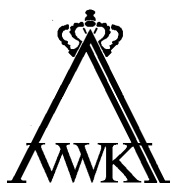




**Royal Belgian Academy Council
of Applied Science**

**INDUSTRIAL BIOTECHNOLOGY AND
SUSTAINABLE CHEMISTRY**

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Industrial Biotechnology and Sustainable Chemistry

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EXECUTIVE SUMMARY

At present, a third wave of biotechnology – industrial biotechnology – is strongly developing. This industrial biotechnology (also referred to as white biotechnology) stands apart from the red biotechnology (directed towards health care) and green biotechnology (genetically modified crops). Industrial biotechnology uses biological systems for the production of useful chemical entities. This technology is mainly based on biocatalysis (the use of enzymes and cells to catalyse chemical reactions) and fermentation technology (directed use of micro-organisms), in combination with recent breakthroughs in the forefront of molecular genetics and metabolic engineering.

This new technology has developed into a main contributor to the so-called green chemistry, in which renewable resources such as sugars or vegetable oils are converted into a wide variety of chemical substances such as fine and bulk chemicals, pharmaceuticals, bio-colorants, solvents, bio-plastics, vitamins, food additives, bio-pesticides and bio-fuels such as bio-ethanol and bio-diesel.

The application of industrial biotechnology offers significant ecological advantages. Agricultural crops are the preferentially used starting raw materials, instead of using fossil resources such as crude oil and gas. This technology consequently has a beneficial effect on greenhouse gas emissions and at the same time supports the agricultural sector producing these raw materials. Moreover, industrial biotechnology frequently shows significant performance benefits compared to conventional chemical technology, such as a higher reaction rate, increased conversion efficiency, improved product purity, lowered energy consumption and significant decrease in chemical waste generation. The combination of these factors has led to the recent strong penetration of industrial biotechnology in all sectors of the chemical industry, particularly in fine chemicals but equally so for bulk chemicals such as plastics and fuels. At present, the penetration of biotechnological production processes in the chemical

industry is estimated at 5 %, and is expected to increase to 10 – 20 % by the year 2010.

This strong development is now mainly driven by the laws of market economy, in view of the higher efficiencies obtained by biotechnological production processes. In the future, a number of societal and technological changes are expected to reinforce this trend even further, such as the depletion of crude oil reserves, the increased demand of a growing world population for raw materials and energy, the demand for sustainability and efficiency in chemical production systems and the changes in the European agricultural policy.

The development of industrial biotechnology is of immediate interest to the economically important Belgian chemical industry and agro-industry. Out of the collaboration of these two industries, entirely new chemical activities can be created, as has already been demonstrated abroad. Also, industrial biotechnology may contribute significantly to the future of European agriculture, and as such is very relevant for the sustainable development of our society.

In view of the strategic importance of industrial biotechnology for the future of Belgium and the lack of a consistent policy in this area, the BACAS working group has formulated a number of recommendations directed towards government and industry. In particular, more Research and Development effort is needed in this domain, by specific research programmes both on a national and European level. The BACAS working group further recommends a number of political and fiscal measures, in which the detaxation of bio-fuels is particularly urgent, when comparing the lack of initiative of the Belgian government contrary to its neighbouring countries. Also, efforts to increase the public's awareness about industrial biotechnology are needed, with the added benefit that this is likely to improve the public's perception of biotechnology as a whole, in view of the clear link between industrial biotechnology and the sustainable development of our society.

1. INTRODUCTION

Sustainability is meeting the needs of the present without compromising the ability of future generations to meet their own needs. No one disputes this "Brundtland" definition of sustainable development and the trend in all sectors of society goes clearly in this direction. In view of the growing importance of sustainable production and doing business with respect for nature and future generations, this principle is increasingly employed in the various sectors of agriculture, industry and services.

Within the framework of sustainable development, more attention has recently been given to the importance of biotechnology and the use of renewable raw materials for chemical production as well as energy production. The use of biotechnology and renewable raw materials as an alternative for conventional chemistry based on fossil resources is only one aspect within the broad concept of sustainable chemistry, in which the numerous interactions between the ecosphere (nature as the supplier of raw materials and energy), technosphere (production and processing) and the sociosphere (use and consumer behaviour) come into play.

Conventional chemical production sectors should give more attention to this "green chemistry". As a result of the greenhouse effect, depleting reserves of fossil resources, the implementation of clean technologies and the desired biodegradability of industrial products, many sectors within the chemical industry are seeing the benefits of using renewable raw materials for non-food applications.

Sustainability is also imperative for our energy supply. The European Union has set its objective of obtaining 14 % of our energy supply from sustainable sources by 2010. Up to now, these energy sources cover only 5.8 % of the total energy consumption in the EU. This illustrates that also the energy sector will see important changes in the near future.

Biotechnology will play an increasingly important role in the development of sustainable (green) chemistry and bio-energy production. In this respect, one refers to "industrial biotechnology". This scientific field deals mainly with the use of micro-organisms such as yeast, fungi, bacteria, or their enzymes to produce useful chemical products and materials.

It is the task of scientists and technologists to be critical and to contribute to correct scientific, technological and social developments in this area. The task of the BACAS working group has been to discuss and report the state of the art of industrial biotechnology and to demonstrate its impact on a broad range of industrial applications. In addition to that, the vision of the

experts in the different fields had to be integrated into policy recommendations in order to stimulate, and if necessary correct, these new developments.

Sustainable development

Sustainable development intends to insure primarily the viability of our world in the long run and create a harmonious balance between economic development, the preservation of ecosystems and the improvement of our quality of life. Natural resources are to be distributed and used in a socially equitable fashion. Sustainable development meets the needs of the present without compromising the ability of future generations to meet their needs.

2. SUSTAINABLE CHEMISTRY

The chemical industry produces a broad range of compounds that can roughly be divided into the following groups: fine chemicals, pharmaceutical products, bulk chemicals, plastics and fuels. The chemical industry is a very important production sector, but at the same time a big user of fossil resources and a significant source of waste.

Researchers, chemists and chemical engineers face major challenges for developing sustainable chemical processes that respect the environment, improve our quality of life and at the same time are competitive in the marketplace. This includes the development of new production processes, which reduce or eliminate the use of dangerous or hazardous substances, minimise energy consumption and waste generation and start from renewable raw materials as much as possible. The ultimate goal is the development of a clean chemical technology, starting from renewable raw materials and energy, with minimal waste generation, and maximal productivity and competitiveness.

Sustainable chemistry is based on a range of different technologies, ranging from more efficient conventional chemical processes, the use of better catalysts, innovative separation methods such as membrane processes, recycling technology and last but not least, the use of industrial biotechnology. The latter technology is increasingly impacting the chemical sector, a reflection of the fact that biotechnology is naturally suitable for sustainable chemistry. Whereas the use of renewable raw materials is rather difficult in conventional chemical processes, industrial biotechnology can handle these renewable raw materials with amazing ease. Low waste generation and energy consumption, the use of non-hazardous, harmless and renewable raw materials and the high efficiency guarantee the sustainability of this technology. Industrial biotechnological processes increasingly penetrate the

chemical industry, with very positive results with regard to sustainability as well as competitiveness.

It is important to stress that industrial biotechnology is not the sole technology in this quest for sustainability. The most sustainable chemistry consists of an interplay between different technologies. In fact, it is common to obtain the best results from a suitable combination of conventional chemical technology and industrial biotechnology. New processes increasingly seem to consist of so-called combi-syntheses, consisting of a number of chemical and biotechnological steps. Also innovative separation technologies such as membrane technology and the use of supercritical solvents are being increasingly integrated and help to increase the eco-efficiency of this “green chemistry”.

Sustainable chemistry

Sustainable chemistry is chemistry that is environmentally friendly, minimises waste generation and energy use, and preferentially uses renewable raw materials such as agricultural products instead of fossil resources such as crude oil or natural gas.

3. BIOTECHNOLOGY

3.1. Biotechnology: an evolving definition

In a broad sense, biotechnology consists of the controlled exploitation of living cells (plants, animals and micro-organisms) or its components for useful purposes. In that sense, agriculture, forestry as well as animal husbandry and all of their ramifications fall under this broad concept of biotechnology. In fact, this conventional biotechnology has been the largest industrial activity on earth since a very long time and people have relied on it to obtain their food, clothing, construction materials and many other natural products like drugs, paper, rubber, etc. It has been mainly based on products and in vivo (in the field) activities of plants and animals, but also of (mostly invisible) microbial cells in the soil, in digestion processes, composting, etc. where they play an essential role in closing the carbon -, nitrogen-, and sulphur cycles of the natural world.

Modern biotechnology goes much further with respect to control of the biological processes. It intervenes rationally in these processes by directed modification of the genetic material (DNA) of organisms, as well as through in-vitro cultivation (in a bio-reactor or factory) of micro-organisms, plants and animal cells for the synthesis of useful products or for carrying out processes that conventional agriculture or chemistry cannot perform as efficiently or not at all.

Biotechnology

A single clear definition of biotechnology has not been accepted yet, but the European Federation of Biotechnology (EFB) has provided a good definition: “Biotechnology is the integrated application of natural and engineering sciences for the technological use of living organisms, cells, parts thereof and molecular analogues for the production of goods and services”.

Biotechnology thus consists of the use of living organisms or parts thereof, to make or modify products, improve plants and animals, or develop micro-organisms for specific purposes.

Conventional biotechnology consists of traditional methods for breeding animals and plants, and the use of bacteria, yeasts and fungi to make for example bread, beer, and cheese by means of conventional fermentation processes.

Modern biotechnology develops these techniques one step further: it makes use of genetic engineering to adapt the properties of bacteria, plants and animals by directly intervening in the information carrier that is the basis for all properties of each organism: the DNA.

3.2. What is industrial biotechnology

Industrial biotechnology is the application of modern biotechnology for the industrial production of chemical substances and bio-energy, using inherently clean processes, with less waste generation and reduced energy consumption. This technology preferably – but not exclusively – uses renewable raw materials as a starting material. Industrial microbiology mainly uses micro-organisms (genetically modified or not) and their enzymes to make useful products and materials in the areas of chemistry, food, health care, energy supply, etc. These cells are real “nano-scale factories” for the industrial conversion of renewable (mostly plant based) raw materials such as sugars and oils to bulk chemicals, fine chemicals, pharmaceutical ingredients, bio-fuels, bio-plastics, etc.

The term “white technology” is also increasingly used, white being a symbol for clean and sustainable technology. White biotechnology is distinguished from “red biotechnology” (medically oriented) and “green biotechnology” (agriculture oriented, such as the genetic modification of crops).

Industrial biotechnology is a multidisciplinary technology and includes the integrated application of disciplines such as biochemistry, microbiology, molecular genetics and process technology to develop useful processes and products, based on microbial, animal or plant cells, their organelles or enzymes as biocatalysts.

Particularly micro-organisms have received a lot of attention as a biotechnological instrument and are used in so-called fermentation processes. Numerous useful bacteria, yeasts and fungi are widely found in nature, but seldom find the optimum conditions for growth and product formation in their natural environment. In artificial (in vitro) conditions, the biotechnologist can intervene in the microbial cell environment (in a fermentor or bio-reactor), as well as in their genetic material (DNA), in order to better control and direct the cell metabolism during these fermentation processes. Because of their extremely high synthetic versatility, ease of using renewable raw materials, great speed of microbial reactions, quick growth and relatively easy to modify genetic material, many micro-organisms are extremely efficient and in many cases indispensable workhorses in the various sectors of industrial biotechnology.

Industrial biotechnology has long been sort of hidden within a number of sectors of health care, food industry and fine chemistry. At present, this technology increasingly penetrates into areas such as bulk chemistry and energy supply, in a world where sustainable development is the key word.

A McKinsey study has indicated that the market share of industrial biotechnology will strongly increase in all areas by 2010, but particularly in fine chemicals production (Bachmann, 2002). The estimated penetration degree in 2010 is estimated to lie between 30 to 60 % for fine chemicals and between 6 to 12 % for polymers and bulk chemicals. Taken over the whole of the chemical industry, the penetration of biotechnology is presently estimated at 5 % and this is expected to increase to between 10 to 20 % by 2010, and strongly increase even further afterwards. The penetration speed will depend mainly on a number of factors such as the prices for crude oil and agricultural raw materials, technological developments and the political will to support and structure this new technology.

Industrial biotechnology

Industrial biotechnology – also known as white biotechnology – is the application of modern biotechnology for the industrial production of chemical substances and bio-energy, using living cells and their enzymes, resulting in inherently clean processes with minimum waste generation and energy use.

4. INDUSTRIAL BIOTECHNOLOGY AND SUSTAINABLE CHEMISTRY

4.1. Renewable versus fossil resources

The use of renewable resources as raw material for technical (non food) purposes is certainly not new.

People already used such materials from the first civilisations onwards. To meet their basic needs, people have employed plant and animal based raw material, from natural fibres for clothing, wood for heating, animal fat for lighting to natural dyes for art works, etc.

The first industrial activities were also largely based on the use of renewable resources and this continued until the industrial revolution. In the 19th century there was a fundamental change, brought about by the emergence of carbochemistry (based on coal, aromatics and synthesis gas) and in the 20th century by the development of petrochemistry. The use of renewable raw materials declined significantly, mainly as a consequence of the extremely low prices for petrochemical resources. During this period, the strongly developing chemical industry was nearly systematically based on petrochemical resources. Nowadays, a large part of the chemical industry is based on petrochemical resources and our energy needs are also largely met by fossil fuels such as coal, petroleum and natural gas. Currently, 95.8 % of all organic chemical substances produced in Europe (including fuel) are based on fossil resources.

Nevertheless, a fair number of important industries are still based on renewable raw materials. Half of the fibres used in the textile industry are natural fibres (cotton, wool, flax,..), the oleo-chemical industry supplies our daily hygienic needs for soap, detergents based on vegetable oils, the building industry still uses a lot of wood and other natural fibres as construction material, etc. Moreover, petrochemistry does not offer a realistic alternative for the use of renewable raw materials in several important applications. For example, almost all antibiotics are made by fermentation processes, starting from natural sugars and about half of our drugs are still isolated from living organisms.

The oil crisis between 1973 and 1979, when OPEC raised oil prices from 2 to 30 \$ per barrel (1 barrel = 159 l), gave rise to a renewed interest in renewable resources. As a result of this crisis, serious concern grew about our increasing dependency on fossil resources and the fact that these are not infinitely available. This concern was largely channelled politically into the energy question and resulted in many studies concerning the development of alternative energy sources. The results of these studies underlined that renewable raw materials were not (yet) competitive and the enthusiasm for renewable raw materials quickly disappeared when the oil price dropped again and the economy turned back to business as usual.

In the nineties, the discussions around sustainable development and the greenhouse effect as well as the

emergence of the green political parties provided new impulses. The problems related to the food surpluses in the European Union were also an important driving force. Because of the huge costs arising from these food surpluses, the EU strongly intervened into the European Common Agricultural Policy (CAP). For this purpose, the European Union developed the “set-aside” land concept in 1992. According to this principle, subsidies were given to farmers for not planting anything on parts of their land, in order to limit over-production. Then, within the European Common Agricultural Policy, possibilities were created to use this land for non-food applications. Thus, farmers could earn additional revenue from this land.

With the increasing awareness and concern about industrial waste and its effects on the environment, the need arose for better biodegradable intermediates and final end products. These biodegradable products can naturally degrade into components that are absorbed back into the natural cycle, in contrast to persistent products that do not (or only after an unacceptably long period) disappear from the environment or from the food chain. Biodegradability was the focal point of many products and these were frequently based on renewable resources, in view of their intrinsic biodegradability. Such applications are e.g. chemical substances that will almost certainly end up in the environment like lubricating oils for tree saws and agricultural machinery, detergents, etc. Green detergents like alkylpolyglucosides have already achieved a significant market share and are made entirely from renewable resources (fatty acid alcohols and glucose).

The world's crude oil reserves will not last forever. With regard to fossil reserves, we are now faced with the paradoxical situation that while crude oil (petroleum) is being consumed faster than ever, the “proven oil reserves” have remained at about the same level since thirty years as a consequence of new oil finds. Nevertheless, these “proven oil reserves” are located in increasingly difficult to reach places. Therefore, the cost for extracting the crude oil rises continuously, reflected in increasing oil prices. In sharp contrast to this, agricultural raw materials such as wheat and corn are becoming cheaper as a fundamental consequence of the rising agricultural yields. This trend will most likely continue for some time, also as a consequence of the realisations of the “green” bio-technology. This long-term trend may be perturbed by the transitory effects of market imbalances and politics but for a growing number of applications the economic balance is tipping towards the use of renewable resources, also in the segment of (inexpensive) bulk chemicals. The following table of world market prices is very informative in this respect.

Raw material	Average world market price (2000 - 2003)
Petroleum	175 €/t
Coal	35 €/t
Ethylene	400 €/t
Corn	80 €/t
Straw	20 €/t
Sugar	180 €/t

Thus, on a weight basis, renewable resources are about half as expensive as fossil resources. Agricultural by-products such as straw are even 10 times less expensive than petroleum. It is also quite remarkable that the current world market prices for petroleum and sugar are about the same, despite the fact that sugar is a very pure (99.8 %) and refined product and petroleum is a non-refined crude raw material, consisting of a very complex mixture of hydrocarbons and other compounds.

For all these reasons, it is clear that the use of renewable raw materials has significant growth perspectives.

The greenhouse effect and the Kyoto treaty

As a consequence of the increased concentration of the so-called greenhouse gases in the atmosphere, the earth is expected to become warmer on a global scale. The main contributor is CO₂, primarily resulting from the burning of fossil resources such as coal, petroleum and natural gas. Although the increase in atmospheric CO₂ concentration has been recognised for some time and the greenhouse effect was well understood, these problems only caught the public's attention in the early nineties. Especially within the framework of climate conferences (IPCC, International Panel on Climate Change), the greenhouse effect has been scientifically proven and its consequences were measurable.

In the 1997 Kyoto treaty, which Belgium signed, a plan was set up to reduce the emission of greenhouse gases in the short term. The Kyoto protocol obliges us to reduce our greenhouse gas emissions by 2008 – 2012 by 7.5 % relative to the 1990 reference level. At present, emissions have actually increased to 7 % above the 1990 level! Consequently, we have 7 more years to reduce our greenhouse gas emissions by about 14 % in order to meet our Kyoto target.

The main contributor to the increased greenhouse effect is the burning of fossil resources, in addition to other factors like deforestation of tropical forests and increased methane and N₂O production by agriculture and cattle raising. Generally, it is assumed that

the use of renewable raw materials does not disturb the carbon balance of our planet's atmosphere. In fact, when renewable raw materials are burnt, they release the same amount of CO₂ as was absorbed during the growth of the plants. Thus they do not add extra carbon dioxide to the atmosphere. When renewable raw materials are processed into sustainable products e.g. non-biodegradable plastics, there is even a decrease in the quantity of global atmospheric CO₂. The use of renewable instead of fossil raw materials consequently has a favourable impact on the greenhouse effect.

4.2. Renewable raw materials for the industry

Renewable raw materials are essentially based on the use of "biomass", the sum of all substances that the living world is made of. Renewable raw materials thus have a biological origin. Its fundamental basis is the plant production that is fueled by the photosynthesis process, and possibly via the intermediate step of animal production, results in a large variety of available biomass.

The total annual biomass production on our planet is estimated at 170 billion tons and consists of roughly 75 % carbohydrates (sugars), 20 % lignins and 5 % of other substances such as oils and fats, proteins, terpenes, alkaloids, etc. (Okkerse & Van Bekkum, 1999). Of this biomass production, 6 billion tons (3.5 %) are presently being used for human needs, distributed as:

- 3.7 billion tons (62 %) for human food use, possibly via animal breeding as an intermediate step;
- 2 billion tons of wood (33 %) for energy use, paper and construction needs;
- 300 million tons (5 %) to meet the human needs for technical (non food) raw materials (clothing, detergents, chemicals,..).

The rest of biomass production is used in the natural ecosystems (wild animals must also eat), is lost when biomass is obtained for humans (especially by burning) or is lost as a result of the natural mineralization processes.

The renewable raw materials discussed here are almost all provided by agriculture and forestry. The animal breeding sector and fisheries also contribute (mainly animal fat), but are clearly less significant, also in view of the low conversion efficiencies of plant to animal (about 10 to 25 %).

A range of different technologies can be used to industrially convert this available biomass into renewable raw materials or energy carriers. This industrial activity is often linked or connected to the food sector, in view of the fact that food ingredients and renewable raw materials for technical use can be made within the

same factory from the same agricultural raw materials. For example, sugar or glucose are produced for human food use and are also the most important raw materials for industrial fermentation processes.

The following industrial sectors supply the most important renewable raw materials:

- the sugar and starch sector: it produces carbohydrates such as sugar, glucose, starch and molasses from plant raw materials such as sugar beet, sugar cane, wheat, corn, potatoes, sweet cassava, rice, etc.;
- oil and fat processing sector: it produces numerous oleo-chemical intermediates such as triglycerides, fatty acids, fatty alcohols and glycerol from plant raw materials like rape seeds, soybeans, palm oil, coconuts and animal fats;
- the wood processing sector, particularly the cellulose and paper industry: it produces mainly cellulose, paper and lignins from wood.

These industries process plant raw materials in order to break them down into separate components such as sugar, starch, cellulose, glucose, proteins, oils, and lignins. They make use of two technological pillars:

- fractionation technology: this technology is primarily based on physical and chemical separation methods to separate agricultural raw materials into their separate components.
- enzymatic technology: this aspect of industrial biotechnology intervenes during the transformation of agricultural raw materials. In practice, mainly hydrolytic enzymes are used that e.g. hydrolyse starch to glucose.

Although both technologies are clearly very different in nature, the interaction between them is particularly decisive for success. For example, the fractionation technology is strongly influenced by the use of hydrolytic enzymes.

The obtained pure basic products (sugar, starch, cellulose, oils) are then converted into a very broad range of products, employing physical, chemical and biotechnological processes. For example, starch and cellulose are chemically modified to derivatives that find many uses in our daily lives. Sugars like sucrose and glucose are chemically coupled to oleo-chemicals to obtain detergents and emulsifiers.

With respect to industrial biotechnological processes, the fermentation technology needs to be specially mentioned. This very important key technology makes use of micro-organisms (bacteria, yeasts, and fungi) to convert basic raw materials such as sugars and oils into an almost unlimited range of products. By simple use of another production organism, the raw material (for example sugar) can be converted to totally different products, ranging from products with a chemical

structure that is very close to the raw material (e.g. gluconic acid from glucose) to products that have virtually nothing in common with the starting material (for example, antibiotics, enzymes,...).

This whole chain of different process steps, implying the use of very different technologies often takes place within the same factory or industry complex. These are increasingly referred to as "bio-refineries", analogous to the petrochemical crude oil refineries.

For orientation, the estimated world production figures and indicative world market prices of a number of renewable and petrochemical raw materials are given in the following table. The comparison clearly shows that their volumes and prices are quite comparable.

	Estimated world production (million tons per year)	Indicative world market price (euro per ton)
Renewable raw material		
Cellulose	320	500
Sugar	140	180
Starch	55	250
Glucose	30	300
Bio-ethanol	26	400
Glutamic acid	1	2000
Petrochemicals		
Ethylene	85	400
Propylene	45	350
Benzene	23	400
Terephthalic acid	12	700
Isopropanol	2	700
Caprolactam	3	2000

Bio-refineries

Bio-refineries are large industrial factory complexes, in which agricultural feedstocks are processed, fractionated into intermediate basic products and converted into final products. These products often have little in common with the original plant feedstock. Bio-refineries use physical, chemical, and biotechnological processes, whereby particularly fermentation technology and biocatalysis play a major role. This technology uses micro-organisms and their enzymes to convert basic resources such as sugars and oleochemicals to products that often having nothing in common with the feedstock. By simply using another production organism, the same renewable raw material can be converted into totally different products.

As an example, the following products have been made industrially from a single feedstock as corn :

- glucose (a natural sugar) as a raw material for foods;
- citric acid as a food additive;
- bio-ethanol as a motor fuel;
- bio-plastic (polylactate), used as a packaging material and as a textile fibre;
- starch carboxylate as an ingredient of washing powders;
- lysine as an animal feed additive;
- antibiotics as a pharmaceutically active substance for drugs;
- vitamins for human food and animal feed use;
- bio-colorants for the food industry;
- xanthane biopolymer as a viscosity control agent in numerous applications

The organisational structure of these large factory complexes is comparable with that of the chemical industry. They are also often connected to a closely located integrated petroleum refinery. The fundamental difference is that bio-refineries use renewable agricultural feedstock, whereas petroleum refineries and the conventional chemical industry start from fossil feedstock such as crude oil and natural gas.

Renewable raw materials in Belgium: fact or fiction?

When discussing whether the use of renewable raw materials makes sense or not for Belgium, one is often faced with narrow approaches to this question. One must inevitably conclude that there is not enough farmland in Belgium and particularly in Flanders to produce the required amount of renewable raw materials. However, using this conclusion to minimise or even deny the importance of renewable raw materials for Belgium boils down to ignoring vast opportunities. If this approach had been employed in the past, petrochemistry and the closely connected chemical industry would never have gotten off the ground in Belgium, as its reserves of crude oil are inexistent. In practice, in spite of the lack of crude oil in Belgium, the chemical industry which processes this raw material, turned out a very important part of the industrial fabric of the country. The chemical industry presently accounts for 14 % of Belgian exports and for 6.1 % of employment in Belgium.

Indeed, farmland is limited in Belgium and it is mainly used to produce food in view of its high population density. Nevertheless, Belgium can play a very important role in processing these (imported) agricultural raw materials and converting them into a broad range of chemical substances or bio-fuels. The added value is mostly created in the high-tech industrial processing of these raw materials, rather than in the primary production of these raw materials via agriculture.

Our country possesses a strongly developed chemical industry, as well as a well developed agro-industry, which forms the industrial basis for the use of renewable raw materials. Geographically, Belgium is located in the centre of the European agro-industry. With respect to renewable raw materials, the starch sector is very active and innovative. Belgium has a very favourable starting position here: the starch sector in Europe consists of an oligopoly of 4 large groups, of which the two largest players (Cerestar and Amylum) have set up their headquarters and central research centre in Belgium (in respectively Vilvoorde and Aalst). The French company Roquette is the third player with its headquarters and central research centre just over the French border in Lestrem near Lille. AVEBE, player number four, is located further away in Groningen (the Netherlands). As a result of mergers, take-overs and other shifts within the sector, the R & D activity of almost the entire European starch sector is now concentrated in Belgium and its direct vicinity. The societal and industrial base for a strong development of this technology is thus certainly present in Belgium.

The sugar industry traditionally has a strong presence in Belgium and has shown courage and innovative spirit with respect to developing new products such as inulin from chicory and lactic acid from sugar (Orafti, Warcoing Industries and Galactic).

The oleo-chemical industry is also cutting edge with respect to the use of renewable raw materials. One of the most important European players in this domain is based in Belgium (Oleon in Ertvelde and Oelegem).

The assertion often heard that renewable raw materials may be of interest in countries like the USA or Brazil because of favourable prices (contrary to the European situation) can be vehemently disputed. Current prices for the most important agricultural raw materials are quite similar in Europe and the USA. For example, even with the high euro exchange rate, wheat costs almost the same in Chicago as in Rouen (May 2003). The farmland required to produce these agricultural products is largely present in Europe, particularly in view of the imminent integration in the European Union of 10 East-European countries with huge agricultural potential. Eastern Europe is far closer than the Middle East from which we import most of our crude oil.

acid, citric acid, vitamins, amino acids, solvents, antibiotics, bio-polymers, bio-pesticides, industrial enzymes, bio-colorants, bio-surfactants, alkaloids, steroids, etc. Industrial fermentation is the main technology here, whereby micro-organisms (bacteria, yeasts, and fungi) are cultivated that efficiently convert sugars into useful products. It is the only industrial production method for some of these products and some are produced in very significant quantities. The table below compiles the production figures and prices for a number of these fermentation products. The range varies from inexpensive bulk products to very expensive fine chemicals.

	World production (ton/year)	World market price (€/kg)
Bio-ethanol	26,000,000	0.40
L-Glutamic acid (MSG) . . .	1,000,000	1.50
Citric acid	1,000,000	0.80
L-Lysine	350,000	2
Lactic acid	250,000	2
Vitamin C	80,000	8
Gluconic acid	50,000	1.50
Antibiotics (bulk products) . .	30,000	150
Antibiotics (specialities)	5,000	1,500
Xanthan	20,000	8
L-Hydroxyphenylalanine	10,000	10
Dextran	200	80
Vitamin B ₁₂	10	25,000

Thanks to recombinant DNA-technology, one can now specifically intervene into the genetic material of these micro-organisms. On the one hand, the metabolism of micro-organisms can be modified or even completely changed (so-called "metabolic engineering"). On the other hand, genes from higher organisms (plants and animals) or other micro-organisms (yeast, bacteria, virus, algae) can be inserted into industrial micro-organisms and brought to expression. Thus, new direct gene products can be made or new metabolic pathways can be created to produce chemical substances with high efficiency via industrial fermentation processes.

In practice, well-known, productive and harmless production organisms are used that, equipped with the new genetic information, will produce the desired chemical products in high yield and efficiency. A major advantage is that these genetically modified micro-organisms do their work under controlled conditions in a fermentor or bio-reactor, carefully contained and separated from the outside world. They cannot escape from the factory and ecological problems or concerns with regard to the release of genetically modified organisms in the environment are thus avoided altogether.

4.3. Bioprocesses in industrial biotechnology

4.3.1. Fermentation processes

Industrial biotechnology is used to produce a wide variety of bulk and fine chemicals like alcohol, lactic

4.3.2. Enzymatic processes

Enzymes are natural accelerators of (bio)chemical reactions; they are also referred to as biocatalysts. Enzymes are catalytically active proteins that have evolved and were perfected over billions of years of evolution. As very specific and efficient catalysts, they direct the chemistry of life without needing extreme temperatures, high pressures or corrosive conditions as often required in chemical synthetic processes. Enzymes are the machinery of the living world and their amazing properties are increasingly used for industrial applications. This technical discipline is referred to as biocatalysis.

Enzymes have become very important in a wide range of industrial sectors to carry out biocatalytic reactions. Typically microbial enzymes are used, produced by the previously mentioned fermentation processes. New technologies such as specific site-directed mutagenesis and "gene shuffling" allow new enzymes to be tailor-made. These developments can strongly improve this technology or even expand it to totally new applications.

Conventional applications are the large scale use of enzymes in the starch sector, not coincidentally the sector at the source of glucose, one of the most important renewable raw materials. A key enzyme is α -amylase, a very thermostable enzyme used to hydrolyse starch at a temperature of 105°C. Such thermostable enzymes allow bio-reactions to take place at high temperatures, considerably increasing the reaction rate. Glucose isomerase is another important enzyme in this sector. This enzyme converts glucose to fructose. It is used in immobilised form and maintains its catalytic activity up to two years when used industrially. Last year, the world production of fructose with the help of this enzyme passed the mark of 15 million tons per year.

The detergent sector is another big application area for enzymes. Here, proteases and lipases are used to break down protein and fat stains on clothing.

The animal feed industry is another important market. For example, phytase from the fungus *Aspergillus niger* is employed to release phosphate from phytic acid in animal feed. Thus, less additional phosphate has to be added to animal feed, with considerable environmental benefits. Other enzymes strongly improve food conversion, with equally positive ecological benefits.

Enzymes are increasingly penetrating the chemical industry to catalyse numerous reactions. The specificity of the enzymatic reaction is very important here. When compared with conventional chemical catalysts, this specificity is often very high. Besides a high

degree of reaction specificity, chirality has also provided strong impulses to the application of biocatalysts in the chemical industry (see frame). The use of enzymes (used in free or immobilised form) for very specific organochemical reactions is rapidly developing. These are mostly one-step reactions, carried out with high efficiency, specificity and reaction rate. This scientific domain is often referred to as "biocatalysis" and the processes used are described as "bioconversions" or "biotransformations". These bioconversions are normally performed at normal temperatures and pressures, whereby no dangerous intermediate products are needed nor dangerous waste products generated. Typically, the reactions take place in "green" solvents such as water, ethanol or supercritical CO₂, though enzymes are also active in "conventional" chemical solvents such as methanol, acetone, chlorinated solvents, etc.

Chirality and biocatalysis

Chirality means that a chemical substance exists in two forms, i.e. a "left" and a "right" form. Without going into much detail, these can be compared to human hands of which there are left and right hands that are identical in all other aspects. This also applies to certain chemical molecules, whereby their biological action can differ strongly depending on whether they are "left" or "right". A conventional example of this chirality of molecules is found in the drug "softenon". This drug was used in the sixties as a cure against pregnancy vomiting. Unfortunately, the other chiral form also present in the drug caused very serious birth defects (so-called softenon babies). The biological world is fundamentally chiral such that the chirality of molecules is of major importance for their biological action.

Chemical catalysts typically cannot distinguish between these chiral forms and they catalyse the chemical reaction of both forms. Especially in the case of the synthesis of active substances that intervene in the biological world such as drugs and pesticides, typically only one form is active. At best, the other form is harmless and can be considered as a simple loss but in the worst case the other form may be very harmful as has been sadly demonstrated by the softenon case. In such case, the harmful form must be separated, which is technologically not at all easy. Furthermore, it leads to a considerable reduction in efficiency and the generation of dangerous waste.

Biocatalysis uses enzymes, catalysts originating from the biological world that can handle such reactions much more efficiently. They are nearly always chirally selective and synthesise only one or the

other form. There is a strong drive to use such chiral molecules, especially in the pharmaceutical and agro-chemical industries. Therefore, it is not surprising that biocatalysis now has the strongest penetration level in the chemical synthesis of drugs, agro-chemicals and their intermediate products.

4.4. Products based on industrial biotechnology

A vast range of useful products can be produced by industrial biotechnology. These fall within the categories of fine chemicals, pharmaceuticals, food additives and supplements, colorants, vitamins, pesticides, bio-plastics, solvents, bio-plastics, bulk chemicals and bio-fuels. These products range from very cheap bulk chemicals (e.g. ethanol: 26 million ton/year at 400 €/ton) to extremely expensive fine chemicals (e.g. vitamin B₁₂: a few ton/year at 25,000 €/kg). Whereas industrial biotechnology is already well established in the production of fine chemicals and pharmaceuticals, also bulk chemicals such as e.g. bio-fuels and bio-plastics are now increasingly produced by industrial biotechnology. In some cases a polymer building block is produced from fossil resources using enzymatic technology. In other cases a completely biodegradable bio-plastic can be obtained from renewable resources, e.g. the biodegradable bio-plastic PLA can be produced from corn.

Industrial biotechnology can either intervene in a single step in a chemical synthesis route or replace an entire cascade of chemical synthesis steps with one single fermentation step. The synthesis of vitamin B₂ (riboflavin, 4,000 t/y) is a fine example of this. The conventional production process consisted of a combined chemical-biotechnological synthesis route of no less than 8 steps. This combined synthesis route has recently been replaced by the complete biotechnological synthesis of riboflavin in one single fermentation step with the help of bacteria, yeast or fungi (respectively by Roche, ADM and BASF). The production cost of the new biotechnological process is 40 % lower than the conventional process.

Annexe A at the end of this document describes a fair selection of such products produced by industrial biotechnology. Both production technology, application, as well as some market data are described.

4.5. The economical and ecological advantages of industrial biotechnology

Introducing biotechnological process steps into chemical syntheses often results in significant ecological advantages such as considerably reduced waste generation, reduced energy requirement, decreased use of solvents, elimination of dangerous intermediate

products, etc. However, these ecological advantages are typically not the reason for the technology switch. The technological process improvements and accompanying cost reduction are almost always the driving force for such decision. The ecological advantages are a pleasant side effect. By themselves, they are not sufficient to motivate decision makers to introduce a new technology (with associated failure risk). The way industrial biotechnology combines both economical and ecological progress is quite typical: the increased efficiency and reduced production cost of such biotechnological processes nearly always results in a greatly decreased ecological impact and generally leads to an improved competitiveness.

In a 2001 OECD report ("The Application of Biotechnology to Industrial Sustainability"), 21 such case studies were presented. Each case study convincingly illustrates the economic and ecological advantages offered by industrial biotechnology. It should be mentioned that in most cases, the processes described have been implemented in industrial practice and are competitive, and in no way limited to theoretical studies or research projects.

In this context, the E-factor should be mentioned. This factor measures the efficiency of a chemical process in terms of kg waste products per kg of product. Bulk chemicals have an E-factor of less than 1 to 5, the E-factor for fine chemicals is typically between 5 and 50 and for certain pharmaceutical substances it even goes up to 100!

The company Biochemie (Austria) provided data on the conversion of cephalosporin C to 7-aminocephalosporanic acid (7-ACA), a very important intermediate during the synthesis of semi-synthetic cephalosporin antibiotics. In the conventional multi-step chemical process toxic reagents as well as chlorinated solvents were used. Most waste streams were burnt because they poison normal biological waste water treatment systems. The process took place at very low temperatures, associated with a high energy consumption. In 1995, the management of Biochemie decided to leave the chemical synthesis and replaced it with a biotechnological route. This bio-process uses two naturally existing enzymes and takes place at room temperature. Solvent use is almost completely eliminated. The most important waste products are aqueous waste flows from the fermentation that can be processed in the biological waste water treatment system without problems and waste mycelium that is employed as a fertiliser. Incineration to process the toxic waste was reduced from 31 kg/kg of 7-ACA to 0.3 kg/kg of 7-ACA, an improvement factor of 100!

Also the company DSM (the Netherlands) has provided figures for the production of 7-ADCA (7-amino-deacetoxy cephalosporanic acid), an intermediate for

the synthesis of the semi-synthetic cephalosporin antibiotic cephalixin. From 1975 to 1985, cephalixin was produced in a ten-step process with conventional chemistry and a waste stream of 30 - 40 kg per kg of final product. In 1985, the waste/product ratio was reduced to 15, as a result of long and tedious optimisation of the chemical process and the introduction of recycling. In 1995, DSM-Chemferm introduced a biocatalytic process, comparable with the one used at Biochemie. This resulted in further waste reduction to less than 10 kg per kg final product. This went even further as DSM developed a fermentation process for the direct production of 7-ADCA in a single fermentation step. Cephalixin is produced from this intermediate product by chemical synthesis. Thus, the original ten-step process for cephalixin was reduced to a four-step process, with 50 % cost reduction and 65 % less energy consumption. Besides the considerable reduction in the quantity of waste produced, the toxicity of this waste has been significantly reduced. The new process emits only aqueous waste with some harmless inorganic salts. The further use of modern biotechnology (such as metabolic engineering) can possibly reduce the number of steps even further. The production organism is to be modified such that cephalixin or a direct precursor are produced by means of fermentation, with the potential of further reduction in waste down to 2 - 5 kg/kg of cephalixin.

4.6. Bio-energy

4.6.1. *The perspectives of bio-energy*

In our society, a large proportion of the available resources are used to make energy. About 85 % of our energy needs are still met by fossil resources such as petroleum, natural gas and coal. The use of these resources for energy production is subjected to the conditions already mentioned, i.e. the depletion of these raw materials and their negative environmental effects, particularly the greenhouse effect. Additionally, there is an intensive search for renewable energy sources, such as hydraulic power, solar energy, wind energy, tidal energy, geothermal energy and also energy from biomass. Renewable raw materials can play an important role in the production of biomass, using green biotechnology, as well as with respect to the conversion of that biomass into useful energy carriers or bio-fuel through industrial (white) biotechnology.

A number of conversion processes based on industrial biotechnology seem to be particularly important for transforming biomass into useful fuel. The value of a fuel is not only defined by its energy content, but also by its physical form and ease of use. For example, in principle cars could run on firewood. However, this would be quite inefficient and not at all user friendly. In contrast to that, the production of bio-ethanol from e.g.

sugar beet leads to a compact and user friendly liquid fuel that can be mixed with normal gasoline and employed without having to adapt the car engine. The use of bio-ethanol thus fits perfectly into the current concept of mobility based on motorised vehicles, powered by liquid fuel and supplied over gasoline stations. In addition to that, the current agricultural practice (sugar beet cultivation) remains essentially unchanged.

These conversion processes are essential. They are performed with the help of industrial biotechnological processes in the aforementioned "bio-refineries". Again, the analogy with the petrochemical industry is striking: an automobile does not run on petroleum, but on a refined product derived from it, i.e. gasoline. The production of bio-fuels is expected to provide a strong stimulus for the development of bio-refineries. Once the infrastructure for the production of bio-fuels has been created in the bio-refineries, new possibilities will open up for producing more complex chemical substances. This is very similar to the early development of the petrochemical industry: initially it produced primarily fuel but subsequently an entire chemical industry developed around it.

Annexe B at the end of this document describes the production technology, application and market data of the main types of bio-fuels. Bio-ethanol is obtained from sugar beet or cane, wheat or corn by fermentation and can be used in mixtures with normal (petrochemical) gasoline. Bio-diesel is produced from vegetable oils such as rape seed oil and can be mixed without any problem with normal diesel fuel. Biogas is obtained by anaerobic fermentation of waste streams and can be burnt to produce heat and electricity. Conventional renewable fuels such as firewood and other energy plants are not considered here as they do not involve the use of industrial biotechnology. Also the potential of "green" biotechnology for improving crop yields or cultivation conditions is outside the frame of this report.

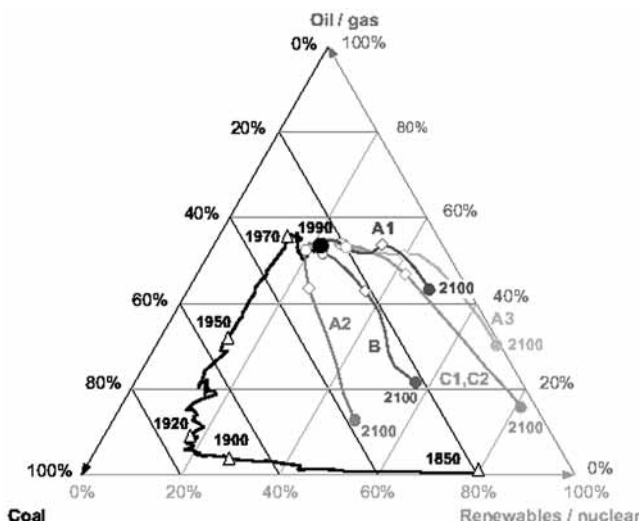
4.6.2. *World energy consumption and the possibilities of biomass for energy supply*

What will our future energy supply look like and will biomass take a realistic place in it? Many studies have been carried out in this respect and we will attempt to provide a short overview.

The various scenarios provide very different results, depending on the uncertainty on how long the current world reserve of fossil resources will last, the growth rate of the human population and the rate of economic development in the developing countries.

It is generally assumed that petroleum will be exhausted in about 50 years, natural gas in 65 years and coal

in about 200 years. The “proven oil reserves” in the world represent less than 40 years of consumption at today’s rate. As these reserves are increasingly found at difficult to reach places, the cost for extracting them, and with it the crude oil price, increases constantly. The same applies to natural gas. In a comprehensive study on global energy outlooks by the World Energy Council, several future scenarios were envisioned and developed. Scenario A assumes strong technological progress and resulting fast economic growth. Scenario B assumes a more modest, maybe more realistic technological progress. Scenario C is the most ecologically oriented and assumes strong economic development of the third world. All scenarios assume a worldwide population of 10 billion people in 2050 and 12 billion people in 2100. For scenario C, the world’s energy needs triple by 2050 (relative to 1990) and increase by five times by 2100. The result of scenario C1 is shown in the following graph from IIASA (International Institute for Applied System Analysis).



One can see from the graph that in scenario C1 the consumption of oil and coal clearly decreases from 2000 on. Only natural gas keeps up at an almost constant consumption over the coming 50 years, after which this source also dries up. At present, nuclear energy meets only a small percentage of our energy needs and will completely disappear by 2050. From 2000 on, solar energy and biomass strongly gain importance. By 2100 they will meet more than half of our energy needs. The proportion of traditional renewable fuels such as firewood diminishes continuously.

The next graph compiles the path of the other scenarios (A,1, A2, B, C1, C2). It is clear that no matter how the boundary conditions of the scenarios differ, they all clearly aim at the same corner of the graph in which all energy is produced by renewable energy sources (and to a limited extent by nuclear energy).

So basically, we are heading back to where the journey started in 1850.

The present (2003) worldwide energy consumption is estimated at 450 EJ (E = exa = 10^{18}) per year for 6 billion people, with an average energy consumption of 2,200 W per person ($1 \text{ W} = 1 \text{ Js}^{-1}$). This average energy consumption differs greatly (e.g. Bangladesh: 30 W per person versus the USA: > 10,000 W per person, the OECD average is 6,500 W per person, Belgium: 7,600 W per person).

At present, 79 % of the global energy consumption is met by fossil energy sources, divided over crude oil (35 %), natural gas (23 %) and coal (21 %). 9 % is provided by hydraulic power and nuclear energy. The remaining 12 % is met by renewables, particularly conventional firewood, wind energy and solar energy.

It is predicted that by 2040 the yearly global worldwide energy consumption will have increased to 935 EJ for 10 billion people; 3,000 W will be consumed per person on the average.

Solar energy radiation is the fundamental source of all energy on the earth (except for nuclear energy): it is the gigantic amount of energy of $2.8 \cdot 10^{24}$ J per year. This is 3,000 times the annual energy consumption of mankind in 2040. Hence, the amount of solar energy is enormous; the usable amount however is only a fraction of this and it will strongly depend on the technology how much of this energy could be harvested in the future. Solar energy can be directly recovered via solar collectors and photovoltaic cells. The indirect method to collect this solar energy uses photosynthesis for the production of biomass, which is far easier to store than direct solar energy in the form of electricity or heat. Photosynthesis is unfortunately a low efficiency process: the maximum theoretical yield is 6.6 %, in practice one obtains only 1 to 3 %. The direct use of solar energy can reach higher efficiencies. As can be seen from scenario C1, both forms of solar energy (direct and indirect) are expected to develop in parallel and should play an equal role in 2100. However, biomass reaches a fundamental limit at a certain point whereas the direct use of solar energy can continue to grow.

Okkerse & Van Bakkum (1999) assumed rather optimistic presumptions of strongly increased biomass yields per ha and a big increase in the amount of arable farmland. They obtained a total annual quantity of biomass for people of 65 billion tons (on 5 billion ha of land) by 2040. This is about 10 times more biomass than is harvested today worldwide!

These 65 billion tons can be consumed as follows:

- the production of food for 10 billion people in 2040 would require 15 billion ton of biomass per year, produced on 2 billion ha of farmland;

- 5 billion ton of biomass would be used for the synthesis of approximately 1 billion ton of organic products, fully meeting our demands for chemicals;
- the remaining 45 billion ton of biomass would be available for energy generation.

The use of these 45 billion ton of biomass for energy purposes can provide around 200 EJ. Estimating that the worldwide energy consumption in 2040 will be around 935 EJ, biomass is expected to supply a maximum of approximately 20 % of the worldwide energy demand. In a more optimistic scenario with “only” 8 billion people and an energy consumption of 1500 W per person, biomass is to provide approximately 50 % of the energy required.

Although this is not a small contribution, it is also clear that the entire energy supply for a future population of 10 billion people, at 3,000 W per person, can not be covered by biomass alone. Thus, biomass will never meet the total worldwide energy consumption, but can make a substantial contribution!

Thus, our future energy needs are to be met by different simultaneously measures:

- the development of bio-energy based on biomass;
- the development of solar energy via collectors and photovoltaic cells;
- the development of other renewable energy sources like hydraulic power, wind energy, geothermal energy;

and also – and particularly – by:

- reducing our energy consumption by means of improved energy efficiency;
- limiting the growth of the world population as much as possible.

The enormous effect of animal production is remarkable. Today, 40 % of all farmland in the world is used to produce animal food. This animal food is then used in animal husbandry to produce meat, a wasteful food conversion process with (very) low efficiency. If people were to switch over to a more vegetarian diet, a large quantity of agricultural raw materials would become available for energy production. In a future overpopulated world without fossil raw materials, people may perhaps need to choose between eating meat or driving automobiles...

It is very clear from the calculations and simulations that we are quickly approaching the limits of our natural resources, no matter what scenario we look at. Thus, the sustainable management of our mother earth is now needed more than ever.

4.6.3. *European directives, agricultural policy and detaxation of bio-fuels*

On April 8th 2003, the European Parliament approved the first of two directives, aimed at stimulating the

development of bio-fuels in Europe. Especially the fuel consumption of motor vehicles is tackled. The policy is to promote particularly liquid bio-fuels, i.e. bio-ethanol and bio-diesel. The transport sector accounts for no less than 32 % of total energy consumption in the EU.

The first European directive (2003/30/EC) sets the objectives: 2 % of the total engine fuel consumption in Europe is to be met by bio-fuels by 2005. After that, this minimum percentage is to increase progressively up to 5.75 % by 2010. The Commission further sets a long term goal of 20 % substitution by 2020. Today, the bio-fuel percentage in the EU is only 0.3 %, and is very unevenly distributed over the different member states. In order to reach these objectives, some 9,3 million tons of bio-ethanol are to be produced in Europe in 2010, requiring about 3.7 million ha of wheat and sugar beet. This figure should be compared with the 5.6 million ha of set-aside farmland, for which the European Community pays farmers to produce absolutely nothing!

A second European directive (amendment 92/81/EC) regulates the boundary conditions for the use of bio-fuels, particularly the tax aspects. These are very important in view of the fact that the taxation of petroleum based engine fuels in Europe is traditionally very high and accounts for more than half of the price at the gas station. Supporters of bio-fuels (typically farmers) consider that no (petrol) tax whatsoever should be levied because the bio-fuels are not made on the basis of petroleum. The opponents (typically the petrochemical concerns) point to the unfair competition when petrochemical fuels are to be taxed more heavily than bio-fuels. The directive deals with this matter and opens up the path for tax breaks (detaxation) for bio-fuels. The directive leaves it up to its member states to opt for complete or partial detaxation of bio-fuels, depending on their political will. Each member state must determine how it will best achieve the objectives of the first directive. In practice, the different implementation of this European directive into national legislation results in strong local differences. For example, Germany has opted to completely exempt bio-fuels from taxes, such that the use of bio-fuels is strongly stimulated. Other countries such as France have introduced partial detaxation, whereby bio-fuels are taxed less than petroleum based fuels.

It should not be forgotten that at the end of the story, the government pays the bill as it loses important tax revenues. The current difficult economic situation with low economic growth and budget deficits in most European countries is certainly not helpful in this respect. Supporters of bio-fuels claim that the operation is overall positive, also for the government. Expenditures for bio-fuels become ultimately revenue for farmers, who supply the agricultural raw materials for the bio-refineries that convert them to bio-fuel.

This revenue stimulates the domestic economy and ultimately benefits the government. At any rate, this revenue stays in the country, in contrast with the money paid for petroleum, which flows to the Middle East, where other values apply with regard to human rights, working conditions or concern for the environment. On the other hand, if the European farmers are to be helped, it would be wise to let them produce bio-fuels instead of just paying them to set-aside their land and do nothing. In this way, subsidies are replaced by bio-fuel revenue and the agricultural sector may once again become a normal sector in which market mechanisms predominate.

5. EXPECTED CHANGES IN SOCIETY AND TECHNOLOGY

Several changes in both society and technology are expected to happen in the coming years that may seriously modify the present order. Admittedly, these changes are likely to come about gradually, but may nevertheless give rise to some real "shock waves". If we are to overcome these changes, the technological basis of our society will have to change radically. The BACAS working group believes that the development of industrial biotechnology will play an essential role in this transition.

5.1. Changes in the supply of primary raw materials

It is expected that the first effects of the depletion of fossil fuels will be felt in the near future, particularly of crude oil (petroleum). Petroleum is very important for our society as it is the most important supplier of raw materials and our main source of energy at the same time. Therefore there is good reason to believe that the development of alternative raw materials, particularly those based on biomass, will be strongly stimulated. At any rate, there is a strong consensus that this major change will indeed take place, the only question is when and how fast.

5.2. Increased demand of a growing population for raw materials and energy

Currently, around 80 % of all available raw materials and energy are used by approximately 20 % of the world population. Naturally, it can be expected that the other 80 % of the world population will do everything possible to improve their living standards and thus require much more raw materials and energy than before. Also, one should not forget that the world population keeps growing at an alarming rate. The often cited and erroneously interpreted assertion of "reduced population growth" will only take effect in a few generations at the earliest, an unfortunate consequence of the peculiarities of population growth

dynamics. For the time being, the world population continues to grow faster than ever. In particular, the dynamics of China and India must be taken into consideration, in view of the fact that these population-rich countries are expected to improve their living standards in the short term.

All these effects will inevitably lead to a strong increase in the demand for raw materials and energy. A fair redistribution of the available raw materials is unlikely, so that in the end far more raw materials will be used globally. This will deplete the remaining fossil reserves and other raw materials even faster and bring renewable raw materials and energy to the forefront of attention.

5.3. Increased demand for efficiency in chemical production systems

The laws of the market economy provide strong pressures to continuously improve the efficiency of all production systems. Wasteful production systems that produced large quantities of waste were still economic in the past, either because this waste could be dumped into the environment or the cost of clean-up was shifted to the society. Nowadays, the principle of "the polluter pays" means such processes are doomed. Waste costs money, firstly to get rid of it and secondly, waste essentially means a yield loss with all its associated extra costs. Consequently, a high degree of efficiency and performance is required of all chemical processes today. Biocatalytic methods are particularly efficient, specific, with less waste, raw material use and energy consumption as a result. The penetration of industrial biotechnology into the chemical industry is almost always motivated by normal economic principles such as cost saving, increased efficiency, etc.

Furthermore, as it can be expected that the factors discussed previously will inevitably result in further price increases for raw materials and energy, the need for efficient chemical processes will grow even stronger. The further penetration of industrial biotechnology into the chemical industry and its synergistic cooperation with conventional chemical technology will no doubt be strongly stimulated.

5.4. Growing need for sustainability of the production systems

Since the Kyoto treaty, most industrialised nations have been obliged to respect a number of base criteria with respect to raw material use and energy policy. Our country and many others with it have undertaken efforts within the framework of the Kyoto treaty. The negotiability of CO₂ emission rights is now a fact. The first penalties for exceeding the norms soon will become effective. This is expected to result in a

fundamental perception change of the use of raw material and energy consumption. It is clear that renewable raw materials that are CO₂ neutral, such as biomass, will benefit from this development.

5.5. Changing consumer perception and behaviour

There was a time when consumers were only concerned with the quality and price of consumer products. Now, consumers also want to be informed about the production systems by which their products are made and even what happens with them after their use (waste, degradability,...). Production systems that cause ecological damage, cause animal suffering, are based on inequitable trade or exploitation (such as child labour), etc. are being increasingly rejected by consumers, even if they happen far away and strictly speaking do not directly burden or harm them. Consumers are looking for goods and services that are obtained and used under socially and economically acceptable circumstances, and do not raise any ethical or emotional dilemmas.

The European public's refusal of genetically modified crops is of this nature. It must be emphasized that often exactly the same consumers that reject genetically modified crops, use without grumbling and even enthusiastically products made by means of fermentation processes, such as fermented milk products or Quorn, a mycoprotein from fungi.

The consumers' perception of chemical products in their food (preservatives, colorants, anti-oxidants) is also very negative and natural alternatives are demanded. One can therefore expect increasing replacement of these "chemical" products by products obtained by industrial biotechnology.

5.6. Changes in the European Common Agricultural Policy

It is clear that the European agricultural sector will have to adapt continuously to new needs and problems, under the pressure of consumers, governments as well as non-European partners, particularly the USA and developing countries. Also, the expansion of the European Union with a large number of new member states from Eastern Europe will put a lot of additional pressure on the system.

Some guidelines for the change can be clearly distinguished:

- one wants an agriculture that is less oriented to (mass) production but more directed towards high quality agricultural products;
- one wants an agriculture with more respect for the environment;

- one wants to reduce or abolish altogether production-related subsidies for farmers;
- one wants to reduce import taxes for a number of agricultural products in order to allow developing countries to market their products in Europe;
- more diversity of agricultural systems is desired, such as new agricultural crops and new production systems, such as organic agriculture;
- one wants to stimulate the production of agricultural crops for non-food purposes.

The use of agricultural raw materials as a renewable raw material for the chemical industry and for fuel clearly meets the many expectations. Consequently, these developments are warmly welcomed, particularly by the agricultural community that has clearly understood the importance of industrial biotechnology.

6. POLICY RECOMMENDATIONS

In view of the importance of industrial biotechnology for the sustainable development of our society;
in view of the importance of industrial biotechnology for the Belgian industry, particularly the Belgian chemical industry and agro-industry;
in view of the importance of industrial biotechnology for the Belgian agricultural sector;
in view of the dependency of Belgium on imported fossil resources;
in view of the absence of a policy to stimulate industrial biotechnology in Belgium,

BACAS wants to put forward a number of recommendations, directed towards the government, the political and the industrial world. It hopes that a number of recommendations can be translated into an effective policy. Ideally, this can occur in a concerted action by the government, the involved industrial sectors and research institutions. The creation of a "technology platform for industrial biotechnology", bringing together all important stakeholders can make sure that the government, the industry and the academic world cooperate towards a common goal, as will be specifically addressed below.

6.1. Recommendations to the Belgian government

6.1.1. More governmental support for industrial biotechnology

The current governmental attention and its accompanying stimulation policy is almost entirely directed to the red (medical) and green (plant) biotechnology. Although these sectors certainly need attention and governmental support, a third important sector, industrial biotechnology, is and has been almost systematically neglected. Moreover, the public's acceptance of

the green biotechnology is very low at present, to such a point that nearly all new developments in this field cannot be applied within the borders of Belgium or Europe. Although the public's perception might change sooner or later, there are few reasons for optimism at present.

The red biotechnology on the other hand is much better accepted by the general public and is well integrated into large pharmaceutical companies. The very centre of this field is clearly located in the USA and not in Europe.

Industrial biotechnology has its industrial and technological basis in the chemical and agro-industrial industry. In this respect, Europe and Belgium have a good starting position and enjoy major advantages. In fact, Europe is privileged to possess the world's most extensively developed chemical industry. Particularly Belgium enjoys a leading position. Although it accounts for only 2.7 % of the European population of the European Union, the Belgian chemical industry represents:

- **8.2** % of turnover achieved in the European chemical industry;
- **14.0** % of the Belgian export;
- **6.5** % of investment in Belgium;
- **6.1** % of employment in Belgium.

The proportion of the chemical industry within the entire Belgian industry was no less than 24 % in 2002. Also the agro-industry is strongly developed in Belgium and achieved 4.5 % of its Gross Domestic Product (GDP).

The conditions for a strong development of industrial biotechnology in Belgium are therefore exceptionally favourable. The governmental support policy should be guided by the relevance for our society and on the future economic perspectives of the sector that is supported. In that respect, the Belgian government should strongly promote the development of industrial biotechnology, in view of the fact that it is a basic technology for the sustainable development of our society.

The Belgian authorities must urgently develop a long-term strategy and a vision for the development of industrial biotechnology in Belgium. Such a vision is at present lacking. Such a vision and strategy (for the next 20 to 30 years) should ideally be developed within the framework of the technology platform for industrial biotechnology (see 6.4.).

6.1.2. *More support for multidisciplinary applied research*

Industrial biotechnology is an application-oriented and multidisciplinary scientific domain that is based on different disciplines such as biology, microbiology,

molecular biotechnology, chemistry as well as environmental and engineering sciences.

The current research climate within the universities and the institutions for research support, and certainly in the biotechnology area, is strongly oriented towards fundamental research. If applied research in biotechnology is supported, it is limited to established sectors such as pharmaceuticals and the agro/food sector. Moreover, applied research is all too often seen as the exclusive task of companies, whereby governmental authorities are not expected to play an active role. The industry is quite conservative and hardly ventures into new research areas, especially during economically difficult times. Many companies therefore consider R&D in a new area with a technology change as the goal as too ambitious and difficult to accomplish.

The government must therefore also provide support to applied research in industrial biotechnology by means of different mechanisms, either at the company level or within the applied science (engineering) faculties of the Belgian universities.

The government should support the research and development efforts at universities via specific research programs, with research budgets that reflect the relevance for society and industry as well as the future perspectives of industrial biotechnology for our country. Care should be taken that this support covers the entire field of industrial biotechnology and not only the genetic ("genomics") or molecular biological aspects, as is all too often the case. The importance of the study of cell physiology, microbiology, metabolic engineering, biocatalysis, but also engineering-oriented bioprocess technology should not be forgotten.

Additionally, expansion support can be given to those companies that wish to commit themselves to this technology. The institutions for research support can stimulate a fruitful cooperation between the industry and universities by financially supporting common projects, e.g. via "public-private partnerships" and within the framework of the technology platform for industrial biotechnology (see 6.4).

6.1.3. *Attention for critical mass in Research and Development*

Research efforts in our country are strongly fragmented, without a clear coordination between the different technologies and disciplines. Moreover, there is a risk that the approaching reform of higher education (Bachelor/Master reform) results in a further fragmentation of the research efforts. BACAS strongly believes that the universities must continue to function as the most important knowledge and competence centres in this country, such that sufficient critical mass with

respect to Research and Development can be maintained. The consequences of fragmentation are serious: research groups often lack the critical mass with respect to people and funds. Moreover, the stimulation programs for Research and Development frequently finance only one in 10 projects, a waste of energy, time and people.

For a long-term strategy and vision however, more is needed than just critical mass. We have too many research islands, each working separately in their narrow domain. For the stimulation of Research and Development in industrial biotechnology, interdisciplinary research clusters and networks must be created that can grow into real centres of excellence.

6.1.4. *Promotion of knowledge and awareness of industrial biotechnology*

The public's perception of biotechnology is generally negative. It is a new technology, evolving at rocket speed, and often negatively presented in the media (just think about the cloning of people, "Frankenstein food", etc.). As most people have no basic knowledge about this technology, it's very easy to create a negative image.

The possibilities of industrial biotechnology offer an excellent opportunity to improve upon this image, especially in view of the fact that there is a clear connection between industrial biotechnology and the sustainable development of our society.

In this respect, education should receive special attention:

- education about biotechnology should be introduced at the secondary education level. In order to do so, teachers can be encouraged to integrate aspects of biotechnology into their teaching package. This should go beyond the biology course and extend into the subject areas of chemistry, ecology, ethics, etc. Teachers should also be encouraged to obtain general knowledge of biotechnology (both technology and applications). In practice, specific seminars on biotechnology can be organised for teachers.
- higher education should also be better adapted to the future and the needs of corporate life. More interdisciplinarity is clearly required in the curricula. Also basic knowledge about biotechnology must be inserted as part of the curriculum of chemistry, environmental sciences, engineering, physics, but equally so in the social sciences.

The possibilities that are offered by industrial biotechnology should also be communicated to the population as a whole. There is a general lack of awareness and knowledge about industrial biotechnology. It is extremely important that the authorities play a cata-

lytic role. This can be done e.g. by organising lectures and seminars, providing information brochures, etc.

6.1.5. *Development of political and fiscal support measures*

In order to develop and promote the industrial development and application of industrial biotechnology in Belgium, a number of political support measures are needed.

6.1.5.1. *Detaxation of bio-energy in a European perspective*

The different authorities in our country should urgently get together and implement the European bio-fuel directives into national legislation. Full detaxation of bio-fuels seems attainable and should be aimed at. The importance of a fast implementation of the legal framework should not be underestimated. A fast government action creates freedom of initiative to the industry and any delay in this leads to major handicaps for the Belgian industry. At present, there is no Belgian position or action in sight regarding bio-fuels and this keeps our industry in frustrating wait-and-see mode, in sharp contrast to their German and French competitors. These have a much better starting position and can move into action, thanks to the pro-active policy of their governments. Critical mass, increase of scale and efficiency are the keywords in this industry. If one gets on the train too late, one may never catch up.

Also within the framework of the Kyoto treaty, even if Belgium would not be able to respect it, the motto should be "better late than never". The creation of a well structured bio-energy policy in Belgium is urgently needed, all the more since this evolution is sufficiently supported and politically well structured in a European perspective. Belgium does not need to play "cavalier seul" but only needs to follow the crowd. If the current lack of initiative continues, Belgium risks being too late altogether.

A properly structured bio-energy project in Belgium would offer the following advantages:

- the project gets the public's attention as each time a person fills up at the gasoline station, he will be confronted with this aspect of sustainable development;
- the project helps to achieve the Kyoto objectives, to which Belgium has committed itself;
- the project supports the development of industrial biotechnology and will stimulate the creation of bio-refineries in Belgium;
- the project builds bridges between the agro-industry and the chemical industry and can initiate a better understanding and collaboration for new common projects;

- the project creates new revenue sources for the agricultural sector and improves the image of agriculture.

6.1.5.2. Promote market penetration of sustainable bio-products and processes

The commercialisation of technologies and products that have been proven to be environmentally friendly should be promoted through specific (fiscal and financial) support measures. An obligation to use e.g. biodegradable packaging material for certain (disposable) applications can help such products by speeding up their market penetration. Such a support policy can help achieve bigger sales volumes, making them more competitive against conventional packaging materials. Without support, conventional plastics will continue to control the market because of their dominant position, mature technology, depreciated production installations and inexpensive large-scale production.

6.2. Recommendations to the European authorities

6.2.1. *Develop a European policy for industrial biotechnology*

Although many applications are already known, industrial biotechnology is still in its infancy in Europe. A European policy for industrial biotechnology is needed with effective support measures to remove the obstacles for the implementation of this environmentally friendly technology, including support measures to innovate production processes or to make them profitable.

This fits perfectly within the policy framework that the European Union has defined for itself. For example, at the European Council Meeting in Stockholm in February 2001, the European Commission pointed to the enormous economic, social and environmental potential of life sciences and biotechnology and to the strategic and long-term importance for Europe to have access to these sciences, technology and their applications. In a communication from the Commission, "Towards a Strategic Vision of Life Sciences and Biotechnology", the European Commission stated: "Life sciences and biotechnology have entered a stage of exponential growth, opening up a vast potential for sustainable development of our society and economy in Europe and the rest of the world, and for the improvement of the quality of life. They are therefore of strategic importance in Europe's quest to become a leading knowledge-based economy. Europe cannot afford to miss the opportunity that these new sciences and technologies offer". This has resulted in an action plan for the development of biotechnology in Europe: "Life sciences and biotech-

nology: a strategy for Europe", in which particular attention is given to industrial biotechnology.

Also, much attention has been given to sustainable development. In June 2001, the European Council in Göteborg adopted a strategy for sustainable development: "A sustainable Europe for a better world: A European strategy for Sustainable Development", whereby the industry was asked to cooperate on the development and the use of new environmentally friendly technologies. The need to change to more sustainable production processes is also one of the objectives in the 6th Environmental Action Programme of the European Union. These noble intentions should however be translated into concrete actions to promote industrial biotechnology. Unfortunately, until now this is hardly the case.

6.2.2. *More attention for industrial biotechnology in the European Research and Development Programs*

The European Union recently launched the plan to increase European Research and Development efforts to 3 % of GDP (actually 1.94 %) ("EU Action Plan to boost research efforts in Europe"). These noble objectives still need to be translated into concrete action. Furthermore, increasing research budgets will do no good unless these funds are spent at the right places.

The Sixth Framework Programme for Research and Technological Development (FP6) is one of the most important policy instruments of the European Commission for supporting Research and Development. It is striking that industrial biotechnology is virtually absent in FP6. Although it must be recognised that some aspects of industrial biotechnology find a (small) place in FP6 (some aspects of industrial biotechnology and sustainable chemistry are integrated into "nanotechnology", while bio-energy is found under the heading "Sustainable Development"), no real priority is given at all to this technology in FP6 (in contrast to e.g. biotechnology for medical use or ICT). It is therefore essential to give more "space" to industrial biotechnology in the next framework programme. The creation of a technology platform for industrial biotechnology can offer the right framework, a possibility that the European Commission created itself ("Industrial policy in an enlarged Europe").

Time is running short in this respect. Otherwise, we may see exactly the same development for industrial biotechnology as what has already been experienced in other biotechnology areas, where the USA has clearly acquired the technological leadership position. Some companies have already closed their European research departments and moved them to the USA.

6.3. Recommendations to the Belgian industry

6.3.1. *Collaboration between the chemical industry and agro-industry*

Examples abroad (e.g. vitamin C and bio-plastic), clearly indicate that a collaboration between the agro-industry and the chemical industry has much potential. Both industrial sectors are well developed in Belgium, so that conditions are favourable. However, collaboration between these industrial sectors is not easy and they differ greatly with respect to industrial practice, tradition and technological base. Today, there are no “exemplary” Belgian collaborations in this respect and each industrial sector lives its own life.

The Belgian chemical industry is heavily based on petroleum; the massive presence of the petroleum refineries in Antwerp is indicative for this. It would be positive if the chemical industry in Belgium studies the perspectives of industrial biotechnology for their production and development activities. On the one hand, this can lead to lucrative applications and process improvements; on the other hand, it prepares the future with respect to renewable raw materials. This is also one of the recommendations of the exploratory study “The Chemical Industry in Flanders - towards 2010”, carried out by the Flemish Council for Scientific Policy.

The argument that Belgium is not a nation with strong agricultural resources and thus does not offer the right conditions for such activities makes no sense. Agricultural raw materials can be imported just like crude oil and form the basis for new chemical activities.

The Belgian agro-industry is heavily oriented towards producing raw materials for the equally well developed Belgian food industry. This priority is opportune but offers little perspectives for growth, in view of the fact that our human food requirements are largely saturated. A part of the Belgian agro-industry holds a defensive strategy to secure its acquired rights, e.g. the sugar industry, that above all does not want to loose its quotas. A more offensive strategy towards expansion by new activities in the area of industrial biotechnology would be far more constructive. Some good examples of such novel activities exist, e.g. the conversion of old sugar factories to a lactic acid factory (Galactic) or inulin factory (Orafti).

6.3.2. *Invest more in Research and Development*

Industry should invest more in Research and Development, through internal and external cooperation, e.g. with universities. The particularly ambitious research efforts that the USA initiate for industrial biotechnology may lead to devastating effects for our industry if a suitable response from European companies is not found. Industry has an important role to

play with respect to creating interdisciplinary research clusters or by participating in a technology platform for industrial biotechnology.

6.3.3. *Promotion of knowledge and awareness of industrial biotechnology*

Industry also has an exemplary function. In order to promote awareness and knowledge about industrial biotechnology, production units based on industrial biotechnology should be clearly promoted (with brochures, visits, etc.). This can simultaneously improve the image of that industry and biotechnology in general.

6.4. Creation of a technology platform for industrial biotechnology

In order to establish a long term strategy, it is essential to create a “Technology Platform for Industrial Biotechnology”. Such technology platform can make sure that authorities, industry and the academic world get to know each other and cooperate effectively.

Such platform must unite all important stake-holders (industry, academic world and research institutes, representatives from environmental and consumer associations and the different authorities). A long term vision and strategy must be developed within such a technology platform by creating a “think tank”. Attention should focus on Research and Development, but also on a suitable and coherent legislative framework and on the necessary measures for integrating this technology into society. In order to be effective, the programs should be specifically aimed at industrial biotechnology and not towards biotechnology in general.

The European Commission has already clearly indicated the possibilities of such technology platforms in several communications (“EU Action Plan to boost research efforts in Europe” and “Industrial policy in an enlarged Europe”). Local authorities should however also be active, particularly when it comes to developing the specific strengths of the region.

In view of its strong concentration of agro-industry and chemical industry, Belgium should strongly develop the concept of “bio-refineries” and the different technologies of industrial biotechnology. This technology base should cover all aspects of industrial biotechnology such as fermentation technology, enzyme technology, biocatalysis, metabolic engineering, molecular biology, bioprocess technology and control and downstream processing technology. This should be done in a structured, strategic and goal-oriented manner, in order to achieve the world class technology base necessary to develop the sustainable chemical industry of the future.

7. CONCLUSIONS AND PERSPECTIVES

Industrial biotechnology can synthesise a broad range of chemical substances, usually by using useful micro-organisms and their enzymes. The recent wave of new applications seems to indicate that only the tip of the iceberg has been touched. The microbiologist Jackson Foster already predicted in 1964: "Never underestimate the power of the microbe" and has been proven right so far.

This technology, already strongly developed in conventional domains of the food and health care industry, now also strongly penetrates the chemical industry with applications in fine and bulk chemistry, polymer synthesis, pharmaceutical industry and the energy sector. As these processes and products are largely based on renewable raw materials and possess substantial ecological benefits, this provides them with a major advantage in the perspective of sustainable development.

Science, industry and policy people alike should give more attention to this green chemistry and its bio-products. Successful innovation by a biotechnological product or process is never solely defined by technology and science but equally by other factors such as acceptance by the general public, the innovation climate and support by the authorities through a consistent Research and Development policy.

BACAS hopes that this document may serve as a real eye-opener for its readers and that the groups targeted in our recommendations will be motivated to take appropriate action, in order to stimulate the development of this discipline.

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Annexe A

Examples of products produced by industrial biotechnology

A1. Food additives and food supplements

Amino acids are very important natural building blocks of proteins. They are increasingly used as supplements for human food and animal feed. Previously, only a small number of amino acids were made by industrial biotechnology, Nowadays, almost all 20 natural L- amino acids are produced by fermentation or enzyme technology.

These are very large-scale industrial productions. The world-wide production of L-glutamic acid is over 1 million tons a year. It is one of the most important fermentation products with a tonnage comparable with many petrochemical products. Glutamic acid is used in the form of monosodium glutamate (MSG) as a taste enhancer in many foods. L-Lysine (350,000 t/year) is another large-scale produced amino acid, mainly used in animal feed.

L-Phenylalanine is yet another amino acid, taking part in the synthesis of L-aspartame. Aspartame is an artificial sweetener that is 200 times sweeter than sugar. It is used in many foodstuffs, such as "light" beverages. Worldwide, around 15,000 tons of aspartame are produced each year at an approximate world market price of 35 €/kg. The initial synthetic process for aspartame was based on chemical synthesis. But, in the mean time, it is now strongly based on industrial biotechnology. For example, the most important building blocks, L-phenylalanine and L-aspartic acid, are produced by fermentation and biocatalysis, respectively. The Holland Sweetener Company uses enzymatic technology to connect the two building blocks: the amino acids phenylalanine and aspartic acid are very specifically linked to one another by the bacterial enzyme thermolysine. After that, a few more chemical steps are needed to obtain the sweetener aspartame.

L-Carnitin is a vitamin-like natural component in animal tissues that stimulates the lipid metabolism. Initially, L-carnitin was produced via chemical synthesis, but now it is entirely made through a fermentation process, starting from renewable raw materials. The L-carnitin obtained is very pure and is increasingly used. People and animals use L-carnitin as a food supplement to stimulate their fat metabolism (more energy, less fat synthesis and more growth).

Erythorbic acid or iso-ascorbic acid is an anti-oxidant used in food. It is made by a fermentation process from glucose (Pfizer, Roquette). It is a chemical analog of vitamin C, but has no vitamin action. By fermentation with bacteria, glucose is almost quantitatively converted to 2-keto gluconic acid that is then chemically cyclised to erythorbic acid. These are just a few examples from an ever increasing list, not in the least due to the more positive image that such biotechnologically made food additives enjoy, contrary to their chemically prepared counterparts.

A2. Bio-pesticides

The worldwide bio-pesticide market amounts to 130 million Euro. They are also referred to as "Biological Control Agents" (BCA). These biopesticides impose less of a burden on the harvest, are very specific, do not leave any toxic residues and are completely biodegradable. Also their developmental costs are favourable with respect to their chemical counterparts.

Bio-insecticides on the basis of bacteria, fungi and viruses have long been known and have been commercialised. Various *Bacillus thuringiensis* bacteria produce d-endotoxins, proteins that are toxic for insects but totally harmless for human beings. These bacteria are grown in fermentation processes and are used as a bio-pesticide to kill insects in

agriculture and forestry, grain storage, etc. Green biotechnology has further improved the efficiency of this principle, by using recombinant DNA technology to insert the gene that codes for this d-endotoxin into various agricultural crops such as corn, turning them resistant to insect infestation. Also living fungi preparations, insect viruses and even parasitic nematode worms are employed as a selective insecticide because they aggressively infect insects, making them sick and eventually killing them. Such preparations are obtained by fermentation processes and are sprayed on fields and forests.

Bio-herbicides are also receiving lots of attention. Certain weeds can be selectively fought with well-defined phytopathogenic fungi: they are called mycoherbicides. Spores of *Colletotrichum gleosporioides* (COLLEGO) are used against vetch in rice and soy bean cultures; *Phytophthora palmivora* (DEVINE) against choking weed in citrus cultures in the USA. These fungi are produced by fermentation. After the weed has been killed, the fungi also die.

Bialaphos (gluphosinate, a phosphinotricin derivative) is a *Streptomyces* fermentation product, employed as a total herbicide. It was discovered and developed in Japan (Meiji Seika Co). It is a tripeptide analogue that inhibits the glutamine synthesis in plants, leading to their death. It is widely applied in agriculture, but also on rail track beds, roads and squares. On the basis of this knowledge, green (plant) biotechnology has developed transgenic plants that are gluphosinate resistant.

Industrial biotechnology is also increasingly applied in **the synthesis of conventional pesticides**. The synthesis of S-chloropropionic acid, an intermediate product in the synthesis of chiral phenoxypropionate herbicides (2,000 t/year) serves as an example. These herbicides recently became important and industrial biotechnology is particularly suitable for their synthesis. One starts with racemic chloropropionic acid that contains a mixture of R- and S- forms, whereby only the S-form results in an active herbicide. With the help of an R-specific dehalogenase enzyme from *Pseudomonas* bacteria, only the R-form is converted that can be separated and recycled. Another bioprocess starts from glucose, that is converted to D-lactic acid using fermentation. The D-lactic acid is then chlorinated chemically to S-chloropropionic acid.

Thus, so-called homochiral herbicides are synthesised, containing only the active chiral form. In comparison with the conventional herbicide mixtures, containing both forms, only half the amount of chemicals need to be applied on the field for obtaining the same efficacy.

A3. Bio-colorants, flavours and aroma compounds

Bio-colorants are increasingly produced by industrial biotechnology, in particular when these are employed for food, pharmaceutical or cosmetic applications. These substances can often be made by both chemical synthesis and industrial biotechnology, with comparable production costs. Bio-colorants produced by biotechnology have an important marketing advantage with respect to entering the market because consumers disapprove of synthetic substances.

β -Carotene (or provitamin A) is produced by organic synthesis, extraction from roots as well as by fermentation with the fungus *Blakeslea trispora*. Optically active hydroxycarotenoids such as zeaxanthin and astaxanthin are important as animal and human food, and are mainly used in the fish and animal feed industry. The pink pigment astaxanthin is e.g. added to the feed of sea-farm raised salmon to obtain the beautiful pink salmon meat. In nature, salmon get the pigment from their natural diet. Until recently, astaxanthin was mostly synthetically produced, via a complex synthesis route using a combination of chemical and enantio-selective bioconversion steps. Recently, there has been growing interest in the direct fermentative synthesis of this pigment with the help of the red yeast *Xanthophyllomyces rhodozyma*, because the synthetic variant has been criticised in view of the fact that it differs slightly from natural astaxanthin. A blue food pigment – phycocyanin – is produced in Japan with the cyanobacterium *Spirulina* sp. Also the orange red food/drink pigment – monascin – is produced with the fungus *Monascus purpureus* via a fermentation process.

Flavours and aroma compounds can also be produced by fermentation or enzymatic technology. The German company BASF recently started with the microbial synthesis of 4-decalactone, a peach aroma. It is based on a fermentation process with the yeast *Yarrowia lipolytica*, whereby 12-OH-19 octadecenic acid is released from ricinus oil and subsequently metabolised to the desired 4-decalactone. Unilever in England makes the butter aroma, R-dodecanolide, starting from 5-ketododecane acid with the help of baker's yeast as the biocatalyst. Butyric acid and its ethyl-esters have been obtained by fermentation since a long time and are used in cheese aroma, fruit aroma,....

A4. Solvents

The most important "green" solvent today is ethanol, obtained by fermentation from sugar or glucose. Ethanol is a widely used solvent in the chemical industry because it is available in large quantities, is very pure, inexpensive, not toxic and perfectly biodegradable. The world production in 2002 was 26 million tons, of which 63 % was used as a bio-fuel, 12 % for human food and beverages and 25 % in the chemical sector. It is produced for the most part by fermentation. Only 9 % is still being produced by the petrochemical route.

Ethyl lactate is a strong newcomer solvent. Ethyl lactate is made from ethanol and lactic acid, both produced from glucose or sugar by fermentation processes. It is also non-toxic, inexpensive and has other solvent properties than ethanol. A great future is expected for this new solvent but its availability is still a limiting factor, given that it just recently entered the market.

A5. Plastics or bio-plastics

Recently, numerous plastics have been commercialised on a large scale, in which industrial biotechnology has a significant part in their synthesis. The production process typically consists of an intelligent combination of conventional chemical polymer technology and industrial biotechnology. Industrial biotechnology usually participates in the synthesis of monomer building blocks of these plastics. These

monomer building blocks are then converted to plastics by means of conventional (chemical) polymerisation technology.

Mitsubishi Rayon produces acrylamide from acrylonitrile with the help of an immobilised bacterial enzyme nitrile hydratase. Acrylamide is then polymerised to the conventional plastic polyacrylamide. This process was one of the first large-scale applications of enzymes in the bulk chemical industry and replaces the conventional production process that uses sulphuric acid and inorganic catalysts. The enzymatic process has clear advantages with respect to the chemical alternative as indicated in the following table. The efficiency of enzymatic conversion leads to less waste, higher yields and significantly lower energy consumption with consequently reduced CO₂ production, as indicated in the following table:

	Chemical process	Bioprocess
Reaction temperature	70°C	0 – 15°C
Single-pass reaction yield	70 – 80 %	100 %
Acrylamide concentration	30 %	48 – 50 %
Product concentration	necessary	not required
Energy demand (steam and electricity-demand in MJ/kg acrylamide)	1.9	0.4
CO ₂ production (kg CO ₂ /kg acrylamide)	1.5	0.3

Surprisingly, the main reason for introducing the biotechnological route was the much better product quality. No undesired polymerisation occurs when the biotechnological route is employed, resulting in a purer acrylamide that can be better polymerised for high-quality applications. Today, about 100,000 tons of acrylamide are produced yearly via this method in Japan and other countries. In this case, a conventional plastic is produced from petrochemical raw materials with the help of industrial biotechnology; renewable raw materials are not involved here, apart from the enzyme used.

Sorona 3GT is a new polyester synthetic fibre produced by DuPont. 1,3-propanediol is one of the monomers for the production of this polymer. It is made by fermentation from renewable raw materials, i.e. glucose, derived from corn. In a collaborative project between Genencor and DuPont, an *E. coli* production strain has been equipped with 4 foreign genes from other micro-organisms. As a result, the recombinant production organism converts glucose to 1,3-propanediol, which it naturally does not produce. This is a fine example of so-called "metabolic engineering". The monomer is usually produced from the petrochemical raw materials ethylene oxide or acrolein via conventional chemical synthesis, but can now also be produced at comparable cost using biotechnology from renewable resources. The new polymer Sorona is mainly used as a synthetic fibre in the textile industry. It is not biodegradable and is thus a conventional plastic in that sense.

Natureworks bio-plastic (PLA, Poly Lactic Acid) has been made (140,000 t/y) in the USA by Cargill-Dow since 2002 from glucose, derived from corn. In a first step, glucose is

converted to lactic acid by fermentation, which is subsequently polymerised to Poly Lactic Acid (PLA). The properties of the polymer are quite comparable to conventional polymers such as polyethylene or polypropylene and it is used in the packaging and textile industry. The polymer is completely biodegradable (compostable) so that packaging, plastic cups, etc. can be simply put on the compost heap after use, together with the organic waste. In this case, one closely approaches the ideal situation: a completely biodegradable plastic is produced from a renewable raw material with the help of industrial biotechnology. Moreover, the new biodegradable plastic has technological properties

very comparable to those of the conventional polymers, greatly facilitating its market introduction.

Thus, industrial biotechnology penetrates into the production of plastics, either a single step in a conventional polymer synthesis to completely new biodegradable polymers, produced from renewable raw materials. In all cases, these are large-scale productions of inexpensive plastics, a sector, in which renewable raw materials in combination with industrial biotechnology appear to be competitive. The table below compiles the characteristics of above-mentioned processes:

Plastic	Raw material	Renewable raw materials used	Monomer	Biodegradability of the plastic	Technology used
Polyacrylamide	Acrylonitrile	No	Acrylamide	No	Biocatalysis
Sorona [®] polyester	Glucose	Yes	1,3-propanediol	No	Fermentation
Natureworks [®] PLA	Glucose	Yes	Lactic acid	Yes	Fermentation

A6. Vitamins

Vitamins are important fine chemicals that are produced in relatively large quantities. Whereas a number of vitamins can be prepared only via biotechnology such as vitamin B₁₂, an extremely complicated compound, other more simple vitamins can be produced by either a chemical route or a biotechnological route and quite often by a combination of both.

The synthesis of vitamin B₂ (riboflavin, 4,000 t/y) is a good example of this. The conventional process consisted of the synthesis of the building block D-ribose by fermentation with *Bacillus* bacteria, followed by a sequence of chemical reactions to obtain riboflavin. Thus, this was a combined chemical-biotechnological synthesis route of no less than 8 steps. This combined synthesis route has been recently replaced by the complete biotechnological synthesis of riboflavin in one single fermentation step with the help of bacteria, yeast or fungi (respectively by Roche, ADM and BASF). The productivity of these fermentation processes is so high that the product already crystallises out during the fermentation itself! The production cost of the new biotechnological process is 40 % lower than the conventional process.

Another example is the synthesis of vitamin C (ascorbic acid), that is made conventionally via the Reichstein-Grüssner synthesis, a synthesis process starting with glucose and consisting of one fermentation step and 5 chemical steps. Cerestar/BASF recently developed a new process in which a fermentation process takes over the greatest part of the chemical steps. The new synthesis route consists of one fermentation step and two simple chemical steps (via 2-keto-L-gulonic acid). Moreover, one works very hard on a fully biotechnological route that will convert glucose to vitamin C in a single fermentation step.

A7. Fine chemicals and pharmaceuticals

Today, industrial biotechnology has the greatest degree of penetration in the fine chemical and pharmaceutical sector

(15 %), with further strong development underway. Antibiotics and their intermediates are among the most important fine chemicals, with a world market value of about 20 billion euro. They are almost exclusively made by fermentation processes with the help of specially selected micro-organisms. The structural complexity of most antibiotics is so great that chemical synthesis has never been a serious alternative. Only in the case of so-called semi-synthetic antibiotics are the building blocks obtained by fermentation, and subsequently chemically modified to obtain new antibiotic derivatives with improved effectiveness. Nowadays, these chemical modifications are increasingly replaced by biotechnological methods, with excellent economic and ecological benefits (see frame).

Another example in the pharmaceutical sector is the synthesis of Captopril, a so-called ACE inhibitor used to treat high blood pressure. Captopril is built from two building blocks D-b-hydroxy-isobutyric acid and L-proline. These building blocks are both synthesised by fermentation, respectively with the yeast *Candida rugosa* and the bacterium *Corynebacterium* sp. Both building blocks are then linked by conventional chemistry, directly resulting in Captopril.

In the fine chemical sector, Lonza has developed a biotechnological route, starting with 3-cyanopyridine to nicotinamide (niacin or vitamin B₃), nicotinic acid and 6-hydroxynicotinic acid. At present, these intermediate products for many chemical syntheses are made via industrial biotechnology. Conversions are done by means of enzymatic hydrolysis with nitrile hydratase from *Rhodococcus* bacteria or by bioconversion with living bacterial cells. The reactions are very specific and the yields are almost quantitatively.

In the sector of enzymatic conversions, Novozymes has introduced an extremely thermostable lipase from the yeast *Candida (Pseudozyma) antarctica* (Novozyme 435). It is excellently suitable for carrying out specific esterifications in organic solvents. This enzyme is widely used today in different sectors of the chemical industry.

Annexe B

Bio-fuels: technology, application and market

B1. Bio-ethanol

Bio-ethanol (alcohol or ethyl alcohol) is produced by fermenting sugars, usually with the help of yeasts. These sugars can be obtained from numerous raw materials such as sugar beet, sugar cane, wheat, corn or organic waste. In Europe, most alcohol is produced from sugar beet or wheat, leading to an easily fermentable substrate. More research is now directed towards producing alcohol from more difficult substrates such as organic waste, either from agro-industry or domestic waste (e.g. vegetable, fruit and garden waste).

On the one hand, by genetic modification, a number of micro-organisms have been modified such that they can convert more complex substrates like pentose (C5) sugars and cellulose to alcohol. These "superbugs" are then used to convert more complex substrates like bagasse, domestic waste, straw, paper to ethanol. A lot of work has been invested in this technology, particularly in the USA. The whole field has been strongly stimulated by the US government, already providing several pilot installations and some industrial realisations.

On the other hand, much effort has been put into biotechnological research for inexpensively producing and improving the required cellulase enzymes. These "super-cellulases" must hydrolyse cellulose to glucose that can be easily fermented.

After the fermentation process, usually alcohol is obtained by simple distillation from the fermentation liquid, resulting in

a very pure product. This alcohol must be dehydrated before it can be used in motor fuel, usually by membrane processes. This dehydrated alcohol is commercialised as so-called bio-ethanol and can be used in motor fuels in different forms, typically in mixtures with normal gasoline. On the one hand, bio-ethanol can be converted with the petrochemical intermediate isobutylene, and the ETBE (Ethyl Tertiary Butyl Ether) obtained added to normal gasoline. On the other hand, bio-ethanol can be directly added to normal gasoline without any further elaboration, usually up to a maximum percentage (5 % is permitted in Europe). In the current European practice, it is mostly added in the form of ETBE. In the USA and Brazil, ethanol is frequently directly added to gasoline and in higher percentages. The use of these ethanol/gasoline mixtures does not require any engine adaptations up to an addition percentage of 15 %. In fact, adding bio-ethanol or ETBE increases the oxygen content of fuel, leading to a greatly improved combustion. Moreover, ETBE is often added to gasoline to serve as a lead substitute. Thus, ecological advantages are achieved also in this respect. Without realising it, a considerable part of the European population is already driving on a (small) percentage of bio-fuels!

About 1.6 million tons of ethanol was produced in Europe in 2002, of which 225,000 ton is used as a bio-fuel, mainly in Spain, France, and Sweden. Europe stands very weak in this respect compared to the USA and Brazil, where respectively 5.7 and 8.7 million ton of ethanol is produced as a bio-fuel (see table below). In the world as a whole, 26 million ton of ethanol was produced in 2002, 63 % of which was used as a bio-fuel. Hence, Europe only makes about 6 % of the ethanol in the world.

Bio-ethanol production and consumption in 2002

Country	Most important raw material	Total production bio-ethanol (million t/y)	Bio-fuel application (million t/y)	Percentage bio-fuel (million t/y)
Brazil	Sugar cane	9.5	8.7	92 %
USA	Corn	6.4	5.7	90 %
Europe	Sugar beet and wheat	1.6	0.2	14 %
World	various	26	16.4	63 %

Fuel cells are used to convert fuel directly to electricity, typically at higher efficiency than by conventional combustion and associated electricity production. Hence, fuel cells can considerably improve the efficiency of our electricity supply. Particularly the use of fuel cells in motor vehicles is potentially more efficient than present combustion engines. For this purpose, compact, reliable, long-lasting and inexpensive fuel cells must be developed, to mention just a few of the difficulties that this technology has to address. Within the framework of bio-fuels, the development of fuel cells on the basis of bio-ethanol is very promising. The high degree of purity of this fuel creates in principle favourable conditions to develop efficient, reliable and long-lasting fuel cells based on bio-ethanol.

B2. Bio-diesel

Bio-diesel is produced from fats and vegetable oils. Except for recovered fats and oils (e.g. used frying fat), most bio-diesel in Europe is made from rape seed oil. Essentially, bio-diesel consists of methyl esters of C₁₆-C₁₈ fatty acids. After so-called transesterification with methanol, the fat fraction of rape seed oil is separated into its components, bio-diesel and glycerol, that has other useful applications. The technology is based on a simple chemical process (base catalysed trans-esterification). Biotechnology is normally not implicated here although work is performed on a biotechnological alternative for the current chemical production. Lipase enzymes might circumvent the use of alkaline

agents, which can make the whole production process considerably greener.

The bio-diesel obtained can be directly mixed to normal diesel fuel, typically up to 5 %. In France, a 30 % mixture is also used (referred to as diester) and in Germany and Austria even pure bio-diesel is used. Bio-diesel addition requires absolutely no adaptation of the diesel engines. Quite the contrary, the addition of bio-diesel is well appreciated because of its engine-lubricating action. When pure bio-diesel is used, some problems may occur in winter due to cold crystallisation. The production of bio-diesel is growing rapidly and has passed the mark of 1 million ton per year in 2002, mainly produced in Germany, France and Italy.

B3. Biogas

Biogas usually is the result of methane fermentation of biomass. This process uses a consortium of different microorganisms that can transform complex organic material to carbon dioxide (CO₂) and methane gas (CH₄). These molecules distil spontaneously from the liquid and this biogas normally contains about 70 % of methane and has an energy content of approximately 20 – 25 MJ/m³. This biogas can be burnt and used for the production of electricity and heat. Typically 1 kg of dry organic matter (sugar, plants, etc.) corresponds to 1 kWh of electricity and 2 kWh of thermal energy, produced on the basis of biogas.

It must be stressed that this process is very efficient in the sense that around 90 % of the energy content of the raw material is recovered by the collected biogas, even starting with liquid waste such as manure and sludge, which cannot be burnt under normal conditions. The remaining product

after methane fermentation contains only minerals released from the organic material, as well as some residual recalcitrant organic molecules such as lignin. These residues can be brought back to the soil as a fertiliser for plants and as a source of humus. This concept of biomass to biogas has been fully developed and tested in practice. One hectare of corn can produce 48,000 kWh of biogas that can be transformed into 14,500 kWh of electrical energy and 24,000 kWh of useful thermal energy. The economics of the process are at present insufficient as it yields only a gross margin of approximately 400 euro per ha to the farmers, whereby a minimum of 1000 euro/ha is required. In view of the fact that the process is fully optimised and fits perfectly into the framework of sustainable agriculture. Changes in the values of agriculture, soil management and energy prices should occur before this concept can be become operational.

In sharp contrast to the above, the economy of converting organic waste to biogas is very favourable. This process is widely employed to process waste water (mainly from the food industry), excess sludge and vegetable, fruit and garden waste.

Various processes have been developed in Belgium and these are applied on a large scale all over the world. After methanisation, the remaining waste water must be processed, typically by means of an aerobic wastewater treatment.

The next table shows that for a raw material such as straw or bagasse (the residue of sugar cane), their total conversion to electricity becomes economical. At any rate, at an electricity price of about 100 /MWh (with green electricity certificate), the electricity revenue becomes two to three times the raw material price.

Table: Economic estimate of the conversion of biomass to electricity through biogas
(1 kg of sugar equivalent = 1 kWh electricity produced)

Raw material		Fermentation efficiency to biogas	Electricity	Theoretical revenue €/ton Dry Matter	
Type	World market price €/ton Dry Matter			Minimum @ 25 €/MWh	Maximum (with green energy certificate) @ 100 €/MWh
Sugar	180	100	1	25	100
Corn	80	80	0.8	20	80
Bagasse	20	60	0.6	15	60
Straw	20	50	0.5	12.5	50

Hydrogen fermentation is also possible, which is of interest as hydrogen is an energy carrier with favourable environmental characteristics. It can be obtained from organic materials by means of a combination of microbial fermentation processes. As with methane fermentation, the gas separates spontaneously from the liquid (distillation is not needed). In a normal fermentation with organotrophic bacteria, only 30 % of the energy content of the biomass is released as hydrogen. The remaining fraction is contained in fatty acids and can be released by using photosynthetic bacteria and light energy. These processes have been under devel-

opment for several years, but up to now do not lead to interesting perspectives. In any case, in such an integral conversion, 1 kg of carbohydrate provides a maximum of 1.2 m³ of hydrogen gas. At the current market price for hydrogen, this results in a value loss with respect to the starting product. It must be stressed though, that the production of hydrogen gas by means of fermentation is technically perfectly feasible, if the economic context changes. It must also be mentioned that the production of hydrogen from waste is not recommended because the processes relies on pure cultures and well defined raw materials.

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