

## ELECTRICAL ENERGY GENERATION BY MICROBIAL FUEL CELLS WITH MICROALGAE ON THE CATHODE

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**Background.** The possibility of converting organic compounds into electrical energy in microbial fuel cells (MFCs) makes MFCs a promising eco-friendly technology. However, the use of platinum or hexacyanoferrates may increase costs or lead to secondary environmental pollution. The use of microalgae in the cathode chamber is a promising solution to these problems.

**Objective.** We aimed to establish the dependence of electrical energy generation and the efficiency of the application of a specific type of algae on the type and mode of lighting.

**Methods.** In the study, two-chamber H-type MFC with salt bridge was used. Fermented residue after methanogenesis was used as inoculum in the anode chamber, and microalgae cultures *Chlorella vulgaris*, *Desmodesmus armatus*, and *Parachlorella kessleri* were used as inoculum in the cathode chamber.

**Results.** MFCs with microalgae demonstrate the ability to generate current under different light sources. The maximum voltage for the MFC with an anode biofilm and with microalgae in the cathode chamber is 13–15% lower compared to the MFC with an abiotic cathode ( $840 \pm 42$  mV). The maximum current is 2–6% lower than the control ( $480 \pm 24$   $\mu$ A) for the MFC with *Chlorella vulgaris* and the MFC with *Parachlorella kessleri*, and 8% higher for the MFC with *Desmodesmus armatus* compared to the MFC with an abiotic cathode. The MFCs with microalgae are capable of generating electrical energy for an extended period.

**Conclusions.** With a pre-grown anodic biofilm, both the current and voltage maintain relative stability when the light source is changed. The potential use of solar lighting broadens the applicability of the MFCs with microalgae, as it eliminates the need for additional costs associated with artificial light sources.

**Keywords:** microbial fuel cell; biofilm; bioanode; biocathode; microalgae.

### Introduction

Climate change is a global issue that necessitates international cooperation. In addressing this challenge, world leaders convened at the UN Climate Change Conference in 2015 and negotiated the Paris Agreement. This landmark agreement officially came into force on November 4, 2016. As of now, 195 Parties, including Ukraine, have joined the Paris Agreement. The accord establishes long-term goals for reducing global greenhouse gas emissions, signaling a commitment to transition towards a net-zero emissions world, which involves a reduction in the use of fossil fuels for energy purposes [1]. Bioenergy stands out as a promising field capable of replacing fossil resources, including natural gas, oil, and coal. As a renewable energy source, bioenergy plays a vital role in enhancing energy independence, fostering local economic development, and mitigating the environmental impact. Therefore, the research and implementation of bioenergy technologies have become an urgent priority.

One such technology is Microbial Fuel Cells (MFCs), a bioelectrochemical process in which

microorganisms participate in the transformation of organic compounds [2]. The versatility of MFCs, capable of transforming a wide range of organic compounds through mixed microbial associations, makes it suitable for wastewater treatment with simultaneous energy generation [3].

Most of the investigated MFCs consist of anodic and cathodic chambers separated by a membrane. In the anode chamber, exoelectrogenic microorganisms decompose organic compounds, releasing electrons, protons, and carbon dioxide. Electrons are transported to the cathode through an external electrical circuit, while protons pass through the membrane. At the cathode, electrons, protons, and oxygen combine to form water molecules [4]. In these MFCs, platinum or hexacyanoferrate is commonly used. The use of platinum significantly raises the technology's cost, and the use of hexacyanoferrate can result in secondary pollution [3].

Alternatively, artificial aerators can be employed in the MFC cathode chamber, albeit at an additional cost to the technology [5, 6]. Considering that, in addition to producing oxygen, algae can capture carbon dioxide through photosynthesis, increasing biomass that can be utilized for the pro-

duction of biodiesel and other value-added products, the use of algae in the cathode chamber is a promising avenue of research [7, 8].

The photosynthetic production of oxygen relies on light. Bazdar *et al.* [9] determined that under constant illumination with a fluorescent lamp at white light intensities of 3500, 5000, 7000, and 10000 lux, the MFC voltage measures 509, 544, 524, and  $465 \pm 4$  mV, respectively. However, maintaining constant lighting increases the costs of the technology. Don *et al.* [10] determined that when illuminated with a fluorescent lamp in the light/dark mode: 12/12 and 32/32 h, the maximum voltage of MFCs with algae is 340–360 and 375 mV during illumination, respectively, and decreases in the dark. However, the use of artificial illumination for MFCs can increase the cost of the technology, especially at scale. Thus, establishing the possibility of replacing it with sunlight is an important consideration.

Many studies on full microbial fuel cells utilize a single lighting mode and a single species of microalgae, conducted under different startup conditions, making it challenging to compare the effects of different lighting types and modes. The aim of our study was to determine the relationship between the generation of electrical energy and the efficiency of applying a specific type of algae based on the type and mode of lighting.

## Materials and Methods

**Experiment 1.** We utilized a two-chamber H-type MFC with a salt bridge connecting the chambers. Cathode and anode chambers had a volume of  $1 \text{ dm}^3$  and were constructed from polypropylene. The salt bridge was composed of 2.4 g of agar, 8.947 g of KCl, and 120 ml of distilled water [11]. Its length was maintained at  $105 \pm 5$  mm, and the chambers were linked by a polyvinyl chloride tube with a diameter of  $\varnothing 8 \times 3$  mm, ensuring a sealed entry with PG-11 cable entries. The electrode frame was constructed from stainless steel mesh, wrapped with carbon fiber. The entire electrode weighed  $8.5 \pm 0.5$  g, with the carbon thread contributing  $4.5 \pm 0.5$  g. Electrodes, measuring  $100 \times 50$  mm ( $\pm 5$  mm) with a visible area of  $0.01 \text{ m}^2$ , underwent cleaning with 1N HCl and 1N NaOH, followed by rinsing with distilled water and soaking in distilled water for 24 hours before use [12].

The anolyte was composed of a 50 mM PBS buffer solution (pH 6.1) with sodium acetate ( $1 \text{ g/dm}^3$ ) [13],  $\text{FeCl}_3$  ( $200 \mu\text{M}$ ) [14], and 10 ml of vitamins and minerals. The PBS components

were present in the following concentrations (in grams per liter of distilled water): 4.58  $\text{Na}_2\text{HPO}_4$ , 2.45  $\text{NaH}_2\text{PO}_4$ , 0.31  $\text{NH}_4\text{Cl}$ , 0.13 KCl. The concentrations of components in the vitamins and minerals solution were (in milligrams per liter of distilled water): 1.5 retinol palmitate, 0.01 cholecalciferol, 60 ascorbic acid, 13 nicotinamide, 10  $\alpha$ -tocopherol acetate, 5 calcium pantothenate, 1.2 riboflavin, 1 thiamine nitrate, 2 pyridoxine hydrochloride, 0.003 cyanocobalamin, 0.4 folic acid, 20  $\text{Mg}^{2+}$  (magnesium lactate), 15  $\text{Ca}^{2+}$  (calcium hydrogen phosphate), 12  $\text{P}^{5+}$  (calcium hydrogen phosphate), 10  $\text{Fe}^{2+}$  (ferrous fumarate), 3  $\text{Zn}^{2+}$  (zinc sulfate), 1  $\text{Cu}^{2+}$  (copper sulfate), 1  $\text{Mn}^{2+}$  (manganese sulfate), 0.1  $\text{Mo}^{6+}$  (sodium molybdate).

In the first experiment, the MFC with a pre-grown exoelectrogenic biofilm under the conditions of applying an external voltage of 3 V was utilized [15]. Once the biofilm had formed at the anode, no additional external voltage was applied. The catholyte, containing  $\text{K}_3[\text{Fe}(\text{CN})_6]$ , was replaced with Tamiya medium, and microalgae cultures were introduced into different MFCs: MFC 1 – *Chlorella vulgaris*, MFC 2 – *Desmodesmus armatus*, MFC 3 – *Parachlorella kessleri*. In the control MFC, the catholyte containing  $\text{K}_3[\text{Fe}(\text{CN})_6]$  was not replaced. While biofilms were pre-grown in MFC 1, MFC 2, and MFC 3, the control MFC in this experiment had no anodic biofilm. This means that in this study, the MFC with an abiotic anode and abiotic cathode served as the control MFC.

The microalgae cultures, namely *Chlorella vulgaris*, *Desmodesmus armatus*, and *Parachlorella kessleri*, were sourced from the collection of the M.G. Kholodny Institute of Botany of the National Academy of Sciences of Ukraine. Detailed information about these cultures can be found in [16].

The fermented residue obtained after methanogenesis (Department of Bioenergy, Bioinformatics, and Environmental Biotechnology of the National Technical University of Ukraine "Igor Sikorsky Kiev Polytechnic Institute") served as a valuable reservoir of exoelectrogens. This residue, resulting from methanogenesis, comprises a diverse array of microorganisms, particularly exoelectrogens and methanogens. The inoculum is notably enriched with microorganisms from the Methanosarcinaceae family, including *Methanobacterium* sp. and *Methanosaeta* sp. This specific composition was thoroughly documented by Golub *et al.* [17].

The microbial communities present in this particular type encompass various taxa, including Firmicutes and Proteobacteria, with a noteworthy

representation of *Geobacter* sp. *Geobacter* sp. is recognized for its distinctive capacity to utilize electrodes as electron acceptors. The choice of sodium acetate as a carbon source aligns with the preference of *Geobacter* sp. for acetate as their preferred substrate [18, 19].

The Tamiya medium, prepared in gram per liter of distilled water, comprises: 5 KNO<sub>3</sub>, 2.5 MgSO<sub>4</sub>·7H<sub>2</sub>O, 1.25 KH<sub>2</sub>PO<sub>4</sub>, 0.037 EDTA, 0.003 FeSO<sub>4</sub>·7H<sub>2</sub>O, and 1 mL of a mineral solution. The mineral solution includes (in milligram per liter of distilled water): 2.86 H<sub>3</sub>BO<sub>3</sub>, 1.81 MnCl<sub>2</sub>·4H<sub>2</sub>O, 0.22 ZnSO<sub>4</sub>·7H<sub>2</sub>O, 0.15 Co(NO<sub>3</sub>)<sub>2</sub>·4H<sub>2</sub>O, 2.3 NH<sub>4</sub>VO<sub>3</sub>, and 0.01 CuSO<sub>4</sub>·5H<sub>2</sub>O [16]. The cathode chamber was not sealed hermetically, allowing gas exchange between the cathode chamber and the environment. This design facilitated carbon dioxide exchange, crucial for microalgae growth. This approach aligns with previous studies where an open cathode chamber supported gas exchange with the atmosphere [10].

The experiment involving a pre-grown biofilm investigated the impact of different lighting types, including 12 W LED lamps (PPG T5i-900 Agro 12W IP20), 12 W and 8 W lamps (PPG T5i-600 Agro 8W IP20) with 650 nm and 450 nm LEDs, monochrome lighting using blue and red LEDs, and white LEDs, maintaining a light-to-dark ratio of 9:15 hours.

Blue light (400–450 nm) and red light (650–750 nm) are absorbed by pigments bound to chlorophyll. Therefore, blue and red light may have a positive impact on the photosynthetic process. Blue light can activate specific photoreceptor proteins, including phototropin and phytochrome A, while red light has positive effects on the biomass production of algae. White light, encompassing a broader spectrum, leads to higher cell density. Increased photosynthetic activity is associated with higher oxygen production. Given that oxygen serves as the terminal electron acceptor in the cathode chamber, enhanced photosynthetic activity is expected to have positive effects on the voltage and current of MFCs. Therefore, for this study, white, blue, red, and a combination of LED lights were chosen [20–23].

The study also examined the impact of sunlight (daylight duration during the study: 9.5 ± 1.25 hours, coordinates: 50°27'00"N 30°31'24"E, under direct sunlight, indoors) on the bioelectrochemical characteristics of MFCs. In all experiments, the light source was positioned on the side of the cathode chamber.

**Experiment 2.** Considering that the pre-growth of exoelectrogenic biofilm requires additional time and the application of voltage, and also acknowledging that MFCs can generate voltage and current under sunlight conditions, a second experiment was conducted. This involved the simultaneous addition of inoculum (fermented residue after methanogenesis) in the anode and corresponding microalgae in the cathode chamber. A control was also set up, in which the inoculum was added only to the anode chamber. The anolyte contained 50 mM PBS buffer solution (pH 6.1) with sodium acetate (1 g/L), FeCl<sub>3</sub> (200 μM) [24], and 10 ml of a solution of vitamins and minerals, as in Experiment 1. Fermented residue after methanogenesis (dry weight 11 ± 0.5 g) was used as an inoculum for the anode chamber. In the experiment with the simultaneous addition of the inoculum, sunlight was used as a source of illumination (length of daylight during the study was 15.5 ± 1.0 hours).

The catholyte contained Tamiya medium and cultures of microalgae: MFC 1 – *Chlorella vulgaris*, MFC 2 – *Desmodesmus armatus*, MFC 3 – *Parachlorella kessleri*. In the control, the catholyte contained only Tamiya medium. The terminal electron acceptor was oxygen, which was produced by microalgae (MFC 1, MFC 2, and MFC 3) and obtained from the environment, as the cathode chamber was not hermetically closed (all MFCs).

Voltage and current were measured with a multimeter (UT131C). The optical density of the catholyte was measured using a spectrophotometer (ULAB 102) at a wavelength of 680 nm, corresponding to the absorption peak of chlorophyll *a* [2].

Statistical analysis of the obtained MFC data was performed using Microsoft Excel software. Data analysis involved all variables, including voltage, current, optical density, computations, means, and standard deviations, as well as standard error. The mean ± standard error was presented in all graphs.

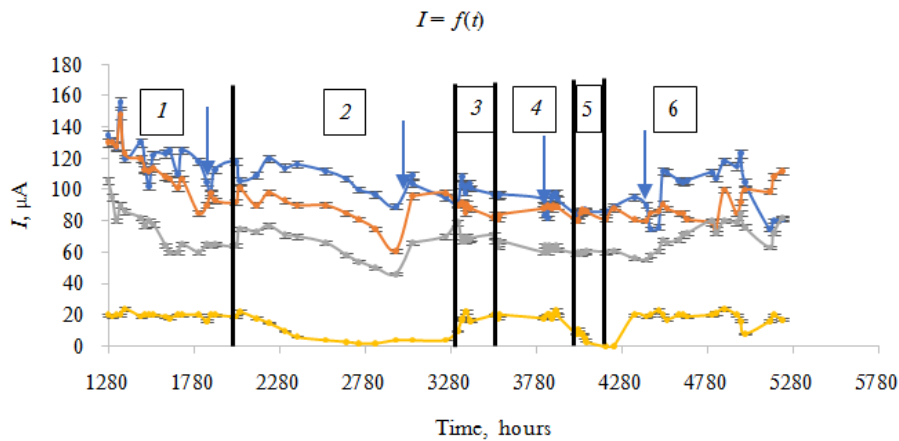
## Results

**Experiment 1.** *Effect of artificial and solar lighting on current and voltage generation of MFC with pre-grown anodic biofilm.* After the pre-growth of the biofilm under external voltage [15], the catholyte in the cathode chamber (catholyte composition described in [15]) was replaced with Tamiya medium containing microalgae, and the lighting was turned on. In the control, the catholyte was not replaced. After 24 hours under illumination (Figs. 1, 2), slight changes in voltage within the margin of error

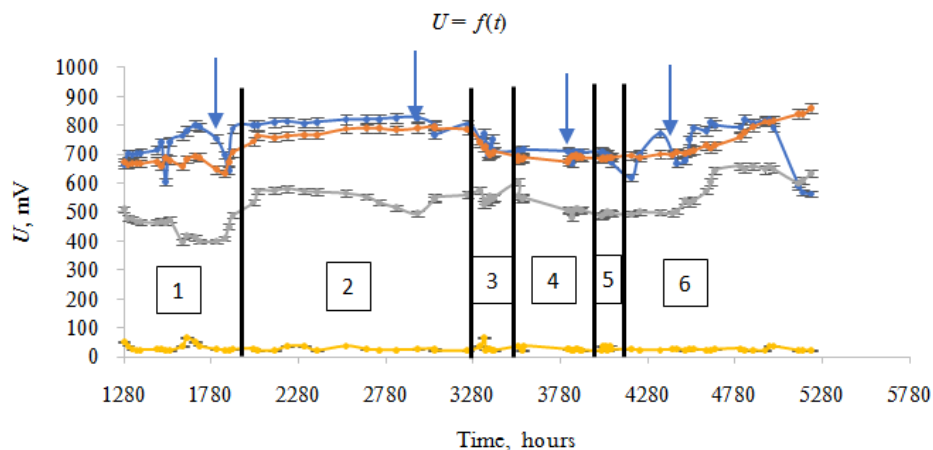
were observed. Specifically, in MFC 1 (*Chlorella vulgaris*), the voltage changed from  $706 \pm 35$  to  $663 \pm 33$  mV, in MFC 2 (*Desmodesmus armatus*) from  $660 \pm 33$  to  $672 \pm 34$  mV, and in MFC 3 (*Parachlorella kessleri*) from  $515 \pm 26$  to  $510 \pm 26$  mV. The current changed from  $142 \pm 7$  to  $135 \pm \mu\text{A}$  in MFC 1, from 130 to  $130 \mu\text{A}$  in MFC 2, and from  $109 \pm 5$  to  $106 \pm 5 \mu\text{A}$  in MFC 3. Different light conditions enabled obtaining varying currents in the MFC with *Chlorella vulgaris* (75–156  $\mu\text{A}$ ), in the MFC with *Desmodesmus armatus* (61–148  $\mu\text{A}$ ), and in the MFC with *Parachlorella kessleri* (46–106  $\mu\text{A}$ ) (Fig. 1),

as well as varying voltages in the MFC with *Chlorella vulgaris* (560–827 mV), in the MFC with *Desmodesmus armatus* (635–858 mV), and in the MFC with *Parachlorella kessleri* (397–660 mV) (Fig. 2). The MFC with an abiotic anode and cathode served as a control in this study. These results indicate that algae can sustain the generation of current in MFCs using different light sources.

**Experiment 2.** *The influence of sunlight on current and voltage generation of MFC with the simultaneously addition of the inoculum into the anode and cathode chambers.* After confirming that electricity



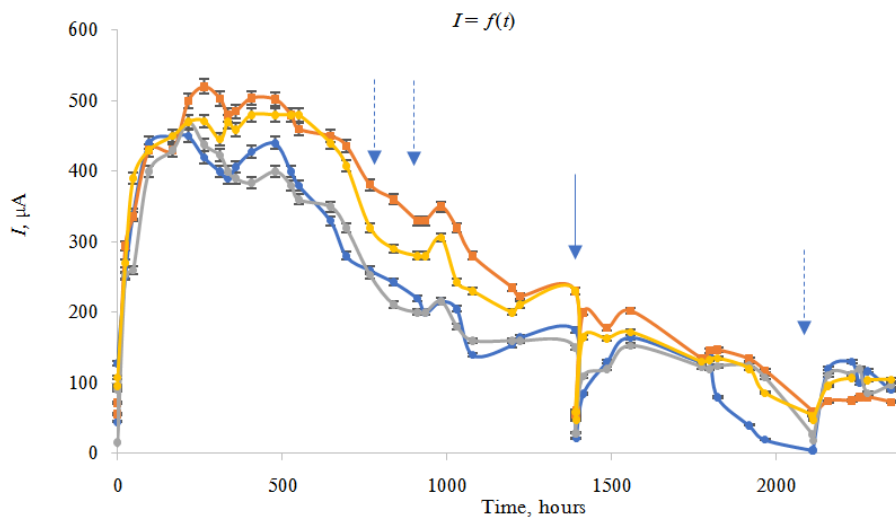
**Figure 1:** Change in the microbial fuel cell current over time. Arrows indicate the addition of  $\text{CH}_3\text{COONa}$  (1 g/L) to the anode chamber. Types of artificial lighting (light:dark = 9:15): 1 – 12 W LED lamp (650 and 450 nm LEDs), 2 – 12 W and 8 W LED lamps (650 nm and 450 nm LEDs); 3 – monochrome lighting with blue LEDs; 4 – monochrome lighting with red LEDs; 5 – monochrome lighting with white LEDs. Natural lighting (length of daylight during the experiment –  $9.5 \pm 1.25$  hours): 6 – solar lighting; ● – *Chlorella vulgaris*, ● – *Desmodesmus armatus*, ● – *Parachlorella kessleri*, ● – abiotic cathode and anode



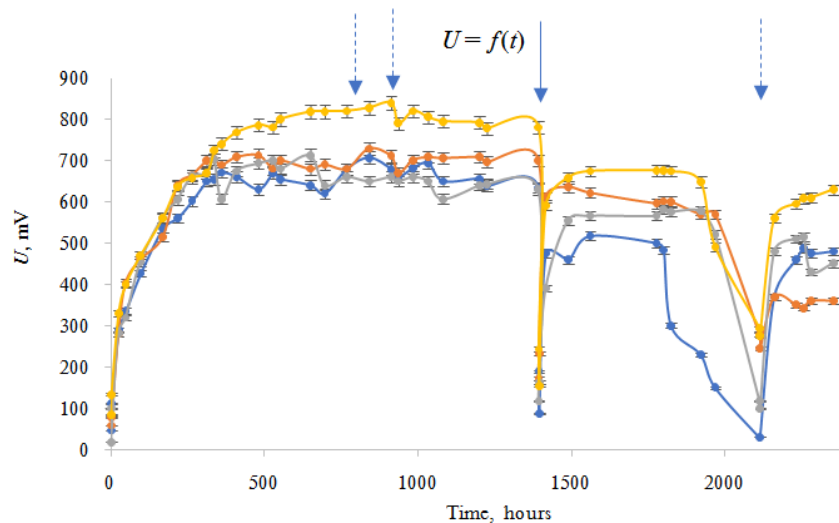
**Figure 2:** Change in the microbial fuel cell voltage over time. Arrows indicate the addition of  $\text{CH}_3\text{COONa}$  (1 g/L) to the anode chamber. Types of artificial lighting (light:dark = 9:15): 1 – 12 W LED lamp (650 and 450 nm LEDs), 2 – 12 W and 8 W LED lamps (650 nm and 450 nm LEDs); 3 – monochrome lighting with blue LEDs; 4 – monochrome lighting with red LEDs; 5 – monochrome lighting with white LEDs. Natural lighting (length of daylight during the experiment –  $9.5 \pm 1.25$  hours): 6 – solar lighting; ● – *Chlorella vulgaris*, ● – *Desmodesmus armatus*, ● – *Parachlorella kessleri*, ● – abiotic cathode and anode

generation can occur not only with artificial illumination of the cathode chamber but also with solar illumination, we conducted a simultaneous addition of fermented residue after methanogenesis into the anode chamber and microalgae culture into the cathode chamber. In the control MFC, fermented residue after methanogenesis was added to the anode chamber, and no microalgae were introduced into the cathode chamber. After just 24 hours from start-up, the current for the MFC with *Chlorella vulgaris* was  $251 \pm 13 \mu\text{A}$ , for the MFC with *Desmodesmus armatus* –  $294 \pm 15 \mu\text{A}$ , for

the MFC with *Parachlorella kessleri* –  $253 \pm 13 \mu\text{A}$ , and for the control –  $270 \pm 14 \mu\text{A}$  (Fig. 3). Meanwhile, the voltage after the same duration for the MFC with *Chlorella vulgaris* was  $290 \pm 15 \text{ mV}$ , for the MFC with *Desmodesmus armatus* –  $280 \pm 14 \text{ mV}$ , for the MFC with *Parachlorella kessleri* –  $280 \pm 14 \text{ mV}$ , and for the control –  $330 \pm 16 \text{ mV}$  (Fig. 4). The maximum current was observed at 168 hours, with  $450 \pm 22 \mu\text{A}$  in the MFC with *Chlorella vulgaris*, at 216 hours with  $470 \pm 23 \mu\text{A}$  in the MFC with *Parachlorella kessleri*, at 264 hours with  $520 \pm 26 \mu\text{A}$  in the MFC with *Desmodesmus armatus*, and at



**Figure 3:** Change in the microbial fuel cell current over time. Dotted arrows indicate the addition of  $\text{CH}_3\text{COONa}$  (1 g/L) to the anode chamber; solid arrow indicates the complete replacement of the anolyte with the addition of  $\text{CH}_3\text{COONa}$  (1 g/L) to the anode chamber. The control contains Tamiya medium without the addition of microalgae culture; —●— *Chlorella vulgaris*, —■— *Desmodesmus armatus*, —▲— *Parachlorella kessleri*, —◆— control



**Figure 4:** Change in the microbial fuel cell voltage over time. Dotted arrows indicate the addition of  $\text{CH}_3\text{COONa}$  (1 g/L) to the anode chamber; solid arrow indicates the complete replacement of the anolyte with the addition of  $\text{CH}_3\text{COONa}$  (1 g/L) to the anode chamber. The control contains Tamiya medium without the addition of microalgae culture; —●— *Chlorella vulgaris*, —■— *Desmodesmus armatus*, —▲— *Parachlorella kessleri*, —◆— control

408 hours with  $480 \pm 24 \mu\text{A}$  in the control (Fig. 3). The current gradually decreases. The addition of new portions of sodium acetate at 768, 912, and 2114 hours contributes to a slight re-increase in the current. A complete replacement of the anolyte (with the preservation of the anodic biofilm on the anode) with the simultaneous addition of a new portion of sodium acetate at 1393 hours leads to a repeated increase in the current. In contrast, after 500 hours during the further study, the current does not increase to peak values, and there is a tendency toward a gradual decrease in current.

The maximum voltage was observed at 840 hours in the MFC with *Chlorella vulgaris* ( $707 \pm 35 \text{ mV}$ ) and in the MFC with *Desmodesmus armatus* ( $729 \pm 36 \text{ mV}$ ). For the MFC with *Parachlorella kessleri*, the maximum voltage was observed at 648 hours ( $713 \pm 36 \text{ mV}$ ), and at 912 hours ( $840 \pm 42 \text{ mV}$ ) for the control (Fig. 4). Unlike the current, the voltage remained relatively constant. A rapid decrease in voltage was observed at 1394 hours (58th day). A complete replacement of the anolyte led to a rapid rise in voltage again. However, the obtained voltage after that was compared to the voltage before  $-72 \pm 4\%$  in the MFC with *Chlorella vulgaris*,  $91 \pm 5\%$  in the MFC with *Desmodesmus armatus*,  $88 \pm 4\%$  in the MFC with *Parachlorella kessleri*, and  $84 \pm 4\%$  in the control. The repeated addition of  $\text{CH}_3\text{COONa}$ , following another rapid voltage drop, yielded similar results (see Fig. 4).

Throughout the study, the optical density of the catholyte increased in all MFCs with algae (Fig. 5). The optical density increases in the catholyte with *Chlorella vulgaris* by 8.3 times, with *Desmodesmus armatus* by 18.5 times, and with *Parachlorella kessleri* by 23.3 times compared to the initial

optical density values of  $0.049 \pm 0.005$ . An increase in the growth rate was observed after 648 hours (27 days). During this period, a gradual decrease in the current in the MFCs was noted (see Fig. 3), while the voltage in the MFCs maintained relatively constant values.

### Discussion

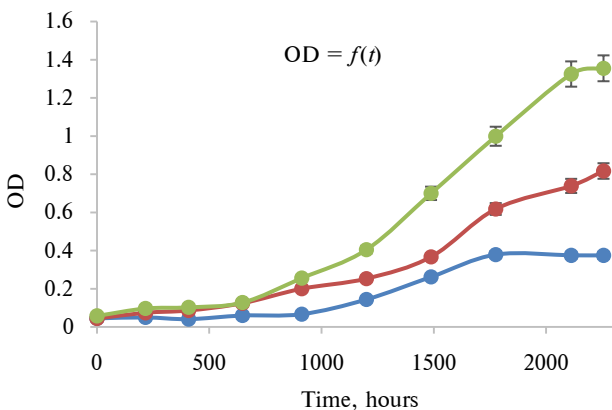
The obtained results indicate the feasibility of using microalgae in MFCs with various light sources. Algae have demonstrated their capability to supply the cathode with oxygen [25]. Furthermore, through photosynthesis, microalgae assimilate carbon dioxide, converting it into biomass. This was evident in our study as an increase in the optical density of the catholyte. The carbon dioxide introduced into the catholyte results from gas exchange with the atmosphere, meaning its quantity is limited by the rate of dissolution of carbon dioxide from the air into the aqueous solution.

The standard redox potential for the  $\text{CO}_2/\text{acetate}$  pair is  $-0.28 \text{ V}$ , and for the  $\text{O}_2/\text{H}_2\text{O}$  pair, it is  $+0.82 \text{ V}$  [26]. Theoretically, the MFC voltage of  $1.1 \text{ V}$  is possible. However, observed voltages higher than  $0.9 \text{ V}$  were not achieved, likely due to losses in electrolytes and electrodes. Additionally, reactions associated with the recovery of anions present in the nutrient medium and algae metabolism by-products may occur.

In the first experiment, the replacement of the catholyte, which contained  $\text{K}_3[\text{Fe}(\text{CN})_6]$ , with Tamiya medium containing microalgae did not induce significant changes in current and voltage. This suggests that the pre-grown biofilm on the anode plays a predominant role in current and voltage production. Microalgae, in this context, can effectively substitute for the abiotic cathode.

In the second experiment, the sustained ability to generate current, even with a complete replacement of the anolyte at 1393 hours, suggests that the exoelectrogenic anode biofilm, rather than the microorganisms present in the anolyte, plays a pivotal role in the generation of both MFC current and voltage.

Despite the need for periodic sodium acetate addition, MFCs demonstrated remarkable long-term viability throughout the study's 95-day duration. A gradual decrease in both current and voltage is observed in MFC research, even when employing an abiotic cathode with a comparable substrate and inoculum [15]. This suggests a gradual overgrowth of the anode, posing challenges for mass exchange



**Figure 5:** Change in the optical density (OD) of the catholyte over time: — *Chlorella vulgaris*, — *Desmodesmus armatus*, — *Parachlorella kessleri*

and electron transport to the anode. This overgrowth contributes to an increased internal resistance in MFCs, consequently reducing the current at constant voltage [10].

Microalgae in the catholyte play a crucial role in sustaining current and voltage generation. Compared to the control (without microalgae), the MFC with microalgae exhibited a 13–15% lower maximum voltage. The maximum current was 6% lower for the MFC with *Chlorella vulgaris* and 2% lower for the MFC with *Parachlorella kessleri*, while the MFC with *Desmodesmus armatus* showed an 8% higher maximum current compared to the control. Notably, the MFC with *Parachlorella kessleri* demonstrated a significantly faster increase in biomass, as evidenced by optical density. This rapid growth may induce self-shading in the microalgae culture [27], potentially impacting the efficiency of photosynthesis. The adverse effect on photosynthesis results in a reduction of oxygen release, the terminal electron acceptor in these MFCs. This phenomenon may account for the lower current and voltage observed in the MFC with *Parachlorella kessleri* compared to the MFC with *Desmodesmus armatus*. Conversely, the slow growth in optical density in the MFC catholyte from *Chlorella vulgaris* may also diminish the efficiency of photosynthesis. Reaching the optical density plateau at 1776 hours (Fig. 5) coincides with a sharp decline in current and voltage (see Figs. 3, 4) in the MFC with *Chlorella vulgaris*. The output of optical density on the plateau suggests an equilibrium between cell division and cell death rates. It is plausible that the accumulation of dead cells may result in the release of compounds from them that could adversely affect the current and voltage of MFCs, either impacting other microalgae or directly influencing the cathode.

The voltage in the control MFC (without microalgae) was observed to be higher than in the MFC with microalgae, potentially due to the occurrence of photorespiration alongside photosynthetic oxygen release in the microalgae. However,

a greater current was observed for the MFC with *Desmodesmus armatus*. This can be attributed to the formation of a biofilm by algae on the cathode, which reduces the internal resistance of the MFC, thereby increasing the current [10]. The gradual decrease in current and voltage during the study can be attributed to the overgrowth of the anode, complicating mass transfer within the biofilm.

Considering the observed growth of microalgae biomass during cultivation, this biomass holds promise for the production of biodiesel and carotenoids [28]. Additionally, it opens avenues for the production of value-added products such as lutein, violaxanthin, astaxanthin, and canthaxanthin. The accumulation of xanthophylls in microalgae induces stressful conditions in MFCs, providing protection of microalgae against oxidation [29].

Bazdar *et al.* [9] reported that under constant illumination with a 15 W fluorescent lamp, the voltage of MFCs ranged from 465 to 544 mV. Meanwhile, Don *et al.* [10] found that the maximum voltage of the MFCs with algae reached 340–375 mV under fluorescent lamp illumination. In our study, the maximum voltage observed was 707–729 mV under sunlight.

The Table presents a comparison of power density in electric current in MFCs with other studies. These findings highlight that utilizing sunlight as a light source for algae in MFCs allows for the attainment of relatively high power density values. These results suggest that sunlight could serve as a viable light source for MFCs. Sivakumar *et al.* [30] emphasized that the MFC with algae could be employed for long-term, low-maintenance power production, especially in applications such as wastewater treatment and living solar cells. Notably, MFCs eliminate the need for toxic and costly catalysts, reducing the risk of pollution. In our study, MFCs demonstrated the potential for long-term use (study duration: 95 days). However, it's observed that both voltage and current decrease over time, necessitating the periodic addition of sodium acetate.

**Table:** Comparison of power density generated in microbial fuel cells with algae

Cell type	Light source	Algae species on biocathodes	Power density, W/m <sup>2</sup>	Reference
Two chambers	Sunlight	<i>Chlorella vulgaris</i>	28.2	This study
Two chambers	Sunlight	<i>Parachlorella kessleri</i>	35.8	This study
Two chambers	Sunlight	<i>Desmodesmus armatus</i>	29.1	This study
Two chambers	Fluorescent lamp	<i>Chlorella vulgaris</i>	13.5	[25]
Two chambers	Fluorescent lamp	<i>Chlorella vulgaris</i>	21.0	[31]
Two chambers	Fluorescent lamp	<i>Chlorella vulgaris</i>	5.6	[32]
Two chambers	Sunlight	Mixed microalgal	57.0	[33]
Two chambers	Fluorescent lamp	<i>Desmodesmus</i> sp.	64.2	[34]

## Conclusions

MFCs equipped with a biocathode and bio-anode exhibit robust performance over an extended duration under various light sources. In the presence of a pre-grown anodic biofilm, both MFC current and voltage exhibit remarkable stability even when the light source is altered. Another promising approach involves initiating MFCs with the simultaneous addition of microalgae to the cathode chamber and fermented residue after methanogenesis to the anode chamber. This strategy proves beneficial in reducing the preparatory stages of the MFC technology. However, it's noteworthy that, in cases where the inoculum is added during the study, there is a gradual decline in current, although the voltage remains relatively constant. The potential utilization of solar lighting broadens the applicability of the MFC with microalgae, eliminating the need for additional expenses associated with artificial light sources. The maximum voltage readings were noted in the MFC with *Chlorella vulgaris* ( $707 \pm 35$  mV), *Desmodesmus armatus* ( $729 \pm 36$  mV), and *Parachlorella kessleri* ( $713 \pm 36$  mV). These values were 13–15% lower compared to the control.

The maximum current was 2–6% lower than the control ( $480 \pm 24$   $\mu$ A) for the MFC with *Chlorella vulgaris* and *Parachlorella kessleri*, and 8% higher than the control for the MFC with *Desmodesmus armatus*. The increased biomass of microalgae can be harnessed to produce value-added products. The complete replacement of the anolyte while preserving the anodic biofilm demonstrates that the anodic biofilm plays a key role in the generation of current and voltage.

## Interests disclosure

The authors have no conflicts of interest to declare.

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#### ГЕНЕРУВАННЯ ЕЛЕКТРИЧНОЇ ЕНЕРГІЇ МІКРОБНИМИ ПАЛИВНИМИ ЕЛЕМЕНТАМИ З МІКРОВОДОРОСТЯМИ НА КАТОДІ

**Проблематика.** Можливість перетворення органічних речовин на електричну енергію в мікробних паливних елементах (МПЕ) робить МПЕ екологічно чистою технологією. Однак використання платини або гексаціанофератів може збільшити витрати або ж призвести до вторинного забруднення довкілля. Використання мікроводоростей у катодній камері є перспективним для вирішення цих проблем.

**Мета.** Встановлення залежності генерації електричної енергії та ефективності застосування певного виду водоростей від типу та режиму освітлення.

**Методика реалізації.** Використано двокамерний МПЕ Н-типу із сольовим містком. Як інокулянт в анодній камері використано ферментований залишок після метаногенезу, а як інокулянт у катодній камері – культури мікроводоростей *Chlorella vulgaris*, *Desmodesmus armatus* і *Parachlorella kessleri*.

**Результати.** МПЕ з мікроводоростями демонструють здатність до генерації струму за використання різних джерел освітлення. Максимальні значення напруги для МПЕ з анодною біоплівкою та з мікроводоростями в катодній камері на 13–15 % менші порівняно з МПЕ з абіотичним катодом ( $840 \pm 42$  мВ). Максимальні значення сили струму на 2–6 % менші за контроль ( $480 \pm 24$  мкА) для МПЕ з *Chlorella vulgaris* і для МПЕ з *Parachlorella kessleri* та на 8 % більші для МПЕ з *Desmodesmus armatus* порівняно з МПЕ з абіотичним катодом. МПЕ з мікроводоростями здатні протягом тривалого часу генерувати електричну енергію.

**Висновки.** З попередньо нарощеною анодною біоплівкою сила струму та напруга залишаються відносно незмінними при зміні джерела освітлення. Можливість використання сонячного освітлення розширює можливість використання МПЕ з мікроводоростями, оскільки не потребує додаткових витрат, пов'язаних із застосуванням штучних джерел освітлення.

**Ключові слова:** мікробний паливний елемент; біоплівка; біоанод; біокатод; мікроводорості.