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The study of the performance of a microbial fuel cell: a progress towards the improvement of low electrical bioenergy output by using an amplification system

Mohammed Benghernit · Mostefa Kameche · Fatima Zohra Zerhouni ·
Fatima Zohra Krim · Tewfik Sahraoui · Christophe Innocent

Abstract

Objective A microbial fuel cell (MFC) has been conceived and constructed for the treatment of the sheep manure wastes and their conversion into clean sustainable renewable energy. The aim of the present investigation was to examine the performance of this bioelectrochemical device, in breaking down the organic matter (pollutant removal) and simultaneously producing electricity. Furthermore, the objective was to enhance the low electric energy by using an adequate amplification system.

Results So, the chemical oxygen demand (COD) removal was increased by 58.7% with the MFC running for 10 days. However, this technology faces practical barriers as it produces low electrical energy. A power management system was therefore elaborated in this respect. It included the MFC, operational amplifier (OA), solar photovoltaic panel and a boost DC/DC converter. The low voltage output obtained was thus increased substantially using the OA prior to its polarization by the solar photovoltaic module. The amplified voltage was sufficiently enough and in consequence, utilized to feed a light emitting diode. The low output voltage 0.5 V was simply harvested, successfully boosted up to approximately 2 V (i.e. 4 times higher) and finally harnessed as a power supply.

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Conclusions The MFCs association shows the positive stacking effect successfully, when the cells were connected in parallel. This novel application is very interesting to utilize the natural bioenergy contained in wastes to supply small electronic devices.

Keywords Environment · Waste treatment · Microbial fuel cell · COD removal · Bioenergy · Power amplification

Abbreviations

MFC	Microbial fuel cell
DC	Direct current
LED	Light emitting diode
ATP	Adenosine triphosphate
tPMET	Trans plasma membrane electron transport
ETC	Electron transport chain
NADH	Nicotinamide adenine dinucleotide hydrogen
CG	Carbon graphite
OA	Operational amplifier
CEA	Common emitter amplifier
EA	Electro active
EPS	Extracellular polymeric substances
EET	Extracellular electron transfer
V _{OC}	Open circuit voltage
I _{SCC}	Short circuit current
MPP	Maximum power point
AC	Alternating current
RedOx	Reduction/oxidation
SCE	Saturated calomel electrode
GJ	Grape juice
COD	Chemical oxygen demand
SHE	Standard hydrogen electrode

Introduction

The botanist Michael Cresse Potter discovered in 1911 that an electrode of platinum placed in yeast or *Escherichia coli* cultures was able to produce an electromotive force or potential difference (Potter 1911). On this basis, the technology of biological fuel cells in particular microbial fuel cell has emerged. The fuel cell revolution started when a research group from Kings College in London (UK), has developed and demonstrated improved biological fuel cells using various microorganisms to enhance both the efficiency of electron transfer and the reaction rate using

mediator systems (Bennetto 1984; Allen et al. 1993). A (MFC) represents an eco-friendly approach to generating electricity while purifying wastewater concurrently, permitting chemical oxygen demand removal and producing electric power densities (Obileke et al. 2021). The system utilizes the metabolism power of bacteria for electricity generation, and application of microorganisms as available and inexpensive biocatalysts (Khajeh et al. 2020). The basic information and description of this novel technology are widely reviewed in the literature. However, scholars and researchers wish to know the problem of low power management and efforts deployed to increase it. Although there have been great advances in improving the performance of the MFC by using bacterial natural metabolisms (Adekunle et al. 2016), the low voltage output produced by such device seems to be the main drawback of this technology. For this reason, reliable system has been considered in order to make it beneficial on the industrial scale. There are thus two ways to undertake for increasing the low power output of a MFC: the modification of electrodes and the use of amplification/management systems.

Recently, it has been proven that commercial application of MFCs can be increased through optimization of microorganism and invention of novel electrodes, which provide a promising option for cost effective bioelectricity generation (Choudhury et al. 2017). Really, due to low electricity generation performance and high cost of operation, the electrodes of a single-chamber air-cathode MFC have been modified with graphene and polymer polyaniline and then their effects on its performance have been assessed positively. Indeed, the modified electrode displayed higher catalytic activity toward oxygen reduction compared with unmodified electrodes (more than 6 times higher) (Wang et al. 2018). Furthermore, Fan et al. studied the effects of the polymer polypyrrole/iron(III) oxide composite modified anode on the electricity generation performance (i.e. the steady-state current density) and the sewage treatment capacity performance (i.e. higher rate of chemical oxygen demand removal) (Fan et al. 2021). Khajeh et al. have also made an investigation on the efficient improvement of a MFC performance by the modification of graphite cathode via electrophoretic deposition of copper/zinc oxide nanoparticles. The MFC performance was evaluated with and without visible light irradiation. A maximum voltage was achieved by

the modified graphite electrode under the irradiation (Khajeh et al. 2020).

Although there have been a great deal of research towards the improvement of power outputs of MFCs, by modifying their electrodes and optimizing the electro active biofilms (biocatalysts), there still a drawback in using this technology in practical applications. For this reason, other researchers and scholar scientists have been focusing on the study the substantial increase of the output voltages of MFCs. Up to now, several MFCs were connected in series or in parallel to overcome the low voltage. Nevertheless, although a serially stacked MFCs unit could provide a higher voltage, it is ineffective due to voltage reversal, leading to a significant overall voltage decay (Kim et al. 2019). Moreover, much effort have been deployed to control the voltage reversal occurrence by connecting individual MFC units with a power point tracking system to charge a stacked polarized capacitor (Papaharalabos et al. 2017). This procedure allowed a maximum voltage which was fair enough to drive only low voltage electronic devices but not for real applications.

As a matter of fact, the power management unit (PMU) utilizing a combination of a DC/DC converter to boost the low MFC voltage to practical uses and a super capacitor to store electrical energy temporarily, has been proposed (Garita-Meza et al. 2018). As a result, a considerable boosted voltage was obtained from a wastewater single-chamber air-cathode MFC equipped with a low voltage booster (Koffi 2019). It was also proposed a new two-step “boost-and-multiply” system, in which the low output voltage was firstly boosted into an alternating current (AC) voltage by a transistor-based self-oscillating low voltage booster circuit. After, the boosted AC voltage was further multiplied and turned back into a direct current (DC) voltage by a multistage Single-Phase Cockroft-Walton voltage multiplier circuit. This newly designed low voltage booster multiplier was tested successfully and yield yielded higher voltage using a single-chamber air-cathode MFC treating domestic wastewater as a power source (Koffi et al. 2020). Specific converter topologies were also required to step-up in another manner the output voltage of a MFC using another PMU for operation at low input voltage and at very low power in a completely autonomous way to capture energy from MFCs with the highest possible efficiency (Khaled et al. 2016). The power

obtained was amply sufficient to supply a low-power temperature sensor.

To the best of our knowledge, there is no previous study on the amplification of the low voltage delivered by a MFC. In the present investigation, we have therefore fixed the goal to improve the performance of the MFC by amplifying its low output voltage in order to feed a Light Emitting Diode (LED). The amplification has been achieved with two electrical circuits by proposing the polarization of the operational with the conventional energy or that of solar radiation using photovoltaic panels. Furthermore, our PMU has been set up with a manner which quite different from that suggested recently by Koffi et al. without utilizing any alternating current (AC) (Koffi 2019). A one-chamber MFC was thus inoculated and powered with leachate of sheep manure wastes, rich of EA bacterial source, that promises a sustainable MFC system. In the beginning, we studied the parameters useful for MFC conception, i.e. electron-generation, EA bacterial biofilm, power generation, and cell configuration. Then, we described all materials used and experimental procedure for energy harvesting, in particular the amplification of low energy generation. Finally, we presented the exploitation of the low bioenergy produced by the MFC, to feed for example the low electric input device LED. This has been achieved successfully by proposing the suitable amplification scheme.

Theoretical background

MFC technology and bacterial medium

In the MFC technology, there are three main constituents: biocatalyst (bacterial medium), substrate (degradable organic matter) and electrodes (electrons carriers). All these three constituents allow treatment of the wastewaters with concomitant bioelectricity production. The biocatalyst of an MFC is generally the microorganisms, which adhere to the electrodes while performing the role of electro-catalyst. They come from a pure culture, mixed or a natural consortium. They are called anodophiles, exoelectrogens and electrochemically active bacteria (Lovely 2008).

As reviewed previously, the activity of bacterial growth in the sea sand/animal manure or sludge mixtures has been proved experimentally (El-Nahhal

et al. 2020). In effect, it was thus clearly reported that *Bacillus* species in such wastes, can be isolated from all manure types. Nearly three bacterial species were characterized in the animal manure. They were grown under aerobic conditions and actively participate in the production of electricity. The microbial community was therefore responsible for the transfer of electrons to electrodes through electron transport chain during the oxidation reduction reactions that took place in the bio electrochemical fuel cell. Moreover, it was revealed the important role of bacteria in harvesting electricity. After electricity generation, the manures had lower chemical oxygen demand due to consumption by bacterial community (Alkhamis et al. 2021). In addition, owing to its potential bioenergy production by underground burial system heated with cascade-controlled solar water heated system, the sheep manure was used as an indicator of biomass potential contribution to power generation in less fossil energy countries (Zhang et al. 2012). On this basis, the sheep manure was thus chosen by us in this study to test its potential electricity generation in a MFC for the production of energy and concomitant preservation of environment.

Organic substrate degradation using bacterial metabolisms

The fuel must be a biodegradable compound and not toxic to microorganisms. The first studies focused on organic compounds of low molecular weight such as glucose (Chauduri et al. 2003). Later, complex fuels were tested such as cellulose. Finally, studies have been developed on the possibility of using domestic or industrial waste (Liu et al. 2004). Complex industrial wastes are generally diluted to avoid undesirable planktonic metabolisms that may compete with the biofilm-catalyzed electricity generation and to decrease fouling on electrode and membrane surfaces as well (Cercado-Quezada et al. 2010a, b). As most organic substrates undergo combustion with the evolution of energy, the bio-catalyzed oxidation of organic substances carried out by means of microorganisms in the presence of O_2 at electrodes, permits the conversion of chemical energy into electric energy. In normal microbial catabolism, an organic substrate is oxidized initially without involvement of O_2 , while its electrons are taken up by an enzyme-active site, which acts as a reduced intermediate.

Electron transfer and bioelectric power generation

In the MFC, the catalytic microorganisms oxidize the organic matter to produce electrical energy. They shuttle the electrons exogenously (called exoelectrogens) to the electrode surface without utilizing artificial mediators. The transfer of electrons, whether direct or indirect, takes place at the end of a cascade of oxidation and reduction reactions, considered as the keys to the catabolic processes necessary for the survival of the bacteria. These processes are the natural metabolisms of fermentation and cellular respiration. These latter are used by the bacteria to break down the organic matter (waste) and extract energy from it which will be stored in the form of ATP considered as a universal source of energy.

Intact microorganisms have been used in the anode compartment of fuel cells in two distinct ways. The 'direct' microbial fuel cell depends on the transfer of electrons from the organism to the electrode, whereas the 'indirect' or 'product' cells utilize microorganisms to generate a fuel such as hydrogen, which is then used as a reductant in the electrochemical reaction at the anode. Such a 'product' cell using *Clostridium butyricum* has been described (Suzuki et al. 1980) but the performance of 'direct' cells (Lewis 1966; Austin 1967; Allen 1972) has been poor in terms of power production and the current densities obtained at the anode.

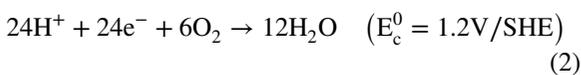
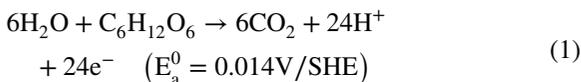
The biodegradation of the organic substrate has been achieved by using the electrocatalytic ability of the bacteria. The electric energy can thus be generated following the two extracellular electron transfer pathways which are the trans Plasma Membrane Electron Transport (tPMET) and Electron Transport Chain (ETC) (Schaetzle et al. 2008). Both pathways allow the generation of the electrons which go out through the bacterial cell. The electro catalytic oxidation of glucose can be performed thanks to NADH oxidation (Mardiana et al. 2015). Electrons are therefore produced and captured by the species under oxidized form. In anaerobic fermentation pathway, the recycling of NADH to NAD^+ is important to keep the glycolysis process continuous (Nelson 2005)]. In all cases, the process results in the generation of electrons, protons and carbon dioxide. In this way, the substrate is biodegraded and the electric power is produced. Furthermore, the microbial growth is supported by the energy derived from the electron

transfer process itself and results in a stable, long-term power production.

The degradation of the organic matter (grape juice) by the microbial metabolism (fermentation), in the absence of oxygen, produces on one hand the energy which is stocked by the bacteria in the form of ATP, and on the other hand the electric energy recovered by the final electron acceptors (bio anode). This latter gives rise to an electric current. However, the bacteria continue to degrade by oxidation process the organic matter contained in the polluted environment by reducing its COD (chemical oxygen demand) without really using an external substrate (glucose). Micro-organisms (bacteria, yeasts, etc.) can oxidize a wide variety of organic molecules (substrate), and thus produced electrons which are transformed into the energy useful for their growth and their metabolism.

Principle of voltage generation

The microbial generated electron can be harnessed by artificially introducing an adequate electrode (i.e. an electron acceptor) into the MFC. This allows the stimulation of the development of the potential difference between the bacterial membrane and the anode, which contributes to the elaboration of the bio anode potential that helps in the electron delivery. The electrons that are drawn to the anode induce positive potential of the cathode. The potential difference between the positive cathode and the negative bio anode, is called the electromotive force or the cell voltage. This potential drives the generated electrons from the anode to the cathode through the external circuit across the load, that can be harvested as electricity (i.e. the electric current flows in the opposite way from the cathode to the anode). For example, the enzymatic-catalyzed oxidation of glucose (fuel) on the anode yields the electric potential versus standard hydrogen electrode (SHE) according to the reaction (Eq. 1). Whilst the reduction of the oxidant at the cathode proceeds according to the reaction (Eq. 2) (Scott et al. 2012).



Experimental

Materials

A quantity of bulk sheep manure was collected directly from a sheep barn located in the town suburb farm. The manure samples were finely grinded and sieved to obtain very small particles in powder form. This latter was then mixed with potassium chloride solution (60 mM) to make the medium more conducting for ionic motion of species which were obtained by oxidation of the organic matter. The mixture was afterwards stirred for 24 h and then filtered through a paper filter. The obtained manure sheep leachate was finally obtained and put inside a plastic cup under room temperature. Three forms of the waste manure were prepared and tested in the present study: solid (compost), sludge and leachate. The bio anode formed in the manure compost showed efficient electro active properties with the grape juice (substrate) which readily oxidized in the presence of the biocatalyst (electroactive biofilm). It thus revealed a rich microbial flora with remarkable electro active ability, as it has been already reported in the literature (Parot et al. 2009).

Furthermore, the grape juice was consequently tested, which generally represent most favorable environmental condition for microorganisms. Diverse metabolites such as formate, succinate, lactate, acetate and propionate, are produced during glucose oxidation in MFC due to its fermentative nature but acetate has been identified as the dominant and most effectively utilized (Kim et al. 2011). As shown above (Eq. 1), when it is oxidized in the presence of the appropriate catalyst (chemical or biological), the substrate glucose main constituent of the grape juice, releases electrons, protons and carbon dioxide.

Methods

Set up of MFC

The one-compartment MFC reactor was used to generate electricity. It was fully filled with leachate of sheep manure wastes. The anode and cathode electrodes made of carbon graphite (CG) were inserted inside the chamber, which was tightly closed with plastic paper to prevent penetration of

oxygen and therefore to avoid unwanted oxidation of present species. As reported by Heilmann et al. (2006), Li et al. (2010), and Kook et al. (2021), the two electrodes were then connected with the nominal external resistance 1000 Ω , to allow the optimum power production, the natural polarization and in consequence the effective growth of the EA biofilm. While using the sheep manure as the inoculum waste in the MFC, El-Nahhal et al. described in details the mechanism of electricity generation (electrochemical potentials of both electrodes and biodegradation of organic matter), oxidation of inorganic molecules and electron transport chain reactions (El-Nahhal et al. 2020). The cell used in this study was operated in a batch mode with view to utilize it in future in the continuous mode for a real application. The generated voltage (V) was monitored using a digital voltmeter. The value of the electric current (I) was thus obtained by taking the ratio between the measured voltage and the discharged resistance (R). Moreover, in order to assess the performance of the MFC, the current density (mA/cm²) was calculated by dividing the electric current by the projected area. The fluctuation in the current density response can be explained by the change in the bacterial population distribution more fermentative or electro genic culture. Nevertheless, when the voltage of the MFC declined, 5 mL of commercial red grape juice of simple sugars concentration (i.e.113 g/L) was added to the leachate to feed the microorganisms which resulted in the generation of electrons and an increase in the current density. The MFC started functioning since last summer and it is still working up to now as long as the electrodes are preserved from corrosion and correctly connected to the discharge resistance.

Chemical oxygen demand (COD)

The MFC technology was used for the first time in 1991 for the treatment of wastewaters coming from different sectors (municipal, agricultural, farming, industrial, etc....) (Habermann et al. 1991). The COD is usually used as a measure of organic pollutants present in wastewater and it is expressed in (mg/L). The most common application of COD is the evaluation of the amount and level of ox disable pollutants found in the wastewater. It makes it possible to determine the concentration of organic matter in the

wastewater through the quantity of oxygen necessary for their total chemical oxidation. A COD analysis was thus performed by us at the end of the MFC experiment to make in evidence the conversion of the polluted organic matter into bioenergy. It was measured according to the ISO 15705 standard (ISO 15705 2002) based on the closed tube small scale method. The tubes were heated at 148 °C for 2 h. Then, the samples were left to cool down for 1 h and diluted with distilled water. After that, both the blank (distilled water) and the experimental sample were analyzed with Mohr's salt solution (0.1 N) by titration using the colored indicator (Ferroine). Then, the COD values (mg L⁻¹) were calculated by using Eq. 3 and subsequently, the corresponding COD removals were, therefore tabulated by using Eq. 4.

$$\text{COD (mg/L)} = 8000 \cdot \frac{(V_1 - V_2) \cdot N_{Ms}}{V_3} \quad (3)$$

$$\text{COD (\%)} = 100 \cdot \frac{(\text{COD}_i - \text{COD}_f)}{\text{COD}_i} \quad (4)$$

where V_1 —volume (mL) of Mohr's salt solution (titrating solution) used for the blank test with distilled water. V_2 —volume (mL) of Mohr's salt solution (titrating solution). V_3 —volume (3 mL) of the sample (titrated solution). N_{Ms} —normality of Mohr's salt solution (0.1 N). COD_i and COD_f —initial and final COD values respectively.

Amplification of low voltage output

In a previous work carried out by Ridvan Umaz on the power management system of the low power obtained from MFCs (Umaz et al. 2020), the proposed system consisted of a fronted converter, a super capacitor, a charge control circuit and a second converter. The maximum power extraction from a MFC was achieved successfully in the fronted converter. In our present investigation, we followed the same principle but we undertook on another procedure for harvesting the maximum output voltage by using an amplifying system polarized with a solar photovoltaic panel. Although the MFC could produce renewable energy from wastes, the generated power was practically unusable. Hence, we used two amplifiers:

operational amplifier and common emitter amplifier. This allowed to extract the usable electrical power.

Operational amplifier (OA)

An operational amplifier is a high gain differential electronic device. It is used to greatly amplify an electric potential difference present at its inputs. Initially, the amplifier was designed to perform mathematical operations in analog calculators: it easily implemented basic mathematical operations such as addition, subtraction, integration, derivation and others. Subsequently, it was used in other applications such as the control of electric motors, voltage regulation, current sources or even oscillators.

We have a positive input signal which becomes negative at output. The gain (G) of the assembly is fixed by the value of the resistors. In effect, in the mesh 1, $V_{in} = R_1 I_1 + \varepsilon$. So for ε small enough, we get $V_{in} = R_1 I_1$. The same reasoning applies for the mesh 2, but this time with a negative sign due to the inversion between the input and the output in this amplification system. Indeed, $V_{out} = (-R_2 I_2)$. This results in the gain which is the ratio between the two voltages. For example, for $R_1 = 100 \Omega$ and $R_2 = 3000 \Omega$, the gain is equal to (-30) . The negative sign highlights the amplification reversal system between the input and the output.

Common emitter amplifier (CEA)

A common emitter amplifier assembly is one of the three elementary configurations for amplifying a bipolar transistor. The other two are called common base or common collector. In this assembly, the emitter (indicated by an arrow) is connected to the common ground or to a reference voltage, while the base of the transistor is connected to the input and the collector to the output load. The analog circuit using a field effect transistor is called a common source amplifier circuit.

The common-emitter amplifier circuit consists of a C_e decoupling capacitor which is chosen to have a low enough impedance, when studied in small signals, to short-circuit the resistor R_e . The presence of C_e makes it possible to significantly increase the gain of the assembly, but on the other hand the circuit has lower input and output impedances. The emitter resistor R_e therefore makes it possible to

create a feedback called degeneration of the emitter, which ensures good characteristics of stability and linearity of the circuit, in particular in response to temperature variations. In the case where C_e is absent, the impedance of R_e reduces the overall trans conductance $G_m = g_m$ of the circuit by a factor $(g_m \cdot R_e + 1)$. Thus the gain in tension is expressed by:

An electronic amplifier is used to amplify AC signals with a very large amplification gain. However, in the microbial fuel cell, it can be applied to increase the low voltages, which are variable over time, in order to use them to power small electronic devices such as LED. The active components used in electronic amplifiers, in particular the transistor, make it possible to control their output current as a function of an electrical quantity (current or voltage), the image of the signal to be amplified. The output current of these active components is taken directly from the amplifier power supply. However, it can be, replaced by a mixed association (series/ parallel) of a set of MFCs. Depending on how they are installed in the amplifier; the active components thus make it possible to increase the voltage and/or the current of the input electrical signal.

Direct current converter

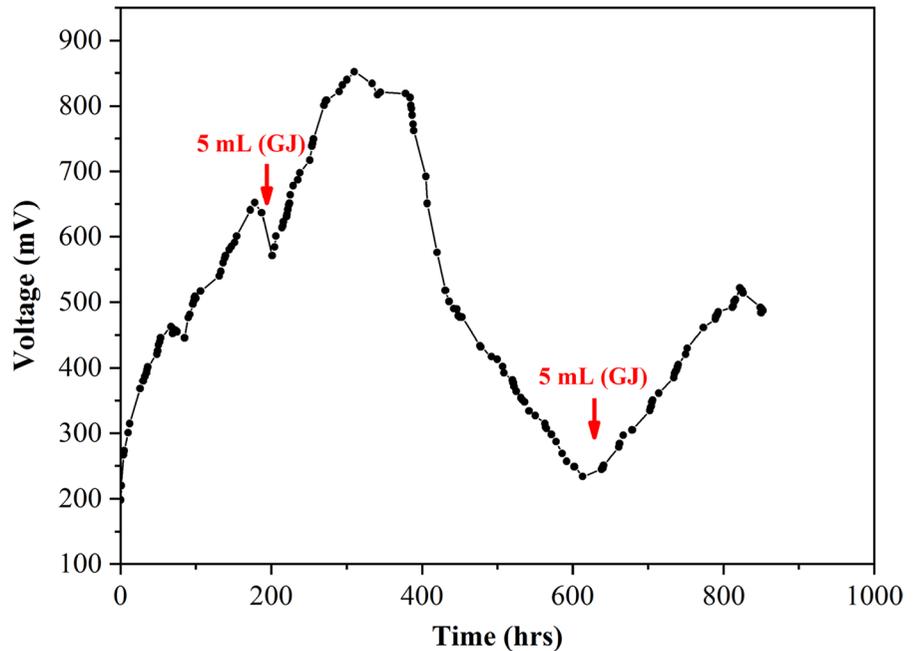
As it is commonly well known, the voltage harvested from the MFC is relatively low (less than 0.5 V) and requires thus an electric management system to step-up the low voltage to acceptable voltage values (i.e. > 1.0 V). This management system contains in general, a charge pump or a direct current (DC) converter to boost the voltage and capacitors to store the energy (Donovan et al. 2011).

Results and discussions

Evolution of MFC voltage in open circuit

The CG electrode was inoculated with leachate of sheep manure waste to build a bio anode. This latter was set up as soon as the MFC started. At the beginning, most of bacteria were still present in planktonic state in the solution. Overtime, the bacteria start to colonize on the anode surface and form

Fig. 1 Evolution of the open circuit voltage of the sheep manure microbial fuel cell (MFC) during 870 h



a biofilm. The colony grows larger and therefore the biofilm becomes visible. With a pure strain such as “*Escherichia coli*”, the performance is much better (Mukhtar et al. 1914). However, the system is very expensive. Moreover, as the MFC is discharged to an electric load (resistance), the self-developed current contributes to the EA biofilm development and consequently the fuel cell is thus polarized naturally. Previous studies reported the formation of the EA biofilm, once the bio anode was polarized by applying a constant stimulating potential (Ojima et al. 2020).

Figure 1 shows a plot of the voltage generation of the MFC versus time. The generated voltage expresses the potential difference between the CG bioanode and the stainless steel plate cathode both dipped in the same chamber containing the leachate of the sheep manure wastes. In the beginning, the observed voltage is very low because the potential difference between the bare CG anode and the cathode is narrow. Along with the biofilm growth, we observe an increase in voltage generation and stability of the MFC (Kosimaningrum 2018). As a result, the potential difference between the bio anode and the cathode becomes wider, mainly due to the formation of EA biofilm on the anode surface. Biofilm attachment involves extracellular polymeric substances

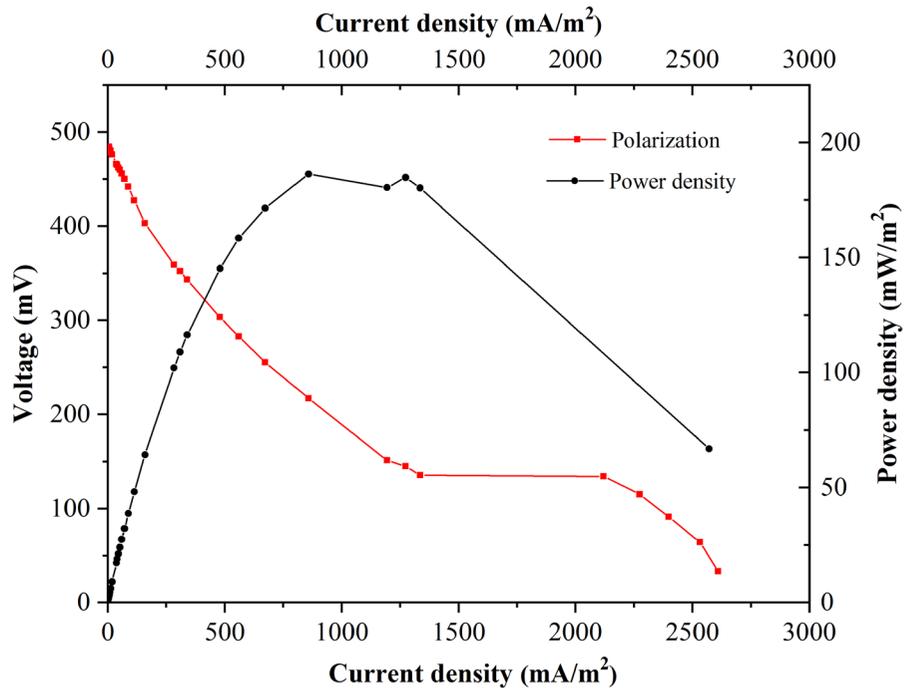
(EPS) that can facilitate electron transfer from the bacterial cell to the anode (Zhang 2008). These substances surround the cell by acting as transient media in the extracellular electron transfer (EET) process. Xiao et al. suggested the electron hopping to be the molecular mechanism of the (EET) through (EPS) (Zhang et al. 2014).

As soon as the EA biofilm was built up and agglomerated around the anode, it catalyzed the oxidation of the organic matter in the leachate inside the MFC chamber; thus, the voltage increased steadily and reached the value 650 mV. Then, it declined as the substrate was consumed around the bio anode. However, upon the addition of substrate (5 mL of grape juice), the voltage of the MFC rose up again drastically and attained its highest value i.e. 850 mV. Regardless the efficiency of the MFC, after a long working period, the voltage decreased naturally, because of the depletion of the substrate and the nutrient in the leachate, leading to lower activity of the bacterial consortium.

Polarization and power curves

The polarization represents the variation of the voltage (V) versus the current density (mA/m^2) by using the projected area of electrode 3 cm^2 . As shown

Fig. 2 Polarization and power density curves during the MFC operation (obtained after discharging to different external electrical resistances)



in Fig. 2, the polarization curve displays regions expressing three types of loss: (1) activation loss due to activation polarization; (2) ohmic loss due to polarization; and (3) concentration loss. At low current density, the potential drops drastically, due to energy loss for oxidation and reduction initiations at anode and cathode respectively, as well as energy loss during electron transfer from the bacterial cell membrane to electrode surface.

At intermediate current density, the ohmic loss originates mainly from the higher resistance of ions travelling inside the leachate (low ionic conductivity), and the electrical connection between the two electrodes. However, in order to reduce the ohmic loss, one should reduce the electrodes spacing and increasing the ionic conductivity of the electrolyte. Whilst at higher current density, the cell performance falls sharply, owing to the mass transport limit of the reactants towards the electrode and subsequently, the products out of the electrode (Lin et al. 2015). The polarization curve was obtained when the cell was connected to an external variable electric resistance in the decreasing order from the highest resistance 10 M Ω down to lowest resistance 1 Ω , avoiding unwanted rapid discharge of the MFC. The voltage produced on the load, was measured with a voltmeter

of higher input resistance, in order to minimize any unwanted current flow in it. The resulting current was therefore calculated by using Ohm's law. As shown in the figure, the MFC presents an open circuit voltage (V_{OC}) of 484 mV (i.e. $R = \infty$) and a short circuit current (I_{sc}) of 2611 mA/cm² (i.e. $R = 0$).

Besides, the power production which was deduced from the polarization curve (i.e. $P = U \cdot I$), is one of the key parameters to determine its performance. The output power was determined from the measured MFC's voltage and the resulting delivered current. It determines the maximum power point (MPP) that can be supplied by the MFC at a specific electric resistance (Logan 2007). At this point, the load resistance can be deduced and should be placed permanently to yield the highest output. Adapting a MFC to the power of the respective load plays a crucial role in practical applications. Whenever possible, the power of the load should always be close to the MPP (Helio-centris GmbH 2000). Table 1 summarizes the most important values obtained with the MFC after having worked for 140 days.

Table 1 The characteristics values of the MFC

Power density at MPP (mW/m ²)	Current density at MPP (mA/m ²)	Potential at MPP (mV)	Electric resistance at MPP (Ω)	Open circuit potential (mV)	Potential ratio (%)	Energy released during 140 days (mJ/g)
184.84	274.76	145	900	484	29.95	0.019

Internal and external resistances of MFC

As suggested in the literature, in the first tests of our experiments, the MFC was often discharged to the nominal external resistance 1000 Ohms, to make the maximum power transfer from the microbial fuel cell. However, one could use another value of resistance, providing current flows in the external circuit. So, when connecting the MFC to this resistance, an electric current flowed and polarized naturally the bioanode by rendering the biofilm more electroactive. This is called the natural polarization. One can thus promote the bacterial adhesion and electroactive biofilm formation, where the electrons can be effectively transferred to the external circuit, and therefore allowing organic matter degradation. Recently, Kook et al. published a research paper in which they stated that the most suitable electrical external loads (resistors) < 1000 Ohms were found and may be appropriate for experiments to assess the optimum power production (Kook et al. 2021). However, the microbial anode can be formed under constant applied potential by electrochemical method. This can be carried out by using a three-electrode cell and a potentiostat. So, under the application of a suitable potential between the working electrode (W-bioanode) and the reference electrode (Ag/AgCl), a current can be recorded between the W and the auxiliary electrode (platinum wire). The resulting current versus time, is called the chronoamperometry (CA). Once the oxidation peaks appear repeatedly upon the addition of the substrate (organic matter), the anode with the biofilm became mature and therefore it can be used as the bio-anode to be readily used in the MFC. This method of potential under the application of potential, has been utilized recently by Oliot et al (2017) and previously by Cercado-Quezada et al. (2010a, b).

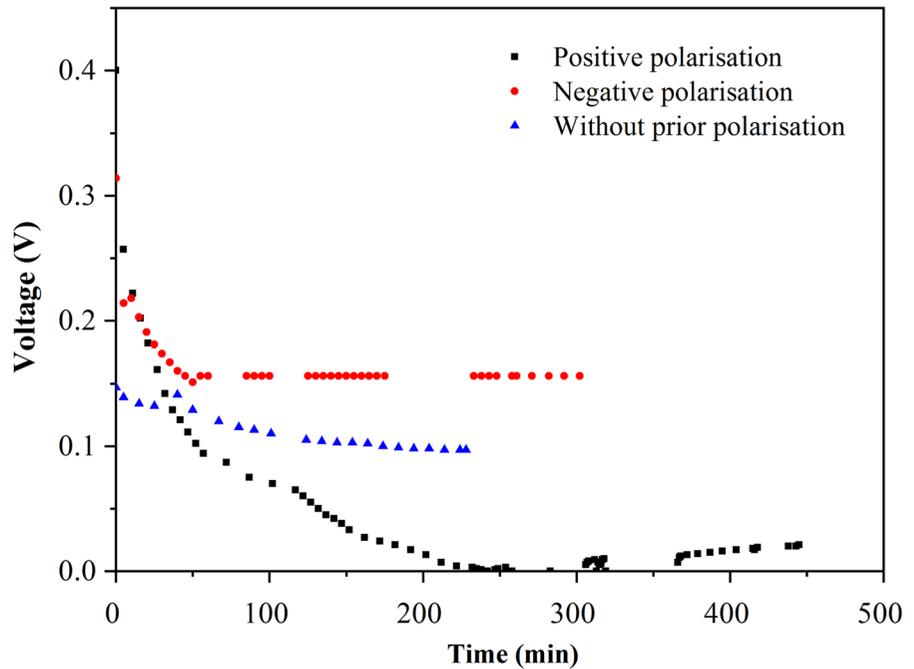
As a matter of fact, a rigorous analysis has been carried out in order to choose the appropriate resistance to be used during the course of the experiment. In effect, as depicted by Eq. 5 the MFC of

electromotive force (E_{emf}) and internal resistance (R_{int}) generates a voltage (E_{cell}) and an electric current (I_{cell}) when it is connecting to an external resistance (R_{ext}). Besides, Kamau et al., pointed out that the slope of the linear region of the polarization curve represents the internal resistance of the MFC (Kamau et al. 2017). This latter generates its maximum power when $R_{int} = R_{ext}$ (R_{ext} being the external resistance that corresponds to the maximum power). Hence, when we set $R_{int} = R_{ext}$ implies $E_{cell} = E_{emf}/2$. This value was found to be quite close to that corresponding to the Maximum Power Point (MPP) obtained from the power curve characteristic (i.e. power density versus current density).

$$E_{cell} = E_{emf} - R_{int} \cdot I_{cell} \quad (5)$$

In our case, from the polarization and power curves for the single MFC operating during 140 days (Fig. 2), the voltage of open circuit (VOC) and current of short circuit are 484 mV and 2571 mA/cm² respectively. Whilst, the power density (PDM), current density (CDM) and voltage (VM) at the MPP are 186 mW/m², 858 mA/m² and 216 mV respectively. Thence, by using Ohm's law and power formula (i.e. $U = R \cdot I$ and $P = U \cdot I$), we found the external resistance (R_{ext}) value 839.16 Ohms using the projected area of the electrode 3 cm². On the other hand, from the polarization curve, we determine the slope of the ohmic region which corresponds to the internal resistance (R_{int}) value 715.33 Ohms. These two values are quite close to each other, which suggests that the voltage of the MFC is approximately a half of its emf, when it is connecting to the external load mentioned above (i.e. 839.16 Ohms). So, if we consider the experimental error, we find that this value is approximately similar to that chosen arbitrary at the beginning to carry out the experiment (i.e. 1000 Ohms). Besides, the ratio E_{cell}/E_{emf} is 0.44 close to one half which proves that the MFC reaches its higher

Fig. 3 Evolution of the MFC discharged to external electric resistance 1000Ω upon positive and negative polarizations



performance and consequently the choice of that value, is quite judicious.

Applied potential to formation of EA biofilm on bio anode

The efficiency of the EA biofilm is characterized by the maximum current density of projected bio anode area and by the faradic yield, it achieves. In effect, an anodic EA biofilm performs well when it is capable of producing a high current density at the lowest possible potential. Thus, the choice of the potential to be imposed on the bio anode generally depends on two major objectives: the increase of the chance of obtaining an EA biofilm and the selection of the most effective one.

It has been described previously that in the same reactor, when microorganisms compete for the consumption of the organic substrate, the biofilm formed under negative polarization compared to the standard hydrogen electrode, made it possible to obtain better performance (Torres et al. 2009). In addition, it has also been shown that imposing a very negative potential for a long period allows the evolution of a pure culture of “*G. Sulfurreducens*” towards a more efficient strain with a density of current produced more than 5 times greater than that of the starting strain (Yi

et al. 2009). The authors showed thus that the negative potential acted as a selective pressure towards the evolution of a more efficient strain. On the other hand, it has been made in evidence that a low negative potential, despite a lower quantity of biomass produced, allowed maximum powers to be obtained much more quickly than higher potentials (Aelterman et al. 2008).

Thus, on this basis of these considerations, we have polarized the bio anode of our MFC with positive and negative potentials successively. The value of this polarizing potential was ± 0.6 V which is quite close to the RedOx potential of $\text{CO}_2/\text{Glucose}$ (i.e. -0.43 V/HSE); Glucose being the main constituent of the grape juice used by us as substrate to feed the existing bacteria in the sheep manure inoculum. As shown in Fig. 3, the evolution curve of the MFC discharged to the resistance 1000Ω , decreases sharply with time during the period of 8 h, reaching the lowest voltage with the MFC polarized positively. The voltage decreases from 400 mV down to less than 1 mV. In contrast, the voltage of the MFC polarized negatively, started with 300 mV and lowers as well but stabilizes readily to a constant value, which is greater (i.e. 155 mV). However, the voltage of the MFC without prior polarization, lies between.

During the positive polarization, as the current arrived to the bio anode, the electrons were pumped out from the EA biofilm and as a result, the voltage of the MFC dropped to lower values. In contrast, during the negative polarization, as the current left out the bio anode, the electrons were added massively to the EA biofilm, inducing an increase of the voltage. This result is in quite agreement with that reported by Ketep et al. while they studied the lowering of the applied potential during successive scratching/re-inoculation for the improvement of the performance of microbial anodes for microbial fuel cells. The microbial anodes were formed under polarization at -0.2 V/SCE on smooth graphite plate electrodes with paper mill effluents (Ketep et al. 2013). The effect of anode polarization on biofilm formation and electron transfer in “*Shewanella oneidensis*” /graphite felt microbial fuel cells, supports also the results obtained in the present study (Pinto et al. 2018).

COD removal

Amongst other applications, the MFC technology allows maximum COD removal and its conversion into bio-energy in a treatment plant. We have adopted this application in the present work, in order to establish the relationship between the production of the bioenergy and the depollution of the medium by assessing the COD removal. The MFC was continuously operated for 10 days, achieving a COD removal of 58.7% with a pH ranging between 7 and 8, similar to those observed in typical biological wastewater treatment processes. However, the COD removal would have been higher, if the MFC had been left working longer. Nevertheless, the overall efficiency observed for the COD removal confirms the effective wastewater treatment process using the MFC technology. Moreover, the increase in the COD removal is well correlated with an increase in the cell voltage, making in evidence the conversion of the biomass into the green eco-electric energy. These findings revealed the anaerobic development of the microorganisms, which were electrochemically active. The same behavior of the COD removal has been observed previously, by studying the effects of the resistance load, the geometry and the design of the MFC (Lee et al. 2016), as well as other effects such as electrodes modification and biofilm scratching. In general, at the end of the experiment, the color of the

Table 2 Characteristics of photovoltaic solar module used for polarization of operational amplifier

Standard irradiation (W/m ²)	1000
Temperature (°C)	25
Current I _{cc} (A)	0.36
Voltage V _{oc} (V)	14.4
Current I _{opt} (A)	0.33
Voltage V _{opt} (V)	12
Power P _m (W)	4
Resistance R _s (Ω)	0.2
N _s	48

leachate of the sheep manure inside the MFC, became very much less darker, almost clear, which means that the maximum COD has been effectively removed, yielding sufficient bio-electrochemical energy conversion. It resulted in an electric current, which was produced after having grown an electro active biofilm that caused the reduction of the COD.

Amplification of low voltage output

In order to extract usable power from it, we have used two amplifiers: operational amplifier and common emitter amplifier. The low output MFC voltage (ca. 0.5 V) was successfully boosted up to 1.9 V, which was the highest voltage that has been ever reported. Moreover, the boosted voltage was stably maintained for more than 2 h. The energy harvesting efficiency increases as the polarization of the amplifier is utilized. These results clearly suggest that the proposed system is an efficient and self-starting energy harvester and storage for low-power generating MFCs. This finding is consistent with another renewable system that concerns the high voltage generation from wastewater by MFCs equipped with newly designed low voltage booster multiplier Nguyen et al. (2019).

As shown in Fig. S1, the low voltage of the single-compartment MFC was exploited by amplification by using an amplifier polarized with the photovoltaic solar module of characteristics given in Supplementary Information (Table 2), for the improvement of output voltage. The low voltage of the single-compartment MFC was exploited by amplification by undertaking five different ways. In the first case, as shown in Fig. 4, the voltage 0.5 V delivered by the

MFC was amplified with the operational amplifier, but unfortunately it generated a low current of $160\ \mu\text{A}$ which lasted just 20 s with a drastic drop in the input voltage to the end (a–a'). On the other hand, the series association of four MFCs made it possible to increase the voltage substantially to 1.64 V, but as before the current drawn was always very low (14 s) and the amplification time was shorter (b–b'). Whilst, the parallel association of the four MFCs made it possible to maintain the average voltage of 0.5 V overall, but the current was increased relatively to $650\ \mu\text{A}$ (c–c'). The advantage of this amplification lies in its long operating time (65 min). Finally, the polarization voltage of the amplifier was biased with 3 V using a current generator in our case it can be replaced by that of microbial fuel cells associated in series and in parallel (d–d'). Thus, it was restarted by delivering a voltage a little higher (0.65 V) but by delivering the same current 650 mA. This polarization therefore results in an increase in the duration of amplification (120 min).

Furthermore, the common emitter amplifier was biased with a photovoltaic panel powered with solar radiation (Fig. 5). The first test was carried out at the end of the day where the LED was illuminated for 135 min. It went off when the sun set down completely. However, The LED could remain illuminated

as long as there is lightening, in particular solar radiation in our case. So, from a renewable energy point of view, this method is advantageous because the polarization of the common-emitter amplifier has been replaced by solar radiation. During the night, one is obliged to polarize with a current generator in order to keep LED on.

Integration of DC/DC boost converter for effective exploitation

Moreover, in order to remedy to the deficiency encountered when using only the amplifier alone, we have included another potential electronic device to harvest the maximum energy delivered by the MFC. The energy harvested from the MFC required thus a power management assembly to step-up the low voltage to acceptable level. It contained thus the power supply (MFC), the amplifier and in addition a DC/DC converter to boost the voltage (Fig. S2).

As a matter of fact, we have therefore thought of the design of the precision amplifier, in particular for continuous signals. We therefore looked for appropriate circuits with very low drift, because the smallest detectable signals are limited by the signals error created by the amplifier. Really, the amplification of AC

Fig. 4 Evolution of input and amplified voltages using different configurations: (a–a') single stack of MFC; (b–b') series association of MFCs; (c–c') parallel association of MFCs; (d–d') parallel association of MFCs with polarization voltage

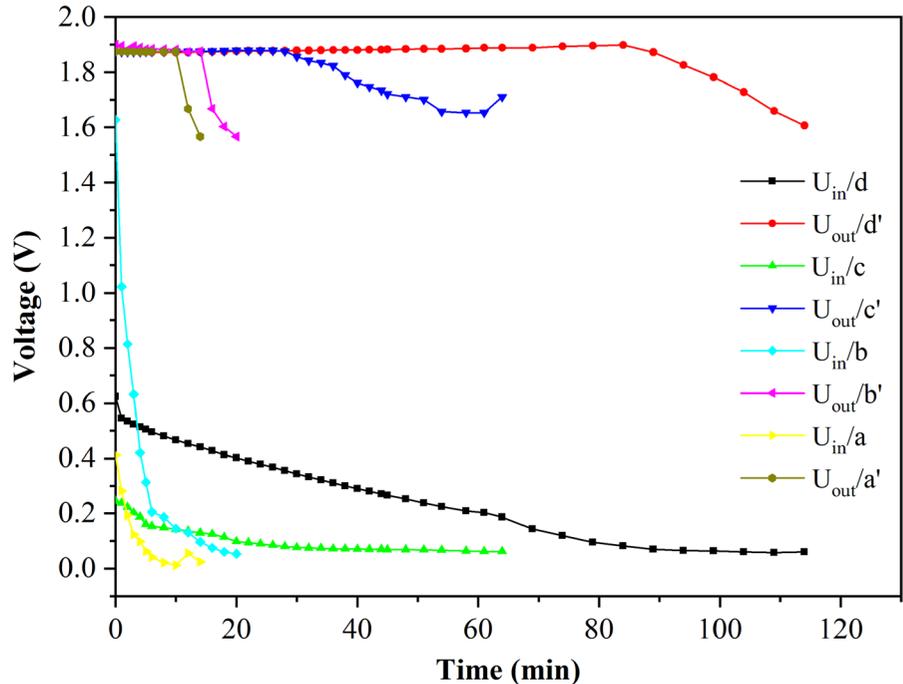
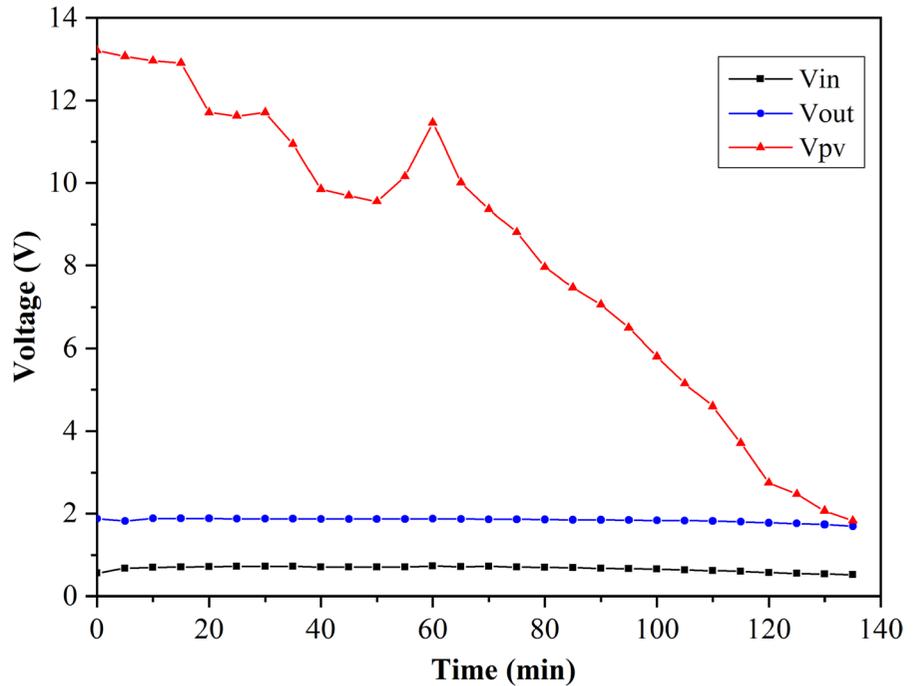


Fig. 5 Evolution of output amplified voltage of the MFC using an amplifier polarized with a solar panel



signals, noise and risk of oscillation can be avoided by just short-circuiting DC errors with capacitive couplings. However, this decoupling is no longer possible if utilizing amplified continuous signals. In this case, the continuous errors due to parasite voltages and currents and their drifts are very restrictive. An optimized boost-type step-up voltage converter (DC/DC) is therefore necessary to increase the voltage delivered by the MFC. So in order to meet the requirements for the real applications, the common emitter amplifier and the DC/DC converter boost should be used, so that the voltage and current of the MFC can be increased.

Figure 6 illustrates the variation in voltage used and the result of manual conversion using the pulses due to the position of the switch (S) of the boost converter. When the switch is closed (on state), the current in the inductor increases, and as a result, energy of magnetic origin is stored. The diode is therefore blocked, and the load is disconnected. On the other hand, when the switch is open, the inductance is then in series with the generator and its voltage is added to that of the generator: called the booster effect. This therefore results in a transfer of cumulative energy in the inductance to the capacitance. In the literature,

there have been investigations on MFC stacks/cascades in series, parallel and series/parallel configurations, and their impact on the stack performance (Ledeżma et al. 2013; Walter et al. 2016; Tremouli et al. 2018; Bongkyu et al. 2020).

Performance of MFCs after amplification

After having been tested successfully in powering the LED, the performance of MFCs has again been tested by plotting the power density curves. In Fig. 7, we compare the power density curves of the single-compartment MFC and the parallel association, after amplification. Whilst, in Fig. S3, we compare the MFCs with parallel and series associations successively. The figure reveals the enormous difference between them. Indeed, the parallel association yields the maximum power density with respect to the series association. Although the parallel association is quite beneficial in powering the LED, the stored energy was exhausted and in consequence, the MFCs stack requires a longer time to recover. Whilst, the series association yields higher voltage, but unfortunately lower current output. Table 3 summarizes the values of current and power densities of MFCs associated

Fig. 6 Evolution of output amplified voltage of the MFC using solar panel polarization and DC/DC boost converter

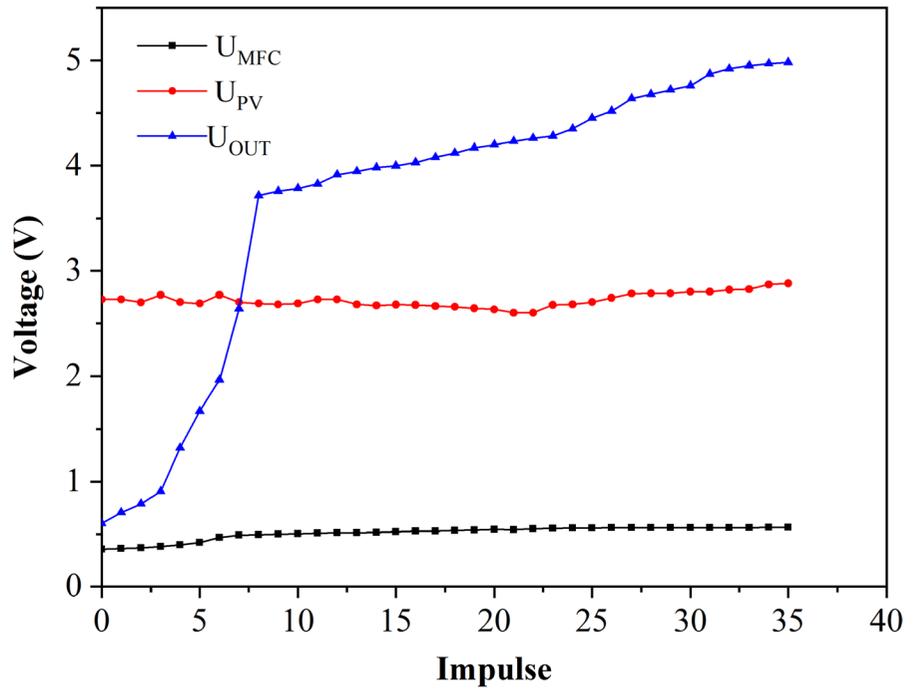
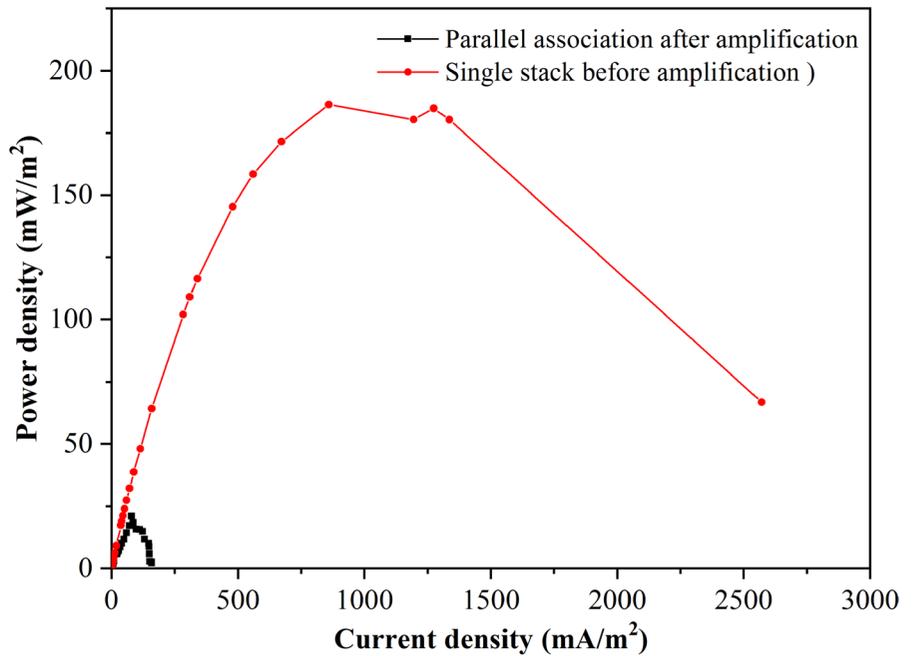


Fig. 7 Comparison between power density curves of MFCs: Single stack before amplification and parallel association after amplification



in series and in parallel after having been used in the amplification.

Statistical analysis of MFCs used as single stack either in series or in parallel

As it was reported in the literature, Oliot et al., had

already investigated before the effect of association of 02 MFCs connected in series, and found that the experimental voltage at maximum power point (i.e. 0.21 V) displayed relatively lower performance when the individual MFC₁ (i.e. 0.24 V) and MFC₂ (0.23 V) were connected in series in the same stack. It was revealed that the difference of 25 mV in voltage was not at all quite negligible. This constituted for them a suitable background for validating their numerical model (Oliot et al. 2017).

On the above-mentioned basis, a statistical analysis has been carried out with the three identical MFCs that were used as a single stack either in series or in parallel in the amplification system. The aim was to study the replicate aspect of the present work, in order to be statistically significant, eventually to see the resulting effects of two associations. For that reason, a further statistical experiment has been carried out for this purpose, by making measurements with individual MFCs and with stack of 3 identical MFCs either in series or in parallel.

The 3 MFCs namely MFC₁ MFC₂ and MFC₃ were first characterized individually (Fig. S3a). The power curves showed maximum values of the same order of the magnitude (i.e. 3.192, 3.210 and 3.269 mW/m²) respectively, allowing an average value of 3.223 mW/m². In absolute terms we must realistically allow that our values could all be subject to a systematic error up to about 1% (i.e. $3.223 \pm 0.0.030$ mW/m²). Whilst, the values of the CDM were 14.384, 19.263 and 16.584 mA/m² allowing an average value of 16.743 ± 1.679 mA/m². As a result, the cell voltages measured VMs also showed similar values (i.e. 221.91, 166.64 and 197.18 mV) with the average value 195.24 ± 19.07 mV.

The 3 MFCs were then electrically connected in series (Fig. S3b), the same current intensity flows through the three modules and the three cell voltages add up. The theoretical power curves of the stack composed of MFC₁, MFC₂ and MFC₃ connected as individual cells, is obtained by multiplying the values of current intensity by the sum of the MFC₁, MFC₂ and MFC₃ voltages. A value of PDM of 9.671 mW/m² was thus expected with a theoretical stack VM of 585.73 mV. In contrast, the measured PDM displayed drastically lower performance when the 3 MFCs were connected in series. The PDM measured experimentally was 3.141 ± 0.027 mW/m² quoted with estimated precision of less than 1%. The corresponding

stack CDM and VM were 5.041 ± 0.559 mA/m² and 650.26 ± 70.29 mV respectively. This experiment confirmed that connecting 3 MFCs in series, results in a considerably lower PDM than the sum of those of the individual cells. This drop may be caused by the electrical connection between the cells' electrodes, as it has also been observed previously by Zhuang et al. (2012) and Ledezma et al. (2013)

Furthermore, another stack of 3 MFCs connected in parallel, was set up (Fig. S3c). In contrast to the series association, the results obtained with the parallel association were remarkably higher 16.196 ± 0.184 mW/m² (i.e. ~ fivefold) showing the positive stacking effect when using the parallel association (Fig. S3). The results reported in Table 3 were obtained from the parabolic smoothing of both series and parallel associations. It may be conclude that the parallel association is much better than the series association when proceeding to the improvement of the lower bioenergy produced from MFCs for practical applications (Fig. 8).

Finally, the scalability of MFCs appears a key to the development of stacks. A recent study has shown that self-stratifying membraneless MFCs could be scaled down to 2 cm without performance deterioration. Really, it has been shown that the MFC of electrode 2 cm high, yielded power 5.45 mW per cascade, grater that with electrode 12 cm high (i.e. 1.48 mW) (Walter et al. 2020). It is therefore worthwhile to undertake in our forthcoming investigation, a research work on the MFC stack configuration and scalability for specific utilizations.

Conclusions

The initial findings of this study established the applicability of the MFC technology for the treatment of organic wastes and simultaneously the production of electrical energy. Furthermore, the objective was to enhance the low electric energy by using an adequate amplification system. The energy harvested from the MFC was demonstrated from the biodegradation of the sheep manure solid waste by using the bacterial catalyst. The output voltage de the MFC was substantially increased upon the addition of the GJ substrate (i.e. from 650 to 850 mV) making in evidence the electro activity of the bio-film. Really, the bio anode was much more effective

Fig. 8 Comparison of power density curves between single MFC units and a 3-MFC stack connected in parallel configuration; Inset (series configuration)

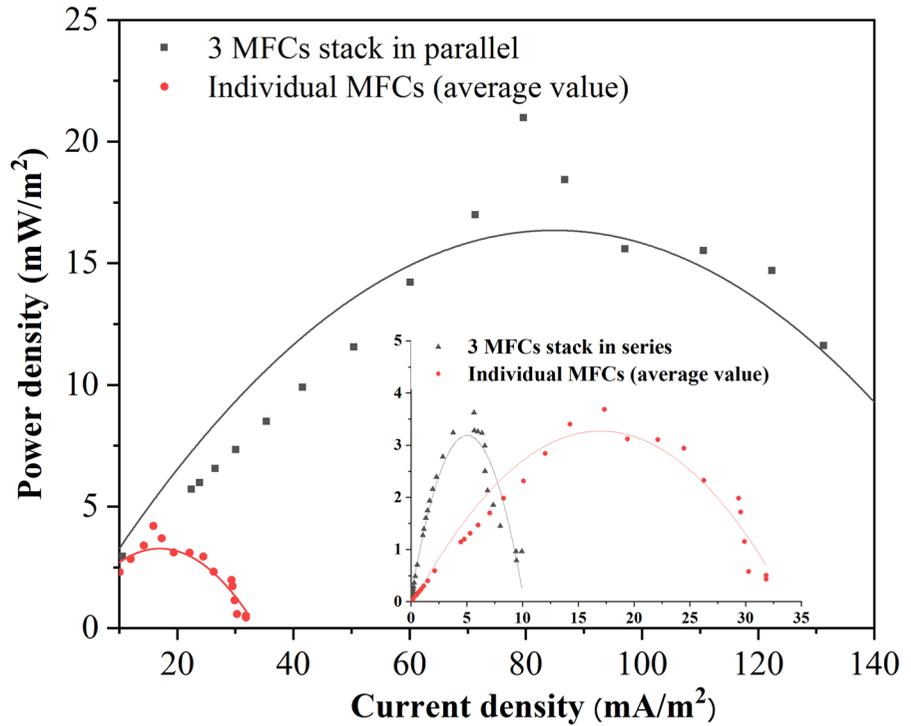


Table 3 Values of current and power densities of MFCs associated in series and in parallel after having been used in amplification and showing the positive stacking effect

Type of association	PDM (mW/m ²)	CDM (mA/m ²)	VM (mV)
Individual MFCs (average value)	3.223 ± 0.030	16.743 ± 1.679	195.24 ± 19.07
3 MFC stack in series	3.141 ± 0.027	5.041 ± 0.559	650.26 ± 70.29
3 MFC stack in parallel	16.196 ± 0.184	78.566 ± 4.653	206.91 ± 10.94

The values were obtained from the parabolic smoothing

in current production when it was polarized negatively (from 300 down to 155 mV) than positively (from 400 mV down to less than 1 mV). This allowed the possibility of adapting the microorganisms, rendering them more electro active and thus producing higher current densities. The COD removal of 10 days running MFC increased by 58.7% showing simultaneously an increase in the cell voltage. Furthermore, a power management unit including the MFC and an operational amplifier was utilized to power supply a light emitting diode (LED). The initial relative low output voltage 0.5 V was successfully boosted up to approximately 2 V (i.e. 4 times higher) and effectively harnessed as a power

supply. The amplifier was tested satisfactorily with parallel association of MFCs. However, both single stack and series association of MFCs were not able to achieve the required current for functioning the LED. The common emitter amplifier biased with a photovoltaic panel powered with solar radiation, allowed to feed the device as long as it was illuminated. The MFC stacking was beneficial, when the individual cell units were connected in parallel. The sheep manure MFC technology could therefore be implemented at larger scale to ensure electricity to offshore sheep barns farms. Whilst at smaller scale, it could be a promising alternative to supply with eco-friendly bioenergy the monitoring and measuring electrical systems.

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Declarations

Conflict of interest The authors declare no conflict of interest regarding the publication of this paper.

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