties are being selected and new varieties were to become available in 2009-2010 (*Le Monde*, 24 December 2008, p.4).

In the United States, for decades, sweet potatoes have been a holiday favourite. Since the late 1990s, they have popped up increasingly at restaurants catering to diners eager for something new. Per-capita production of sweet potatoes, a close approximation of consumption, has gradually risen 30% over the last decade according to the data

from the US Department of Agriculture : approximately 5.5 pounds per capita in 2008-2009. The percentage of American restaurants offering at least one sweet-potato meal more than doubled by 2009 from 2005 – to about 13% – according to survey research from restaurant tracker Datassential (Brat, 2010).

So far, however, none of the country's largest restaurant chains has consistently offered sweet potatoes nationwide. That was partly because no major foodmaker could quickly produce fries and other products from sweet potatoes to meet a chain's demand, stated Jeffery DeLapp, president of ConAgra's – a company in Omaha, Nebraska – Lamb Weston unit. Executives of ConAgra have thought of building a factory to handle sweet potatoes exclusively, while for years, the company had shipped sweet potatoes from the South and Southeast to its potato-processing factory in Washington State (Brat, 2010).

ConAgra Foods Inc. hopes to make the sweet potato a modern equivalent of the russet Burbank potato variety. In the mid-1940s, entrepreneur J.R. Simplot developed the frozen French fry, thus transforming the russet potato from kitchen staple into multibillion-dollar franchise. In 2007, ConAgra started to work with scientists at the Louisiana State University AgCentre and elsewhere to change some characteristics of sweet potatoes, which are not ideal for machines designed for russet potatoes. The company was spending US$155 million in 2010 to build a sweet-potato processing plant in Delhi, Louisiana, with the help of a federal income tax credit and more than US$30 million from the State of Louisiana (Brat, 2010).

When it opened in the fall of 2010, ConAgra's first new American plant in years was turning sweet potatoes into French fries, waffle fries and other products. ConAgra thought it was the first factory devoted to sweet potatoes in North America. H.J. Heinz Co., McCain Foods Ltd. and other companies also produced sweet-potato fries and other products, but used standard potato-processing plants (Brat, 2010).

ConAgra's executives hoped that new, improved sweet potatoes would fuel growth and profit in its US$2.2-billion potato business as well as its Healthy Choice and other retail brands, where sweet potatoes increasingly are part of the mix. Chief executive Gary Rodkin stated that including sweet-potato fries on restaurant menus also helped boost total fried-potato sales, driving ConAgra sales in both products. ConAgra's Lamb Weston division began offering sweet-potato products to restaurants in 2001. Sales took off, growing about 50% a year during the five-year period 2005-2009. The national trend and incentives to eating healthier

food helped; sweet potatoes, rich in provitamin A and fiber, are widely perceived as healthier, although when fried it is debatable whether they are healthier than regular potatoes (Brat, 2010).

The knobby sweet potato shape remains a disadvantage. The pointy end of the tuber is snipped, wasting about 10% to 15% or so of the flesh. Because sweet potatoes vary in size, sweetness and colour, extra workers are required to cut, taste and sort samples before sending loads through processing machines. Cutting tools sometimes tear through harder, more fibrous sweet-potato fries, wasting more food, according to Lamb Weston's senior director of manufacturing. In their efforts to remodel the sweet potato, researchers and ConAgra's food scientists first sought to determine what mix of sugars and other traits would yield sweet-potato fries that are crispy on the outside but soft and sweet inside. Restaurants and consumers wanted potatoes that are deep orange throughout, without the pale portions that sweet potatoes have. Also, the sweet potatoes would have to maintain the traits while stored for up to a year, perhaps twice as long as they are currently (Brat, 2010).

From the 20,000 sweet potato lines they evaluate annually, the University of Louisiana researchers selected and then bred the ones that had as many of the traits as possible. They also began testing whether planting sweet potatoes farther apart and leaving them in the ground longer before harvest would produce larger potatoes, commented Don LaBonte, Louisiana State University AgCenter breeder. One line of sweet potato that includes many characteristics desired by ConAgra has been bred. Dubbed 07-146, the variety can grow 25% heavier than those on farms currently. Local sweet-potato farmers were eager to work with the new ConAgra plant. For many of them who lost a great proportion of their harvest of sweet potatoes in recent years due to harsh weather, selling more to ConAgra may well be a matter of survival (Brat, 2010).

Maca

Lepidium meyenii (maca), an Andean tuber species, also contains nutrients that could contribute to a healthy diet : essential amino-acids (lysine and arginine), non-saturated fatty acids (oleic and linoleic), phytoestrogens, flavonoids, calcium and iron, and glucosinolates (GLS). These GLS are transformed into isothiocyanates and thiocyanates that may play a role as anticancer agents and against proliferation of cancerous cells. The GLS content in maca tubers (the plant looks like a turnip) varies between 3.8 and 69.5 µmol per g of dry weight, compared with 34.9 µmol/g in Brussels cabbage and 16.7 µmol/g in broccoli. Maca is exported mainly

in the form of flour by some 50 companies : about 300 tons per year, with an annual value of US$3.3 million. The area cultivated is estimated at 2,000 hectares. While the price per kg of dried tubers has been evaluated at US$1.5-2.1, by 2004, the price rose to US$6 when the tubers were converted into flour and to US$30 when the flour was sold in capsules or even US$70 when the extract was commercialized (Roca, 2006).

Yacon

Smallanthus sonchifolius (yacon), an Andean root species, is considered a major source of fructo-oligosaccharides (FOS) : 9% to 12% of dry weight of roots compared with 15%-20% of inulin in Jerusalem artichoke (*Helianthus tuberosus*). In 2005, nutritional studies on mice have shown that very high doses of FOS in yacon flour do not induce toxic effects, while lower doses could reduce the concentration of triglycerides. It was concluded that yacon flour could be used as food supplement that does not rise the level of glucose in the bloodstream (Roca, 2006).

Açai

Açai is a small red-purple fruit – the size of a grape –, which one could compare with blueberry or black currant. It is the fruit of a palm, bearing the same name (*Euterpe oleracea*), growing in the swampy and flooded plains of the Amazon region. It is collected as bunches at the top of the tree, and consumed in the form of marmelades in all the small restaurants and food shops of Belem, Para State. It is also used to make beverages, sherbets, biscuits, sweets and pills, and even a strong spirit (Tuquoi, 2009).

Big companies producing soft drinks and cosmetics have shown interest in *açai*. The latter is sold not only in Belem, but also in California, Japan, Austria, and probably in Europe in the near future. An advertisement campaign launched by the company Sparky Superfoods on one of the three brands of *açai* juice, introduced in the United Kingdom, highlighted the properties of the fruit which tastes like a blackberry, but sugarless, and has a black chocolate after-taste. In Rio de Janeiro, for the last ten years, *açai* has been a fashionable drink, generally mixed with *guarana*, another fruit with stimulating properties. A mixture of *açai* and strawberry juice has been sold in the United Kingdom at Waintrose and Sainsbury's stores. Internet sites that publicize *açai* are numerous, but the market of the product is mainly a Brazilian one (Tuquoi, 2009).

In the port of Belem, fruit bunches harvested in the Amazonian forest are unloaded and sold. Fruits, once dehulled (they are surrounded by

a shell), are mixed in water in rudimentary blenders, and the resulting pulp is mixed with cassava flour or with fried fish. Tradition attributes many health properties to *açai* : effective against anaemia, supportive for physical and sexual activities, effective against tissue ageing and some forms of cancer. Physicians prescribe *açai* for infants who are older than six months (Tuquoi, 2009).

Ana Vánia Carvalho who heads a laboratory at EMBRAPA (the Brazilian Agriculture and Livestock Research Corporation), stated that "scientific studies on *açai* were recent and the results were still fragmented. Research was carried out on laboratory animals and not on human beings : the first results confirmed that *açai* was one of the fruits that contained high amounts of antioxidants, which are effective against premature ageing. It is also rich in fibers and is recommended to sustain physical exercise; other qualities or beneficial effects were not yet scientifically based, but *açai* was a fashionable product" (Tuquoi, 2009).

In fact, in 2006, *açai* was unknown outside the Northeast of Brazil. Later on, it became the fashionable beverage among athletes in Rio de Janeiro and was sold in the form of sherbets on Copacabana and Ipanema beaches. Then it was exported to Florida and California, when it was consumed as pure juice or mixed with other exotic fruit juices, sold in bottles or in small bags at a high price. In California, it was imported by the Belizza company and commercialized under labels that publicized its healthy properties : "Beverage full of antioxidants, vitamins and which provides energy to active people for hours and not minutes" (Tuquoi, 2009).

Local entrepreneurs have seized the new opportunity offered by *açai*. One of them has created a company at the end of the 1990s, which was the first exporter of the product. Nowadays, it has been exporting more than 100,000 tons of *açai* since 2007 to the United States, New Zealand, a few European Nordic countries, Switzerland and the United Kingdom. Its objective is to export *açai* to Europe via Portugal. This company, Amazon Fruit, hires hundreds of workers to harvest the fruit bunches, and from November to March (period of full harvest), employs about 50 persons in its factory situated in a small island of the Amazon delta, Muratuku. According to the owner of Amazon Fruit, Ben Hur Borges, a former civil servant of the State of Para Institute of Agrarian Reform, *açai* production has contributed to the economic development of the region : this was also the State's agriculture secretary's opinion. The production chain provides an income to one of ten inhabitants of the State and contributes 10% of agricultural exports of Para. The forecast was to increase the annual production of *açai* from 500,000 to 700,000 tons.

Most of these quantities are harvested in natural forests and it is therefore a good example of biotrade (Tuquoi, 2009).

To move towards an industrial production, when the nutritional and health properties of *açai* are demonstrated on the basis of statistically significant scientific studies, the palm should be domesticated, improved genetically and planted in large-scale plantations. The governor of the State of Para (the area of which is equivalent to 2.5 times that of France) was planning the reforestation of that part of the Amazonian forest and she wanted to develop *açai* plantations. Her project has been criticized by environmental associations that feared the negative implications of a monoculture. The socialist governor stated that one billion trees will be planted during the five-year period 2009-2013 in the State of Para, and part of them will be *açai* palms (Tuquoi, 2009).

Baobab fruit

Adansonia digitata (*baobab*) is a typical tree of the Sahelian ecosystems (semi-arid sub-Saharan belt). Its fruit (monkey's bread) contains six times more vitamin C than one orange, two times more calcium than milk, important quantities of iron and phosphorus, as well as antioxidants. It is usually consumed in African countries (Le Dref, 2008).

The British not-for-profit association Phyto Trade Africa has been struggling for three years and invested about €220,000 into research in order to introduce *baobab* fruit on the European markets. For the first time, the European Commission has accepted this introduction because the fruit has been consumed for decades by African people without any negative impact on their health. In September 2007, the French Agency of Food Sanitary Safety (AFSSA) stated that "because of the traditional consumption of this plant in many regions of Africa, the consumption of this new ingredient does not raise any toxicological risk" (Le Dref, 2008).

Phyto Trade Africa is trying to sign contracts with large agrifood groups and to obtain the fair-trade label for the product. It also works with African producers and helps them to manage their stocks. The product will be processed in Africa and commercialized only as a powder (the fruit pulp that is protected by a thick and very resistant shell does not lose any of its nutritional qualities). In Europe, the powder will be incorporated into beverages and cereal bars. The association believes that in five or ten years the processing of the fruit would support the local economy and the daily subsistence of some 10,000 people in the African countries concerned (Le Dref, 2008).

Fragrances and flavours

A promising global market

On 5 June, 2007, representatives of more than 50 top fragrance-and-flavour companies met at the World Perfumery congress in Cannes, France, where Gilles Andrier, CEO of Givaudan, the world's leading fragrance company, spoke on *The noses of tomorrow*. The latest robotic smell mixers were on display. International Flavours and Fragrances (IFF), Givaudan's closest rival, flew in most of its 96 top scent developers separately to the congress; their noses are so precious that IFF prohibits more than two from ever travelling on the same aircraft (Caplan, 2007).

Technological advances, consolidation and the race to conquer new markets are sparking up the scent industry and business. Givaudan and IFF accounted for about 30% of the US$18 billion global market (2006) for flavours and fragrances. Givaudan launched ahead during the spring of 2007 by buying Quest, which had been the market's fifth leading company. Some experts warned that as the industry was heating up, its traditions of artistry and creativity could be eroded. For now, IFF and Givaudan are thriving. The global market for flavoured packaged food topped US$1 trillion (2006), and consumers spent hundreds of billions on scented cleaning and hygiene products. In the flavour-and-fragrance industry, the toughest battles between companies are fought not in the perfumes area but on grocery-store shelves. Within an hour of waking, many Americans interact at least five times with companies most of them have never heard of. Lather up with any popular brand of soap or shampoo in the shower; apply deodorant; brush teeth; and put on sun block, skin cream or hair gel; and eat a flavoured packaged food; and chances are they are relying on creations by IFF, Givaudan or smaller competitors like Firmenich and Symrise. IFF's five largest customers, according to a JP Morgan report, are Procter & Gamble, Unilever, Colgate, Estée Lauder and PepsiCo (Caplan, 2007).

Research and development, and innovation

The only thing more closely guarded than its client lists are the formulas in IFF's manufacturing process. No two production operators have a complete recipe, and the employees who compile ingredients are not privy to the name – or even the type – of a final product being prepared. In 2006, IFF devoted 9% of its revenue, or US$185 million a year, to research and development (R&D), employing more than 100 scientists

whose task is how just to develop new smells and flavours but also to rethink how smells and flavours are embedded in products. That investment is a sunk cost for fragrance companies; they have a return only when a manufacturer buys the finished product as an ingredient. So IFF makes money on volume : a soda maker, for instance, would buy vats of flavourings for every batch of a popular drink. But unlike other raw ingredients, like high fructose corn syrup or carbonated water, flavours are unique (Caplan, 2007).

Scent treks

To innovate, fragrance-and-flavour companies send their scientists on *scent treks*. On a trip to Papua New Guinea, for instance, Roman Kaiser, director of smell research for Givaudan, collected more than 50 samples, including a rare hoya plant, the scent of which "reminds you of dark chocolate, with olfactory notes rarely found in flowers," stated R. Kaiser – 65 years in 2010, from Saint-Gall (Switzerland), and trained as a chemist. He has been working for Givaudan since 1968 and has participated since 2000 in scent treks to collect fragrances of plant species threatened with extinction or of very rare exotic species, e.g. in Hawaii, in the Cape region (South Africa), Vietnam mountains, Mexico's Sierra Madre. He overflies the canopy of the Amazonian forest sitting in balloons. He also walks through the alleys of Bartram's Garden, the oldest botanical garden of Philadelphia. He captures the scents of flowers without cutting them, nor altering their fragrance; he uses a glass trap around the flower, which is connected to a sophisticated device, that absorbs air and humidity, but not the molecules of the fragrance; each operation lasts between 15 minutes and three hours (Vulser, 2010).

Up to 2010, Roman Kaiser has been able to capture 2,700 different scents from 9,200 that he has recorded, and 590 of these fragrances have been reconstituted chemically, like those of violet or lily of the valley. For instance, he has been able to detect gardenia fragrance in *Brighamia* of Hawaii, fruity scents in Guyana's *Vouacapoua*, the sweet iris scent in a very rare Mexican orchid species, or the fragrances of clover and violet in the flowers of *Franklinia*, a species that lasts only one day. R. Kaiser found that the most magic fragrance he discovered was from a South African cedar, called *Widdringtonia* (Vulser, 2010).

R. Kaiser visits Givaudan's headquarters in Paris once or twice a year to show samples of the scents he has discovered. He published in 2010 his third book, titled *Scent Of The Vanishing Flora* (Verlag Helvetica Chimica Acta and Wiley – VCH), where are described the plants threatened with

extinction and are presented the chemical profiles of their olfactive identities (Vulser, 2010).

To create authentic flavourings Givaudan's researchers go on *taste treks* to gourmet restaurants and popular street stands. On a trip to a hot-pot restaurant in China's Sichuan province, they sampled a spicy noodle soup that they hoped to reproduce in a broth. Subha Patel, R. Kaiser's counterpart at IFF, has travelled to Kenya and Guangzhou (China). In southern India, she drew samples from cardamom flowers, local tea and fresh red clay. Instead of bringing back buckets of samples or dead flowers, S. Patel recorded her findings chemically. Her primary tool, a solid-phase microextractor, is a US$100 penlike device that can record the specific molecules present around anything with a smell. Scent notes of a Japanese ginger, Indian mango, lantana leaves, evening maiden orchids and pickled *jalapeño* peppers could appear in the next generation of products (Caplan, 2007).

Products and new opportunities

One of the biggest opportunities for flavourmakers is the push by food manufacturers to market healthier packaged foods. A team of five researchers at IFF spent more than three years coming up with a low-sodium flavouring that could reproduce the salty taste of canned soups. Givaudan is also developing low-sodium, low-sugar and low-fat flavours intended to replicate the taste and texture of their full-figured counterparts (Caplan, 2007).

As "taste and smell are cultural" according to Givaudan's spokesman, cultural sensitivity is a crucial competitive advantage. The fastest-growing markets are in Eastern Europe, Latin America and Asia, and fragrance giants hope their staff's noses and palates are global enough to understand their new customers. However tough the competition, the industry is united in one concern : development is endangering its raw material. For instance, India has lost thousands of acres of its sweet-smelling sandalwood trees over the past decades. Such a trend would make even harder for fragrance-and-flavour companies to develop new products. Already, for every five to ten samples perfumers dream up and improve, just one generates a sale (Caplan, 2007).

For now, the giants of the fragrance-and-flavour industry continue their quest for the scents and flavours yet to be discovered. In particular, they trek to ever more remote spots. This is where lie the challenges and opportunities for developing countries, whose large biological diversity has a lot to offer.

But it needs good economic evaluation of the potential markets, intelligent bioprospecting and prioritized research and development (Caplan, 2007).

Black truffle flavour

Black truffle of Périgord, France (*Tuber melanosporum*) is the fructification organ of a fungus that lives in association with the roots of certain tree species. The highly-valued fungus had long been thought to lead an asexual existence, but Francis Martin and his colleagues at the University of Nancy, France, have found that it has two mating types. The information is of great significance to truffle growers whom F. Martin advises to infect roots with both mating types of truffle species. The fungus then benefits from the combination of genes and diversity (Wade, 2010).

On 28 March 2010, F. Martin and his colleagues, and also Italian researchers, reported in the online edition of *Nature* that they had decoded the genome of the black truffle with the help of the French organization, Genoscope. This is a large genome made up of about 7,500 genes. It will take many years to figure out the role of each gene, but there are already some useful informations (Wade, 2010).

Located underground in the roots of its oak-tree host, the fungus has a series of genes for detecting light. These genes may help the truffle to avoid sunlight and to stay safely beneath the soil surface, or to help it sense the cycle of seasons. Other genes control the exchange of nutrients between the fungus and its host. Truffle-infected trees can often be recognized because a patch of bare earth develops around the trunk. The fungus may eliminate competitors of its tree host, perhaps by producing some plant toxin. The corresponding genes have not yet been identified and they may be unique to the truffle fungus (Wade, 2010).

The spread of fungus spores (ascospores) raises some difficulty for an organism living underground. Biologists suggest that the truffle produces smells that will compel above-ground organisms to search for it, eat it and distribute its spores afterwards. For instance, sows are attracted by it because the truffle secretes androstenol, a hormone produced by boars before mating; dogs are also used to detect truffles; squirrels are attracted by the compounds secreted by the fungus, which mimick their own sex hormones. F. Martin and his colleagues have been able to trace most of the genes which control the synthesis of these chemicals (Wade, 2010).

There are also the truffle flies that lay their eggs in the truffle. The insects are another way of spreading the spores of the fungus. The latter attracts

them by releasing anisole and veratrole, two insect pheromones, when the truffle has reached maturity. Therefore, truffles will likely carry eggs and larvae, adding proteins and aroma to the truffle, according to F. Martin (Wade, 2010).

There are regional varieties among truffles that differ in terms of aroma and texture. People of Périgord, Provence and other regions of France each claim their truffles are the best. F. Martin and his colleagues hope to develop genetic methods for identifying these regional varieties, now that the overall genome has been decoded. Each truffle could have a Certified Geographic Indication similar to those that protect local wines or cheeses in France. In other words, in addition to phenotypic traits (including morphology, texture and aromas), genomics of the fungus will help differentiate its local or regional varieties (Wade, 2010).

Truffles being a very expensive food item, truffle oil is increasingly used to bring aroma to various culinary preparations. Most of these truffle oils are made by mixing olive oil with one or more compounds like 2,4-dithiapentane (the most prominent of the hundreds of aromatic molecules that make the flavour of white truffles so exciting) that have been synthesized in the laboratory (Patterson, 2007).

Truffles have become a luxury brand, because of scarcity and high value. This has created an environment ripe for fraudulent behaviour. Thus, French regulatory agencies conduct chemical analyses of black truffles to ensure that they are not inferior to Chinese or Spanish truffles soaked in truffle oil or juice. Chefs in the United States use black truffle oil to enhance the flavour of real black truffles, for instance in a pasta dish, and thus make it available at a reasonable price. Shea Gallante of Cru in Manhattan stated : “Price is definitely a factor… If I didn’t use the two drops of oil I would have to add another 8 to 10 grams of truffle,” making the dish too expensive (Patterson, 2007).

Conclusions : biological diversity ownership, cooperation for research and development, and mutual benefit sharing

Countries with high biological diversity, often called “hotspots”, e.g. those sharing the Amazon ecosystems, rainforests, highlands and mountains, and some arid or desert areas, have a lot to offer in terms of biological diversity of plants and crops, as well as in the wide range of all kinds of microorganisms. This diversity is to a large extent unknown, but the work of farmers over millennia and traditional knowledge or ethnoscience (e.g. ethnobotany) have

drawn attention to useful properties of a large number of lower and higher plants, indigenous crop varieties and microbes that have or may have nutritional and health benefits. The major challenge is, in addition to a steady and long-term effort of research focused on increasing knowledge of biodiversity (taxonomy, biochemistry, microbiology, genetics), to select those plants or crop varieties that show promising characteristics, to study them in depth and to establish, including through clinical trials, their real nutritional and health value. When the plant is a wild one, and if it is proved to be beneficial for human health and/or nutrition, its domestication should be envisaged, using the tools of molecular biology, genomics and advanced breeding to speed up the process.

There is in all these fields of research and development ample room for international and bilateral cooperation, e.g.

- in carrying out tests and clinical trials on products that show promise and can be incorporated into functional foods to be commercialized on national, regional and international markets, in full compliance with existing regulations;
- in domestication trials of the relevant plants by joint research teams, relying on advanced breeding technologies and respecting intellectual property rights as well as the rights of indigenous communities (Bonn guidelines), i.e. sharing the benefits from any successful commercialization of end products in an equitable way;
- in involving small, medium and large corporations, including multinationals, in the costly clinical tests concerning products or raw materials with commercial interest, and thereafter sharing the benefits on a fair basis in case of success.

In all these forms of cooperation, the issue of biological diversity ownership (and of the products derived from it) is often a thorny one. It is debated in international fora, within the framework of the Convention on Biological Diversity (CBD), but also under the rights of indigenous peoples who claim that it is part of their heritage. The following two examples illustrate, with respect to staple food crops, the conflicting views and situations regarding biological diversity ownership.

In 1994, Larry Proctor and his wife, Enola, who were on holidays in the Mexican State of Sonora, bought a bag of beans on a local market place and brought them back to the United States. These common beans, called *mayacobas*, of Andean origin, were grown by L. Proctor and their seeds were selected so as to keep only those with a stable yellow colour. In 1996, he filed a patent request on this yellow bean variety, christened Enola. In April 1999, L. Proctor was granted patent no.5894079 and

acquired the rights on all yellow beans. He wrote later on to all importers of *mayacoba* beans in the United States in order to inform them that these beans were his property and that royalties should be paid to him by those who would commercialize that variety : 12 cents per kg. By the end of 1999, L. Proctor did the same with 16 American farmers. Also Mexican dealers had to pay royalties to L. Proctor. Consequently, export sales plummeted (-90%) and 22,000 farmers in northern Mexico were affected by this policy.

In January 2000, ETC Group, a non-governmental organization involved in the defence of biological diversity ownership by indigenous peoples, denounced the patent and called for its deletion. In June 2000, the patent was rejected by the US Patent and Trademark Office (PTMO), but L. Proctor appealed in courts to cancel this decision. By mid-2007, there was not yet a decision made on the issue. Pat Mooney, founder and executive director of ETC group, considered the case of "Enola" beans a striking and revolting case of biopiracy. He stated : "Even the US PTMO admitted that it made a mistake. But L. Proctor who has been contesting the decision for ten years, keeps cashing royalties and prevents Mexican farmers from selling their beans to the United States. If he loses, he would not even have to pay for the damage caused."

In northern India, near Dehra Dun (Uttaranchal State), *basmati* rice varieties with scented long grain that does not stick during cooking, are stored in a seed bank, created by a 64-year-old woman, Bija Devi, who has been working in a farm since the age of seven. The bank includes 250 rice varieties, 13 of them being *basmati* rice, e.g. *Punjab basmati* with slightly yellow long grain, *Kasturi basmati* with mint and lemon scents, *Todal basmati* with a slight balsamic flavour, and the famous *Desi basmati*, with a scent of white flowers and sandalwood, that should be soaked for more than 12 hours before being prepared (Géné, 2007 a).

In 1998, two rice varieties called *Kasmati* and *Texmati*, developed by Rice Tec in Texas, generated furore among Indian farmers because this company wanted to patent this rice as *basmati* in the United States. Due to the absence of an official registration of Indian *basmati* rice, according to the rules of the World Trade Organization (WTO), traditional growers of *basmati* in Dehra Dun might be obliged to pay a royalty to Rice Tec if the latter held the exclusive right. That was also considered an example of biopiracy. However, after several years of legal disputes, the US Patent and Trademark Office (USPTO) decided to authorize Rice Tec to sell its products as a *basmati* variety without granting it the right to a trade

mark; this decision enabled Indian farmers to continue to grow their own varieties. Despite this decision, Indian authorities did not obtain a guarantee similar to that applied to Scotch whisky or French champagne, prohibiting the use of appellations outside the territory of origin (Géné, 2007 a).

Under the leadership of Vandana Shiva, who created the Navdanya movement in 1986 (about 200,000 members at that time) in order to promote organic agriculture, fair-trade, women's rights and forbid the cultivation of GM crops, Indian farmers have created about 20 seed banks in some 15 Indian States. These include 2,000 rice varieties, hundreds of cereals, vegetables and medicinal plants that are carefully stored against the greediness of food and cosmetics companies (Géné, 2007 a).

The stewardship of global biological diversity, as well as the access to, and equitable use of, bioresources and traditional knowledge concerning these resources may improve in the future after the signing of an agreement on 30 October 2010 at the Conference of Parties of the Convention on Biological Diversity (CBD, Nagoya, Japan, 18-29 October 2010). The 10th Conference of Parties attended by 193 countries reached an agreement and made a series of commitments on the preservation of biological diversity by 2020. A protocol on the Access and Sharing of the Advantages drawn from the exploitation of genetic resources was also signed (Mesmer, 2010 b).

The overall agreement signed in Nagoya opens new prospects and is a new approach to the preservation of biological diversity, according to Izabella Teixeira, Brazil's environment minister and, as such, leader of developing countries within the group of 77 (G77). The international community, except the United States which never signed the 1992 Convention on Biological Diversity, has now adopted a new framework for the protection of the biosphere. After the failure in 2002 of the commitments made in order to slow down the disappearance of species by 2020, the participants in the 10th Conference of Parties have adopted a *Strategic Plan* with 20 items. The plan makes a strong plea for slowing down the pace of biological diversity reduction in natural ecosystems, and it foresees the increase in terrestrial protected areas from 13.5% to 17% of total surface, as well as the area of protected marine ecosystems up to 10% of ocean surface, compared with 1% nowadays (Mesmer, 2010 b).

The protocol on the Access and Sharing of the Advantages drawn from the exploitation of genetic resources is the outcome of eight years of negotiations. The fair and equitable sharing of these advantages was one

of the three objectives of the CBD. In Nagoya, the final text adopted by the participants has been softened considerably but it was considered "fair" by most parties. The protocol recalls the "sovereignty of States concerning their natural resources." The protocol also concerns traditional knowledge related to the use of genetic resources – a consistent claim of indigenous peoples. Three issues have been raised during the debates. The first one concerned the compensations. The text adopted remains vague in this respect, as the products derived from a gene have been defined in the protocol, but they have not been dealt with in the protocol articles. The second issue concerned the creation of "national agencies" in charge of delivering the authorizations for bioprospecting, which is mentioned in the protocol, but not the issue of retroactivity of the framework. This seems to have been balanced by the creation of a fund called Multilateral Mechanism of Advantages Sharing; the fund will be replenished mainly by enterprises. Revenues drawn from the use of genetic resources will be, to the largest extent possible, devoted to the preservation of biological diversity (Mesmer, 2010 b).

Claudio Chiarolla of the French Institute for Sustainable Development and International Relations (IDDRI) stated that this protocol was "a good compromise, even though it tended to neglect the interests of the weakest, particularly indigenous peoples" (Mesmer, 2010 b).

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