A biofuel cell (BFC) is a bioelectrochemical device that can directly produce electricity or biohydrogen as a result of highly efficient “cold” fuel combustion. Nowadays there is no unified definitive classification and terminology, because BFCs are complex devices and they are still at research stage.

Ukrainian scientists have proposed a classification of BFCs based on nature of the biological component in the anode chamber, type of enzymes, presence of mediators, etc. Such classification is still relevant today, but due to the expansion of research areas and promising fields of BFC implementation and creation of hybrid and integrated systems there is a need to expand the review of existing BFC. The aim of the work was to study the current state of development of different BFC types and prospects for their implementation.

Results of the analysis of modern publications in the field of BFC research have revealed a wide range of variations and possible promising fields of BFC application.

Further research and implementation of these devices as environmentally friendly fuel for autonomous operation of robots, in biosensors and for wastewater treatment etc. should be based on the study of biotechnological parameters of biofilm formation and operation of BFC.

**Key words:** biofuel cell, biofilm, biosensor, microbial fuel cell, biocathodes.
In our review a biofuel cell is defined as a term for all bioelectrochemical systems, i.e., such electrochemical systems that have a biological component in at least one (anode or cathode) chamber. Pandey et al. [3] and domestic scientists [1, 4] identify basic (main) components of BFCs as the following: 1) electrodes, which are made of carbon-based materials (graphite, graphite felt, carbon paper, carbon fabric), stainless steel, platinum etc; 2) anode and cathode chambers, which can be made of glass, plexiglass, polycarbonate and other materials that are inert to wastewater, microorganisms, buffer solutions and are not toxic.

**Classification of biofuels and areas of use**

Domestic scientists [4] proposed a classification of BFC. This classification was based primarily on the nature of biological component in anode chamber in enzymatic or microbial fuel cells (EFC and MFC), type of enzymes, presence of mediators, etc. This classification is relevant today, but due to creation of hybrid and integrated systems there is a need to expand the review of existing BFC (Table).

Zhou et al. [5] proposed to classify BFC according to a design. Firstly, the configuration is one-, two- and multichamber. Secondly, the structure of reactor is planar, disk, tubular (cylindrical) and concentric. Planar is more appropriate to use in biosensors with the view to reduce the impact of concentration resistance. According to the presence of membrane BFC’s are divided into membranes (anion-exchange membranes and cation-exchange membranes) with a salt bridge and membraneless one. According to the mode of operation there are non-flowing and flowing BFCs. According to the type of cathode the division is as follows: air cathode, biocathode and abiotic cathode.

As Pandey et al. [3] noted such proton exchange membranes (PEM) as Nafion®, Ultrex®, polyethylene, polystyrene-divinylbenzene, salt bridge, porcelain partition, terracotta, ceramics [6], etc. are widely used among the membranes. Additional components are catalysts: Fe³⁺, Pt, polyaniline, MnO₂, etc. [3].

Flow-type BFC can be promising on an industrial scale, for example at wastewater treatment plants, where water is supplied constantly. However, a technological complication may be the need to provide an oxygen-free environment in the anode chambers, because the wastewater entering the water treatment plant contains dissolved oxygen. The possibility of wastewater treatment of both industrial (pig farm effluents) and domestic wastewater was demonstrated. At the initial values of COD 60 g/dm³ in industrial effluents single-chamber BFC with bioanode and air cathode reduce the value of COD by 76–91% while producing electricity with a capacity of 1–2.3 W/m² [5].

The use of BFC with a bioanode is limited by the fact, that abiotic cathodes are coated with catalysts, such as platinum, or use mediators such as iron (II) phthalocyanine to reduce activation resistance. If this technology is used for wastewater treatment, the disadvantages are high cost of platinum and secondary water pollution. The use of biocathodes and electrodes with a developed surface is proposed as a solution. Among electrodes with developed surface there are the following variations: formation of electrodes in the form of brushes and use of carbon materials and nanomaterials [7].

Hays [8] suggested the use of cathodes made of activated carbon. However, the question of influence of the developed surface on electricity generation is still under study [7].

BFC could also be classified according to electron acceptors in the cathode chamber. Oxygen is the most attractive, environmentally friendly and cost-effective electron acceptor, but its disadvantage is its limited solubility, being 8 mg/dm³ as noted by Rabaey [7]. It should be noted that solubility changes with changes of temperature and pressure. If technology is used for wastewater treatment, then climatic conditions of the area, where the wastewater treatment takes place, should be taken into account as external conditions will affect both solubility of oxygen, metabolism of microorganisms and the technology as a whole.

Rabaey et al. [7] noted that anions such as NO³⁻, SO₄²⁻ could be acceptors. Thus, BFC can perform denitrification and purify wastewater from sulfates. Denitrifying bacteria can be used to reduce NO₃⁻ in the cathode chamber [9]. However, wastewater should be added to the cathode chamber only after pre-treatment and mixing with buffer. Oxygen can be spent on the oxidation of organic compounds when using untreated wastewater.

One of the types of BFC is MDC (Microbial Desalination Cell). These are BFC that are able to desalinate water [10]. This type of BFC involves the supply of liquid-containing salts between the electrode chambers. Cathode chamber is separated from liquid-containing salts by anion exchange membrane while
the cathode chamber is separated by cation exchange membrane. This type of BFC can be used to desalinate seawater or wastewater into drinking water quality in areas where access to drinking water is limited.

Electron acceptors in cathode chamber can be not only oxygen molecules, but also MnO$_2$, Fe$^{3+}$, which, with the participation of bacteria and oxygen, are converted into Mn$^{2+}$, Fe$^{2+}$ [24].

The fundamental possibility of using BFC-based biosensors, implanted in macroorganisms, was demonstrated. The study was performed on mice. Biosensor implants with electrodes volume of 0.1 cm$^3$ showed a power of 1.8 μW at a current of 5 μA. These devices can measure the content of glucose in blood in real time, responding to changes in glucose content by level of electrical signal [11]. The problems of use are in determining the acceptable size of biosensors in the body, which could provide required threshold of power density, long service life, stability and reproducibility. There is also a need to use biocompatible materials in order to avoid rejection of such devices, as well as to provide their long-term operation. Non-invasive BFC-based biosensors are possible too [12]. It has been shown that the combination of such nanomaterials as graphene and gold nanoparticles can solve the problem of miniaturization of devices and increase their efficiency. Such BFCs have the power of $1.10 \pm 0.12$ W/cm$^2$ [12], which is many times more than ordinary BFCs. Because the power of BFCs is affected by the concentration of glucose in saliva, these devices can be used as non-invasive as the power of BFCs is affected by concentration of glucose in saliva.

In addition, BFC can be used as biosensors in bioreactors. In this case, they will work as “biocomputers” [13]. The essence of their work: BFCs respond to the concentration of substrate, co-products, toxins, etc. in real time by changing electric current. The change of current sends a signal to the automation and control system, which is able to change the parameters of cultivation (supply of substrate, temperature change, selection of culture fluid, supply of detoxifiers, etc.). However, the stability of such systems and their reproducibility for the purpose of industrial implementation is still being studied [20]. BFCs have this feature due to the wide range of substrates that can be used in them as electrolytes. Butyrate, acetate, glucose, ethanol, as well as mixtures of glucose-lactate-cellulose or glucose-glutamate etc. were used as substrates. In addition to model solutions, wastewater was also used as a substrate [25].

| Types of BFC | Purpose | References |
|-------------|---------|------------|
| MDC         | Desalination of sea water | [10]       |
| Biosensors based on BFC (including EFC) | Development of invasive and non-invasive biosensors for medical use | [11], [12] |
| “Biocomputers” | Response to changes in parameters in bioreactor in real time using current; integration with automation systems | [13] |
| BFC-based robots | Generating current for autonomous operation of robots in remote areas (space, deserts, etc.) | [14] |
| MCC         | Wastewater treatment and reduction of carbon dioxide emissions | [5] |
| SMFC:       | Bioremediation of soils with current generation | [15]       |
| SB-SMFC     | Treatment of reservoirs and wastewater with electricity generation | [16]       |
| FME-MFC     | Autonomous current generator for sea buoys | [17]       |
| BMFC        | Wastewater treatment with current generation | [18]       |
| CW-MFC      | Reduction of carbon dioxide emissions, transformation of electrical energy into chemical; fermentation | [19], [20] |
| BFC with MBR | Reducing the level of membranes’ overgrowth | [21]       |
| PBECs       | Generation of current and production of biohydrogen | [22]       |
| MFC         | Generation of current | [23]       |
| MEC         | Getting energy carriers (biohydrogen) | [1]         |
| MEC         | Getting energy carriers (biohydrogen) | [1]         |

Types of biofuel cells and their purpose

| Types of BFC | Purpose | References |
|-------------|---------|------------|
| MDC         | Desalination of sea water | [10]       |
| Biosensors based on BFC (including EFC) | Development of invasive and non-invasive biosensors for medical use | [11], [12] |
| “Biocomputers” | Response to changes in parameters in bioreactor in real time using current; integration with automation systems | [13] |
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| PBECs       | Generation of current and production of biohydrogen | [22]       |
| MFC         | Generation of current | [23]       |
| MEC         | Getting energy carriers (biohydrogen) | [1]         |
BFCs are being considered as a source of energy for autonomous robots. Nowadays, the works of EcoBot and Gastrobot have been developed [14]. Gastrobot was the first robot to run on BFC energy. EcoBot is an autonomous robot; today there are 4 types of this one. Their development has been in progress since 2002. Model of EcoBot-I BFC contains E. coli as a biological agent, and sucrose is used as a substrate. BFC in EcoBot-II model contains activated sludge, which can use flies and rotten fruit as a substrate, so the robot based on such BFC is more autonomous than the previous model. The EcoBot III model is able to capture water and nutrients from the environment. The EcoBot IV model is in the process of development. Such robots can be used in areas, where autonomous work is required and human intervention is not possible, e.g., in the study of water bodies at great depths, in space, in desert areas, etc. [26].

Enzyme fuel elements (EFC) are also identified as a separate type of BFC [27]. The possibility of using oxidoreductases in both the anode and the cathode chambers is considered. Enzymes such as glucose oxidase and glucose dehydrogenase were investigated in the anode chamber, and laccase and bilirubin oxidase were investigated at the cathode chamber, the latter using oxygen as an electron acceptor. Before use enzymes are immobilized on nanomaterials, from which electrodes are made. Such EFCs show power of 155 μA/cm² [27]. However, EFCs require high-cost pre-production and purification of enzymes.

Biocathode with microalgae may be used as an alternative to abiotic cathodes. Dissolution of oxygen through the water surface is limited, so the solution may be artificial aeration. On the other hand, this requires an increase in energy consumption, microorganisms that are able to produce oxygen, such as microalgae, can be cultivated so in the cathode chamber. In addition, microalgae are able to purify water from nitrates, sulfates, heavy metals, etc. [9]. Separately identify among biocathod, another type of BFCs, MCC (Microbial carbon capture cell) is also a separate case of biocathodic BFCs. Such BFCs contain microorganisms, such as microalgae, in the cathode chamber, these microorganisms are able to use carbon dioxide from the anode chamber or one that is bubbled from other sources. The advantages of such BFCs, when using carbon dioxide from the anode chamber, are the closure and reduction of CO₂ emissions, which is released during the biochemical decomposition of organic matter with the simultaneous generation of electricity [5].

Freguia et al. [28] proposed to treat wastewater in the cathode chamber after the anode chamber. In this case, both cathodophilic and heterotrophic bacteria are present in the cathode chamber. The following efficiency of purification of model solutions was demonstrated: the concentration of acetate before purification in the anode chamber was 500 mg/dm³, at the outlet of the anode chamber — 10–200 mg/dm³, after the cathode — less than 5 mg/dm³. Thus, the treatment efficiency is more than 99%. It should be noted that not all organic compounds are subject to biodegradation in real wastewater, so the efficiency in real wastewater may be lower. The integration of a photobioreactor with microalgae with BFC was studied in addition to direct usage of algae in anode or cathode chambers [29, 30].

Other types of BFC are sediment MFCs (SMFCs), including SB-SMFCs (soil-based), with FME-MFC ecosystem (floating macrophytes-based ecosystem) and BMFCs (benthic) [30]. In a BFC of this type, as noted in [20], there is no need to use a membrane, because the natural barrier (soil or thick layer of water) plays such a role, but these systems may require additional energy. The existence of redox potential in soil was known in the first half of the XX century [31], but the usage of this energy was not considered. In addition to generating electricity, SB-SMFCs are able to treat groundwater in situ. It has been shown that the usage of SMFC in soil increases the efficiency of remediation in comparison to a similar bioremediation system that does not contain BFC i.e. from 12.3% to 90.1%. Such devices are capable of developing power of 101.52 mW/m² [15].

Macrophytes can be used in the FME-MFC. It is worth mentioning that power increased from 80.08 mW/m² to 179.78 mW/m² with the decrease of COD. This proves the importance of COD value in such systems and the need for pre-treatment of wastewater [16].

BMFC is a type of BFC that is able to use the marine environment to generate current. In this case, benthic deposits such as organic substances and sulfides act as substrates for anode microorganisms. The usage of graphite electrodes allowed increasing the power from 0.12 mW/m² to 30 mW/m² [17]. This type of BFC is used for sea buoys.

CW-MFC are similar to FME-MFC. CW (Constructed Wetland) is widely used and studied for the purpose of wastewater treatment. At the same time, such systems have aerobic and anaerobic zones, separated
by layers of water, which is suitable for creation of CW-MFC [30]. The power of electricity generation was obtained, being 43 m W/m² in case of CW-MFC. It should be noted that CW is placed on sloping sites, so wastewater moves in these systems under the influence of gravity, and therefore all the electricity produced is an energy gain [9].

In CW-MFC flow-type systems with additional aeration, in which *Elodea nuttallii* was used in the cathode part, the maximum power was obtained — 184.75 ± 7.50 m W/m³, and 204.49 W/kg of COD. In addition, such systems can remove nitrates an ammonium. The possibility of using macrophytes *Canna indica*, *Ipomoea aquatica*, *Phragmites australis* was demonstrated [18]. However, the usage of such BFC can be limited by climatic conditions, because the metabolism and reproduction of macrophytes slows down in the absence of sunlight and lowering the temperature.

Hydro-, wind- and solar power plants can produce more electricity than it can be consumed, so consequently there is a need to store it. This electricity with the carbon dioxide can be used in BFC, while conserving electrical energy in the form of chemical bond energy. It has been demonstrated that wastes from one production, such as glycerol, can be processed into ethanol, i.e. biofuels. If metabolism of these substances occurs by bioelectrocatalysis (Figure, a), BFC can be used for production of organic substances. It is possible to place microorganisms both on cathode and anode. When using a biocathode, water is electrolyzed in the anode with the help of additionally applied voltage (Figure, b). Advantages of producing organic compounds in BFC are presence of biofilm and stereospecificity of products. However, the disadvantage is, firstly, the energy consumption of the process, and secondly, the need to add a substrate for growth, because the substrate, from which the target product is formed, is not used by microorganisms for their own growth and development. Carbon dioxide for biochemical production of acetate can be used as a substrate [19]. Thus, it is possible to reduce carbon dioxide emissions with the simultaneous production of organic matter, and electrical energy can be stored in organic compounds by microorganisms [20].

Another type of BFC usage in integration is its combination with MBR systems. This idea was proposed by Logan et al. [21] with the view to reduce overgrowth of MBR with activated sludge, because this problem is a deterrent to the widespread use of systems of this type. It was demonstrated that integration of MBR with BFC due to the electric field, produced in BFC, as well as production of hydrogen peroxide at the cathode helped to reduce membrane overgrowth. The power of such MPE is 0.0197 kWh/m³.

Taking into account the fact that solar energy is inexhaustible, studies

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**Types of biofuel cells:**

*a* — bioelectrochemical control of fermentation; *b* — microbial electrosynthesis

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have been conducted on the usage of BFC in combination with photosensitive semiconductors, which can be used as photoanodes and photocathodes depending on the nature of these semiconductors. Lee et al. [32] described the possibility of combining a photoanode and a bioanode by TiO₂ nanotube. At the same time, oxygen was reduced to water at the cathode. Zubchenko [22] studied the type of the integrated systems of BFCs photobioelectrochemical systems (PBECS). In PBECS there is a process that is similar to the photolysis of water, but the proton donors are organic matter, not water. Integrated systems consist of 4 electrodes, including a bioanode, a cathode for hydrogen reduction, an abiotic anode and a photocathode, consisting of semiconductors. The need to use additionally applied voltage is reduced and simultaneous production of both current and biohydrogen with such integrated systems becomes possible.

However, despite the great attractiveness of BFC and the wide range of their use, among the problems, which hinder industrial implementation, Kadier [33] highlighted the following: high cost of abiotic cathodes in particular due to the need for platinum and catalysts; insufficient ability to scale BFC into sustainable and long-term operation. Another reason for the lack of industrial implementation was the insurmountable power gap. Thus, chemical fuel cells, currently used in industry, have a power of 1−10⁸ W, while today the power of BFCs ranges from 10⁻⁶−10⁻² W [34]. That is, today the level of electricity generation by BFC is still is at the level of laboratory research, and, according to Rabaey [7], is insufficient for implementation on an industrial scale so far. BFC requires further study, namely the peculiarities of microbial metabolism of exoelectrogens, use of biocathodes, microbiological communications, etc., for the industrial implementation. It is important to determine the parameters of usage, substrates and different types of membranes, electrodes and directions of application [10].

Today BFCs have a wide range of variations and possible promising areas of application. Such devices can provide bioremediation, generation of green energy and energy carriers. The use of biocathodes can reduce carbon dioxide emissions along with obtaining of organic matter. BFC-based biocomputers, integrated with an automation system, have the potential to determine online the content of substrates or toxicants in bioreactors and the signal to automation systems. The usage of such devices in robots (as an ecological source of energy) is promising for the autonomous operation of robots. Invasive and non-invasive biosensors, based on BFC, are able to expand the spectrum of detection of compounds in human blood, saliva and simplify the transmission of electrochemical signals. Taking into account the wide prospects for the usage of BFC, as well as environmental friendliness of devices of this type, there is an urgent need for further study and implementation. It is advisable to study the formation and operation of anode and cathode biofilm with the view to increase the power of BFC along with the selection of biocorrosion-resistant, non-toxic and electrically conductive materials that will reduce the cost of technology for widespread introduction of BFC with bioanode and biocathode with the purpose of cleaning the environment from contaminants such as wastewater, carbon dioxide and etc.

The production of energy sources, such as hydrogen, will expand the range of production of environmentally friendly fuel from waste. The use BFC with membrane technologies will reduce the level of membrane overgrowth, which is promising, given the labor-intensive process of membrane regeneration. MDC water desalination is promising both for production of fresh water in arid areas and for treatment of highly concentrated wastewater.

A wide range of biochemical reactions allows the usage of BFC in many industries, but for the widespread implementation of BFC it is necessary to resolve such technological issues as optimal biotechnological parameters of exoelectrogenic biofilm, long-term operation of flowing and non-flowing BFC, search for mathematical models that describe the processes of current generation, search for ecologically clean mediators, study the influence of electrode modification on the formation of the biofilm and on the metabolic processes that take place in it, etc.

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Біопаливний елемент (БПЕ) — біоелектрокіосмічний пристрій, який унаслідок високо-ефективного «холодного» горіння палива без-посередньо може продукувати електроенергію або біоводень. На сьогодні не існує єдиної ви-значеної класифікації та термінології, оскіль-ки БПЕ — складні пристрої, що перебувають на стадії дослідження.

Вітчизняними вченими було запропонова-но класифікацію БПЕ за природою біологічної складової в анодній камері, видом ензимів, на-явністю медіаторів тощо. Така класифікація є актуальною і сьогодні, однак через розши-рення галузей дослідження та перспективних сфер впровадження БПЕ створення гібрид-них та інтегрованих систем, виникає потреба збільшити обсяг огляду існуючих БПЕ. Метою роботи було дослідити сучасний стан розро-блення різних видів БПЕ та перспективи їх упровадження.

Результати аналізу сучасних публікацій у галузі досліджень БПЕ дали змогу виявити ве-ликий спектр варіацій та можливих перспек-тивних сфер їх застосування.

Подальше дослідження та впровадження таких пристріїв як екологічно чистого палива для автономної роботи роботів, у біосенсорах, для очищення стічних вод тощо має базувати-ся на дослідженні біотехнологічних параме-трів формування біооплівки та функціонування БПЕ.

Ключові слова: біопаливний елемент, біооплів-ка, біосенсор, мікробний паливний елемент, біокатоди.