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Improvement of Microbial Fuel Cell Performance by Selection of Anodic Materials and Enrichment of Inoculum

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Abstract: *This work aimed at evaluating the effect of four anodic materials and the use of enriched inocula on the microbial fuel cell (MFC) performance. The anodic materials were granular activated carbon (GAC), graphite rod (GR), triangles of graphite (GT) and graphite flakes (GF). When loaded with a sulfate-reducing inoculum (SR-In) the internal resistance (R_{int}) obtained were 273, 410 and 795 Ω for GF, GT, GR, respectively and higher than 10 000 Ω for GAC, whereas the maximum volumetric power ($P_{V,max}$) were 1326, 2108 and 3052 mW m^{-3} for GR, GT and GF, respectively. We observed a decrease of R_{int} and an increase of $P_{V,max}$ with the increase of the log of A'_s of the graphite anodic materials that was consistent with a mathematical model previously reported by our Group. The use of the Fe (III)-reducing inoculum significantly enhanced the MFC performance; $P_{V,max}$ was up to 5000 mW m^{-3}, 40% higher than the power obtained with SR-In whereas the R_{int} was 140 ohms. Highest P_Vs of our MFC were close to values of electricity power derived from the anaerobic digestion of municipal wastewaters. In this regard, results of this work point out to a promising approach to further tapping bioelectricity from organic wastes that previously have yielded biohydrogen.*

Keywords: microbial fuel cells; anodic materials; volumetric power; enriched inoculum

1. INTRODUCTION

Fossil fuels scarcity and the associated costs, as well as the deleterious effects of their use on the environment and health have fostered the research on bioenergies as well as other renewable energy sources [1,2]. For instance, in biohydrogen production from dark fermentation of organic wastes, the complete conversion of wastes to energy is not possible [3]. Dark fermentation typically leads to partial degradation of the organic matter; a large amount of organic metabolites still remain in the spent liquors or leachates of dark fermentation that can be used as “fuels” in microbial fuel cells (*MFC*s) [4-6].

In this context, *MFC*s have emerged as a promising process for sustainable production of low intensity electrical energy and treatment of wastes such as the leachates and liquors of biohydrogen dark fermentation, thus constituting key contributors to sustainable development of our societies in the near future [7]. A *MFC* is an bioelectrochemical device that is capable of converting organic matter into electricity [5,6,8-10]. However, there are still some bottlenecks that can affect the electric energy production in a

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ABBREVIATIONS

$A_{el,}$	electrode surface area
AnD	anaerobic digestion of effluents
ANOVA	analysis of variance
A'_s	specific surface area of the electrode
C	conductance
COD	chemical oxygen demand
E	cell voltage
EAB	electrochemically active bacteria
E-In	enriched inoculum
E_{MFC}	measured voltage in a *MFC*
$E_{MFC,max}$	voltage maximum potential at the maximum power
$E_{MFC,OCV}$	maximum voltage in open circuit potential
GAC	granular activated carbon
GF	graphite flakes
GR	graphite rod
GT	triangles of graphite
I	current
I_{MFC}	current produced by a *MFC*
I_{max}	maximum current intensity
m_p	average weight of a particle of the given size fraction
M	total mass of anodic material loaded into the *MFC*
MFC	microbial fuel cell
NAFTA	North American Free Trade Agreement
OCP	open circuit potential
P	power
$p(F)$	probability of the F statistic in the ANOVA test of the regression
P_{max}	maximum power
P_{cath}	maximum power density based on surface area of electrode (cathode)
$p(T)$	probability of the t-Student statistic in significance test of the coefficients in the regression
P_V	volumetric power
$P_{V,max}$	maximum volumetric power
R_{ext}	external resistance
R_{int}	internal resistance
RVC	reticulated vitreous carbon
SI	supporting information
SR-In	sulfate-reducing inoculum
V_{cell}	geometric volume of the cell chamber

Greek characters

e	enrichment factor
$Ö_s$	shape factor of the particle defined as the quotient of the area of a sphere equivalent to the volume of the particle divided by the actual surface of the particle
$ñ$	actual density of the material
$ó$	electric conductivity in our work

MFC, such as the nature of the biocatalysts, the type and materials of electrodes, electrode catalysts, cell configuration, and architecture, among others [11-14]. Performance of *MFC*s can be restricted by ohmic overpotentials that related to the internal resistance (R_{int}). This effect translates into a decrease of cell voltage. The ohmic losses typically sum up the resistance to the flow of ions through the membrane and the anodic and cathodic electrolytes (only anodic electrolyte in a single chamber *MFC*) as well the resistance to the flow of electrons through the electrodes and interconnections as well as [15-17]. Thus the ohmic losses reduction or mitigation is crucial for improving the electrochemical characteristics and performance of the *MFC* [18]. Also, the value of R_{int} is related to the eventual operation of the *MFC*, since Jacobi's Theorem demonstrated that the maximum power output of an electromotive force is achieved when it is connected to an external resistance (R_{ext}) equal to its R_{int} [15,19-22]. According to the latter, it is recommendable to load the *MFC*s with an external resistance close to the value of the R_{int} that is determined previously in the characterization stage of the cell.

The R_{int} of a *MFC*, in turn, depends on some factors such as the surface area of electrodes, distance between electrodes, anodic material conductivity, the presence and type of membrane, the type of electrolyte(s), *inter alia* [15,23,24]. Indeed, the anodic material plays an important role on R_{int}. A good anodic material should have the following properties: high electrical conductivity, strong biocompatibility, chemical stability and anti-corrosion, large surface area and appropriate mechanical strength and toughness [14,15]. Thus, in order to reduce the R_{int} of the cell some materials and designs have been evaluated, such as new anodic materials, replacement of the salt bridge by membranes, choosing membranes with high protonic conductivity or building membrane-less *MFC*, increasing solution conductivity and reducing the pH, reducing electrode spacing, among others [5,6,25-31]. Regarding the effect of anodic materials, there has been much research on the use of graphite anodes in *MFC* [12-15,30-40].

Biological catalysts (microorganisms) play an important role on the performance of the *MFC*. Some microorganisms that collectively have received the denomination of electrochemically active bacteria (*EAB*) (also known as exoelectrogens or anodophilic) such as *Shewanella oneidensis*, *Geobacter sulfurreducens, Geobacter metallireducens, Clostridium butyricum, etc.*, possess the ability to foster the transfer of electrons to the anode in the *MFC* [41].

Several studies have used pure cultures of *EAB* to ensure a good electron transfer in cells. However, for practical applications in *MFC*, pure cultures do not seem to be a good option because of expensive sterilization of the influent and equipment is typically required. Instead of pure cultures, microbial mixed cultures or consortia from brewery wastewater, activated sludge, marine sediments, methanogenic and sulfate-reducing cultures, domestic wastewater and preacclimated bacteria from an active *MFC* could be used as inocula [6,24,42,43].

It has been reported that several *EAB* are dissimilatory metal reducers, *i.e.*, they can use Fe (III) and Mn (IV) as electron acceptors [44-47]. Since this type of microbes could improve the performance of a *MFC*, a strategy based on enriching natural inocula by the selective pressure of serial transfers in a medium with either Fe (III) and Mn (IV) has been reported in the literature with a good degree of success in obtaining an *EAB* mixed culture well suited for *MFC* seeding [47,48].

To the best of our knowledge studies on leachate treatment in *MFC*s seeded with enriched inocula are very scarce in the open literature [49]. There are, however, a couple of articles of our own group that deals with leachate treatment in *MFC* using more conventional inocula (aerobic, methanogenic, sulphate-reducing consortia) as well as research carried out by others whose results in general show low powers and moderate to adequate pollutant concentration removal (as chemical oxygen demand (*COD*)) (see review in appendix A, Supporting Information (SI)) [5,6,50-53].

Thus, the objective of this work was to evaluate the effect of four anodic materials and the use of enriched inocula on the microbial fuel cell (*MFC*) performance.

2. MATERIALS AND METHODS

2.1. Experimental design

The first experiment consisted of the characterization of the *MFC* fitted with granular activated carbon (*GAC*), graphite rod (*GR*), triangles of graphite (*GT*) and graphite flakes (*GF*) in an *MFC* loaded with a sulfate-reducing inoculum (S*R-In*), with two replicates. In a second experiment, the effect of an Fe (III)-reducing inoculum (*E-In)* on a *MFC* fitted with an anode made with *GF* was evaluated. The main response variables were the $P_{V,max}$ and the R_{int} of the *MFC*s. The experiments were carried out in a single compartment, air-cathode *MFC*. The cells were operated at ambient temperature.

2.2. Microbial fuel cell

The single chamber *MFC* consisted of a horizontal cylinder built in polyacrylated 8.0 cm long and 5.7 cm internal diameter [6,54,55]. The anodic chamber was packed with the different anodic materials, i.e., *GAC*, *GR*, *GT* and *GF* with corresponding surface areas of 0.2, 8.89×10^{-4}, 0.06 and 0.28 m^2, respectively (Table 1).

The cathode of our *MFC* was a flexible carbon-cloth containing 0.5 mg cm^{-2} platinum catalyst (Pt 10 wt%/C-ETEK). On the air side, the cathode was limited by a perforated plate of stainless steel 1 mm thickness. In the liquid side, the cathode was in contact with a proton exchange membrane (Nafion® 117) [6,55]. The current intensity collector used in all cases was a graphite rod of 5.7 cm length and 0.5 cm diameter.

The *GAC* was supplied by Filtrantes and Refacciones Industriales S.A. de C.V., Mexico City, Mexico. The *GR* was purchased to Lumen S.A. de C.V., Mexico City, Mexico; they report that this material was imported from the Czech Republic. The *GT*s were fabricated by conveniently slicing cylindrical graphite bars of 38 mm diameter in disks of 5 mm thickness. Each disk, in turn, was cut in 8 quasi triangular parts. The graphite bar was purchased to Brunssen de Occidente S.A. de C.V., Guadalajara, Jalisco, Mexico.

The specific surface area A'_s of the anodic materials and the net volume of the only chamber in our *MFC*s was calculated according to Hernández-Flores *et al*. [56] according the Eq. 1 bellow.

$$A'_s = \frac{\frac{M}{\Phi_s}\left(\frac{6^2 m_p{}^2 \pi^3}{m_p{}^3 \pi^2 \rho^2}\right)^{1/3}}{V} = \frac{\frac{M}{\Phi_s}\left(\frac{36\pi}{m_p \rho^2}\right)^{1/3}}{\left(V_{cell} - \frac{M}{\rho}\right)} \tag{1}$$

where

$\ddot{O}_s$	shape factor of the particle defined as the quotient of the area of a sphere equivalent to the volume of the particle divided by the actual surface of the particle
m_p	average weight of a particle of the given size fraction
M	total mass of anodic material loaded into the *MFC*
$\tilde{n}$	actual density of the material
V_{cell}	geometric volume of the cell chamber

The net volume of our single chamber *MFC* was estimated as described above in the denominator of Eq. 1.

2.3. Inocula

2.3.1. Sulfate-reducing inoculum *SR-In*

In the first experiment, the *MFC* was seeded with a *SR-In* sampled from a sulphate-reducing complete mix reactor. The biomass concentration in the inoculum was *ca.* 1280 mg VSS L^{-1}. The complete mix bioreactor was operated at 37 °C. The feed to the sulphate-reducing reactor contained sucrose as carbon source, and was fed at a flow rate of 120 mL d^{-1}. Its composition was (in g L^{-1}): sucrose (5.0), acetic acid (1.5), $NaHCO_3$ (3.0), K_2HPO_4 (0.6), Na_2CO_3 (3.0), NH_4Cl (0.6), Na_2SO_4 (11.0).

2.3.2. Enriched inoculum

An enrichment procedure based on selective pressure using Fe (III) as an electron acceptor and sodium acetate as carbon source was implemented [44,55]. The departing inoculum consisted of sediments sampled from a Nogales lagoon at a depth of 2 m. This lagoon is close to the volcano Citlaltepetl, Veracruz, México (geographic coordinates are 18° 49′ 13′′ N latitude and 97° 9′ 52′′ W longitude; the altitude is 1925 m above sea level.)

The *E-In* was obtained with the method of serial transfers from an original sediment sample (See appendix B, SI) [41,47]. We defined a convenient enrichment index (ε) to assess the performance of serial transfers in the enrichment process, given in appendix B, SI.

Table 1. Selected physical characteristics of anodic materials.

Anodic material	Working net volume (m^3)	Anodic actual surface (m^2)	A'_s [a] (m^2 m^{-3})	Conductance (S) [b]
GAC [c]	3.59×10^{-5}	0.2 ± 0.95	5571	< 0.01
Graphite rod	2.03×10^{-4}	$8.89\times10^{-4} \pm 2.5\times10^{-5}$	7.3	0.20 ± 0.04
Triangles of graphite	6.64×10^{-5}	0.0618 ± 0.001	931	0.61 ± 0.02
Graphite flakes	7.22×10^{-5}	0.28 ± 0.078	1302	0.13 ± 0.04

[a] Relationship between the anode surface area to cell volume, also known as specific surface area of the anode. [b] Electrical conductance of the materials, expressed in Siemens. [c] Granular activated carbon.

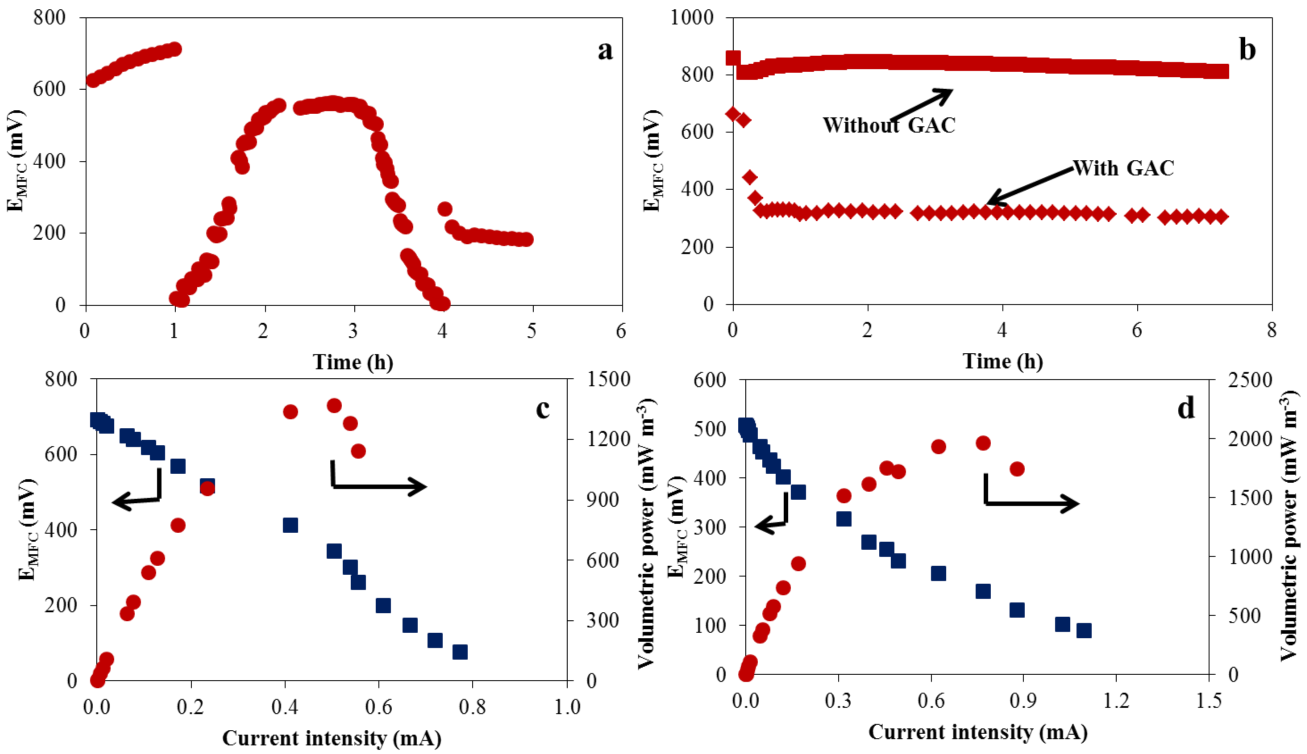

Figure 1. Characterization of the microbial fuel cell fitted either with granular activated carbon, graphite rod and graphite triangles as anodes, and using sulphate-reducing inoculum: (a) Variation of microbial fuel cell potential with the external resistance for granular activated carbon; (b) effect on the open circuit potential for granular activated carbon; (c) Polarization curve and volumetric power for graphite rod as anode; (d) Polarization curve and volumetric power with graphite triangles as anode. Keys: *GAC*, granular activated carbon; E_{MFC}, measured voltage in the *MFC*.

2.4. Leachate

The *MFC* was loaded with 6 mL of a leachate similar to that produced in the hydrogen fermentation of the organic fraction of the municipal solid wastes [57]. The model leachate was concocted with a mixture of simple organic acids and solvents (in g L^{-1}): acetic, propionic and butyric acids (4 each) as well as acetone and ethanol (4 each) and mineral salts like $NaHCO_3$ and Na_2CO_3 (3 each) and K_2HPO_4 and NH_4Cl (0.6 each) [58,59]. The *COD* of this stock leachate was 16 800 mg L^{-1}.

The initial *COD* and biomass concentration in the cell liquor were *ca.* 1300 mg O_2 L^{-1} and 1280 VSS L^{-1}, respectively. The pH and the electrical conductivity were 7.03 and 1385 µS cm^{-1}, respectively. The liquor in the *MFC* was a mixture of inoculum and leachate.

2.5. Determination of internal resistance of the cell

The R_{int} of the cell was determined by duplicate for each anodic material, using the polarization curve method by varying the R_{ext} and recording both the voltage and the current intensity [6,30,31]. The voltage was measured and recorded with a Multimeter ESCORT 3146A. The current was calculated by the Ohm's law and the R_{int} was calculated as the slope of the linear section of the curve voltage versus the current intensity [6,15].

2.6. Analyses

The *COD* and *VSS* of the liquors of sulphate-reducing seed bioreactor and cells were determined according to the Standard Methods [60]. In addition, the individual concentrations of volatile organic acids and solvents in the model extract were analyzed by gas chromatography in a chromatograph Perkin Elmer Autosystem equipped with a flame ionization detector as described elsewhere [6].

Statistical data processing was performed with the tool Analysis of Data/Regression of Excel software, Microsoft Office 2010 (Microsoft, Seattle, WA, USA).

3. RESULTS AND DISCUSSION

3.1. Characterization of the cell using four anodic materials and sulphate-reducing inocula

Table 1 exhibits some properties of the anodic materials whereas Figure 1a shows the polarization curve of the *MFC* fitted with *GAC* anode, when the operation started at open circuit potential (*OCP*). The maximum voltage of the cell ($E_{MFC,max}$) achieved was 710 mV at the end of the first hour, and at the end of procedure the *OCP* was only 180 mV. This voltage indicated that the system did not recover appropriately and results would be unreliable according to Watson and Logan, who reported similar patterns of *OCP* that were discussed as power overshoot [61]. They evaluated a brush anode constructed from carbon fibers and found an unexpectedly large drop in cell potential.

In order to gain further insight on power overshoot in our work, we operated two *MFC* in *OCP* conditions for 7 h. Figure 1b shows that the cell packed with 32 g *GAC* (0.2 m^2 anodic surface) suffered the power overshoot at the first operation hour (from 640 to 320 mV). On the other hand, the *MFC* with no *GAC* and working only with the *GR* did not suffer the overshoot and the voltage achieved up to 860 mV. It was concluded that *GAC* was not a suitable material for the anode of the *MFC*. The poor performance of *GAC* as anodic material could be due to low *s* of the *GAC* (or *C* less than 0.01 S, Table 1).

When using *GR* as anode, the maximum *OCP* was 800 mV in the first hour of the characterization; at the end of the procedure the *OCP* was 600 mV (Fig. 1c). The R_{int} was obtained from the slope of graph voltage *vs.* current intensity and gave a value of 795 Ω (Table 2). With *GT* as anode, the estimated R_{int} was 410 Ω and the $P_{V,max}$ reached 2108 mW m^{-3} (Fig. 1d), 50% lower than the R_{int} obtained by the *GR* (Table 2).

Regarding the *GF* used as anode, the R_{int} and $P_{V,max}$, decreased and increased respectively with respect to *GT* as anode (Fig. 2a, Table 2). The R_{int} was 35% lower than *GT* and the $P_{V,max}$ reached 3052 mW m^{-3}, 45% more than *GT*. It is important to note that in this anode, the relationship anodic surface/cell volume increased 4 times from *GT* to *GF* (Table 1).

The larger surface area of *GF* compared to the other anodic materials would be an advantage since the microorganisms would have a greater anodic surface to colonize and transfer the electrons. In this regard, our results and interpretation are consistent with findings of other researchers (see discussion in appendix C, SI) [33-37].

Also, the better characteristics of *MFC* fitted with graphite anodes in this work (*i.e.*, *GR*, *GT* and *GF*) compared to *GAC* could be explained by the high electric conductivity of graphite compared to the low conductivity of *GAC* (Table 1).

Also, other phenomena could explain the lower performance of the *GAC* anode:

(i) a poor current collection due to the use of only one graphite rod inserted in the middle as current collector in our work that likely was not sufficient to provide a full contact between anodic material and collector. In this regard, in the literature have been proposed other types of anode design that could overcome this effect [62]. We should highlight, however, that similar collector rods were used with *GAC* and *GF* anodes in our work. Since the *GF* results were significantly better (with the same collector), it seems that the determining factor is the anodic material, not the collector device;

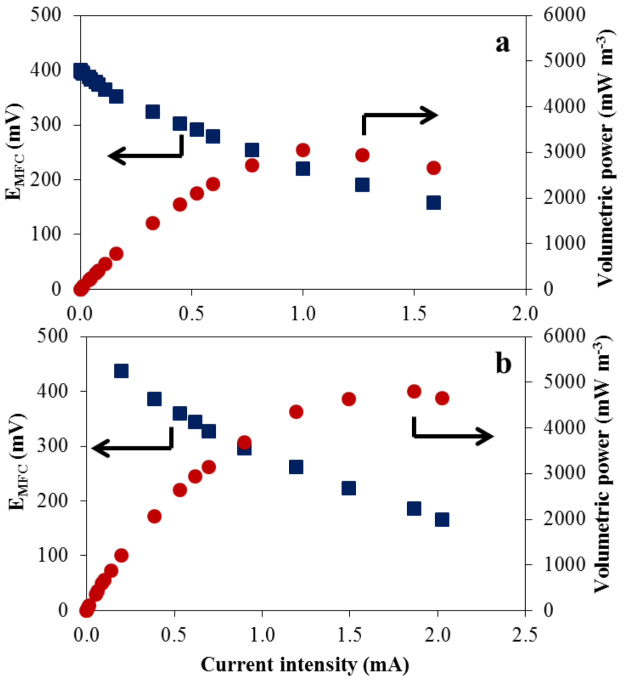

Figure 2. Polarization curve and volumetric power with graphite flakes as anode using (a) Sulfate-reducing inoculum; (b) enriched inoculum. E_{MFC}, measured voltage in a *MFC*.

(ii) The *GAC* used in our work did not receive any chemical/thermal treatment in order to improve its electrochemical and physical characteristics, as discussed in the open literature [63].

We fitted the P_V and R_{int} results of our *MFC*s equipped with graphite anodic materials to models based on Tafel equation that relates those two electrochemical variables to the log of the specific surface of the anodic material A'_s [56].

Table 2. Results of characterization of the microbial fuel cells.

Parameters	Graphite rod	Triangles of graphite	Graphite flakes	Graphite flakes
Inoculum	*SR-In*[a]	*SR-In*	*SR-In*	*E-In*[b]
R_{int} (Ω)	795 ± 147	410 ± 22	273 ± 153	140 ± 15
P_{cath}(mW m^{-2})[c]	65.4 ± 0.12	54 ± 0.1	86.4± 0.7	135 ± 3
$P_{V,max}$(mW m^{-3})[d]	1326 ± 72	2108 ± 174	3052 ± 23	4820 ± 110
P_{max}(mW)	0.17 ± 0.01	0.14 ± 0.01	0.22 ± 0.005	0.35 ± 0.01
I_{max} (mA)[e]	1.53 ± 0.3	1.92 ± 0.6	3.50 ± 0.6	4.5 ± 0.8
$E_{MFC,max}$ (mV)[f]	700 ± 1	500 ± 1	402 ± 1	555 ± 2
$E_{MFC,OCV}$ (mV)[g]	800 ± 120	600 ± 140	575 ± 33	577 ± 114

[a]Sulphate-reducing inoculum. [b]Enriched inoculum. [c]Maximum power density based on surface area of electrode (cathode). [d]Maximum volumetric power. [e]Current intensity value at the maximum power. [f]Potential value at the maximum power. [g]Open circuit potential.

$$P_{V,max} = 842.9 + 633.2 \times \log A'_s \tag{2}$$

with $P_{V,max}$ in mW m^{-3}, A'_s in m^{-1}
The statistical parameters were the following:
$R^2 = 0.8872$; $p(F) = 0.005$; Ranking = 100%
$110.72 \leq a_o \leq 1575.14$ at 95% confidence, $p(T) = 0.0330$
$319.71 \leq a_1 \leq 946.77$ at 95% confidence, $p(T) = 0.0050$
For R_{int}, the model fitting lead to

$$R_{int} \cong 933.5 - 211.6 \times \log A'_s \tag{3}$$

with R_{int} in ohms, A'_s in m^{-1}
The statistical parameters were the following:
$R^2 = 0.8850$; $p(F) = 0.005$; Ranking = 100%
$686.11 \leq b_o \leq 1180.79$ at 95% confidence, $p(T) = 0.00047$
$-317.51 \leq b_1 \leq -105.70$ at 95% confidence, $p(T) = 0.00517$

R^2 is the determination coefficient of the regression, Ranking was defined as the ratio of the number of significant coefficients of the regression to the number of total coefficients and multiplied by 100 [56].

The largest standardized residual was 1.16, whereas the smallest was -1.63. Thus, all the residuals fell in the interval (-2.0, 2.0) meaning that no outliers were found [64].

3.2. Characterization of the cell using graphite flakes and enriched inoculum

Time course of Fe (II) concentration in the last two serial transfers of the enrichment procedure are shown in Figure D1, Appendix D, SI. It can be seen that a maximum concentration of Fe (II) of nearly 8 mM was attained in both cases. This suggested that the inoculum was ready to use. Furthermore, this was confirmed by the similar values of å, *i.e*., 6.4 and 6.5 at the end of serial transfers 6 and 7, respectively (See Figure D2, Appendix D, SI).

The *MFC* fitted with *GF* anode and seeded with *E-In* was characterized by the polarization curve method (Fig. 2b). The R_{int} was very low (*ca.* 140 ohms, Table 2) whereas the $P_{V,max}$ was outstandingly high (*ca.* 5000 mW m^{-3}), 40% higher than the obtained with sulfate-reducing inoculum and the same anode (Fig. 2a). It seems that the combined approach of using an enriched inoculum and a particulate graphite anode lead to the best results in this work (See further discussion in Appendix D, SI) [47,48,65-68].

3.3. Bioelectricity from microbial fuel cells and energy thresholds of other bioenergy-producing processes

Regarding the energy harvested in our *MFC* and other bioenergy processes our highest power value was quite close to the threshold indicator for bioenergy process feasibility if we consider as a reference the electricity generated from biogas of the anaerobic digestion (*AnD*) of municipal effluents (Table 3) [69-72].

Table 3 shows a comparison of the power delivered by *AnD* of diluted and concentrated effluents and our *MFC* with leachate. In order to perform a fair comparison, we have to point out to two main differences between *MFC* and *AnD*. First, the *MFC* operated at ambient temperature whereas *AnD* typically operates at 35 °C or higher [69,70]. Thus, *AnD* needs heating (for carrying the influent temperature from ambient to 35 °C and to compensate for heat losses of the bioreactors and ancillary equipment) that could represent up to 20% of the generated bioenergy in *AnD* (a range of 15 to 20%, with lower percentages for high organic load effluents [73]. So, the net energy threshold of *AnD* will be lower than the maximum energy threshold calculated by the mere enthalpy of combustion of the methane generated by the *AnD*.

Second, the bioenergy provided by *AnD* is not electricity but the total heat content of the net methane. The net methane (produced methane minus heating requirements) should be converted to electricity and this conversion is limited by the 2nd law of Thermodynamics for thermal engines that states that the typically 30 to 40% of the heat value of fuel can be obtained as useful work or electricity [71,72].

The electrical energy that could be derived from *AnD* of several typical effluents is depicted in the last row of Table 3. *It is apparent that the energy indicator of our MFC is quite close to the net energy threshold of AnD treating municipal wastewater (between 60 to 100%).* On the other hand, the energy indicator of our *MFC* is nearly 10% of that of *AnD* of vinasses (distillery slops from the alcohol fermentation in the sugar cane industry) [74]. Indeed, in order to compete with the power delivered by *AnD* of vinasses, a stacked array of several *MFC*s could reach that threshold, based on trends of *MFC* improvement reported elsewhere [75-78].

Moreover, the abnormally high price of the kilowatt-hour in Mexico of Mx $ 2.772 (USD 0.213; the highest price in the *NAFTA* countries) is a powerful drive for further development and application of *MFC* [79].

So, with caution due to particular cases, our *MFC*, or better some stacked array based on our *MFC*s could become competitive with the well established *AnD* of effluents [78,80-82]. Undoubtedly, this could have a positive impact on the management and treatment of leachates and other effluents in Mexico.

4. CONCLUSION

Our work has shown that the type and size of anodic material had a significant effect on the R_{int} and $P_{V,max}$ of *MFC*s in our work: *GAC* proved to be unsuitable for its use as anodic material. On the

Table 3. Electrical power thresholds of anaerobic digestion of effluents and our microbial fuel cell.

Category Output (W m^{-3})	Anaerobic digestion of municipal waste water [75]	Anaerobic digestion of vinasses in fluidized-bed reactor [74]	Anaerobic digestion of concentrated effluents [77]	Our Microbial Fuel Cell
Methane heat power	20 to 30	180	400	NA*
Energy loss due to heating influent and reactors	-4 to -6	-27	-60	NA
Loss due to the second law of thermodynamics	-10.4 to -15.6	-99.5	-221	NA
Net electrical energy	5.6 to 8.4	53.5	119	4.8

*NA: not applicable

other hand, graphite anodes showed good results. The different performance between *GAC* and graphite anodes was ascribed to the relatively high electric *C* of graphite, superior to that of *GAC*.

Comparing the graphite anodes, the $P_{V,max}$ increased and the R_{int} decreased in three materials (*GR*, *GT* and *GF*) with the increase of the log of the A'_s of the anode, according to models based on Tafel equation and reported elsewhere by our Group. $P_{V,max}$ and R_{int} obtained with the graphite-based anodes adequately fitted those models, with determination coefficients higher than 0.85.

Use of *E-In*, directed to select for Fe(III)-reducing bacteria, along with graphite flakes anode, lead to the lower R_{int} and an outstandingly high $P_{V,max}$ *ca.* 5000 mW m^{-3} in our one-compartment, air cathode *MFC*. Its value was 40% higher than the power obtained with *SR-In*. Highest P_Vs of our *MFC* were close to values of electricity power derived from the anaerobic digestion of municipal wastewaters. In this regard, results of this work point out to a promising approach to further tapping bioelectricity from organic wastes that previously have yielded biohydrogen.

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REFERENCES

[1] D. Das, T. N. Veziroglu, Int J. Hydrogen Energy, 26, 13 (2001).

[2] Y. Cheng-Dar, L. Chung-Ming, E. M. L. Liou, Energy Policy, 29, 951 (2001).

[3] I. Valdez-Vázquez, E. Ríos-Leal, F. J. Esparza-García, F. Cecchi, H. M. Poggi-Varaldo, Int. J. Hydrogen Energy, 30, 1383 (2005).

[4] I. Valdez-Vazquez, E. Rios-Leal, A. Carmona-Martinez, K. Muñoz-Páez, H. M. Poggi-Varaldo, Environ. Sci. Technol., 40, 3409 (2006).

[5] H. M. Poggi-Varaldo, A. Carmona-Martínez, A. L. Vázquez-Larios, O. Solorza-Feria, J. New Mat. Electrochem. Systems, 12, 49 (2009).

[6] A. L. Vazquez-Larios, O. Solorza-Feria, G. Vazquez-Huerta, F. J. Esparza-Garcia, E. Rios-Leal, N. Rinderknecht-Seijas, H. M. Poggi-Varaldo, J. New Mat. Electrochem. Systems, 13, 219 (2010).

[7] H. M. Poggi-Varaldo, K. M. Munoz-Paez, C. Escamilla-Alvarado, P. N. Robledo-Narváez, M.T. Ponce-Noyola, G. Calva-Calva, E. Ríos-Leal, J. Galíndez-Mayer, C. Estrada-Vázquez, A. Ortega-Clemente, N. Rinderknecht-Seijas, Waste Management Research, 32, 353 (2014).

[8] Z. Du, H. Li, T. Gu, Biotechnol. Adv., 25, 464 (2007).

[9] O. Lefebvre, A. Al-Mamun, W. K. Ooi, Z. Tang, D. H. C. Chua, H. Y. Ng, Water Sci. Technol., 57, 2031 (2008).

[10] S. Ouitrakul, M. Sriyudthsak, S. Charojrochkul, T. Kakizono, Biosens. Bioelectron., 23, 721 (2007).

[11] P. Belleville, P. J. Strong, P. H. Dare, D. J. Gapes, Water Sci. Technol., 63, 1752 (2011).

[12] B. E. Logan, J. M. Regan, Environ. Sci. Technol., 5172 (2006).

[13] Y. Yang, G. Sun, M. Xu, J. Chem. Technol. Biotechnol., 86, 625 (2010).

[14] M. Zhou, M. Chi, J. Luo, H. He, T. Jin, J. Power Sources, 196, 4427 (2011).

[15] B. E. Logan, B. Hamelers, R. Rozendal, U. Schröder, J. Keller, S. Freguia, P. Aelterman, W. Verstraete, K. Rabaey, Environ. Sci. Technol., 40, 5181 (2006).

[16] S. Sevda, X. Dominguez-Benetton, K. Vanbroekhoven, T. R. Sreekrishnan, D. Pant, Chem Eng J. 228, 1 (2013).

[17] A. ElMekawy, S. Srikanth, K. Vanbroekhoven, H. De Wever, D. Pant, Journal of Power Sources. 262, 183 (2014).

[18] H. Rismani-Yazdi, S. M. Carver, A. D. Christy, O. H. Tuovinen, J. Power Sources, 180, 683 (2008).

[19] D. Halliday, R. Resnick, J. Walker, *Fundamentals of Physics*. 7th ed. John Wiley & Sons, Inc. New York, 2005.

[20] D. Jiang, B. Li, Wat. Sci. Technol., 59.3, 557 (2009).

[21] B. E. Logan, Water Sci. Technol., 52, 31 (2005).

[22] H. M. Poggi-Varaldo, A. Vazquez-Larios, O. Solorza-Feria, Microbial fuel cells. In Rodríguez-Varela F. J., Solorza-Feria O., Hernández-Pacheco, E. (Eds). *Fuel cells.* Book Livres, Montréal, Canada, 2010.

[23] F. Li, Y. Sharma, Y. Lei, B. Li, Q. Zhou, Appl. Biochem. Biotechnol., 160, 168 (2011).

[24] C. I. Torres, A. K. Marcus, H. S. Lee, P. Parameswaran, R. Krajmalnik-Brown, B. E. Rittmann, Bioresour. Technol., 102, 9335 (2010).

[25] K. Rabaey, G. Lissens, D. S. Steven, W. Verstraete, Biotechnol. Lett., 25, 1531 (2003).

[26] H. Liu, B. E. Logan, Environ. Sci. Technol., 38, 4040 (2004).

[27] B. Min, S. Cheng, B. E. Logan, Water Res., 39, 1675 (2005).

[28] H. Liu, S. Cheng, B. E. Logan, Environ. Sci. Technol., 39, 5488 (2005).

[29] S. Cheng, H. Liu, B. E. Logan, Environ. Sci. Technol., 40, 2426 (2006).

[30] B. E. Logan, Microbial fuel cells. John Wiley-Interscience. New Jersey, USA, 2007.

[31] D. Jiang, B. Li, Biochem. Eng. J., 47, 31 (2009).

[32] J. Wei, P. Liang, X. Huang, Bioresour. Technol., 102, 9335 (2011).

[33] S. Hays, F. Zhang, B. E. Logan, J. Power Sources, 195, 8293 (2011).

[34] Y. Feng, Q. Yang, X. Wang, B. E. Logan, J. Power Sources, 195, 1841 (2010).

[35] A. Larrosa-Guerrero, K. Scott, K. P. Katuri, C. Godinez, I. M. Head, T. Curtis, Appl. Microbiol. Biotechnol., 87, 1699 (2010).

[36]Y. Liu, F. Harnisch, K. Fricke, U. Schröder, V. Climent, J. M. Feliu, Biosens. Bioelectron., 25, 2167 (2010).

[37]T. Jin, L. Zhou, J. Luo, J. Yang, Y. Zhao, M. Zhou, J. Chem. Technol. Biotechnol., 88, 2075 (2013).

[38]L. Hsu, B. Chadwick, J. Kagan, R. Thacher, A. Wotawa-Bergen, K. Richter, *RSC Adv*., 3, 15947 (2013).

[39]A. Dewan, H. Beyenal, Z. Lewandowski, Environ. Sci. Technol., 42, 7643 (2008).

[40]A. L. Vazquez-Larios, O. Solorza-Feria, J. Barrera Cortes, E. Ríos-Leal, M. T. Ponce-Noyola, F. Esparza-García, R. G. González-Huerta, N. Rinderknecht-Seijas, H. M. Poggi-Varaldo, Proceedings "Battelle 9th International Conference on Remediation of Chlorinated and Recalcitrant Compounds", Monterey, CA, USA, May 19-22, 2014.

[41]K. Juarez, B. C. Kim, K. Nevin, L. Olvera, G. Reguera, D. R. Lovley, B. A. Methe, J. Mol. Microbiol. Biotechnol., 16, 146 (2009).

[42]M. Sharma, N. Aryal, P. M. Sarma, K. Vanbroekhoven, B. Lal, X. Dominguez Benetton, D. Pant, Chemical Communications. 49, 6495 (2013).

[43]A. ElMekawy, H. M. Hegab, K. Vanbroekhoven, D. Pant, Renewable and Sustainable Energy Reviews. 39, 617 (2014).

[44]D. R. Lovley, E. J. P. Phillips, Appl. Environ. Microbiol., 51, 683 (1986).

[45]D. R. Lovley, E. J. P. Phillips, Appl. Environ. Microbiol., 53, 1536 (1987).

[46]D. R. Lovley, E. J. P. Phillips, Appl. Environ. Microbiol., 54, 1472 (1988).

[47]A. C. Ortega-Martínez, K. Juárez-López, O. Solorza-Feria, M. T. Ponce-Noyola, J. Galindez-Mayer, N. Rinderknecht-Seijas, H. M. Poggi-Varaldo, Int. J. Hydrogen Energy, 38, 12589 (2013).

[48]K. Sathish-Kumar, O. Solorza-Feria, J. Tapia-Ramírez, N. Rinderkenecht-Seijas, H. M. Poggi-Varaldo, Int. J. Hydrogen Energy, 38, 12600 (2013).

[49]D. Pant, G. Van Bogaert, L. Diels, K. Vanbroekhoven, Bioresour. Technol., 101, 1533 (2010).

[50]A. L. Vázquez-Larios, O. Solorza-Feria, G. Vázquez-Huerta, F. Esparza-García, N. Rinderknecht-Seijas, H. M. Poggi-Varaldo, Int. J. Hydrogen Energy, 36, 6199 (2011).

[51]J. N. Zhang, Q. L. Zhao, S. J. You, J. Q. Jiang, N. Q. Ren, Water Sci. Technol., 57 (7), 1017 (2008).

[52]J. Greenman, A. Gálvez, L. Giusti, I. Ieropoulos, Enzyme Microb. Technol., 44, 112 (2009).

[53]A. Galvez, J. Greenman, I. Ieropoulos, Bioresour. Technol., 100, 5085 (2009).

[54]G. Hernández-Flores, H.M. Poggi-Varaldo, O. Solorza-Feria O, T. Romero-Castañón, E. Ríos-Leal, J. Galíndez-Mayer, F. Esparza-García, International Journal of Hydrogen Energy (2015), In press. http://dx.doi.org/10.1016/j.ijhydene.2015.06.057.

[55]G. Hernández-Flores, Interim Report. Sc D Thesis, CINVESTAV-IPN, México, D.F., 2013.

[56]G. Hernández-Flores, H.M. Poggi-Varaldo, O. Solorza-Feria O, M. T. Ponce Noyola, T. Romero-Castañón, N. Rinderknecht-Seijas, International Journal of Hydrogen Energy (2015). In press. http://dx.doi.org/10.1016/j.ijhydene.2015.06.119.

[57]H. M. Poggi-Varaldo, L.Valdés, F. J. Esparza-García, G. Fernández-Villagómez, Water Sci. Technol., 35, 197 (1997).

[58]H. M. Poggi-Varaldo, N. Rinderknecht-Seijas, Acta Biotechnologica**,** 23, 271 (2003).

[59]H. M. Poggi-Varaldo, L. M. Alzate-Gaviria, A. Perez-Hernandez, V. G. Nevarez-Morillon, N. Rinderknecht-Seijas, Waste Manage. Res., 23, 270 (2005).

[60]APHA. Standard methods for examination of water and wastewater. 17th ed. APHA-AWWA-WEF, Washington DC, 1989.

[61]V. J. Watson, B. E. Logan, Electrochem. Commun., 13, 54 (2011).

[62]Y. Alvarez-Gallego, X. Dominguez-Benetton, D. Pant, L. Diels, K. Vanbroekhoven, I. Genné, P. Vermeiren, Electrochimica Acta. 82, 415 (2012).

[63]X. Zhang, D. Pant, F. Zhang, J. Liu, W. He, B. E. Logan, ChemElectroChem. 1, 1859 (2014).

[64]D. Montgomery, Design and analysis of experiments. 3rd ed John Wiley & Sons, New York, 1991.

[65]A. Wang, D. Sun, N. Ren, Ch. Liu, W. Liu, B. E. Logan, W. Wei-Min, Bioresour. Technol., 101, 5733 (2010).

[66]L. Aino-Maija, O. H. Tuovinen, J. A. Puhakka, Bioenergy Res., 5, 481 (2012).

[67]S. Ishii, S. Suzuki, T. M. Norden-Krichmar, K. H. Nealson, Y. Sekiguchi, Y. A. Gorby, O. Bretschger, PLoS ONE 7(2): e30495. doi:10.1371/journal.pone.0030495, 2012.

[68]G. Zhang, Q. Zhao, Y. Jiao, K. Wang, L. Duu-Jong, N. Ren, Water Res., 46, 43 (2012).

[69]C. Garibay-Orijel, E. Rios-Leal, J. Garcia-Mena, H. M. Poggi-Varaldo, J. Chem. Technol. Biotechnol., 80, 1180 (2005).

[70]J. Galíndez-Mayer, J. Ramón-Gallegos, N. Ruiz-Ordaz, C. Juárez-Ramírez, A. Salmerón-Alcocer, H. M. Poggi-Varaldo, Biochem. Eng. J., 38, 147 (2008).

[71]Y. A. Çengel, M. A. Boles, Thermodynamics: An Engineering Approach, 5th ed, McGraw-Hill, 2006.

[72]H. M. Poggi-Varaldo, G. Hernandez-Flores, Estimation of energy thresholds in microbial fuel cells and other commercial bioenergy processes. Internal Report, CINVESTAV del IPN, Mexico, 2013.

[73]H. M. Poggi-Varaldo, L. M. Alzate-Gaviria, A. Perez-Hernandez, V. G. Nevarez-Morillon, N. Rinderknecht-Seijas, Waste Manage. Res., 23, 270 (2005).

[74]C. D. De Bazua, M. A. Cabrero, H. M. Poggi-Varaldo, Bioresour. Technol., 35, 87 (1991).

[75]H. M. Poggi-Varaldo, L.Valdés, F. J. Esparza-García, G. Fernández-Villagómez, Water Sci. Technol., 35, 197 (1997).

[76]P. Aelterman, K. Rabaey, T. H. Pham, N. Boon, W. Verstraete, Environ. Sci. Technol., 40, 3388 (2006).

[77]T. H. Pham, K. Rabaey, P. Aelterman, P. Clauwaert, L. De Schamphelaire, N. Boon, W. Verstraete, Eng. Life Sci., 6, 285

(2006).

[78]D. Stolten, B. Emonts. Fuel cell science and engineering: materials, processes, systems and technology, 1st ed., Wiley-VCH Verlag GmbH & Co. KGaA, 2012.

[79]http://app.cfe.gob.mx/Aplicaciones/CCFE/Tarifas/Tarifas/Tarifas_casa.asp?Tarifa= DACTAR1&anio=2013.

[80]I. Valdez-Vazquez, H. M. Poggi-Varaldo, International Journal of Hydrogen Energy, 34, 3639 (2009).

[81]J. Galíndez-Mayer, J. Ramón-Gallegos, N. Ruiz-Ordaz, C. Juárez-Ramírez, A. Salmerón-Alcocer, H.M. Poggi-Varaldo, Biochemical Engineering Journal, 8, 147 (2008).

[82]J. F. Buenronstro-Zagal, A. Ramirez-Oliva, S. Caffarel-Mendez, B. Schettino-Bermudez, H. M. Poggi-Varaldo, Water Science and Technology, 42, 185 (2000).