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Use of Wastewater and Electrogenic Bacteria to Generate Eco-Friendly Electricity through Microbial Fuel Cells

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Abstract: Power generation and wastewater treatment are two great challenges for sustainable development. Microbial fuel cells (MFCs) are a sustainable alternative that can generate bioelectricity in the bioremediation process of wastewater. For this reason, the objective of this research was to generate bioelectricity through double-chamber microbial-combustion cell systems from wastewater from the Covicorti Wastewater Treatment Plant (PTARC) in the anodic chamber and electrogenic bacteria such as *Stenotrophomonas maltophilia*, *Acinetobacter bereziniae*, and *Achromobacteria xylosoxidans* in the cathode chamber, respectively. Measurements of the voltage, current, power density, current density, and optical density of the bacteria and biochemical oxygen demand (BOD) were made. In addition, a metagenomic analysis of the wastewater sample was performed. It was shown that the MFC with *A. xylosoxidans* generated the highest voltage peak (1.01 ± 0.06 V) on day 24, while the MFC with *S. maltophilia* generated the highest current value (0.71 ± 0.02 mA). The pH levels were slightly alkaline, and the maximum anodic conductivity value was presented by the MFC with *A. cerevisiae*, with a peak value of 81 ± 2 mS/cm on day 24. On the other hand, a maximum power density and current density of 195,493 ± 4717 mW/m^2 and 4987 A/cm^2, respectively, were obtained in the MFC with *A. xylosoxidans*. Finally, the metagenomic analysis identified the predominant phyla of Proteobacteria present in wastewater samples capable of generating electrical energy as *Bacillota*, *Pseudomonadota*, *Bacteroidota*, *Actinomyketone*, and *Campylobacterota*.

Keywords: microbial fuel cell; wastewater; bioelectricity; electrogenic bacteria

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1. Introduction

Currently, one of the latent problems is the increase in energy demand [1]; in 2021, a rise of 4.5%, equivalent to more than 1000 TWh [2], was reported. One of the primary sources of energy generation is fossil fuels, which generate air pollution [3]. Their use intensifies the generation of greenhouse gases such as carbon dioxide, contributing to problems such as global warming [4]. Moreover, fossil fuels are a nonrenewable resource, so the reserves of this fuel are being depleted at an accelerated rate [5]. On the other hand, both municipal and industrial wastewater still present challenges worldwide, contributing to the water crisis and environmental pollution because they contain substances highly toxic to any form of life and, in some cases, usually need pretreatment [6–8]. This leads to the search for sustainable and energy-efficient treatments [6].

To face this problem, new-solution alternatives have emerged, such as microbial fuel cells (MFCs). MFCs are considered an emerging new technology, capable of using a number of organic fuel sources, such as wastewater, to generate electricity by oxidizing substrates using microorganisms [9–11]. These systems consist of essential elements for their operation, such as the anode, in which microorganisms oxidize organic matter releasing electrons that are transported to an external cable. Another important element within MFCs is the cathode, where electron acceptors react with electrons and protons to create reduced molecular compounds. On the other hand, the electron transport system, fuels, and microorganisms play an essential role in the performance of these systems as well [12,13].

MFCs have various configurations; among those known are double-chamber MFCs, composed of two perplexed chambers (anode and cathode) separated and joined by a cation-exchange membrane. Similarly, single-chamber MFCs are another configuration presented as a cube with two electrodes located at opposite ends. Their design is straightforward, so they require fewer materials for their elaboration compared to other MFC designs [14]. Within this context, it is worth mentioning that the electricity generation in these bioelectrochemical systems is influenced by various factors, including the substrates used, considering their type and concentration in the anodic section [15]. The use of wastewater as fuel has been reported because it contains organic substrates that can be easily degraded, so they are a good source of energy generation. In addition, within wastewater can be found various electrochemically active microorganisms, so they can be used directly as the inoculum in an MFC [16].

There is a wide variety of electrogenic microorganisms capable of generating electricity that can be used in MFCs, such as bacteria, archaea, and fungi. These electroactive microorganisms can oxidize the substrates and then transfer them to the anode by three different mechanisms [17,18]. First, electrons can be transferred to the anode through a soluble mediator in the solution that bathes the electrode. Secondly, electrons can be transferred directly to the anode via proteins found in the bacterium's outer membrane. On the other hand, bacteria form a thick film on the cathode, so the pili or nanowires may transmit the electrons to the anode [19]. Microorganisms such as *Geobacter*, *Shewanella*, *Pseudomonas*, and *Rhodoferax* have been extensively studied [17]. These microorganisms form biofilms at the anode, and based on the type of substrate, different microbial communities are formed. Zhao et al. [20] conducted a study based on sequencing the 16S rRNA gene. They found that an anodic biofilm had a complex microbial composition distributed in *Bacteroidetes* (39.4%), *Firmicutes* (20.1%), *Proteobacteria* (11.5%), *Euryarchaota* (3.1%), *Deferribacteres* (1.3%), *Spirochaetes* (1.0%), *Chloroflexi* (0.7%), *Actinobacteria* (0.5%), and others (22.4%). Rojas et al. (2022) demonstrated electricity generation from onion residues, generating maximum peaks of 4.459 ± 0.0608 mA and 0.991 ± 0.02 V of current and voltage, respectively. The species they identified at the molecular level were *Pseudomona aeruginosa*, *Acinetobacter bereziniae, Stenotrophomonas maltophilia,* and *Yarrowia lipolytica* adhered to the anode electrode [21]. Meanwhile, Bose et al. (2019) identified six species of exoelectrogenic bacteria that degrade organic matter in wastewater. The first species detected, *Achromobacter xylosoxidans*, a form of betaproteobacteria, can generate bioelectricity and decompose high concentrations of chromium and organic acids [22]. The selection of the bacteria *S. maltophilia*, *A. bereziniae*, and *A. xylosoxidans* is beneficial because as they are bacteria isolated from the natural sediment of wastewater, they are adapted to survive and metabolize the organic matter present. *S. maltophilia* is highly resistant to heavy metals and antibiotics and can degrade a wide range of compounds, including contaminants [23]. Its morphological characteristics, such as the fact that it has polar flagella, give it the ability to adhere and form biofilms on different surfaces, such as cathodes [24]. On the other hand, a plasmid in Acinetobacter strains has been demonstrated, which confers resistance to antibiotics. Heavy metals such as arsenic, added to their chromosome-encoded metabolic potential, ensure their survival in environmental stress conditions, making them ideal to be used in bioremediation processes as their electrogenic potential has been evaluated [25]. Likewise, it was shown that *Achromobacter xylosoxidans* can be applied in bioremediation and wastewater treatment, especially in detoxifying catechol waste in pulp and paper industries and other compounds [26].

Regarding the background, Rossi et al. (2022) developed an MFC on a pilot scale using air cathodes for the production of electricity and domestic wastewater treatment. In their study, an 850 L capacity MFC was used, and the residual water was processed during a hydraulic retention time of 12 h through a sequence of 17 brush anode modules and 16 cathode modules; later, the effluent was treated in a biofilter. It is worth mentioning that the monitoring was carried out for six months to evaluate the electrochemical and wastewater treatment performance. The results showed that the energy produced was of an average of 0.46 $\pm$ 0.35 W and a current of 1.54 $\pm$ 0.90 A with a Coulombic efficiency of 9%; on the other hand, in terms of wastewater treatment, up to 91 $\pm$ 6% of the COD and 91% of the BOD5 were eliminated, as well as certain bacteria (*E. coli*, 98.9%; fecal coliforms, 99.1%). It was concluded that these systems possess the capacity to generate electricity and produce electricity effectively [27]. Likewise, Yang et al. (2021) investigated electricity production in an MFC without a membrane using watermelon peel and wastewater containing nitrites; the results showed a maximum voltage production with an internal circulation of nitrite in a feeding mode. By batches of 294 mV, in terms of microorganisms, the genus Lactobacillus predominated in the cathodic biofilm, while in the anodic biofilm, it was the genus Clostridium and Bacteroides. Thus, microorganisms, watermelon-rind residues, and wastewater contributed to generating electricity [28]. Within this context, it is worth mentioning that the presence of sugars such as glucose in wastewater sludge improves the conductivity property of MFCs [29]; in this sense, many species of microorganisms can produce currents in an MFC using glucose as a substrate [30], which serves as a carbon source [31]. This research has an impact because it offers a clean and sustainable energy alternative to the demand for fossil fuels that cause greenhouse gases. At the same time, wastewater accumulates and flows into the sea, representing a risk to public and environmental health.

Two global concerns, the energy crisis and environmental pollution, can be addressed using MFC technology. It provides additional value to organic substrates such as wastewater generated in large cities, and these are mostly not adequately treated. In this way, this research used wastewater from a wastewater treatment plant to produce bioelectricity through a double-chamber MFC and, likewise, to improve the performance of MFC electrogenic bacteria (*S. maltophilia*, *A. bereziniae*, and *A. xylosoxidans*). In the current literature, the use of microorganisms as biocatalysts on different types of MFC substrates is being immediately investigated; for this reason, this research has the potential to observe how selected electrogenic bacteria can improve the performance of these electronic devices, thus finding MFC systems with improved performance so that they may soon be scaled to more commercial sizes, providing more sustainable electrical values at the level of cost and electricity production.

2. Materials and Methods

2.1. Sample Collection

The wastewater samples were collected from the Covicorti Wastewater Treatment Plant (PTARC) in Trujillo, Peru. Different sampling points were randomly located, and wastewater with floating sludge was collected per sampling point [32]. Subsequently, the samples were placed in 5 hermetic flasks of 1L, previously labeled, taking into account the following data: place, time, and date. Then, the samples were transported in a cold chain at 4 °C to the Science and Technology Research Institute of the César Vallejo University laboratory for the respective tests.

2.2. Dual-Chamber MFC Design and Operation

The MFC was designed according to the Huarachi-Olivera et al. (2018) [33] model with some modifications. The double-chamber MFC consisted of two 500 mL glass containers, whose anodic and cathodic chambers had a volume of approximately 500 mL. Both vessels were joined by a 10 cm long tube containing the potassium-hydroxide-based proton-exchange membrane (PEM) and agar in a volume of 15 mL, which was sealed with liquid silicone. The electrodes used were copper (Cu) and zinc (Zn) since this type of material shows better resistance and long-term performance, see Figure 1. Subsequently, the wastewater was placed in the anodic chamber. At the same time, the pure cultures of *Stenotrophomonas maltophilia* (10% of the volume) were inoculated in the cathodic chamber with a minimum-salt medium. The same procedure was followed for the pure cultures of *Acinetobacter cerevisiae* and *Achromobacter xylosoxidans*. Three cells were used for each bacterium (nine cells in total) and three cells as control.

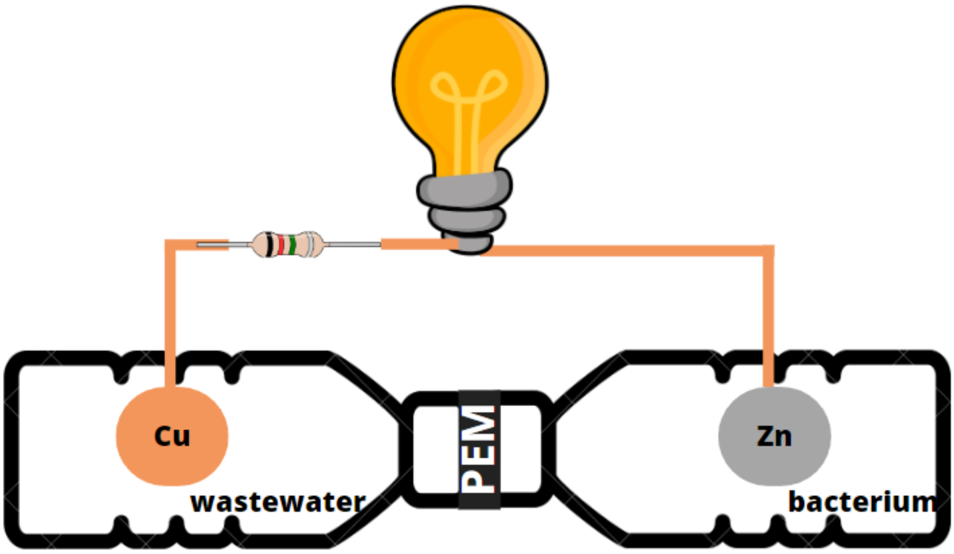

Figure 1. Schematization of MFCs.

2.3. Bioelectrochemical Analysis

The following analyses were carried out: a multiparameter (Prasek Premium PR-85, Lima, Peru) determined the voltage and electric current, while the current density and power density were determined via the method used by Rojas-Flores et al. (2023) [34]; the biochemical oxygen demand (BOD-Trak II Apparatus) and the optical density of electrogenic bacteria were determined by reading the absorbance in a spectrophotometer (Jenway, Liverpool, UK) for 30 days.

2.4. Obtaining Electrogenic Strains

The strains *S. maltophilia*, *A. bereziniae*, and *A. xylosoxidans* were donated by the Institute for Research in Science and Technology of the César Vallejo University, Trujillo, Peru, and were identified by molecular techniques; the identity percentages being 99.93%, 99.93%, and 99.32%, respectively.

2.5. Reactivation of Electrogenic Strains

A pure-culture inoculum of *S. maltophilia* was taken and seeded in BHI Broth (Brain Heart Infusion) and incubated at 35 °C for 24 h. Then, it was seeded in nutrient agar and incubated at 35 °C for 24 h, to obtain isolated colonies which were stained Gram and subsequently stored in tubes with sloping nutrient agar [35]. The same procedure was performed for *A. cerevisiae* and *A. xylosoxidans*.

2.6. Inocula of the Electrogenic Strains in the Cathode Chamber

From a pure culture of *S. maltophilia*, a suspension was made in 50 mL of BHI Broth, incubated at 35 °C for 24 h, then adjusted with nephelometer No. 1 (3×10^8 CFU/mL). Subsequently, the inoculum was added in 450 mL of minimum-salt medium to the cathodic chamber. In the same way, the inocula of *A. cerevisiae* and *A. xylosoxidans* were prepared in triplicate.

2.7. Metagenomic Analysis of Wastewater from the PTARC

Under sterile conditions, a sample of residual water was collected from an oxidation pond of the PTARC in Trujillo, Peru. The sample was transferred in a cold chain to Ecobiotech Lab S.A.C for its subsequent analysis. Genomic DNA was extracted with the E.Z.N.A.® Soil DNA kit (Omega Bio-Tek Inc., Norcross, GA, USA), following the manufacturer's recommendations. Subsequently, the DNA was quantified and verified in a microvolume spectrophotometer (EzDrop1000 Blue-Ray Biotech) and stored at −30 °C. Subsequently, aliquots of extracted DNA were analyzed at MR DNA (Molecular Research LP, Shallowater, TX, USA) in the USA by the genetic sequencing of bacterial diversity (16S rRNA sequencing) using the bTEFAP® Illumina Diversity Assay technology. Diversity and abundance percentages were obtained using Mothur and Excel Professional Plus 2019 software [36].

2.8. Statistical Analysis

During the statistical treatment, the data obtained for voltage, electric current, power, current density, and power density presented a normal distribution, so the mean difference was analyzed via a test of ANOVA using SPSS Software version 22.0.

3. Results and Discussion

Figure 2a shows the voltage values obtained from the monitoring carried out, where the voltage values increase from the first day, with the cell with *A. xylosoxidans* generating the highest of the voltage peaks (1.01 ± 0 06 V) on day 24, while the cell with *A. bereziniae* managed a peak voltage on the fourth day (0.92 ± 0.051 V) and the cell with *S. maltophilia* generated its peak voltage on day 27 (0.87 ± 0.081 V); after reaching their maximum peak, all the cells showed a decrease in their values until the last day of monitoring. The use of biocatalysts managed to increase the efficiency of the microbial fuel cells; due to their intrinsic properties, the use of microorganisms boosts these types of devices, specifically in biofilm formation [37,38]. On the other hand, Figure 2b shows the values of the electrical current of the MFCs, where the cell with *S. maltophilia* (0.71 ± 0.02 mA) offers the highest of the current discounts, followed by the cells of *A. bereziniae* (0.23 ± 0.03 mA) and *A. xylosoxidans* (0.15 ± 0.029 mA), whose values decreased until the last day of monitoring. The increase in electrical values is mainly due to the fact that the bacteria begin to proliferate in the MFCs; the growth depends on each of the microorganisms, and the time it takes to reach its highest point is due to the intensity of the chemical reactions that occur inside the cells achieving the formation of the biofilm [39,40]. Meanwhile, the decrease in the current and voltage values is due to the sedimentation of the microorganisms in the final stage. The electrodes used are an essential factor since being made of copper, they have a negative effect on the performance of the MFCs in the final stage. However, in the final stage, they are also one of the leading causes of the high values, because by using metallic electrodes, it helps the passage of electrons from one chamber to the other by almost not offering resistance and generating a greater flow of electrons [41–43].

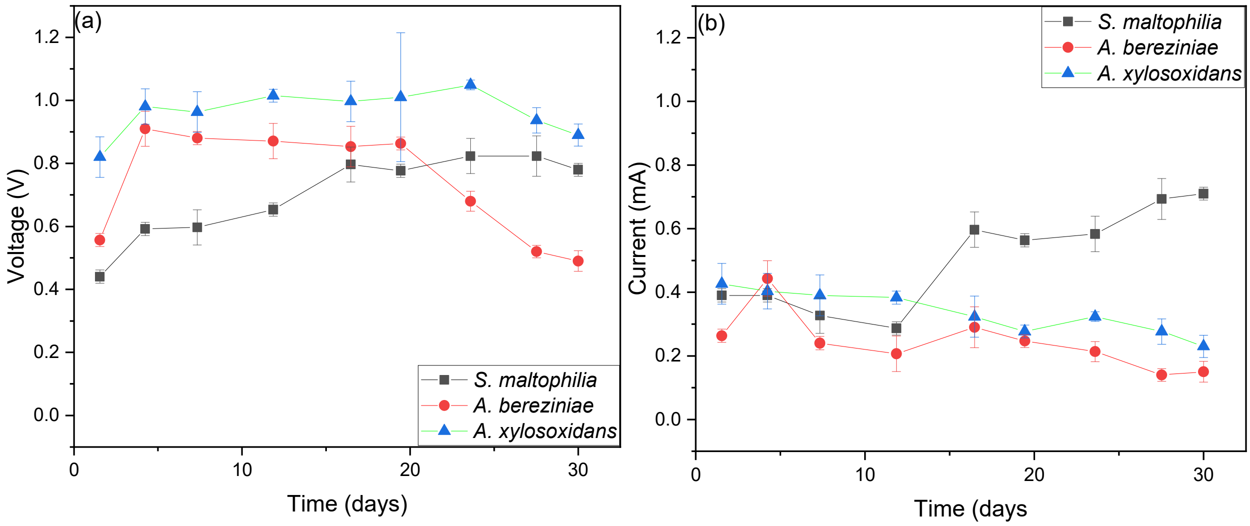

Figure 2. Values of (**a**) voltage and (**b**) electrical current obtained during the monitoring of the MFCs.

Figure 3a shows the average pH values of the anode chambers of each MFC, which were slightly alkaline, where it can be seen that the pH (7.3) remained almost constant during the operation of the MFCs (30 days). MFC-1 started with an average pH of 7.12 ± 0.03 and increased to 7.36 ± 0.09, with a peak of 7.53. On the other hand, MFC-2 began with a pH of 7.17 ± 0.03 and reached a peak of 8.03 ± 0.14, before falling slightly on day 30 to 7.20 ± 0.04. Finally, MFC-3 began with a pH of 7.12 ± 0.01 and slightly increased to 7.63 ± 0.03 on the last day of evaluation. The anode pH values of all the MFCs were somewhat alkaline, ranging between 7 and 8. The slightly alkaline pH is because the protons cross the salt bridge toward the cathode chamber, where it will reduce oxygen [44]. On the other hand, these values are mainly required for the optimal growth of bacteria and biofilm formation on the anode [44,45]. A relationship of the average pH values (Figure 3a) with the intermediate voltages (Figure 2a) can be observed in the three bioelectrochemical systems. This demonstrates the critical role of the anode pH in the metabolic activity of microorganisms and, therefore, bioelectricity generation. Similarly, two studies show similar slightly alkaline pH values when wastewater generates electricity in MFCs [46,47].

Figure 3b shows the turbidity values. It can be seen that the MFC with *S. maltophilia* generates a higher turbidity peak, with a value of 66.44 ± 8.41 UNT on day 16, followed by the MFCs with *A. bereziniae* and *A. xylosoxidans* on days 16 and 12, respectively. The values continuously decreased until the last day in all MFCs; in all cases, there was a decrease in turbidity. In MFC-1, it decreased from 41.25 ± 3.07 to 14.86 ± 7.08 UNT. Similarly, in MFC-2, the initial turbidity value of 22.76 ± 0.57 UNT slightly reduced to 19.54 ± 1.91 UNT. The turbidity of the anode chamber in MFC-3 decreased from 14.06 ± 0.03 to 10.83 ± 0.35 UNT. MFC-1 and MFC-3 present a considerable increase in turbidity, with mean values of 66.44 ± 48.34 UNT and 43.45 ± 15.66 UNT, respectively, whereas the increase in turbidity of wastewater in MFC-2 is slight. The turbidity of wastewater is due to the solids suspended in it and is a quality parameter that must be measured regularly [48,49]. For this reason, the reduction in the turbidity of the residual water in an MFC was investigated, finding that in all three cases, it was considerably reduced. This is possibly due to the retention time of the MFC wastewater and the biological degradation of the colloidal particles, as pointed out in the study by Mardanpour et al. [50].

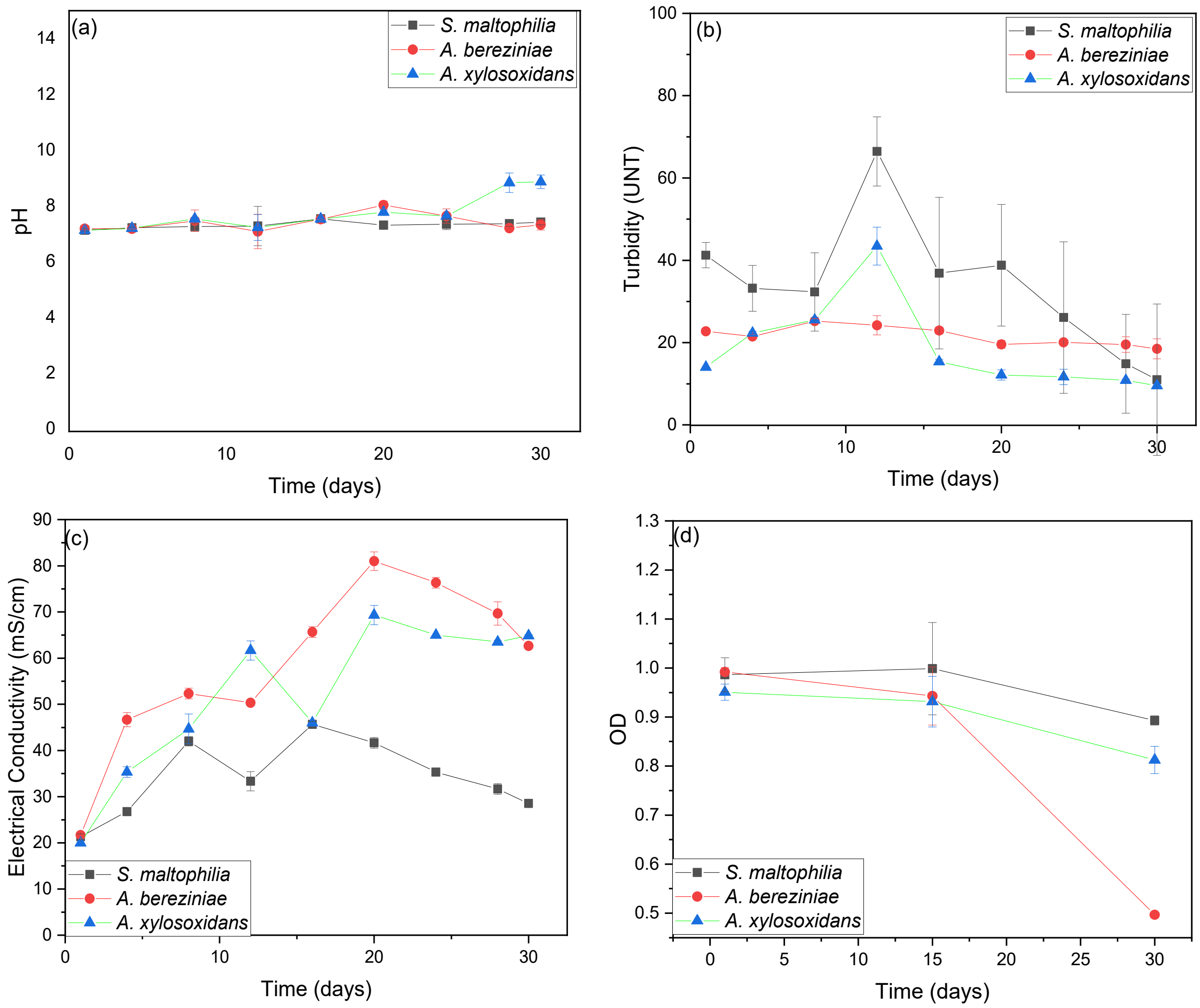

Figure 3. Values of (**a**) pH, (**b**) turbidity, (**c**) electrical conductivity, and (**d**) optical density obtained during the monitoring of the MFCs.

Figure 3c shows the values of the electrical conductivity of the substrate used in the MFCs, where it is observed that the values increase up to a specific time and then begin to decrease. Thus, the MFC with *A. bereziniae* obtained a peak conductivity value of 81 ± 2 mS/cm on day 24, while in the MFC with *A. xylosoxidans*, the maximum peak was reached on day 20 with a value of 69.33 ± 2.08 mS/cm. Finally, in the MFC with *S. maltophilia*, it was possible to generate a maximum value on day 16 (45.67 mS/cm); after reaching their maximum values, the values decreased until the last day. The conductivity values obtained in the three MFCs are because the wastewater presents a great variety of salt content, such as the chloride ion, which comes from the human diet [51,52]. Possibly, the drop in conductivity is due to the precipitation of chlorides due to the retention time. Figure 3d shows the optical density (OD) values measured in three parts of the monitoring, observing a decrease from the beginning. The MFC with *S. maltophilia* retained the highest value until the end of the monitoring (with a value of 0.893 ± 0.009), followed by the MFCs with *A. xylosoxidans* (0.814 ± 0.027) and *A. bereziniae* (0.497 ± 0.049). In this sense, the downward trend in the growth curve of the bacteria is attributed to the reduction in the concentration of nutrients over time and the subsequent increase in acid and toxic substances in the medium, leading to the logarithmic phase of death [53,54].

Figure 4 shows the values of current density (DC) and power density (PD). The PD values were 162.02 ± 5.314, 139.723 ± 7.84, and 195.493 ± 4.717 mW/m^2 in the DC values of 4.291, 5.025, and 4.987 A/cm^2 with a peak voltage of 876.166 ± 10.648, 826.689 ± 8. 647, and 952.332 ± 6.754 mV for the MFCs with a substrate of *S. maltophilia*, *A. bereziniae*, and *A. xylosoxidans*, respectively. The results obtained show that the MFC with *A. xylosoxidans* managed to generate the maximum power density peak of 195,493 ± 4717 mW/m^2 at a current density of 4987 A/cm^2. In this context, it is worth mentioning that Zafar et al. [55] reported the use of MFCs in the generation of electricity, and their results showed an increase in power density from the enrichment of various bacterial species, predominantly the species *S. maltophilia*, results similar to those shown in this study. Therefore, this increase is attributed to the formation of biofilms in the MFCs over time. Likewise, the power density is closely related to other factors, such as the degradation rate of organic substrates, the resistance of the device circuit, as well as external operating conditions [56]. On the other hand, as can be seen in the graphs, the current density reaches a maximum point and subsequently presents a tendency to decrease due to the increase in the operating time of the MFCs [57].

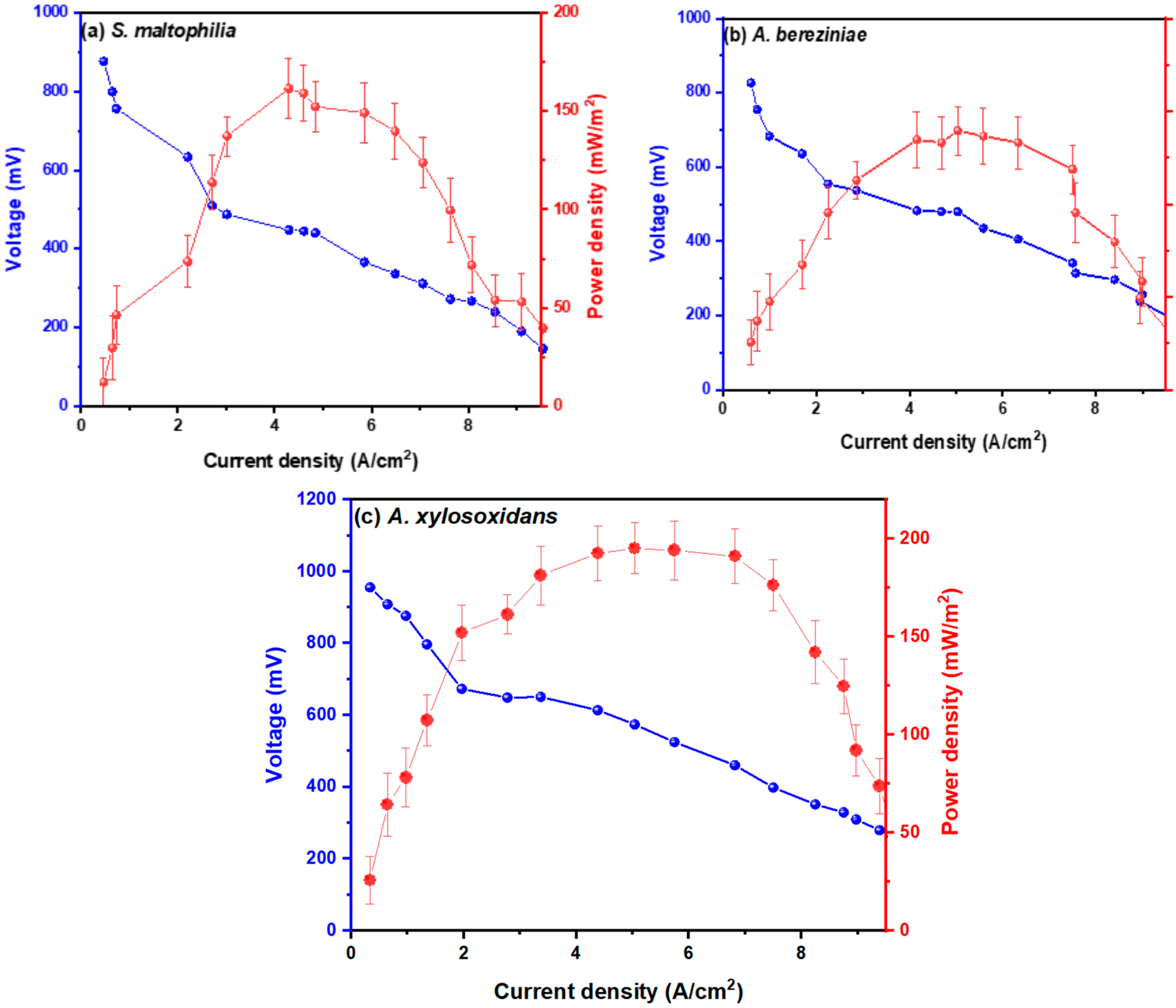

Figure 4. Graphs of the power density and current density in the MFC systems with electrogenic bacteria in the cathode chamber: (**a**) *S. maltophilia*, (**b**) *A. bereziniae*, and (**c**) *A. xylosoxidans*.

Table 1 shows the BOD values in samples of residual water contained in the anode chamber of the three MFC systems with electrogenic bacteria *S. maltophilia*, *A. bereziniae*, and *A. xylosoxidans*, respectively, for 30 days. They presented an initial value of 640 mg O_2/L. These values decreased notably in the MFCs with *A. bereziniae* and *A. xylosoxidans* systems, reaching final values of 3 mg O_2/L, respectively, on day 30. Akinwumi et al. (2022) demonstrated that the generation of bioelectricity, as well as the decrease in COD and BOD, was highly influenced by the pH and the presence of fruit waste. They evaluated MFCs at different pH values (4.5–7.5) and fruit waste (pineapple, orange, and mango) residues as co-substrates, which were subjected to bioelectrochemical remediation for 168 days. Their results found that the highest open circuit voltage (820 mV) and maximum power density (994.78 mW/m^2) correlated with the highest Coulombic efficiency (32.79%) for COD or the 64.75% efficiency removal of BOD, COD (96.28%) and BOD (98.25%). The Nernst–Monod and bioelectrochemical kinetic models developed by Nernst–Monod–Moser adequately described the bioelectrochemical kinetics, whereas the COD and BOD removal kinetics followed first-order and zero-order kinetics [58]. Meanwhile, Bose et al. 2023 evaluated bioelectricity generation and pollution removal over several cycles. According to their results, the MFCs developed an open circuit voltage of 870 ± 20 mV and a power density of 563 ± 30 mW/m^2 with external resistors, with a current density of 0.79 mA/m^2. The chemical contamination of the wastewater was reduced from 1201 ± 60 mg/L to 240 ± 31 mg/L with an average removal of 81%, with a BOD removal efficiency of 86%. The performance of the system was stable. This is attributed to the absence of the carboxylic acid functional group on the cathode surface and the sufficiently developed bacterial biofilm on the anode [59,60].

Table 1. Biochemical oxygen demand (BOD) values obtained during the monitoring of the MFCs.

	Day 1	**Day 15**	**Day 30**
System *	DBO (mg O_2/L)	DBO (mg O_2/L)	DBO (mg O_2/L)
MFC-1	*640*	*65*	5
MFC-2	*640*	*20*	3
MFC-3	*640*	*20*	3

* MFC-1: *S. maltophilia*, MFC-2: *A. bereziniae*, MFC-3: *A. xylosoxidans*.

Several studies have identified a wide diversity of microbial communities in wastewater that could influence the microbial ecology of ecosystems surrounding urban areas and even contaminate agricultural areas and harm people's health [61–63]. Based on metagenomic analysis, the existence of complex microbial communities was revealed in the domestic wastewater sample collected from the PTARC of the city of Trujillo. A total of 17 bacterial phyla, 75 families, 148 genera, and 254 bacterial species was found (Figure 5). The predominant phyla were *Bacillota*, *Pseudomonadota*, *Bacteroidota*, *Actinomycetota*, and *Campylobacterota*, very common in domestic wastewater whose main composition is based on the microbiota of human and animal waste, vegetables, food, and soil [64]. Similar studies show that the phyla *Bacillota* and *Bacteroidota* are very common in animal fecal samples, while *Actinomycetota* predominates in domestic wastewater treatment plants and hospital effluents [62,65,66]. Likewise, Chu et al. (2018) indicate that *Proteobacteria*, *Bacteroidota*, and *Firmicutes* are dominant in wastewater from a treatment plant in Wisconsin, USA. Considering the relative abundance of bacteria by class, it was observed that *Clostridia*, *Bacilli*, *Bacterioidia*, *Gammaproteobacteria*, *Negativicutes*, *Coriobacteria*, *Erysipelotrichia*, and *Betaproteribacteria* are the most abundant [67]. These results agree with the results obtained by Jankowski et al. (2022) in studies of the composition of metagenomic communities in a wastewater treatment plant in Manitoba, Canada [68]. On the other hand, the presence of pathogenic and opportunistic pathogenic bacterial species of humans (*Escherichia coli*, *Klebsiella* sp., *Pseudomonas aeruginosa*, *Stenotrophomonas maltophilia*, *Aeromonas* sp., *Campylobacter coli*, *Enterococcus faecalis*, *Burkholderia* sp., and *Prevotella* sp.) was observed, with relative abundances lower than 1%; these are values that are similar to those obtained by Yasir (2020), with the exception of *P. aeruginosa* which had a value above 1% [62]. It should be noted that Logan et al. 2006 mention that MFCs are a promising technology for wastewater treatment, such as in power generation. Being the bacterial communities that grow in these devices, Proteobacteria predominate in the sediments up to communities made up of α, β Proteobacteria and Firmicutes. These bacteria are capable of transferring electrons that are used to generate electrical energy [69]. Figure 6 shows the circuit of the electrical system generated by MFC systems with wastewater at the anode and electrogenic bacteria at the cathode, generating a value of 8.58 V, which managed to turn on a 6-watt LED bulb.

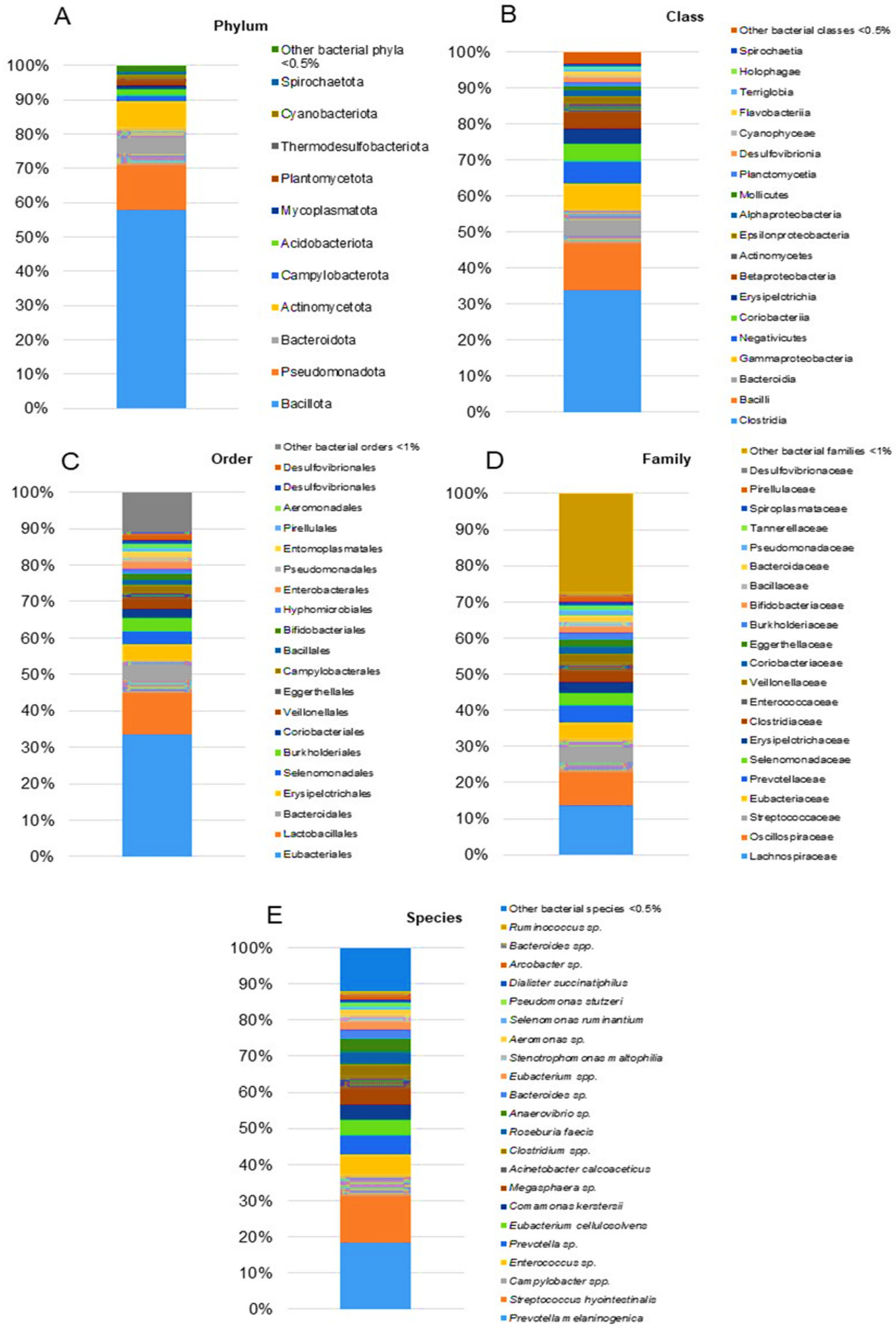

Figure 5. Taxonomy of bacterial communities in a sample of domestic wastewater from the WWTP-Covicorti: (**A**) percentage abundance of phyla, (**B**) class, (**C**) order, (**D**) family, and (**E**) relatively dominant species. The X axis shows the taxa, and the Y axis indicates the abundance in percentage.

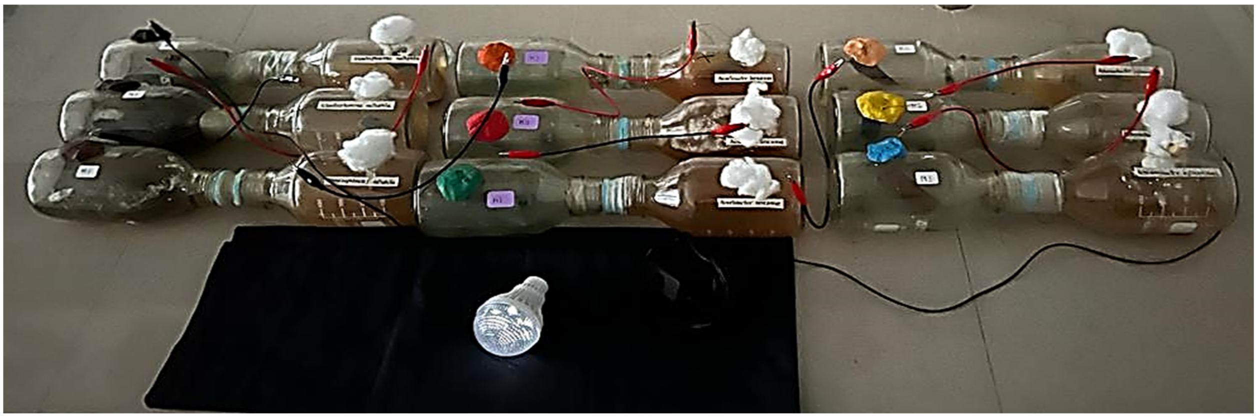

Figure 6. Circuit of MFCs connected to a 6-watt LED bulb.

4. Conclusions

Electrical power was generated by laboratory-scale double-chamber MFCs using wastewater in the anode chamber and electrogenic bacteria such as *S. maltophilia*, *A. bereziniae*, and *A. xylosoxidans*. The bioelectrochemical system that generated the highest voltage was MFC-3 (*A. xylosoxidans*), where 1.01 ± 0.06 V was generated. Regarding current, the highest value (0.71 ± 0.02 mA) was developed in the MFC-1 system (*S. maltophilia*). The operating pH of the three systems was slightly alkaline throughout the evaluation time. The maximum conductivity (81 ± 2 mS/cm) occurred in the MFC-2 system (*A. bereziniae*). The cell where *A. xylosoxidans* was used as a substrate managed to generate the maximum power density value of 195,493 ± 4717 mW/m^2 at a current density of 4987 A/cm^2. The metagenomic analyses showed that the predominant species of the wastewater in the anode chamber were Bacillota, Pseudomonadota, Bacteroidota, Actinomycetota, and Campylobacterota, which can contribute to the transfer of electrons to the cathode plate and therefore to the generation of bioelectricity. Finally, through a series system of 9 MFCs, a maximum voltage of 8.58 V was generated, which managed to light a 6-watt LED bulb for more than 30 days. These results are encouraging in terms of the sustainable generation of electricity using WWTP wastewater as fuel, as well as being eco-friendly since they generate polluting compounds.

The results observed in the investigation reveal that *A. xylosoxidans* and *S. maltophilia* are the microorganisms with the greatest potential to obtain higher values of voltage and electric current than that based on the theory of power that an electrical system can have, and by connecting their MFCs in series, we could improve the output power of the electronic device. Thus, in future work, we can create an electrical device capable of storing this energy so that it can be consumed or applied to some need. This research provides a great contribution in the area of biocatalysts in MFCs since two microorganisms capable of generating considerable electric values for future applications were identified. With this technology, a medium-scale generator could be built for devices that do not require high amounts of electrical energy, such as cell phones, etc. The limitation is that it works on a laboratory scale; the challenge is to carry it out on a larger scale through a pilot plant.

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