CHAPTER 8

NATURAL ECOSYSTEMS: FIGHTING HUNGER, ENERGY AND RAW MATERIALS DEFICITS IN THE MODERN WORLD

Kichura D. B., Sobechko I. B.
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INTRODUCTION

Today, half a billion people on planet Earth do not have enough food. To produce a sufficient amount of vegetable protein, from which farm animals create high-quality animal protein, agriculture needs large amounts of nitrogen fertilizers, which are produced by industry. The modern history of microbiological protein production began during the First World War in Germany – the use of yeast. Due to food shortages, baker's yeast was grown on an industrial scale and "stuffed" mainly sausage products, some dairy products and instant soups with it. Yeast has a significant advantage, feeding on cheap sugar solutions, while converting sugar into high-value protein. During World War II, thousands of people were saved from starvation with the help of yeast "flakes", but the hard times passed, and the experience was forgotten. Only in the 60s did they start building plants for the production of protein using microbes again, since humanity needed more and more of it. Along with the use of oil alkanes and alcohol, wood is a source of nutrients. But at the same time, wood cellulose is not broken down by yeast and must first be destroyed using acid into building "blocks" – sugars. Agricultural waste such as straw, cotton residues, potato waste, vegetables and fruits, containing no more than 5 % protein, can also be converted by microbes into valuable feed.

The pharmaceutical business today ranks third in terms of investments and profits after the drug trade and arms. For example, if in 2010 the profits of pharmaceutical companies reached 300 million dollars, then when the pandemic took place in 2002, 2003, 2009 the profits reached 1…2 billion dollars, as we see an impressive difference of 1000 times. Let's ask ourselves, who will miss such an opportunity to make money? Profits during the 2019 pandemic reached more than 200 billion dollars and are growing to this day, as more and more new strains of viruses appear, which require new research, and therefore further development of vaccines, antibiotics and other auxiliary materials. Nevertheless, the main idea of developing biological energy sources, which, unlike oil and coal, are constantly renewed, has a great future. In many countries, agricultural waste unsuitable for other needs is rationally processed into alcohol. When processing sugar beets and sugar cane, they accumulate in the form of molasses, and in cheese production – in the form of sugar-containing whey. But alcohol is used not only to obtain energy, it is also used in industry as a solvent, as a raw material for the production of dyes, artificial fibers, adhesives and cosmetics.

1. Edible microbes

First of all, there is a shortage of products containing complete protein, such as: meat, fish, eggs, milk, legumes (beans, peas, soy). The problem is exacerbated by the fact that the Earth's population is increasing by approximately 70 million people annually; in order to cover at least the additional need for 2 million tons of protein caused by population growth, it would be necessary to grow protein-rich soy on an area of 40 million hectares. It is precisely in regions where there is a shortage of protein foods that the highest rates of population growth are noticeable with little development of agriculture and industry. To obtain 1 kg of animal protein, 5–10 kg of vegetable protein are needed, thus, already at this stage part of it is "lost", and this is in addition to the colossal losses of products that are caused by the "work" of agricultural pests, carelessness in harvesting, transportation and storage. Microorganisms could effectively help solve the food problem of mankind. They not only produce medicines, wine and cheese – they are also edible! They contain high-quality proteins, fats, sugars and vitamins.

As early as 1521, after the conquest of Mexico, the Spaniard Bernal Díaz del Castillo reported that the Aztecs ate strange little "pies" similar to cheese. Today it is known that these "pies" were made from unicellular algae that lived in Mexican lakes. Thousands of kilometers away from Mexico, the African natives living on the shores of Lake Chad, the Kanevu tribe, have also eaten unicellular blue-green algae of the genus Spirulina since time immemorial. This algae grows in significant quantities in Lake Chad, it is caught, dried and eaten like vegetables. In fact, algae are good protein producers, they double their mass in just 6 hours. For example, cereals need 2 weeks for this, chicks need 4 weeks, piglets need 6 weeks, and calves need 2 months. Therefore, in many countries, science is making a lot of efforts to create an "algae farm". This requires quite numerous reservoirs, namely pools with a large surface area of water. There, algae will be able to be sufficiently irradiated with sunlight, with the help of which they form sugar and then protein from carbon dioxide, water and mineral nutrients.

The need for light and air does not require any financial costs, and to encourage algae to grow abundantly (numerously), only cheap mineral supplements are needed. On the same area, Spirulina algae produces 10 times more protein mass than wheat, and with a higher protein content. When harvesting, the algae are simply "strained" through a mesh, later they are dried in the air and substances are added to them that improve the taste. After that, the product is ready for consumption and goes on sale. Why haven't similar large-scale farms been created in areas where the population is starving? Only because even such a simple technology is missing there, and in many regions, water supplies are very limited, which is quite expensive[1, 2, 3].

2. "Homemade liver" from microbes

Bacteria, yeast and other lower fungi grow even faster than algae. Bacteria double their mass in 20 minutes to 2 hours, and the bacterial mass can consist of 70 % protein. It has already been said that algae synthesize protein 100,000 times faster than a cow. At the same time, a cow gives us in the form of meat about 10 % of the nutrients that the cow itself consumes in the form of plant food, 0.9 % of the cow's feed for human nutrition is lost! In bacteria, yeast and fungi, almost the entire mass of nutrients is converted into proteins, sugars and fats suitable for use by humans and animals. Over time, it was discovered that microorganisms are able to consume not only sugar-containing nutrient solutions, but also assimilate oil components – alkanes (saturated hydrocarbons of the general formula $C_n H_{2n+2}$). Inedible for humans and animals, solid alkanes – paraffins – are only microbes able to utilize and convert into valuable protein. In 1963, the first experimental installations began to operate on pre-purified oil samples, growing yeast of the genus Candida, which consumed alkanes and at the same time multiplied very quickly, forming protein. About 1 ton of oil was obtained, containing 600 kg of protein. Moreover, much higher quality diesel fuel was obtained from the oil that remained and did not contain alkanes. At the very beginning of the production of yeast from alkanes, many doctors and veterinarians expressed concerns that the protein obtained in this way would be toxic to higher mammals. But many years of research have shown that yeast protein is harmless, thanks to many years of experiments this protein is one of the most studied food and feed products. The discussion between scientists and

[1] Біотехнологія : підручник / В. Г. Герасименко, М. О. Герасименко, М. І. Цвіліховський та ін. ; під ред. В. Г. Герасименка. Київ : ІНКОС, 2006. 647 с.

[2] Юлевич О. І. Біотехнологія : навчальний посібник / О. І. Юлевич, С. І. Ковтун, М. І. Гиль ; за ред. М. І. Гиль. Миколаїв : МДАУ, 2012. 476 с.

[3] W.-D. Fessner Biocatalysis: From Discovery to Application. 2000. 268 p.

manufacturers about the possibility of widespread use of microbiological protein (vitamin protein concentrate VPC) as a feed additive in livestock, poultry, and fish farming has taken on a truly dramatic color today. Scientists associate the difficult ecological state of some regions of countries and the population with the location of VPC production plants in these areas[4], [5].

Yeast protein exceeds all fodder plants in terms of its nutrient content. Experiments have shown that 1 ton of yeast can replace 7-8 tons of feed grains. The first large-scale enterprise for the production of yeast based on alkanes began operating in 1973, with a capacity of 70,000 tons per year. Subsequently, 8 giant factories for the production of "alkane" yeast operated. Such enterprises, where valuable protein was produced from oil, operated in Germany in the city of Schwedte (the final point of the Druzhba oil pipeline), as well as in Romania. A similar "protein factory" was also built in the People's Republic of China, since the oil fields of this country are characterized by a high content of alkanes. Even Arab oil-exporting countries are now showing great interest in this biotechnology, because the production of feed protein from their own oil, with its colossal reserves, is economically profitable, because then there is no need to cultivate land in the deserts that occupy the main territory of these countries. With proper biotechnological production, there is no need for expensive imports of grain, beans, soybeans or fish meal. In the mid-70s, as a result of an unexpected rather strong increase in oil prices, countries that do not have their own oil production were forced to look for another, cheaper source of nutrients for microbes that produce protein. Methyl alcohol, which was obtained in pure form from coal or oil, was proposed as such a substitute. In the UK, Bacterium Methylophilus methylotrophus "works" at one installation for the synthesis of microbial protein from methanol; The annual productivity of this bacterium is 50,000 tons of protein feed "prutin", which is used mainly in the cultivation of broiler chickens and fattening calves. A bioreactor with a volume of 150,000 liters of absolutely sterile nutrient solution in which the bacteria live at 35 °C, consuming exclusively methanol, ammonia and oxygen from the air. Bacteria are continuously released from the bioreactor; later they are treated with hot steam (the vital activity of the bacteria stops), and the obtained biomass in the form of rather large lumps is dried. As a result, a granular product is obtained, which has the color of burnt sugar; the brand name is prutin.

Since 1985, microbial protein has also been used in the food industry for the production of various dishes and semi-finished products. In the UK,

4 Roger L. Lundblad Biochemistry and Molecular Biology Compendium. 2007. 422 p.

5 Lilia Alberghina Protein Engineering For Industrial Biotechnology. 2003. 374 p.

specialized stores sell puff pastry with filling, similar in appearance and taste to beef. You can feel the "meat fibers" in this dish! The new bioproduct mycoprotein (from the Greek mykee – mushroom, protein – protein) is made from the Fusarium fungus. It contains 45 % protein and 13 % vegetable fat, that is, it is not inferior in nutritional value to many varieties of meat. The mushroom threads (mycelium) are so "interwoven" with each other that an external analogy with meat fibers appears. And as you know, fibrous food is extremely important for good digestion. Fusarium grows on all sugary substances: in Europe, for example, potato waste is used for this, in America – cassava roots (cassava (manioc) is a plant of the Euphorbia family, which is grown in areas with a tropical climate, flour is obtained from its roots), fruits or sugar cane[6]. Along with "beef", "chicken meat" is also made from Fusarium. Soon, stores will be able to buy at least 10 types of pies, minced meatballs, salads and delicacies made with the addition of microbial protein. In Finland, with the help of lower fungi that grow on toxic wastewater from a pulp and paper factory, 10,000 tons of valuable feed protein are produced annually. Without "microbial treatment", these wastewaters cause mass fish deaths in lakes and rivers. In this case, biosynthesis solves two problems at once – obtaining protein from non-expendable nutrient solutions and protecting the environment[7, 8].

3. Plants that fertilize themselves

Nitrogen, along with carbon, hydrogen and oxygen, are the main components, the so-called small "building blocks", that make up the compounds found in all living things, proteins and hereditary material. The air contains 78 % by volume of nitrogen, but neither humans nor animals can absorb gaseous nitrogen. The same applies to a significant part of plants, which are only able to absorb chemically bound nitrogen in the form of ammonium salts, nitrates or urea.

The ancient Romans already knew that growing legumes, such as clover, lupine, alfalfa, beans and peas, increases soil fertility. When legumes were first cultivated in a field, they were first spread on it with soil from fields where plants of this family were already growing. Of course, the Romans could not yet know that they owed their agricultural successes to nitrogen-fixing nodule bacteria that live in the root systems of certain types of legumes. In symbiosis with the plant, these microbes convert atmospheric nitrogen into

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[6] Ehud Gazit, Anna Mitraki Plenty of Room for Biology at the Bottom: An Introduction to Bionanotechnology. 2013. 216 p.

[7] Т. Пирог Біохімічні основи мікробного синтезу Київ : Ліра, 2019. 349 с.

[8] Трохимчук І. М., Плюта Н. В., Логвиненко В. П., Сачук Р. М. Біотехнологія з основами екології : навчальний посібник. Кондор, 2019. 539 с.

ammonia, one of the nutrients that the plant absorbs. Thus, the bacteria supply nitrogen fertilizers to the soil, and in return receive other nutrients from the plant. Ammonia is also produced in industry by the so-called Haber-Bosch process: atmospheric nitrogen at 550 °C and a pressure of several hundred atmospheres is combined with hydrogen to form ammonia, which is then applied to the soil as a "nitrogen fertilizer" (ammonium salts). The chemical method of producing ammonia requires enormous energy costs, and energy is becoming increasingly expensive, which is why fertilizer prices have increased significantly. As a result, nitrogen fertilizers become inaccessible to agriculture precisely in those countries where increasing yields is a priority task in order to feed a half-starved population[9].

To this should be added that, generally speaking, plants absorb only less than half of the amount of fertilizers that are introduced into the soil by humans. A significant part of the fertilizers is washed out of the soil with rainwater and later "oversaturates" lakes and rivers. As a result, microorganisms multiply excessively in water bodies, which completely consume the oxygen dissolved in the water, after which significant numbers of these microorganisms die. Along with the bacteria, fish, crayfish and all other living creatures that require oxygen die[10].

Bulbous plants form ammonia at normal temperature and pressure. For example, red clover produces 100–150 kg of nitrogen "fertilizers" per 1 ha of crop with the help of its own bacteria. In general, microbes remove approximately 100 million tons of ammonia from the air annually, compared to 40 million tons produced industrially at "hellish" temperatures and high pressure. Moreover, tuber bacteria once again demonstrate the advantage of biological processes that save energy. The formation of "nitrogen fertilizers" from the air by microbes has another advantage: they are not washed away by rain – therefore, they are completely preserved for plants and do not pollute water bodies. Thus, in 2000, American scientists invented a type of red worm that devours garbage with the formation of beneficial microflora: this creature is able to digest 1 kg of garbage per day and obtain 250 g of humus rich in useful and nutritious substances. This method of garbage processing has become interested in Japan, where entire islands have been constructed from garbage, on which people live and work. In many countries, experimental attempts are being made to colonize other non-legume crops with nodule bacteria. Genetic engineers are also trying to transfer genes from nitrogen-fixing bacteria into cereal cells.

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[9] Пирог Т. П. Загальна мікробіологія : підручник. Київ : НУХТ, 2004. 471 с.

[10] Яворська Г. В., Ґудзь С. П., Гнатуш С. О. Промислова мікробіологія. Львів : Вид. центр Львів. нац. ун-ту ім. І Франка, 2008. 256 с.

4. New plants from a test tube

Before cereals "learn" to extract nitrogen directly from the air, a lot of research needs to be done. At the same time, new high-yielding plant varieties are already being propagated in a test tube using biotechnological methods. First, scientists cultivate plant cells (in principle, by analogy with the cultivation of microbes) in nutrient solutions. Later, using enzymes, they carefully dissolve the cell walls. Such a "naked" single cell is able to divide and multiply, forming a cluster of cells (callus) in the nutrient solution. After adding certain specific nutrients and growth-promoting substances, full-fledged plants emerge from this cluster of cells after some time! In this way, thousands of plants can be grown in a test tube from 1 g of plant cells. Asparagus, pineapple, strawberries, alfalfa and ornamental plants have already been propagated in a similar way. Now, on 1 m^2 of laboratory space, 100,000 such plants can be grown in a short time, all from the cells of one "superplant". The resulting offspring are called clones (from the Greek clon – branch). All of them – like identical twins – have the same hereditary material. For example, through cloning, it was possible to obtain 1,000 young olive palms, which were planted in Southeast Malaysia. These palms are direct descendants of one palm tree, which turned out to be extremely resistant to diseases, and also produced 20–30 % more palm oil than ordinary palm trees. At the first stage of cloning, a leaf is cut from the plant that is proposed for propagation. In a solution containing enzymes that destroy cell walls, thousands of single "bare" cells (protoplasts) are formed that do not have walls. In a nutrient solution, the protoplasts form new cell walls, and the cell begins to divide. After about 2 weeks, each individual cell forms a cluster of cells (callus). The callus is placed on a special nutrient medium, where it develops to its full potential and begins to form a shoot. On the second nutrient medium, the shoot grows into a small plant with roots, which is later planted in the ground. Thus, from a single leaf, thousands of new plants with the properties of the mother plant can be grown in the shortest possible time.

If plant cells can be cultivated in nutrient solutions, like microorganisms, why not also try to use them using genetic engineering methods? Plants that are resistant to drought and plant protection agents (herbicides), that grow even on saline soils and are characterized by a large amount of protein – these are the goals set by genetic engineering specialists. Unfortunately, plasmids cannot be directly introduced into plant cells, as was the case with bacteria. But scientists have found a "snake" for genes here too: the widespread soil bacterium Agrobacterium tumefaciens. This bacterium infects plant cells and causes them to grow uncontrollably and form galls (disease-causing growths on plants). In the galls, the bacterial genes enter the plant cells with the help of bacterial plasmids. Later, the modified plant cells of the galls are cloned in a test tube, as described above, and whole plants are grown from them again. Thus, it is indeed possible to transfer such properties to cultivated plants as increased protein productivity, nitrogen fixation from the air, or resistance to drought and pests. Out of pure scientific curiosity, an attempt was made to incorporate firefly genes into tobacco plants. Tobacco plants subjected to such manipulations emitted a greenish-yellow glow in the dark – proof that the experiment was a success. Soon it will be possible to create plantations of trees that grow quickly specifically for the needs of the pulp and paper industry. The basis for them can be transgenic aspen, trees with introduced genes outgrew control ones in growth by 5–10 times.

Foreign genes are introduced into the plant using special agrobacteria or by "shelling" with a gene gun. In this case, a microscopic "ballistic projectile" is covered with a thin layer of gold or tungsten particles, on which genetic material is placed. After firing, the particles penetrate the plant cells, introducing genetic constructs with the necessary genes. The developed design of a gene gun using compressed air turned out to be very successful. With its help, it was possible to obtain more than 30 transgenic plants – wheat, potatoes, tomatoes, cucumbers, peas, soybeans, strawberries, rapeseed, aspen, which was already mentioned, and cedar.

All these plants have many unique features – increased yield, resistance to weather vagaries, intensive growth. But the specialists are in no hurry to transfer the resulting plants to production, although they work only with those genes that are primarily present in all plants. They want to make sure that they are completely safe and harmless to humans, and that the process of attenuation of the expression of the introduced genes will not begin in the "alien environment". To this end, the biochemical and physiological features of the transgenic plants created here are being studied in the newly created laboratories of the institute.

In 1999, the American The Monsanto company offered 4 regions of Ukraine to grow GM potatoes (Volyn, Vinnytsia, Rivne, Khmelnytskyi). Two potato crops that they harvested were buried in Cherkasy region, but it must be borne in mind that firstly, not the entire crop was buried, and secondly, pests remained that had developed immunity to herbicides and became much more dangerous and resistant to the effects of pesticides. They wanted Ukraine to become a springboard for growing GM plants. Although, if you believe today's official data, 40 % of GM corn, wheat, potatoes are already grown in our country, 80 % of rapeseed and soybeans in Ukraine are also GM. Despite the fact that Ukraine has a law prohibiting the use of GM plants in food production and even to meet the needs of the chemical, fuel or energy industries.

In 2010, 150 million hectares – about 10 % of the area on which agricultural crops are grown – were used for the production of GM plants. GM fruits and vegetables have no taste or smell, and during their storage, which lasts up to 3–6 months, they do not lose their consumer properties at all. The disadvantage of their storage is that the longer they are stored, the more proteins accumulate in them, which negatively affect the human body. In 2020, more than 210 million hectares in more than 30 countries around the world are occupied by GM plants. In 2008, the American subsidiary of Monsanto Engine encroached on the most sacred, medicine, proposing to introduce a gene into a banana that has rubella antibodies in order to kill two birds with one stone. On the one hand, to relieve children of the fear of vaccination, and on the other, to support its production[11].

5. Microbes against pests

In some African countries, insects or rodents destroy about 60 % of the harvest. In Europe, crop losses are from 25 to 40 %. In addition, in tropical countries, insects are carriers of dangerous viral diseases, such as malaria (Anopheles mosquito) or sleeping sickness (Tsetse fly). Malaria is the first disease to affect 300 million people every year. In terms of its scale, malaria ranks first in the world among other diseases on Earth.

Various chemicals called insecticides are used to combat insects; annual costs for these products are approximately $ 2.5 billion. But not only pests die, but also all other insects that come into contact with insecticides. Therefore, insecticides disrupt the common habitat of animals and plants, and animals that consume these insects, especially birds, are gradually poisoned. Insecticides eventually get into water and food. In addition, insecticides are no longer effective against approximately 400 insect species. Because these species have become resistant to the drugs used. In order to combat such resistant species, it is necessary to either increase the dosage of poison or use new insecticides. Therefore, many countries are searching for environmentally friendly biological methods of pest control.

Today, in the USA and other countries on different continents, millions of tiny (no more than 1 mm long) Trichogram riders are already grown by the "flow method" and transferred to fields affected by pests. Each Trichogram female pierces up to 300 eggs of other insects and lays her eggs in them.

[11] Мельничук М. Д., Новак Т. М., Кунах А. В. Біотехнологія рослин. Київ : Поліграф Колсантинг, 2003. 520 с.

Trichogram caterpillars, which are formed from the eggs of parasites, eat the contents – the eggs of the host and thus protect the plant.

Bacteria, fungi and viruses are also used to combat pests. Thus, Bacillus thuringiensis, which became known in Thuringia as a moth larva exterminator, has proven itself well as a caterpillar "killer". Fields are sprayed with aqueous suspensions of these microbes, and the caterpillars eat them along with their food. The microbes form poisonous protein crystals that destroy the caterpillar's intestines, causing their death. Bacillus thuringiensis is currently grown in bioreactors. One ton of the microbial preparation is enough to rid 300 hectares of forest, beet fields, cotton crops or fruit tree plantations of pests! It was also possible to transfer the gene that controls the formation of poisonous protein crystals from Bacillus thuringiensis to bacteria that inhabit the roots (Pseudomonas florescens). Now, the caterpillars of the winter scoop, which damage cereals by gnawing at the roots, die if they only ingest Pseudomonas bacteria transformed by genetic engineering methods with their food.

The combined use of Trichogramma riders and bacilli against cabbage moths has been tested. In this case, the "riders" reduce the number of butterfly eggs, while the bacilli kill only the caterpillars that have hatched. The new bacterial strains are specifically effective against the Colorado potato beetle. Insects that consume the leaves of forest trees, such as the oak borer or various types of silkworms, can be eliminated in the same way as houseflies, goldfinches or Anopheles mosquitoes, without harming other insects, including bees. In Eastern European countries, viruses are successfully used against the odd-toed silkworm and the nun, and in the USA against the spiny worm. Microscopic fungi are used to combat the Colorado potato beetle, apple fruit borer and insects. The so-called biological protection of cultivated plants is specifically aimed at harmful insects that are characterized by mass reproduction. In this case, the balanced coexistence of all living things is not disturbed and, in addition, no toxic substances enter our natural environment.

6. "Frost-resistant" bacteria

In addition to pests, unexpected night frosts in spring or early autumn also cause significant crop losses. But cold damages plants only when ice crystals form on them and inside their various parts (roots, stems, leaves).

It was discovered that billions of bacteria of the Pseudomonas syringae species live on plants and secrete proteins. Ice crystals form around the "ice-forming" bacteria and their proteins at sub-zero temperatures. Experiments have shown that if you get rid of these bacteria, then ice crystals do not form on plants almost down to -8 °C. Conversely, in the presence of "ice-forming" bacteria, ice crystals appear on plants at temperatures close to zero, which damage plant tissues. These "harmful" bacteria have found a useful application: they are added to water in "snow cannons": even with a slight cooling to create a snow cover in order to provide conditions for winter sports.

But another idea is more interesting: an attempt was made to transform bacteria "ice-forming" by genetic engineering methods, namely, to cut out from bacterial DNA the gene that controls the formation of protein from "ice". After this "operation" the bacteria can no longer produce protein from "ice". And indeed, ice crystals no longer formed around these modified bacteria! For the test, these bacteria were used to spray land plantings. It turned out that "frost-resistant" bacteria suppressed the development of bacteria – "ice-forming" and warned the plant against sudden drops in temperature. In this case, the strawberry bushes were only experimental plants, since the main task is to protect orange, grapefruit and lemon trees from frost. It is possible that in the future these plants, which are sensitive to sub-zero temperatures, can be grown in areas much further north [12].

These possibilities are constantly being tested. The fact is that some researchers fear that new microbes can spread throughout the world and disrupt the natural balance of various species. In this regard, biotechnology bears a special responsibility, because it is necessary to guarantee that new microbes will be completely safe and will not harm nature and humans.

7. Clean water thanks to the work of microbes

In 1892, the residents of Hamburg thought that they could take drinking water directly from the Elbe and Alster rivers. But such ideas had a tragic end – the residents of Hamburg were "mowed down" by cholera. The water has long ceased to be clean. It is a habitat for microbes, including the most dangerous pathogens of cholera. In large cities, wastewater treatment has become mandatory. Today, each resident of the city "produces" 150–200 liters of wastewater annually. When producing 1 ton of paper, the volume of wastewater is approximately the same as the volume of domestic wastewater of 1,000 people, and often the wastewater of one chemical plant corresponds in volume to the wastewater of a city with a million inhabitants.

The natural ability of rivers to self-clean, due to the activity of microorganisms, is now no longer enough, so wastewater must be decomposed with the help of microorganisms in giant treatment plants so that the wastewater can be discharged into rivers and lakes without harming the natural environment. When treating wastewater, microorganisms do

[12] Річард Доккінз Наука для душі нотатки раціоналіста / пер. з англ. Д. Прокопик. Київ : Наш формат, 2019. 384 с.

a particularly hard job. Consuming oxygen in the process of breathing, they use it to decompose substances (carbohydrates, proteins, fats) in wastewater to carbon dioxide and water, and on this basis grow and build their new cells. Treatment plants provide them with the best conditions for development, reproduction and destructive activity. These are giant "biofactories", and their "bioproduct" is clean water. When treating wastewater, macropollution is first removed from it: paper, pieces of wood, remnants of matter. Heavy particles of sand are deposited in sand traps. Only after this, the lighter suspended particles are concentrated at the bottom of the sedimentation tank in the sludge trap. This wastewater enters the aeration tank – an aerated tank, where ideal conditions for the life of microbes are maintained. In the aeration tank, bacteria, yeast and fungi form large flakes ("activated sludge") with wastewater substances, which are not decomposed due to the mucus (through mucus) secreted by the bacteria. The main problem of this bioprocess is related to the supply of oxygen to microbes. Microbes spend more than 1 g of oxygen to break down 1 g of sugar, and the solubility of oxygen in water at normal temperature is only 9 mg/l. Therefore, very soon after the start of the microbiological process, all the oxygen contained will be completely consumed by microorganisms. Thus, for the success of the business, wastewater in the aeration tank must be constantly mixed and enriched with oxygen. By the way, a person produces so much wastewater per day that microorganisms consume 54 g of oxygen for its decomposition. To ensure aeration of wastewater, the aeration tank is equipped with rotating brushes and a pipe through which air is pumped into the water. The air constantly "swirls" the activated sludge flakes, due to which they do not reach too large sizes, remain suspended and are well supplied with oxygen. Microbes of the flakes absorb wastewater substances, decompose them during respiration and multiply at the same time. Some of the microbial flakes are later deposited in secondary settling tanks in the form of sludge. A smaller part is returned to the aeration tank in order to have a sufficient number of microbes to oxidize the wastewater that is coming in again. All the sediment collected from the first and second settling tanks is decomposed in septic tanks or methane tanks (structures for fermenting sewage sludge with the help of microorganisms) by methanobacteria until the formation of "biogas" (methane). This gas can be used very efficiently by burning to obtain heat. The residual "fermented" sludge is dried and often used as fertilizer.

The tanks of treatment plants occupy large areas. This condition, however, is difficult to fulfill in industrial areas. Therefore, in recent years, biotechnologists have developed "small-sized" tower bioreactors with a height of about 15–20 m for wastewater treatment. Wastewater from industrial enterprises often contains substances and poisons that are difficult to decompose, such as cyanide and mercury compounds. These substances are not decomposed by "normal" microbes, they even kill many microorganisms. Therefore, a search is underway for new highly productive strains of microorganisms that could utilize such toxic impurities. For example, Indian biotechnologist Ananda Chakrabarti has isolated "superbacteria" capable of breaking down highly toxic agents 2,4,5 – T (2,4,5 – trichlorophenolacetic acid), which the USA used during the Vietnam War to cause "leaf fall" in large areas of the jungle. Since the chemical compounds of this group of pesticides cause leaf aging – artificial leaf fall, they are known as defoliants. They are used in agriculture in the period before harvesting for defoliation of plants (removal of leaves), mainly in cotton. Professor Chakrabarti has also identified true "eaters" of oil. These bacteria "greedily" eat toxic oil residues and are used to quickly decompose oil in the event of disasters with oil tankers, when large areas of water are under threat of pollution by oil products[13, 14].

Over time, microorganisms that have multiplied in considerable quantities are eaten by other marine animals and thus quickly disappear. But disasters that occur with tankers are only a small percentage of oil pollution. Every year, about 6 million tons of oil enter the sea, a quarter of this amount is a result of cleaning empty tankers in the open sea, a third is with sewage brought by rivers. By the way, Professor Chakrabarti's "oil eaters" are the first in history "newly formed" living beings for which a patent has been issued. At the Institute of Biotechnology in Leipzig, strains of bacteria have been isolated that absorb and deposit mercury in their cells. If these "mercury collectors" are deposited on a filter, then mercury can be recovered from wastewater. Algae also have the ability to accumulate mercury, lead and silver in their cells and thereby purify wastewater. And yet, the shortest and easiest way to protect our planet's water basin is to develop production processes that do not emit any harmful products into the environment! This is exactly the goal that biotechnologists want to achieve.

8. Biosensors: Microbes signal pollution

How, in fact, do we know how much wastewater is polluted? The content of toxic and decomposing substances in wastewater can be signaled by microbes themselves. First, the oxygen concentration is determined in each sample, then microbes that live in wastewater are added to the samples. The

[13] Daniela Cardinale, Thierry Michon Enzyme Nanocarriers. 2015. 266 p.

[14] Ehud Gazit, Anna Mitraki Plenty of Room for Biology at the Bottom: An Introduction to Bionanotechnology. 2013. 216 p.

sample bottles are tightly closed and after 5 days, the oxygen content in each bottle is determined again. Samples with a high degree of pollution contain many nutrients, so microorganisms will consume more oxygen here than in samples with "clean" wastewater. But 5 days is a rather long period. During this time, heavily polluted wastewater manages to enter rivers in significant quantities. Therefore, an earlier "preliminary signal" is needed!

Over the past 10 years, "biosensors" have been developed – biological measuring probes that show the degree of pollution of wastewater in a few minutes. A biosensor is an electrode connected to an electronic display, which displays data on the oxygen content in a given liquid. A thin layer of microbes that live in wastewater and are retained on the electrode by a dense filter are applied to the surface of this electrode. By immersing such a biosensor in a liquid, it is possible to directly measure the "breathing" of microbes. In the case when the biosensor is in clean water, the microbes almost do not breathe, since too few nutrients are at their disposal and the biosensor gives a very weak (weak) signal. On the contrary, if the biosensor is immersed in a sample of wastewater containing many nutrients (i.e. heavily polluted), then the microbes receive a lot of "food", they breathe more intensively, consume more oxygen, and the biosensors respond with a signal of great strength. With this method, wastewater control can be carried out in one minute. With the help of such biosensors, it is also possible to determine the concentration of nutrients in the bioreactor. There are biosensors that work on the basis of enzymes secreted from microbes. For example, a device with a biosensor that carries the glucose oxidase enzyme on the surface of its electrode can determine with great accuracy in 1 hour more than 100 blood or urine samples whether they contain an increased amount of sugar compared to the norm, that is, diagnose diabetes. Biosensors are particularly suitable for serious tests. With their help, it is possible to check the health of 1000 people in a short time[15].

9. Renewable energy sources

In Brazil, hundreds of thousands of cars run on "new fuel". This fuel is ethanol! The impetus for the introduction of alcohol as a car fuel was the energy crisis that grew in the 70s, when oil prices on the world market suddenly rose sharply. Brazil had little oil, so its import quickly became a burden for the country. In search of a way out of this situation, plans were made to produce alcohol from vegetable raw materials.

[15] Дзядевич С. В., Солдаткін О. П. Наукові та технологічні засади створення електрохімічних біосенсорів / під ред. Г. В. Єльської Київ, 2005. 250 с.

Due to the considerable length and tropical climate of Brazil, growing sugar cane on large areas did not pose any technical difficulties. In this most powerful biotechnological project in the world, cane sugar is fermented with the help of yeast, forming ethyl alcohol. In 1995, 10 billion liters of alcohol were obtained! Thus, Brazil became the largest producer of alcohol in the world.

In addition to sugarcane, which grows well only on fertile lands, it is also proposed to introduce into production a starch-containing raw material – the cassava plant (manioc). Unlike sugarcane, potato-like cassava plants grow on depleted soils. But in them, it is first necessary to break down starch using amylase into sugars, which are then fermented by yeast into alcohol. Until now, it was believed that the Brazilian version of the biotechnological solution to the fuel problem was the best. But it turned out that in Brazil, significant areas of arable land are sown with "car feed". To produce enough alcohol to meet the annual fuel needs of cars with a mileage of 12,000 km, it is necessary to collect plants from 13,000 m^2 of arable land. Meanwhile, for the annual consumption of one person, only 800 m^2 is needed. In other words, one car takes food from about 18 inhabitants! But in Brazil, millions of people suffer from malnutrition! To this should be added environmental problems: for economic reasons, industrial effluents from distilleries are discharged into rivers untreated, turning them into dead waters, and tropical forests are cut down in vast areas – that is, the pursuit of alcohol only leads to violations of an already rather fragile and unstable natural balance. Wood waste can also be broken down using enzymes into sugars. In some countries rich in forests, so-called "energy plantations" are even being established. These are fast-growing tree species, the wood of which is regularly "harvested" every 3–5 years and processed into sugar, which is used as a source of nutrition for microorganisms. True, for many developing countries, wood has become as scarce as food in other countries. In some arid regions of Africa, the annual need for firewood per capita for cooking alone is only 0.5 m^3. The last forests are "burning", the desert is advancing further and further. But what will people use to heat their fireplaces tomorrow? Biotechnology offers a solution here too: the production of biogas.

10. Bioethanol Project in Ukraine

Ukraine cannot fully provide itself with energy resources and is forced to import a significant part of them, which is a significant import item and hinders the development of our economy. Many countries are in a similar situation, but economically developed countries have something to offer oil-exporting countries, and first of all, these are industrial products. States that buy “black gold” abroad, despite the temporary drop in prices on the world market for oil, are forced to develop systematic measures to save energy consumption and look for alternative energy sources. It is also known that, based on the assessment of global oil reserves, the era of its depletion is approaching. Naturally, this will significantly exacerbate the energy problems of most countries in the world.

For Ukraine, the search for new alternative energy sources is of paramount importance today. In this regard, the role of biotechnology in providing humanity with alternative sources is increasing. After all, it is biotechnology that helps to utilize waste or plant biomass, as well as solid municipal waste (garbage) for energy recovery. Various forms of energy can be used to generate electricity, as well as as fuel for transport. There are several types of transport fuels that can be obtained from biomass – biogas (mainly methane), biodiesel and bioethanol. It is believed that fuel ethanol has the greatest potential, given the inexhaustible sources of its production. These can be herbaceous plants and wood, waste from agriculture and the wood processing industry. As well as household waste. Ethanol is the oldest product of biotechnology, which was born no less than 4000 years BC in Ancient Egypt and Babylon. In this technology, sugars (glucose and fructose and some others) are fermented anaerobically by yeast. Until recently, all ethanol obtained in this way was used to produce alcoholic beverages. Only small amounts of it, obtained chemically, have been used in industry, but over the past 25 years the situation has changed radically. Today, more than half of the world’s ethanol production is used as an additive to fuel for internal combustion engines (gasoline), and only 15 % – for the production of alcoholic beverages. For example, in 2000, world production of ethanol was 35 billion liters, food ethanol (alcohol needed for the production of alcoholic beverages) was produced 4 billion liters, ethanol used in the chemical industry – 10 billion liters, and fuel ethanol – about 20 billion liters. Only 7 % of ethanol is obtained by chemical synthesis, and 93 % – using yeast fermentation. About 60 % of the latter is produced from sugar, the rest – from grain. Interestingly, the production volumes of edible alcohol have remained unchanged since 1975, while the production of fuel ethanol has been increasing year by year (from 2 billion liters in 1975, to almost 20 billion liters in 2000, 30 billion liters in 2010, 35 billion liters in 2020).

Today, all fuel ethanol is produced biotechnologically – by fermentation, there are government programs to support the production of fuel ethanol. In 2000, Brazil produced 6.5 billion liters of fuel ethanol, which provided 13 % of its total energy needs and 19 % of its liquid fuel needs. This saved $36 billion that would otherwise have been spent on the purchase of petroleum products. Previously, Brazil produced mainly anhydrous ethanol, which was used as fuel for cars with special engines. But recently, Brazil has been using additives as fuel in which the ethanol content is 26 % in gasoline and 3 % in diesel fuel. Such mixtures do not require changes in the design of internal combustion engines and diesel engines. The United States is also a significant industrial producer of fuel ethanol. Thanks to its production, the USA saves $1.5 billion annually on the import of petroleum products. In large cities with a population of more than 1 million people, in the winter they use only gasoline containing 10 % ethanol, the so-called UE gasoline, or gasohol. The sales volume of which in the USA is 12 % of the total gasoline sales. EU countries produce 2 billion liters of ethanol annually, but today they use less than 10 % as fuel. But the EU has adopted a bill that provides for the addition of 5 % ethanol to all types of gasoline by 2010, and up to 10 % by 2020. In addition to saving currency, the use of fuel ethanol in the form of gasoline-ethanol mixtures allows you to significantly reduce the content of harmful components in exhaust gases (carbon monoxide, oxides and oxides of nitrogen, and other volatile toxic emissions). Back in 1994, the USA adopted a special law, according to which gasoline must contain at least 2 % (by weight of oxygen) of oxygen-containing additives (mainly ethanol) to reduce exhaust toxicity. The positive effect of using bioethanol as fuel is extremely important, even global, because carbon dioxide, which is released during combustion, has a primary atmospheric origin. That is, it can be assimilated again by plants, which in the future will become a source of fuel ethanol. And when using minerals as fuel, CO_2 is released, which is an additional source of the greenhouse effect.

Fuel ethanol obtained from sugar cane fully pays for itself. Its production in Brazil has been growing by 4 % annually since 1990, and the cost price has decreased by 3 %. The reason for this is the scientific and technological progress of the industry, new varieties have appeared, sugarcane cultivation technologies have improved, new technologies for sugar extraction, fermentation and distillation have been developed. Ethanol production from corn, practiced in the USA, despite the reduction in the cost of fuel ethanol by 2/3 over the past 15 years (since 1990), is less profitable. But even today the USA provides serious tax discounts on fuel ethanol to make it cheaper than gasoline. According to the law that came into force in 2010, the discount is $0.014 per liter of gas. Such a tax policy is to some extent due to the constant support of low prices for petroleum products. Meanwhile, the real cost of petroleum products is much higher than the price at which they are sold (for example, in the USA, 4 times higher, taking into account the indirect costs of the US state for monitoring and cleaning the environment from harmful emissions during the production and use of petroleum products, as well as for maintaining security in the main oil production areas in the Middle East).

Even in the USA, where corn is very cheap, maintaining the profitability of fuel ethanol production requires government subsidies, while for other countries, creating a profitable production process (so that ethanol competes with imported petroleum products in price) is possible with a further significant reduction in fuel ethanol prices. The main ways to reduce the cost of this product are alcoholic fermentation technologies that can replace raw materials for production or radically change the technology. The replacement of raw materials consists in using biomass of whole plants such as grass, wood, including agricultural waste, wood processing, and household waste instead of grain. Basically, such dry biomass consists of cellulose (glucose polymer), hemicellulose (glucose and xylene polymer), and lignin (aromatic alcohol polymer), abbreviated as lignocellulose. The use of such non-traditional materials makes the raw material base for producing fuel ethanol practically inexhaustible. Calculations show: using only agricultural waste, wood processing industry and municipal waste for fuel ethanol production will allow the USA to replace 40 % of gasoline with ethanol. Specially cultivating certain woody (alder, aspen, etc.) and herbaceous (in particular sorghum) plants for further conversion into ethanol could provide the remaining 60 %.

The problem of alcoholic fermentation of lignocellulose has been studied for at least 20 years. Various international conferences are attended. Special scientific periodicals are published “Biomass and Bioenergy”, “Renewable Energy”, “Bioresource Technology”, etc. But the problem turned out to be quite complicated. Lignocellulose consists of three biopolymers – cellulose, hemicellulose and lignin, the first two are hydrolyzed to the corresponding monomers, sugars, among which the main ones are glucose (up to 40 %) and xylose (up to 30 %). Methods of such hydrolysis: chemical or enzymatic are still far from optimal. Well-known baker’s yeast, which effectively ferment glucose to alcohol, but would be able to ferment xylose to ethanol, were not described at all until recently. Since there is no free xylene in nature. Nevertheless, such yeasts have been discovered, although they accumulate small amounts of ethanol.

Today, the most effective way to obtain ethanol from plant raw materials is simultaneous enzymatic hydrolysis with subsequent fermentation of free sugars. Enzymes – fungal cellulose and hemicelluloses, which effectively hydrolyze crushed lignocellulose, but function at a relatively high temperature of 45 °C, while known yeasts ferment sugar only at 30 °C. The cost-effectiveness and profitability of such production largely depends on the sale of ethanol. If ethanol is sold at $0.33 per liter, a fuel ethanol plant will not be profitable without producing a second product, furfural. Even if the cost of transporting waste is \$0.19 per ton of dry matter per 1 km (taking into account the idling in the opposite direction) and only 10 % of the waste is processed. The optimal plant size should process 4360 tons of waste per day. Significantly larger or smaller plants are unprofitable. The capital investment for building such a plant is \$455 million. The annual profit would be \$281 million, and the costs would be \$173 million, so the annual profit (before taxes) would be \$108 million.

If we conduct an economic calculation of ethanol production using herbal plants, the price of ethanol would be \$0.62 per liter. At this price, ethanol production is profitable without the simultaneous production of other products. However, only practice can give a decisive answer regarding the profitability of ethanol plants. Since 2000, the first industrial plant for the production of fuel ethanol from bagasse – waste from sugarcane processing, mainly – the remains of plant stems and leaves, was launched in Brazil, with a capacity of 75 million liters of ethanol per year, the price is as close as possible to gasoline. The operation of such a plant was the first technological and economic test of the profitability of fuel ethanol production from lignocellulose in the conditions of using today's technologies. A positive result will allow the processing of the entire (whole) sugarcane plant into alcohol, and not just extractive sugar, without increasing the cost of transporting raw materials.

Ukraine has wide possibilities for industrial production of such ethanol. Since the state produces many agricultural products, annually accumulating considerable amounts of agricultural waste – straw, corn cobs, sunflower husks. In addition, significant amounts of solid household waste are collected, mainly consisting of lignocellulose. Ukraine is a large producer of edible alcohol, at the same time many alcohol plants are not operating at full capacity or are idle at all. In the future, the capacity of these plants can be used for biofuel production, but such production must be profitable. The cost of biofuel will depend on the tax policy of the state, its cost, which in turn is dictated by the production technology. Despite the fact that the creation of the latest technologies for producing biofuels is very relevant, until recently, scientific developments in this direction were not carried out at all in Ukraine. But while there are unresolved scientific problems of a more general nature, the solution of which would make the production of fuel ethanol more efficient.

One of these problems is the need to reduce the cost of the process of distilling ethanol from the fermentation liquid. The classical process is quite energy-intensive (63 % of all energy costs are distillation costs). This is due to the relatively high boiling point of ethanol (78 °C). Several years ago, American researchers proposed stopping the process of alcoholic fermentation at the stage of acetaldehyde (boiling point 21 °C), which can be converted into ethyl alcohol in the process of chemical catalysis at room temperature. But so far, effective methods of stopping alcoholic fermentation at the stage of acetaldehyde formation have not been technologically developed. Scientists at the Lviv Institute of Cell Biology of the National Academy of Sciences of Ukraine first proposed using mutant strains of methylotrophic yeast, specially obtained at the institute, for fermentation, along with general cultures of alcoholic yeast. This indeed leads to the accumulation of acetaldehyde, a very volatile compound, as the final product, not ethanol, but acetaldehyde, a highly volatile compound that evaporates spontaneously at room temperature without any distillation.

All known industrial processes of alcoholic fermentation are periodic, that is, they require the fermentation process to be resumed after the accumulation of 10 % ethanol, which makes the latter highly toxic to yeast. Attempts to conduct continuous fermentation, in which the accumulated ethanol would be constantly removed and the sugar substrate for fermentation would be constantly added, were unsuccessful. The idea mentioned above – to use compatible yeast cultures for alcoholic fermentation with the formation of acetaldehyde as the final product – can also solve the problem of continuous alcoholic fermentation. If the fermentation is carried out at a temperature higher than the boiling point of acetaldehyde, the latter will not accumulate, but will evaporate spontaneously, and the process of its formation from sugars can be continuous.

As already mentioned, technologies for alcoholic fermentation of lignocellulosic raw materials are being developed all over the world, which would significantly reduce the price of fuel ethanol. But alcoholic yeast cannot ferment up to 40 % of the sugars of such hydrolysates. Other yeast strains have been found that can ferment all the sugars of hydrolysates, although the efficiency of such a process is very low. In Ukraine, unfortunately, no one has been engaged in alcoholic fermentation of hydrolysates and other waste until recently. But in recent years, Ukrainian and Belarusian scientists have achieved serious success in solving these problems. Domestic scientists today have unique yeast strains, as well as genetic structures (plasmids) that contribute to the genetic engineering improvement of such yeast. Ukrainian biologists were the first to discover thermotolerant strains of methylotrophic yeasts that can ferment both glucose and pentoses (primarily xylene) to alcohol. Such strains were of interest for the process of simultaneous enzymatic hydrolysis of lignocellulose with subsequent fermentation of the sugars formed to ethyl alcohol or acetaldehyde, as mentioned above. Therefore, the new approaches to fermentation of the starting material developed by scientists will make this process more efficient and cost-effective.

11. Biogas will save tropical forests

“Wandering lights”, known from fairy tales and sagas, were lit by witches and spirits in medieval German mythology to lure and destroy good travelers. The folk epic is based on real facts: in swamps, when there is a lack of oxygen, parts of dead plants, with the participation of microorganisms, form swamp gas – methane, which is capable of self-ignition, and it is then that a “wandering light” appears. If you “shake” the sludge in any dredge, you can smell biogas.

Methanobacteria, which produce biogas, are very sensitive to oxygen. It is assumed that these bacteria lived in the Earth’s primary atmosphere. Then there was no oxygen in the atmosphere, but there was carbon dioxide, hydrogen, and the atmosphere had the temperature just necessary for the development of methanobacteria. If today they were given the same conditions in a swamp or an artificially created bioreactor, they would also produce methane. However, they would depend on the preliminary preparatory work of other bacteria, which, using their enzymes, break down wood, starch, proteins and fats into the building blocks of which they are composed and ferment the latter to acetic or butyric acids, hydrogen or carbon dioxide. In nature, approximately 800 million tons of methane are produced annually as a result of the activity of bacteria. This is approximately the same amount as the amount of natural gas extracted by mankind. In the People’s Republic of China, there are currently 11 million small, technically very simple installations that supply the extracted “biogas”. These “biogas” installations are presented in the form of small, hermetically sealed tanks in which animal and human excrement and plant waste are shaken. When the tanks are heated, bacteria produce methane, which is excreted and used for household needs. Biogas is a valuable fuel, 1 m^3 of it gives as much energy as we get from burning 1 liter of fuel oil. It turned out that instead of the senseless burning of dry manure, plant residues and forest wood, energy can be obtained in the form of biogas. Such biogas burns with the formation of carbon dioxide and water, and in the “bioreactors” remains a natural fertilizer – sapropel. Sapropel – contains extremely important nutrients for plants – nitrogen, phosphorus and potassium salts. Due to which it is possible to reduce the import of fertilizers. In India, 50 million tons of cow dung and 30 million tons of other waste are still burned, because many villagers are too poor to buy a “biogas” installation themselves, and, in addition, to obtain sufficient quantities of biogas, it is necessary to keep several cows. Here in the People's Republic of China, large "biogas" plants are often operated collectively by villagers from the entire village. These plants are also used for health protection: after all, hermetically sealed tanks with pathogens of various diseases are killed by high fermentation temperatures.

So, "biogas" plants are beneficial for residents of small settlements, districts and villages, especially in hot countries. They not only do not take away food and the best arable land from people, as is the case with the production of alcohol from plants, but even increase income for agriculture. Their widespread use can save tropical forests, stop the spread of deserts and make remote areas fertile with the help of fertilizers.

In Europe, such reactors would solve the problem of waste in large livestock complexes. The fact is that liquid animal feces (manure) can only be partially used as fertilizer, otherwise they will significantly overload the soil. After all, up to 45 liters of manure accumulates from a dairy cow or 10 pigs per day! But from this amount you can get 2 m^3 of biogas! For example, a "biogas" plant near Leipzig produces methane from the feces of more than 1,000 cattle. The energy obtained is enough to heat the cowsheds all year round[16].

12. Silent mining

Recently, copper mining has been carried out so intensively that copper deposits with a high copper content have become a rarity. Mining is moving to deeper and deeper areas. Energy costs for mining are increasing dramatically. As early as the 18th century, the Spaniards extracted copper in their mines near Riotingo largely from copper-containing mine water. The water had already leached the copper salt from the rock. But only 50 years ago, no one knew that bacteria were involved in this process: they help convert the poorly water-soluble "copper" into a water-soluble salt form.

Nowadays, microbes are purposefully used to leach copper, and they already account for 20 % of all global copper production, primarily from rocks with a low copper content. It is the bacteria of the genus Thiobacillus (from the Greek theion – sulfur) that help extract metal from the rock. They feed not on sugar, but on sulfur and iron! They are able to multiply even in the presence of strong acids, moreover, they themselves are able to produce sulfuric acid. During biological leaching of ore, millions of tons of waste (overburden) rocks containing sulfur and copper are first transported to collection points.

[16] Яблонський В. А. Біотехнологія відтворення тварин : підруч. / В. А. Яблонський. Київ : Арістей, 2005. 296 с.

Overburden rocks (overburden) are rocks that cover and contain the body of a mineral in the massif and are subject to extraction during open-pit mining of the deposit. These rocks contain small amounts of valuable copper. along with iron. There are known dumps with a height of more than 400 m, which contain 4 billion rocks that would have previously remained unused. They are irrigated and soaked with thousands of cubic meters of water. It is not necessary to pre-contaminate the heaps with Thiobacillus bacteria, because they are present everywhere. In 1 g of sulfur-containing rock, more than 1 million cells of Thiobacillus bacteria live. As water penetrates, the bacteria multiply. These tiny helpers first convert poorly soluble iron salts into a readily soluble salt – iron sulfate – with the concomitant formation of sulfuric acid. Iron sulfate, with the help of sulfuric acid, converts water-insoluble copper sulfide contained in the rock into copper sulfate. After that, a blue solution of copper sulfate begins to seep out at the foot of the heap, which is collected in large collections. Now, using some technical method, it is easy to obtain pure copper. The residual liquid formed during the leaching of ore, which does not contain copper, but contains sulfuric acid and bacteria, is sprayed over the heap again.

It is quite possible that in the future, using "bio-development" of deposits, it will also be possible to obtain uranium, lead, nickel and cobalt. Apparently, coal with a high sulfur content can also be "desulfurized" by the interaction of sulfur bacteria and thus make its use safer for the environment. Even more promising than leaching of rock heaps is ore leaching directly underground. For this, the rock does not need to be removed to the surface at all. The adit is simply sprayed or flooded with water. "Bio-development" of deposits is also beneficial in old, already abandoned mines. Bacteria can probably help make mining safer. Serious disasters constantly occur in coal mines due to a terrible gas – methane, which accumulates at the bottom of the excavations, which ignites as a result of an accidental spark. Thus, bacteria were "sprayed" on some mines and pits, which in 2-4 weeks almost completely converted methane into non-flammable carbon dioxide[17, 18].

Today, microbes can also be useful in oil extraction. To date, only a third of the total reserves of oil fields have been extracted, since oil very rarely forms large underground "lakes" and is often firmly bound in the pores of rock. Therefore, today, along with real wells, water is pumped into oil formations in order to squeeze out the remaining oil. But water is too

[17] Козлова І. П., Радченко О. С., Степура Л. Г. та ін. Геохімічна діяльність мікроорганізмів та її прикладні аспекти. Київ : Наук. думка, 2008. 520 с.
[18] Peter Grunwald Industrial Biocatalysis 2014. 1232 p.

"liquid", it easily flows through the oil "placers" of rock. It is necessary to make the water more "viscous", and for this they found a means – a sugar-containing substance secreted by bacteria of the genus Xanthomonas, called xanthan. But first, the water is mixed with a surfactant (soap), the mixture is pumped into oil rocks to release it. By the way, such "soap" can also be formed by bacteria themselves. After that, xanthan water is given, which squeezes the oil out of the well under high pressure. There are also projects to "pump" microorganisms directly into oil-bearing strata, provided they are provided with oxygen: the action of bacteria produces gases that, due to the pressure created, will force oil sources to emerge more vigorously from the ground, for example in the form of a fountain. Xanthan is one of the very first products obtained in a bioreactor, and was previously unknown. So far, the young biotechnological industry supplies only some of these new "bioproducts". But already today they demonstrate the grandiose possibilities of the bioindustrial future[19, 20].

CONCLUSIONS

Biotechnology plays an extremely important role in the greening of industrial production based on the creation of waste-free processes: the use of biotechnological processes for water purification, biological methods for destroying crop pests are confidently displacing seemingly competitive chemical insecticides. Thanks to biotechnology, energy – and resource-saving production has been developed and implemented. Biotechnological processes are the basis for obtaining feed and food protein: today in the world a considerable part of it is produced using microbiological oil processing. Renewable energy sources are obtained using biotechnology. Biogas plants will not take away food, land from residents, but will even increase agricultural incomes and are quite profitable and quickly pay for themselves. Their use will stop the spread of deserts and make remote lands fertile and preserve significant forest areas of our planet. Biotechnological stages of production processes are implemented using living organisms. It is worth emphasizing that the basis of most classical methods of biotechnology is enzymatic processes and in most cases the objects of research are microorganisms. However, other living organisms are also of great importance – plants and animals, the improvement of which is carried out using traditional methods of genetics, selection, physiology, biochemistry, etc.

[19] Andreas S. Bommarius, Bettina R. Riebel-Bommarius Biocatalysis: Fundamentals and Applications. 2007. 634 p.

[20] Nicholas J. Turner, Luke Humphreys Biocatalysis in Organic Synthesis: The Retrosynthesis Approach. 2018. 416 p.

The universal nature of modern biotechnology is manifested in the widespread use of methods of cellular and genetic engineering, which are based on the development of molecular genetics, which provides opportunities for genetic reconstruction of living organisms in the directions desired by researchers. The main goal of these studies is to obtain the greatest possible diversity of organisms through genetic reconstruction, which could be used not only for the production of qualitatively new products, but also for the processing of various organic and inorganic substances.

Humanity expects the creation of such cell cultures, with the help of which it will be possible to produce valuable medicines, fight a number of hereditary, cancer, cardiovascular and other diseases, contribute to the purification and improvement of the ecological state of the environment. Particularly promising is the development of new disease-resistant, highly productive plant forms with improved product quality. The pace of biotechnology development can currently be compared to the impressive progress of computer technology more than 20 years ago, and the impetus for this was the birth of genetic and cellular engineering.

The processing of plant waste into energy sources has a significant economic effect. Biofuel production is becoming purposeful, although it was previously a by-product of the production of feed and food products. To obtain biofuels, not only plant or animal waste is used, but also cellulose and hydrocarbon and industrial waste. With the help of the yeast Saccharomyces cerevisiae, ethanol is obtained from plant raw materials (sugar cane, cassava, sugar beets, molasses, potatoes, wheat grains, corn).

SUMMARY

On an industrial scale, biotechnology is a bioindustry that includes, on the one hand, industries in which biotechnological methods replace traditional ones, and on the other, industries in which biotechnology plays a major role. Among the first in the chemical industry is the synthesis of artificial seasonings, polymers and raw materials for the textile industry, in the energy sector – the production of methanol, ethanol, biogas and hydrogen, in the biometallurgy sector – the extraction of metals. In the second group of industries, biotechnology is involved in the production of products (cultivation of yeast, algae, bacteria to obtain proteins, amino acids, vitamins, and for the use of their enzymes), increasing agricultural productivity (cloning and selection of plant varieties based on tissue and cell cultures, production of bioinsecticides); the pharmaceutical industry (development of vaccines, synthesis of hormones, interferons and antibiotics); environmental protection and reduction of its pollution (wastewater treatment, household waste processing, composting, as well as the production of compounds that are broken down by microorganisms). The oldest biotechnological process is fermentation, improving, increasing the efficiency, and studying the many biochemical reactions inherent in microorganisms, went in parallel with the release from the cells of bacteria and fungi of substances that replaced synthetic products, namely hypotensive, anti-inflammatory drugs and antiparasitic agents. Modern biotechnological processes are based on recombinant DNA methods, the use of immobilized enzymes, cells and cell organelles. The technology of immobilized enzymes has been successfully used for the production of semi-synthetic penicillins, fructose concentrate, even more effective immobilized cells and cell organelles containing the necessary genes for the synthesis of complex compounds. Biotechnological processes related to microbiology and enzymology play an important role in the cultivation of viruses, in the production of vaccines, for the production of interferon, as well as for the synthesis of monoclonal antibodies by hybridoma cells. This development has also affected plants, the cultures of which are used for the synthesis of various substances, both common: alkaloids, and other secondary metabolites, and exotic idioliths.

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Information about the authors:
Kichura Dariia Bohdanivna,
Candidate of Technical Sciences,
Associate Professor at the Department of Organic Substances,
Institute of Chemistry and Chemical Technologies,
Lviv Polytechnic National University
12, Stepana Bandery street, Lviv, 79013, Ukraine

Sobechko Iryna Borysivna,
Doctor of Chemical Sciences,
Associate Professor at the Department of physical,
analytical and general chemistry,
Institute of Chemistry and Chemical Technologies,
Lviv Polytechnic National University
12, Stepana Bandery street, Lviv, 79013, Ukraine