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Maximum anode chamber volume and minimum anode area for supporting electrogenesis in microbial fuel cells treating wastewater

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During scale-up of microbial fuel cell (MFC), a proportional increment in power does not usually occur and in design determining the maximum possible anode chamber volume (V_{an}) is important to exploit electrogenesis to achieve maximum energy recovery. A systematic approach is proposed for determination of the optimal single anode chamber volume and the minimum anode surface area (A_{an}) of a MFC. The optimal anode chamber volume was estimated based on the substrate required to produce a defined maximum current that is likely to be produced from the basic electromotive force equation. The A_{an} was obtained by considering the area required for biofilm formation, the substrate utilization rate by electrogens, the MFC polarization curve, charge transfer kinetics and mass transport overpotential. Based on theoretical bio-electrochemical considerations, the maximum V_{an} and minimum A_{an} required for each anode chamber are proposed for electrogenesis to dominate. A single V_{an} of a few litres will only be optimal for treating wastewater. With wastewater of chemical oxygen demand (COD) of 5 g L⁻¹ and considering a Coulombic efficiency and a COD removal of 80% each, a V_{an} of 2.02 L is optimum for a single anode chamber to produce a current up to 750 mA; which is the maximum possible current estimated from electromotive force equation. Any additional volume provided will leave substrate unused by electrogens and encourage methanogenesis. Adopting this volume for each anode chamber in a MFC stack is recommended for treating wastewater under the assumptions of the analysis.

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Charge transfer kinetics dominate the minimum A_{an} required, which satisfies the area required for biofilm formation, MFC polarization and mass transfer. This minimum A_{an} should be provided in a MFC to ensure the dominance of electrogenesis.

I. INTRODUCTION

A microbial fuel cell (MFC) is a device that generates power through the metabolic activity of electrochemically active bacteria using organic matter as the fuel substrate. Only limited studies have addressed scale-up of MFCs to enable this technology to move from laboratory studies to full scale implementation. Most of the scale-up studies suggest that the power output of a MFC does not increase directly in proportion to an increase in anode chamber volume.¹ When operating MFCs with larger anode chamber volume, limitations in performance are expected from electrochemical thermodynamics, electrode reaction kinetics, ionic and mass transport limitations, as well as variations in microbial activity.

Developing a mathematical model for MFC is not a straightforward process and needs better understanding of biochemical reactions and associated electrochemistry.² Different MFC models have been reported to study the effects of operating parameters on the performance of MFCs. Marcus *et al.*³ developed a dynamic conduction model to explain electron transfer based on both substrate oxidation and endogenous respiration and reported that the conductivity of biofilm is one of the major limiting factors influencing the current produced by a MFC.

A mathematical model for MFC, integrated with anaerobic digestion, showed that using a smaller external resistance (R_{ext}) increased the growth of electrogenic bacteria on the anode rather than methanogenic bacteria and enhanced the power output of MFC.⁴ Wen *et al.*⁵ developed a voltage, current density model based on polarization curves and showed that

activation and concentration losses had a greater influence on performance of the MFC, as compared to the Ohmic losses.

An optimal anode volume was considered by Fornero *et al.*⁶ from an economic point of view, when using a MFC for treatment of wastewater. Dewan *et al.*⁷ established a relationship between the surface area of the anode and power density of MFCs and reported that there was a decrease in power density as the surface area of the anode was increased. Merkey and Chopp⁸ created a two-dimensional model to study the relationship between the configuration of the MFC and power output and concluded that an increase in the number of anodes, while keeping a constant anode surface area, had no significant effect on current production.

For commercial applications of MFCs to treat a given wastewater flow, it is required to have information on maximum possible volume of the anode chamber (V_{an}) to support electrogenesis and the minimum surface area required for the anode (A_{an}), so that the number of anode chambers required can be estimated. Considering the bio-electrochemical reactions occurring in the anode chamber, the present literature has not really addressed the maximum single V_{an} and minimum A_{an} requirements, to satisfy the electrochemical phenomenon occurring in MFC.

This study describes an approach to determine the maximum possible volume for the anode chamber and the minimum anode surface area required to ensure that electrogenesis dominates in MFC. Using the background knowledge of electrode kinetics and their limitations, as reported in the literature for scalable MFCs and the results of the best performing MFCs, this study investigated the effect of non-catalyzed and catalyzed cathode materials on required anode chamber volume and anode surface area. The electrochemical considerations are further discussed by referring to results obtained after evaluating MFC

performance with different anode surface areas in presence or absence of catalyst on the cathode.

II. THEORETICAL CONSIDERATIONS AND EXPERIMENTAL WORK

The maximum current generated in a MFC can be estimated from the predicted working voltage based on thermodynamics and electrochemistry, after systematic deduction of losses from overall cell electromotive force (or voltage) equation. For supporting this current, a required substrate utilization rate has been considered to estimate V_{an} assuming a 12 h hydraulic retention time (HRT); whereas, A_{an} is estimated to satisfy: (a) the area required for biofilm formation to achieve this substrate utilization rate, (b) polarization behaviour, (c) electrode kinetics, and (d) mass transfer limitations. Nature of the electrode material influences both V_{an} and A_{an} , hence they are determined for catalyzed as well as non-catalyzed cathodes for treatment of wastewater having different chemical oxygen demand (COD) values.

A. Voltage generation in MFC

A MFC basically works as a power source with an internal resistance. In a MFC, the actual voltage output (V) is smaller than the theoretical thermodynamic value. The internal resistance (R_{int}) of MFC vary with electric current (I).⁹ The working voltage of a MFC can be estimated by subtracting voltage losses from its electromotive force using Eq. (1).

$$V = E_t - \eta_{act} - \eta_{ohmic} - \eta_{conc} \quad (1)$$

where, V is the voltage, V; E_t is the electromotive force, V; η_{act} , η_{ohmic} and η_{conc} are voltage losses due to reaction kinetics, Ohmic polarization and mass transport, V, respectively. If we substitute $IR_{int} = (\eta_{act} + \eta_{ohmic} + \eta_{conc})$, then Eq. 1 can be modified as Eq. 2:

$$V = E_t - I R_{int}. \quad (2)$$

where, I is the current (A) and R_{int} is the internal resistance (Ω).

Eq. 2 is the cell voltage equation, which enables to estimate the maximum possible current that a single MFC will generate from the best reported value for E_t in literature, the working voltage V , and lowest achievable R_{int} in MFCs. In practice, E_t is not the maximum voltage possible, as the potential at open circuit is much lower due to several factors such as the heterogeneous nature of the electrode, fuel and oxidant crossover and other non-ideal behavior; hence best reported value of E_t in the literature is used.

B. Substrate utilization in MFC

A series of biological reactions occur inside the anode chamber of a MFC with participation of both suspended biomass and biomass attached to the anode. Electroactive bacteria require a substrate/electron donor present in the wastewater as an energy source for their metabolism. Depending upon the type of substrate, the energy released will vary and affect microbial growth rate and the power output. The substrate loading to the anode chamber is defined by the organic loading rate (OLR) and sludge loading rate (SLR), which define the quantity of COD applied, per unit volume of anode chamber per day and per unit mass of microorganisms per day, respectively. Theoretically, the maximum amount of electric charge generated from a wastewater with a certain COD value can be estimated from Faraday's law of electrolysis using Eq. (3).¹⁰

$$C_{th} = F b_{es} M_s \quad (3)$$

where, C_{th} is theoretical amount of electrical charge produced (Coulombs), F is Faraday's constant (96485 C mol^{-1}), b_{es} equals to 4 moles of electron produced per mole of substrate measured as COD, M_s is the mole of COD added.

The C_{th} can be expressed in terms of the current produced (assuming constant in this case) to obtain the amount of COD required for the desired current or vice versa using (Eq. 4):

$$C_{th} = I t \quad (4)$$

where, t is the time in second.

Combining Eq. 3 and Eq. 4, the relation between M_s and I can be established (Eq. 5):

$$M_s = \left(\frac{I t}{F b_{es}} \right) a \quad (5)$$

Where, a is a multiplying factor used based upon the assumption that some of the substrate supplied to anode chamber will be utilized by electroactive and fermentative microbes for cell metabolism and synthesis and the remaining substrate is used for electrogenesis. This latter substrate consumption can be considered to be up to 20% of the total substrate supplied and hence ' a ' is considered here as equal to 1.2. Therefore, to estimate the amount of COD utilized to produce maximum possible current (as per Eq. 2) from a MFC, Eq. 5 can be expressed in terms of 'g' COD (Eq. 6):

$$\text{'g' of COD} = \left(\frac{8 I t}{F} \right) 1.20 \quad (6)$$

C. Estimating the optimal volume of anode chamber

The optimal anode chamber volume can be estimated on the basis of the substrate required to produce a maximum current (from Eq. 2) for a given wastewater COD and substrate utilization rate by electrogens. Any additional COD supplied in excess of substrate required for electrogenesis will favour methanogenesis. The COD removal efficiency expected in the anode chamber of MFC can be considered as 70-80%. Once the maximum possible single anode chamber volume is determined from the appropriate COD removal efficiency (80% considered here), the number of anode chambers required for treatment of a given wastewater flow can be estimated by dividing the total COD load present in the wastewater by the COD load handled by each anode chamber.

The minimum working voltage to be generated by the MFC is considered as 0.30 V which for a MFC is usually in the Ohmic region of the polarization curve.¹¹ To determine the maximum limit of the substrate that can be utilized for electrogenesis, a maximum current of

16 mA ($E_t = 0.81$ V, $R_{int} = 31.87 \Omega$) reported for a scalable MFC using non-catalyzed cathode material is considered.¹² Moreover, a best possible current of 150 mA ($E_t = 0.850$ V, $R_{int} = 3.67 \Omega$), as reported by Ter Heijne *et al.*¹³, is considered to estimate substrate utilization limit for electrogenesis. This MFC had titanium mesh electrodes coated with platinum and iridium for both anode and cathode respectively, and ferricyanide solution was used as the catholyte.

D. Estimating the minimum anode area required

1. Based on substrate utilization and biofilm formation

Biofilm formed on the anode of MFCs is generally thin in nature, ranging from a monolayer to a maximum thickness of around 40 μm .¹⁴ The weight of biomass can be estimated by considering the SLR of 0.40 kg COD kg VSS⁻¹ d⁻¹¹² and the volume of biomass can be estimated by assuming a specific gravity of 1.03 g/cc for a typical bacterial cell mass. Finally, the anode area required for biofilm formation can be estimated by dividing the volume of biomass by the biofilm thickness (40 μm); although, in reality this anode biofilm thickness will not be uniform.

2. Anode surface area required as per polarization curve

In a MFC, all the electrochemical phenomena are considered to occur at the electrode-electrolyte interface. In a full scale MFC stack, to maintain power output at any given time during fuel cell operation, the average working voltage and minimum cell voltages are considered as 0.525 V and 0.3 V, respectively.¹¹ Therefore, in the present study the lower current density (I_{min}) at 0.5 V and the upper limit of the current density (I_{max}), corresponding to 0.3 V are considered to calculate the A_{an} from the current/current density ratio.

3. Anode area required to satisfy Butler-Volmer reaction kinetics

In an electrochemical system, transport of charge occurs across the interface between an electronic conductor (an electrode) and an ionic conductor (an electrolyte). When an electric potential (E) is applied, charge is transported through the movement of electrons through the electrode and ions through the electrolyte.¹⁵ The exchange current density (i_0) defines the rate at which electrons are transferred between an electrode and electron shuttle or donor present in the electrolyte.¹⁵ The overpotentials at the anode and cathode can be estimated from Eq. 7 and 8:¹⁶

$$\eta_a = -\frac{RT}{n\alpha_a F} [\ln(i_0) - \ln(i)] \quad (7)$$

$$\eta_c = -\frac{RT}{n\alpha_c F} [\ln(i) - \ln(i_0)] \quad (8)$$

where, η_a is anodic activation loss, η_c is cathodic activation loss, R is the universal gas constant in J/mol.K, T is the temperature in K, n is the number of electrons involved in the charge transfer reaction, F is the Faraday's constant in C mol⁻¹, α_a and α_c are charge transfer coefficients for anode and cathode and i_0 is the exchange current density.

To estimate the overpotentials at anode (η_a) and cathode (η_c) to support maximum current density (I_{cd}), low values of charge transfer coefficient and higher i_0 values are required to be considered. For estimating the required anode surface area for a MFC with non-catalyzed cathode, α_a of 0.32, α_c of 0.53, and i_0 of 0.52×10^{-3} mA m⁻² and 2.25×10^{-3} mA m⁻² are considered for the anode and cathode, respectively.¹² While for MFC with catalyzed cathode, $\alpha_a = 0.05$ and $\alpha_c = 0.23$ and i_0 of 1.83 A m⁻² and 1.0 A m⁻², respectively for anode and cathode, are assumed to estimate overpotentials.^{2, 17}

4. Effect of mass transport overpotential on electrode area

Mass transfer is an important process that occurs in an electrochemical cell, which significantly affects performance. To obtain high energy conversion efficiency in a direct electrochemical process, it is necessary to improve performance of the two sequential steps:

first the mass transport rate of electrochemically active species to the electrode surface and second, the rate at which they are oxidized or reduced.¹⁸ When a high current is drawn from a MFC, a rapid consumption of substrate creates a difference in concentration between the bulk liquid and electrode-solution interface, which can limit mass transport by diffusion to the electrode surface. The expression of the mass transport overpotential is given by Eq. 9:¹⁶

$$\eta_t = \frac{RT}{nF} \left[\ln \frac{I_{a,d}}{-I_{c,d}} + \ln \frac{-I_{c,d} + I}{I_{a,d} - I} \right] \quad (9)$$

where, $I_{a,d}$ and $I_{c,d}$ are the rate limiting diffusion currents for the anodic and cathodic reactions, respectively.

The maximum value of I_L can be calculated from Eq. 10:¹⁵

$$I_L = n F k C_0 \quad (10)$$

where, I_L is the limiting current density in $A\ m^{-2}$, k is the mass transfer coefficient in $m\ s^{-1}$, C_0 is the concentration of electroactive species in solution in $mol\ m^{-3}$.

The I_L is further used to estimate actual current density (I_{cd}) that can be obtained at a particular electrode using Eq. 11, and thus area required for anode can be obtained by dividing desired current (as per Eq. 2) with current density.¹⁶

$$I_{cd} = I_L \left(1 - e^{\frac{-nF\eta_t}{RT}} \right) \quad (11)$$

where, η_t is mass transport overpotential in V.

E. Performance evaluation of MFCs using catalyzed and non-catalyzed cathodes

Experiments were performed using MFCs with catalyzed and non-catalyzed cathodes to measure current output and wastewater treatment efficiency for the MFCs. The performance obtained was validated using the bio-electrochemical relations discussed earlier.

1. Microbial fuel cell construction and operation

A set of eight air cathode MFCs were fabricated using clayware ceramic cylinders. The observed performance was cross checked with earlier explained bio-electrochemical

behaviour to understand limitations in performance. The anode chamber working volume was 0.175 L for MFC NC-1, MFC NC-2, MFC NC-3 and MFC NC-4 having non-catalyzed cathodes; and the anode chamber working volume was 1.61 L for MFC C-1, MFC C-2, MFC C-3 and MFC C-4 with catalyzed cathodes. Carbon felt (Panex® 35, Zoltek Corporation USA) was used as the anode in all the MFCs. In MFC NC-1, MFC NC-2, MFC NC-3 and MFC NC-4 the separator electrode assemblies (SEAs) were used as cathode without any catalyst (Fig. 1). The SEAs were made by coating the outer surface of the ceramic cylinder with a conductive carbon ink prepared using Vulcan XC-72R carbon powder (Cabot corporation, India) with a loading of 0.5 mg/cm², without any catalyst and polyvinyl alcohol (PVA) as binder at a loading of 0.5 mg/cm². The same carbon ink incorporated with palladium catalyst (Sigma-Aldrich, India) at a loading of 0.25 mg/cm² was used to make the catalyzed cathodes in MFC C-1, MFC C-2, MFC C-3 and MFC C-4 (Fig. 1).

FIG.1. Schematic diagram showing operating condition of different MFCs

MFC NC-1 and MFC NC-2 were operated in duplicate with A_{an} of 290 cm²; while, MFC NC-3 and MFC NC-4 had values of A_{an} of 145 cm² and 435 cm², respectively (Table I). MFC C-1 and MFC C-2 were operated in duplicate and had values of A_{an} of 420 cm²; while MFC C-3 and MFC C-4 were provided with A_{an} of 210 cm² and 630 cm², respectively.

The MFCs were inoculated with pure culture of electrogenic bacteria *Pseudomonas aeruginosa* (MTCC-12307, Chandigarh, India) having a cell mass (dry weight) concentration of 0.6 g/L in liquid medium. 34 mL and 322 mL were used in the anode chambers of MFCs with non-catalyzed and catalyzed cathode, respectively (Table I). The performance of the MFCs was evaluated for a total duration of three months under a fed batch mode with a fresh feeding interval of 5 days. Synthetic wastewater containing acetate as a source of carbon was used as substrate; which was based on the composition given by Jadhav and Ghangrekar.¹⁹

TABLE I. Experimental operating conditions for MFCs using non-catalyzed and catalyzed cathode

MFC type	V_{an} (L)	A_{an} (cm ²)	Anode	Cathode	Influent COD (g L ⁻¹)
MFC NC-1	0.175	290	C.felt	SEA , No catalyst	7.8
MFC NC-2	0.175	290	C.felt	SEA , No catalyst	7.8
MFC NC-3	0.175	145	C.felt	SEA , No catalyst	7.8
MFC NC-4	0.175	435	C.felt	SEA , No catalyst	7.8
MFC C-1	1.610	420	C.felt	SEA , Pd catalyst	7.8
MFC C-2	1.610	420	C.felt	SEA , Pd catalyst	7.8
MFC C-3	1.610	210	C.felt	SEA , Pd catalyst	7.8
MFC C-4	1.610	630	C.felt	SEA , Pd catalyst	7.8

V_{an} -Anodic chamber volume, A_{an} -Anode surface area, C.felt- carbon felt, SEA- separator electrode assembly made from conductive carbon ink, Pd- Palladium

2. Analysis and calculations

The COD concentrations and pH of the wastewater samples, and the volatile suspended solids (VSS) of the inoculum were measured according to Standard Methods.²⁰

The electrical performance of MFCs was evaluated in terms of V and I , measured using a data

acquisition unit (PicoLog data logger, UK), and converted to power according to $P = V I$, where P = power (W). Polarization studies were performed by changing R_{ext} from 10,000 Ω to 10 Ω in steps using the variable resistance box (GEC 05 R Decade Resistance Box, Renown Systems, Kolkata, India). The voltage across the resistor was recorded every 30 min using a data acquisition unit and polarizations were performed after attaining a stable cell potential.²¹ The Coulombic efficiency (CE) was estimated by integrating the measured current relative to the theoretical current using (Eq. 12).¹⁰

$$CE = \frac{M \int_0^t I dt}{F b_{es} v \Delta COD} \quad (12)$$

where, M is molecular weight of oxygen; I is the current measured (A); F is Faraday's constant = 96485 C mol⁻¹; b_{es} is the number of electrons exchanged per mole of oxygen; v is the volume of the anode chamber of MFC (L); ΔCOD is the difference in the initial and final (COD g L⁻¹).

3. Electrochemical characterization

Cyclic voltammetry (CV) was performed by varying cell potential from - 1 to + 1 V at a slow scan rate of 10 mV/s using a potentiostat (Autolab PGSTAT 302 N, The Netherlands). A three electrode cell arrangement was used, consisting of carbon felt anode as working electrode, Ag/AgCl as reference electrode and a platinum wire as counter electrode. The capacitance of the electrode was calculated according to Khomenko *et al.*²² using Eq. 13.

$$I = C \frac{dE}{dt} \quad (13)$$

where, I is the average current, A; C is the capacitance in Farad and $\frac{dE}{dt}$ is the potential scan rate.

Electrochemical Impedance Spectroscopy (EIS) measurements were performed over a frequency range of 100 kHz to 1 mHz with an AC signal of 10 mV amplitude using Autolab PGSTAT 302N Potentiostat (Metrohm, The Netherlands) and software NOVA 1.9. The

solution resistance (R_s), charge transfer resistance (R_{ct}) and diffusion resistance were determined from Nyquist plot and equivalent circuits used to characterize the EIS.

III. RESULTS AND DISCUSSION

A. Maximum utilizable single anode chamber volume

By adopting the procedure explained in section II.C, the maximum possible single anode chamber volume (V_{an}) was estimated so that electrogenesis can be dominant in the MFC. For treatment of a typical wastewater, having COD concentration of 0.5 g L^{-1} with an applied OLR of 1.0 and $2.0 \text{ g COD L}^{-1} \text{ d}^{-1}$, the anode chamber volume and the required anode surface area for biofilm formation were estimated for MFCs using catalyzed and non-catalyzed cathodes (Table S1).³⁴ Theoretical calculations showed that at 80% CE, the maximum V_{an} of 2.02 L and 0.214 L can support currents of 150 mA and 16 mA, respectively for MFCs with catalyzed and non-catalyzed cathode (Table S1).³⁴ Most of previous research reported higher CE at low V_{an} (few mL), and failed to achieve CE above 20% in scalable MFCs.^{23, 24, 25} Even at 20% CE, the maximum possible V_{an} that can be supported will be 8.06 L and 0.86 L, respectively for MFC with catalyzed and non-catalyzed cathode (Table S1).³⁴ If a greater volume than this is provided, it will reduce the CE because there is potential scope for methanogenes to utilize substrate. Also a larger volume may result in higher mass transport loss. Since the current density supported by the MFC-C is about seven times higher than that of MFC-NC, it can have a larger V_{an} due to higher substrate consumption to producing a higher current.

The quantity of substrate ('g' COD) utilized depends upon the current generated (Eq. 6). When the concentration of COD in the wastewater is changed from 0.5 to 5 g L^{-1} and the OLR is changed from 1 to $5 \text{ g COD L}^{-1} \text{ d}^{-1}$, the current produced can be increased from 150 to 750 mA in the MFC using a catalyzed cathode. Such relatively high current can be

supported by catalyzed anodes, as reported earlier by TerHeijne, who obtained a relatively high current density of 5.0 A m^{-2} with R_{int} of 0.73Ω .¹³ However, V_{an} of 1.61 L at 100% CE remains unchanged due to proportional change in flow rate (Table S2).³⁴ In the case of MFC with a non-catalyzed cathode, this change in OLR and COD concentration cannot increase the current above 16 mA because of the limitations of internal resistance. To reduce the internal resistance further from R_{int} of 31.87Ω , a fivefold decrease in anode chamber volume is required (0.172 to 0.034 L) to achieve 100% CE (Table S1, Table S2).³⁴

At lower CE, more substrate is required in the anode chamber due to a lower fraction of substrate being utilized for electrogenesis; hence the required V_{an} becomes larger for a MFC using both catalyzed as well as non-catalyzed cathode. Use of a catalyzed cathode enables a higher single anode chamber volume; hence, such MFCs will have greater prospects for commercialization due to the lower number of MFCs required for treatment of given wastewater flows. Considering very large number of anode chambers required for MFC using a non-catalyzed cathode, it appears that this type will have very limited application in practice for full scale implementation (Table S2).³⁴

With an increase in COD concentration, the anode chamber volume required to support electrogenesis falls and in the case considered it is inferred that single anode chamber volume of a few litres can only ensure electrogenesis. Hence for the treatment of high strength wastewater (with COD more than 5000 mg/L) using MFCs, a greater number of anode chambers with smaller volume will be required, because of the higher energy content of the feed compared to low strength wastewaters such as municipal wastewater.

The present analysis suggests, large wastewater flows with high strength cannot be economically treated by MFC alone, due to the large number of anode chambers and MFCs required. Therefore, an alternative is to use hybrid reactors, such as up-flow anaerobic sludge blanket (UASB) reactor coupled with a MFC. Such hybrid reactor can produce methane and

also reduce the organic pollutant drastically by methanogenesis and then further “polishing treatment” can be achieved by MFCs. In this case, performance of MFC can also be used as an indicator of performance of UASB reactor for real online monitoring. Hence, considering the maximum volume of each anode chamber and the number of chambers required (Table S2),³⁴ targeting energy recovery only by electrogenesis from higher flow and higher strength wastewater might not be economically feasible using presently available electrode materials. To increase the anode chamber volume, it is necessary to configure MFCs to offer least overall internal resistance.

B. Anode surface area required

When the MFC offers a higher CE, then it can have a lower anodic surface area (Table S1).³⁴ This is due to the fact that anode surface area depends upon the volume of biomass attached to the anode, to support specific SLR. This required biofilm area will increase with lower CE because of the lower substrate utilization rate for electrogenesis. However, the anode surface area required for MFC with non-catalyzed cathode remains unaffected by variation in COD concentration and OLR, since COD utilized (g d^{-1}) per anode chamber is fixed with limiting currents (Table S1, Table S2).³⁴ Whereas, in case of MFC with catalyzed cathode a change in OLR from 1 to 2 $\text{g COD L}^{-1} \text{d}^{-1}$ can increase the current from 150 to 300 mA and hence the required A_{an} will be 0.195 m^2 (Table S1).³⁴ This effect is even more pronounced when the COD is changed from 0.5 to 5 g L^{-1} and the OLR is increased from 2 to 5 $\text{g COD L}^{-1} \text{d}^{-1}$; which increases the current from 300 to 750 mA and demands a higher A_{an} of 0.489 m^2 (Table S2).³⁴

Results obtained from the polarization behavior indicate that areas of 0.029 m^2 and 0.21 m^2 are required, respectively for MFCs with non-catalyzed cathode and catalyzed cathode (Table II). A MFC with a catalyzed cathode requires 7.25 times more anode surface

area than a MFC with non-catalyzed cathode to support 47 times more current. In spite of having a high exchange current, the kinetics require a higher A_{an} of 1.49 m² for MFC using catalyzed cathode to support a higher current (750 mA) as compared to A_{an} of 0.355 m² for a MFC producing a small current (16 mA) using non-catalyzed cathode (Table II). For the values of i_0 and α_a used in this study, the anode area required to satisfy this Butler-Volmer kinetics is higher than A_{an} of 0.029 and 0.21 m² required for polarization and A_{an} of 0.010 and 0.489 m² required for biofilm formation to support substrate oxidation in MFC using non-catalyzed and catalyzed cathode, respectively. Required anode area based on the mass transport analysis is much lower than any other phenomenon discussed above (Table II). Hence, charge transfer kinetics decides the minimum anode surface area required.

TABLE II. Anode surface area required under different approaches to support maximum current generation.

Approach	Parameter	Quantity for MFC with non-catalyzed cathode*	Quantity for MFC with catalyzed cathode**
		$I = 16 \text{ mA}$, CE = 100%	$I = 750 \text{ mA}$, CE = 100%
Substrate utilization	W_{bm}	0.43 g	20.16 g
	A_{an}	0.010 m ²	0.489 m ²
Polarization behavior	I_{max}	0.71 A m ⁻²	5.4 A m ⁻²
	I_{min}	0.54 A m ⁻²	3.6 A m ⁻²
	A_{an}	0.022- 0.029 m ²	0.14-0.21 m ²
Charge transfer kinetics	i	0.045 A m ⁻²	0.502 A m ⁻²
	A_{an}	0.355 m ²	1.494 m ²
Mass transport phenomenon	I_L	61.35 A m ⁻²	61.35 A m ⁻²
	A_{an}	2.61 x 10 ⁻⁴ m ²	2.44 x 10 ⁻³ m ²

W_{bm} – weight of biomass, A_{an} – anode area required, I_{max} – maximum current density (at 0.3V), I_{min} – minimum current density (at 0.5 V), I – electrode current density, I_L – limiting current density

It is well documented that, current density-overpotential relationship in an electrochemical reaction is influenced by the rate of diffusion of a reactant species from the bulk electrolyte to the interface.²⁶ However; anode surface area required is not limited by

mass transport limitations. Since the activation loss is the predominant factor that affects the rate of electrochemical transformation and power output from a fuel cell, hence Butler-Volmer kinetics decides the anode surface area required.²⁷ Therefore, anode surface area is dominated by Butler-Volmer kinetics; hence, it is necessary to select A_{an} obtained from Butler-Volmer kinetics which will support all other bio-electrochemical reactions.

C. Experimental Investigation

1. Wastewater treatment

Average COD removal efficiencies of $81 \pm 3\%$, $80 \pm 4\%$ and $83 \pm 4\%$ were observed in MFC NC-1, MFC NC-2 and MFC NC-4, respectively, at an OLR of $2.0 \text{ g COD L}^{-1} \text{ d}^{-1}$ (Fig. 2). Application of a catalyzed cathode achieved relatively higher COD removals of $82 \pm 5\%$, $84 \pm 3\%$ and $86 \pm 4\%$, in MFC C-1, MFC C-2 and MFC C-4, respectively. However, a lower COD removal of $74 \pm 3\%$ and $76 \pm 4\%$ was observed in MFC NC-3 and MFC C-3, this can be attributed to the inadequate anode area (50% less) available which is even less for substrate oxidation in these MFCs. The results obtained are comparable with COD removals reported in the literature for scalable MFCs.^{12,18}

2. Coulombic efficiency

Coulombic efficiencies of $20.5 \pm 0.8 \%$ and $24.5 \pm 0.8 \%$ were observed in MFC NC-1 and MFC NC-2, respectively (Fig. 2). A higher CE of $27.1 \pm 1.3 \%$ was obtained for MFC NC-4, due to the 1.5 times larger anode surface area provided in this MFC, as compared to MFC NC-1 and MFC NC-2. However, provision of 50% lower anode surface area adversely affected the performance of MFC NC-3, resulting in a CE of only $9.9 \pm 0.4 \%$. A lower CE was due to the lower anode area available for biofilm formation and a thicker diffusion layer formed near the biofilm, increasing the concentration gradient and hence producing lower

current as compared to other MFCs. The CEs obtained in these MFCs were higher than that reported for scalable MFC (9.1%) with V_{an} of 3.75 L using non-catalyzed cathodes.¹²

MFC C-1, MFC C-2, MFC C-3 and MFC C-4 demonstrated CE of $8.9 \pm 0.4 \%$, $7.8 \pm 0.3 \%$, $3.98 \pm 0.39 \%$ and $9.7 \pm 0.7\%$, respectively (Fig. 2). The lowest CE in MFC C-3 was due to less anode area available for bio-electrochemical reactions in the anodic chamber. The current produced by MFC with catalyzed cathode was three times higher than that of MFC with non-catalyzed cathode. However, due to the 9.2 times higher V_{an} of MFC with catalyzed cathode, more than five times increase in Ohmic resistance occurred (as discussed later), than expected to support the higher current close to the theoretically assumed current of 150 mA, and hence the experimentally observed CE values were lower. However, these values of CEs are higher than that reported in literature with scalable MFCs (V_{an} of 26 L and CE = 5.1%; V_{an} = 250 L and CE = $3 \pm 1 \%$) using catalyzed cathodes.^{23, 24}

FIG.2. COD removal and CE observed in different MFCs.

3. Voltage generation in MFCs

MFCs using non-catalyzed cathodes demonstrated open circuit voltage (V_{OCV}) in the range of 512 - 543 mV, whilst MFC with catalyzed cathode showed V_{OCV} in the range of 661 - 674 mV (Table III). The average operating voltages (V_{OV}) of 308 ± 7 mV, 316 ± 5 mV and 337 ± 8 mV across R_{ext} of 100Ω was observed in MFC NC-1, MFC NC-2 and MFC NC-4, respectively. Comparatively, a lower V_{OV} of 240 ± 5 mV observed in MFC NC-3 indicates that inadequate A_{an} might have reduced charge transfer kinetics at the anode, and resulted in increase of overvoltage. MFCs using catalyzed cathode demonstrated V_{OV} of 430 ± 5 mV, 438 ± 7 mV, 343 ± 4 mV and 468 ± 6 mV for MFC C-1, MFC C-2, MFC C-3 and MFC C-4, respectively (Table III). Results from this study indicate that providing 1.5 times A_{an} resulted in a marginal increase in V_{OV} ; whereas, 50% less A_{an} drastically decreased voltage in these MFCs.

TABLE III. Electrical parameters observed under different operating conditions in MFCs.

MFC type	V_{OCV} , mV	V_{OV} , mV	I_{max} , mA	P_{max} , mW	C , μFcm^{-2}
MFC NC-1	522 ± 9	318 ± 7	6.87	1.62	1640
MFC NC-2	531 ± 10	306 ± 5	7.63	1.75	1465
MFC NC-3	512 ± 8	240 ± 5	2.84	0.86	650
MFC NC-4	543 ± 6	337 ± 8	8.71	1.88	3407
MFC C-1	667 ± 7	430 ± 5	21.70	4.71	4150
MFC C-2	661 ± 8	438 ± 7	22.14	5.02	4470
MFC C-3	642 ± 5	343 ± 4	10.63	2.26	2854
MFC C-4	674 ± 9	468 ± 6	24.31	5.92	6305

V_{OCV} – open circuit voltage (mV), V_{OV} – operating voltage (mV), I_{max} – maximum current (mA), P_{max} – maximum power during polarization (mW), C – capacitance from CV (μFcm^{-2})

4. Comparing theoretical and actual current in MFC

A comparison was made between the current production based on theoretical considerations and actual current generated in experimental MFCs using non-catalyzed and

catalyzed cathode. Theoretical current was estimated using Eq. (2) by substituting the observed value of V_{ocv} (Table III) and R_{int} obtained from Nyquist plots during electrochemical impedance spectroscopy for each MFC with non-catalyzed and catalyzed cathodes. The experimentally observed currents of 6.37, 7.63, 2.84 and 8.71 mA in MFC NC-1, MFC NC-2, MFC NC-3 and MFC NC-4 slightly deviated from the theoretical predicted currents of 6.81, 7.94, 2.97 and 9.10 mA, respectively. Similarly, the observed currents of 21.7, 22.14, 10.63 and 24.31 mA in MFC C-1, MFC C-2, MFC C-3 and MFC C-4 marginally deviated from the theoretical value of 22.82, 23.20, 13.50 and 25.64 mA, respectively. These results indicate that the actual currents obtained in these MFCs are in close agreement with the theoretical current predicted using Eq. 2.

5. Polarization and power overshoot study

During polarization, MFC NC-1 and MFC NC-2 produced maximum powers (P_{max}) of 1.62 mW and 1.75 mW, respectively (Table III). However, MFC NC-3 demonstrated lower P_{max} of 0.86 mW; whereas, comparatively higher P_{max} of 1.88 mW was observed in MFC NC-4 (Fig. 3A). MFC C-1 and MFC C-2 demonstrated P_{max} of 4.71 mW and 5.02 mW, respectively. MFC C-4 delivered the highest P_{max} of 5.92 mW, whilst MFC C-3 gave relatively lower P_{max} of 2.26 mW (Fig. 3B).

Power overshoot is an important phenomenon that generally occurs in MFCs at higher current density during polarization. When a current limiting electrode (anode) limits supply of electrons to the cathode, a sudden voltage drop occurs and a double back is seen in the power density curve called power overshoot.²⁹ It is interesting to note here that power overshoot was not observed in MFC NC-1, MFC NC-2, MFC NC-4, MFC C-1, MFC C-2 and MFC C-4. However, in MFC NC-3 and MFC C-3 a double back observed in current-power plot indicated occurrence of power overshoot in these MFCs. Further, CV analysis also confirmed the presence of lower bio-capacitance in MFC NC-3 (650 $\mu\text{F}/\text{cm}^2$) and MFC C-3

(2854 $\mu\text{F}/\text{cm}^2$) as compared to MFC NC-1 (1640 $\mu\text{F}/\text{cm}^2$) and MFC C-1 (4150 $\mu\text{F}/\text{cm}^2$), due to reduced electron generating capacity of former MFCs due to 50% lower anode surface area (Table III).

FIG. 3 Polarization and power density curve for MFCs using (A) non-catalyzed cathode, (B) catalyzed cathode.

6. Electrochemical analysis

Cyclic voltammetry was carried out to characterize the electron transfer interactions between electroactive microbes and carbon felt anode in all the MFCs. The voltammogram shows a maximum bioelectrocatalytic activity with anodic peak current of 39.4 mA in MFC

NC-4. This is followed by MFC NC-1 and MFC NC-2 with respective oxidative current of 26.7 mA and 25.8 mA (Fig. 4A). Lower oxidative current (16.4 mA) recorded for MFC NC-3 indicates decreased electron producing capacity of anodic biofilm due to 50% lower A_{an} than MFC NC-1 and MFC NC-2, to support electrochemical activity at anode. The highest bio-electrochemical current of 127.3 mA was observed in MFC C-4, which confirms that anode surface of this MFC was adequate to support the bioelectrochemical reactions (Fig. 4B). As compared to this, MFC C-1 and MFC C-2 demonstrated lower anodic peak current of 53.4 mA and 44.5 mA, respectively (Fig. 4B); whereas, MFC C-3 supported a current of only 31.7 mA due to 50% lower A_{an} than MFC C-1 and MFC C-2. Cyclic voltammetry also confirmed the significance of A_{an} required for bioelectrochemical reactions and it is a critical factor influencing electricity generation in MFCs.

FIG. 4. Cyclic voltammogram for MFCs using (A) non-catalyzed cathode, and (B) catalyzed cathode.

7. Voltage losses in MFCs

From the polarization curve, (Fig. 3) it is clear that activation loss was dominating in MFC using non-catalyzed cathode, with η_{act} in the range of 200 - 230 mV; while the MFC using Pd catalyst on cathode showed η_{act} of 160 - 170 mV. To identify the main factors contributing to activation loss (η_{act}), anode and cathode overpotentials were evaluated separately using Tafel analysis. Anode overpotentials of 57 ± 7 mV and 48 ± 5 mV were obtained from the Tafel plot for MFC with non-catalyzed and catalyzed cathodes, respectively. In comparison, high cathode overpotentials of 168 ± 8 mV and 102 ± 6 mV were observed in MFC with non-catalyzed and catalyzed cathodes, respectively. Maximum anode exchange current densities (i_0) of 7.6×10^{-3} mA m⁻² and 18.2×10^{-3} mA m⁻² were determined for MFC NC-4 (with non-catalyzed cathode) and MFC C-4 (catalyzed cathode), respectively. MFC with Pd catalyst cathode demonstrated higher i_0 of 34.6×10^{-3} mA m⁻² (at cathode) compared to i_0 of 8.4×10^{-4} mA m⁻² at cathode of MFC without catalyst. However, experimentally observed exchange current densities were lower than that assumed in the section II D.3 due to higher overpotential in reality; hence, lower power output was observed in these MFCs.

8. Way forward

In order to enhance the power output and make MFC practicable, it is necessary to minimize η_{act} both at the anode and cathode by developing highly efficient electrode materials. The activation overpotential at the anode can be decreased by using a high surface area electrode material such as with deposited multi-walled carbon nanotube (CNTs) to support higher current density up to 6.0 A m⁻².³⁰ Also, CNTs/polyaniline composite has proven to be a better anode material, because it offers large surface area and excellent

conductivity. Additionally, application of polyaniline may also improve the electro-catalytic activity.³¹ To decrease the activation overpotential at the cathode, platinum (Pt) is most effective to reduce the cathodic activation energy and increase the oxygen reduction reaction rate. However, it is highly expensive and hence its practical application is limited. Therefore, application of cobalt tetramethoxy-phenylporphyrin (CoTMPP) and Fe-phthalocyanine (FePc) as cathode catalyst, which have demonstrated power density of 14.32 W m⁻³ and 13.88 W m⁻³, respectively, should be considered; since these are inexpensive compared to platinum.³²

Another major contributing factor to the voltage drop is Ohmic loss (η_{ohm}) that occurs due to resistance to the flow of electron through the electrode and to the flow of ions through the separator/membrane of MFC. In the present experiments, the Ohmic resistance was in the range of 27 - 41 Ω , and ideally it should be reduced to 15 Ω in MFC using non-catalyzed cathode, while significant reduction in Ohmic resistance from 13 - 19 Ω to 0.49 Ω is required in MFC with catalyzed cathodes to achieve current production close to 750 mA. The Ohmic loss can be minimized by reducing electrode spacing, use of improved conductivity electrode material with conductive coating and selecting good ion conductive membrane, so that the membrane separator selected should offer lower resistance. The ceramic separator (CS) used in the present study can be replaced with CS modified with cation exchanger to enhance proton conductivity³³ and decrease membrane resistance. Mass transport loss occurs at higher current densities and its magnitude is usually lower than η_{act} and η_{ohm} , and it can be reduced by providing suitable system architecture to ensure mixing.²⁵

In order to achieve a maximum CE, say 80%, in the present work using non catalyzed cathode, it is necessary to generate a current of 14.4 mA using anode materials which are capable of supporting a current density of 496 mA m⁻² with R_{int} of 17.2 Ω . The carbon felt anode material in this study can support current densities up to 710 mA m⁻².¹² Using a non-

catalyzed cathode, the maximum anode chamber volume to support a current of 14.4 mA will be 0.214 L at 80% CE, as compared to 0.172 L at 100% CE supporting a current of 14.4 mA (Table S1).³⁴ In MFC with a catalyzed cathode, to achieve a CE of 80%, it is necessary to produce a current of at least 685 mA using anode material supporting a current density of 3261 mA m⁻², which is not possible with bare carbon felt. Hence, a cathode catalyst must be used. Therefore, it is necessary to use an alternative anode material, such as titanium mesh coated with platinum which is reported to support higher current density of 5000 mA m⁻² and reduce R_{int} to 3.08 Ω .¹³ Using such catalyzed electrodes, the maximum anodic chamber volume that can be supported to ensure electrogenesis may increase from 1.61 L (using bare carbon felt anode) to 2.02 L (using catalyzed anode (Table S2)).³⁴

IV. CONCLUSION

The maximum possible single anode chamber volume (V_{an}) is proposed by establishing a relationship between maximum possible current produced, satisfying electrochemistry, and COD consumed to ensure dominance of electrogenesis in the anodic chamber of MFC. It is revealed that a smaller anode chamber can only help in dominance of electrogenesis in MFC; e.g. a V_{an} of 2.02 L is optimal for MFC with a catalyzed cathode to produce current up to 750 mA while treating wastewater with COD of 5 g L⁻¹. Bio-electrochemical phenomenon and experimental observations indicate that charge transfer kinetics dominates the minimum anode surface area required and lower anode area results in power overshoot. Commercialization of MFC is shown to be feasible only through the use of catalysed electrodes, so as to support higher current densities and making it possible to use higher single anode chamber volume so as to minimize number of anode chambers required, for extracting maximum energy in the form of electricity while treating wastewater.

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Appendix A. supplementary data

Supplementary data associated with this article attached in electronic annexure.

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parameters for different electrode materials for treating wastewater flow of 1000 L d⁻¹ with COD of 5 g L⁻¹ at OLR of 5 g COD L⁻¹ d⁻¹.