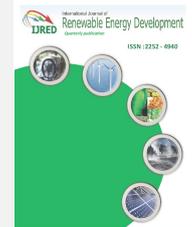




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Research Article

Performance and Techno-Economic Analysis of Scaling-up A Single-Chamber Yeast Microbial Fuel Cell as Dissolved Oxygen Biosensor

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ABSTRACT. The Microbial fuel cells (MFCs) are electrochemical devices that can be utilized as biosensors, specifically Dissolved Oxygen (DO) biosensors. In this research, performance and techno-economic of MFC-based DO biosensors with two sizes, small and large, were evaluated and analysed to determine whether it is more economical to use a small or large reactor. MFC-based DO biosensors were also applied to an irrigation canal. When MFC immersed into distilled water with several variations of DO, the correlation between DO and current density produced equation with R^2 values around 0.9989 and 0.9979 for SYMFC and LYMFC, respectively. The power density for SYMFC and LYMFC was 3.48 and 10.89 mW/m², respectively, in DO 6. Higher power densities are correlated with the electrode surface area, especially the larger cathodic surface area. When applied to the irrigation canal, DO values measured using SYMFC and LYMFC have errors of around 3.39 and 4.42%, respectively, when compared to DO values measured using DO meters. LYMFC requires a capital cost of around \$ 234.22 or 2.57 times higher than SYMFC, although it generates almost similar cost per mW/m², \$ 21.51 and \$ 26.23 for LYMFC and SYMFC, respectively. The results concluded that yeast MFC -based DO biosensors with smaller sizes can achieve more economical compared to larger sizes.

Keywords: Environmental Biosensor, Single-chamber MFC, Economic Analysis, Energy-to-cost Ratio, *Saccharomyces cerevisiae*

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1. Introduction

Microbial fuel cells (MFCs) are devices for oxidizing organic and inorganic materials and producing electricity by using microorganisms as biocatalysts (Logan and Regan, 2006; Rabaey *et al* 2006). Electrical energy can be generated by placing anode in anode chamber and feed them with anolyte which consisting substrates and biocatalysts, and then connecting them via electronic circuits to cathode electrodes containing catholyte in the form of oxygenated water (Bond *et al* 2002; Lowy *et al* 2006). The electrons produced by oxidation from organic matter in the substrate by microorganisms and travel through an external circuit to the cathode electrode. Meanwhile, protons diffuse into the cathode space via proton exchange membrane (PEM), where protons will react with electrons and oxygen molecules to form water (Rezaei *et al* 2007; Tender *et al* 2002).

MFC has a variety of functions and is very broadly applied to various aspects, for example, waste treatment, electricity production, as well as being used as an environmental biosensor (Zhang and Angelidaki, 2011; Schneider *et al* 2016; Yang *et al* 2015). The environmental biosensor is one crucial aspect because it is one of the

devices that can help indicate environmental sustainability. MFC-based environmental biosensors are usually used as indicators of COD, BOD, DO, microorganism activity, or toxic substances (Vishwanathan *et al* 2013; Zhang *et al* 2011; Oh *et al* 2009; Shen *et al* 2013; Tront *et al* 2008). Usually, samples from environmental waters containing organics or inorganics matter flowed into the anode chamber where a consortium of microorganisms placed, make the voltage differences occur, and passed on as a signal. Environmental water samples can also flow into the catholyte chamber when the characterization of the sample is related to oxygen activity.

Dissolved oxygen (DO) is one of the most critical and often used parameters related to water quality, which is an essential indicator of microbial metabolic activity in aquatic samples (Wetzel, 2001; Ansa-Asare *et al* 2000). DO concentrations in waters provide important information regarding biological activities that involve biochemical reactions in the aquatic environment. Therefore, DO can be used to measure the activities of biological processes that occur in environmental waters (Markfort and Hondzo, 2009).

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Yeast MFC is considered an alternative renewable resource that can be utilized as a biosensor. Some of the advantages of yeast as a biocatalyst in MFC include yeast has facilitated extracellular electron transfer, robust, fast-growing, facultative anaerobe, non-pathogen, and temperature tolerant biocatalyst (Hubenova and Mitov, 2015; Rossi *et al* 2015; Rossi *et al* 2016). However, its low output power and voltage are the main limitations of the MFC yeast, which will affect the performance of the biosensor. Yeast MFCs reportedly provide power densities that vary between 0.95- 2771.25 mW/m² (Duarte *et al* 2019, Powell *et al* 2011), depending on the substrate, electrodes, and polarization data collection methods. Several studies have been carried out by several researchers whose base aims to improve the performance of yeast MFC. Modification of anode material, the effect of adding several types of mediators and their reaction studies, as well as optimization of the substrate on the anode side have been carried out (Christwardana *et al* 2018a; Christwardana *et al* 2018b; Christwardana *et al* 2018c; Christwardana *et al* 2019). Considering these matters, it is possible that the MFC yeast can be used as a promising DO biosensor for environmental waters.

Air and water-compatible, good sensitivity and selectivity, scalable, cheap, unique, and environmentally friendly are the preferred biosensor characteristic (Rawson *et al* 1989; Karube *et al* 1995; Dennison and Turner, 1995; Badihi-Mossberg *et al* 2015). Regarding scalability, reactor size is an important issue for the application of MFCs as biosensors, especially DO biosensors. However, there is little information available about the effects of biosensor scaling on its power output. Therefore, an assessment of the effect of biosensor measures on performance must be carried out intensively. The scale correlation of the performance of MFC-based biosensors can be used as a reference for researchers to develop later biosensors that are more compact, accurate, economical, and have good performance.

In this study, we operate an MFC-based DO biosensor, which had small in size, and then compares it with a larger size. With relatively larger the anode surface area and anolyte volume, and without reducing the electrode spacing, the performance between the small and large size

DO biosensors would be compared, and then the economic analysis would be assessed. We hope this study can be used as a reference for determining dimensions and designing MFC reactors as environmental biosensors.

2. Materials and Methods

2.1 MFC-based DO Biosensor Construction

Single chamber acrylic MFC reactor (Phychemi, Hongkong) used as environmental DO biosensors with a liquid volume of 28 mL and a 4 cm electrode space from both electrodes, then called Small Yeast Microbial Fuel Cell (SYMFC). The anode and cathode were made of carbon felt with an active area of 7 cm² and treated Nafion 117 (treated with 3% w/w H₂O₂, 0.5M H₂SO₄, and DI water) acted as a membrane separator between the anode and the cathode (Christwardana *et al* 2018a; Christwardana *et al* 2018b). Meanwhile, a single chamber acrylic MFC reactor with a larger size (home-made) had a higher volume of liquid, which was 154 mL was used as a comparison, with the space of the two electrodes was 4 cm, then called Large Yeast Microbial Fuel Cell (LYMFC). The active surface area of anode, cathode, and membrane separator increased to 38.48 cm² or 5.5 times higher. The photograph of the small and large MFC reactor can be shown in Figure 1a-c.

2.2 MFC-based DO Biosensor Operation

MFC was inoculated using 14 mg/mL yeast *Saccharomyces cerevisiae* (Lessafre, Marcq-en-Baroeul, France) and fresh nutritional media consisting of 14 mg/mL glucose (Merck, Darmstadt, Germany), 5 mg/mL yeast extract (Merck, Darmstadt, Germany), and 2.5 mg/mL peptone (Himedia, Mumbai, India) (Christwardana *et al* 2018c; Christwardana *et al* 2019) was fed into anode chamber every three days for two weeks, to grow biofilm on the anode surface. The system was considered to be operating in a stable condition when the voltage can be reproduced after refilling the reactor with the fresh medium during the incubation.



Fig. 1 a) Lateral, b) front, and c) back views of assembled small and large MFC reactor. While d) is their application in the irrigation canal

Once stable, the MFC yeast system is then put into a container containing distilled water (DW) and operated with different DO volumes. Aeration system was used to increase DO concentrations, while N_2 gas was used to reduce DO concentrations. The anode and cathode are connected to an external circuit with a 1000Ω external resistance. The voltage was taken for 30 minutes by recording every 5 minutes and converted to current density. As a calibration, DO concentration was measured using a commercial DO meter Lutron WA-2015 (Taipei, Taiwan). The equation obtained from the DO vs. current density curve. The equation was used to predict DO concentration using MFC when applied to environmental waters samples.

MFC was applied to an irrigation canal in Salatiga (Central Java, Indonesia) as in Figure 1d, the voltage was measured using a multimeter UNI-T UT61E (Dongguan, China), and DO was measured using a DO meter, for 30 minutes. The current density value, which was the conversion of the voltage, was then entered into the equation obtained previously to obtain the DO value. Then the DO value of the MFC was compared to the DO value of the DO meter.

Finally, a polarization curve was obtained by measuring a stable voltage for 15 minutes, which results in various external resistance values from $5 M\Omega$ to 100Ω .

2.3 Techno-economic Analysis of MFC-based DO Biosensor

The techno-economics of MFC-based DO biosensors were analysed based on the cost of power obtained, both MFC in small and large sizes. The price of each component of MFC is added up (cost per cell), then divided by the power density obtained (mW/m^2). A small cost of power is desirable while still considering its capital cost.

3. Results and Discussion

3.1 Correlation Between DO and Current Density

Both SYMFC and LYMFC were tested in DW with varying DO concentrations and an external resistance of 1000Ω , as illustrated in Figure 2a. The resulting current density increased gradually as the DO concentration raised. Contrary, the current density decreased when the DO level reduced with the addition of N_2 gas. The increase of DO levels made the cathodic potential value shifted in a more positive direction. Assuming the anodic potential has a fixed value, the cell potential or the difference between the cathode potential and the anode will be even more enormous in value. Large potential cell values affect the current density generation. The current density values at the initial DW were 10.1 and 16.7 mA/m^2 for SYMFC and LYMFC, respectively, at DO concentrations of 6 ppm . This value gradually raised when DO was increased to around 8 ppm and produced a current density of around 19.4 and 39.1 mA/m^2 for SYMFC and LYMFC, respectively. Surprisingly, the current density value decreased to 1.2 and 2.0 mA/m^2 for SYMFC and LYMFC, respectively, when N_2 gas was injected into DW to make anaerobic conditions. The reducing DO level makes the cathode potential shifted more negatively, so that the potential difference between the anode and cathode (cell potential) becomes small, as well as its current density value.

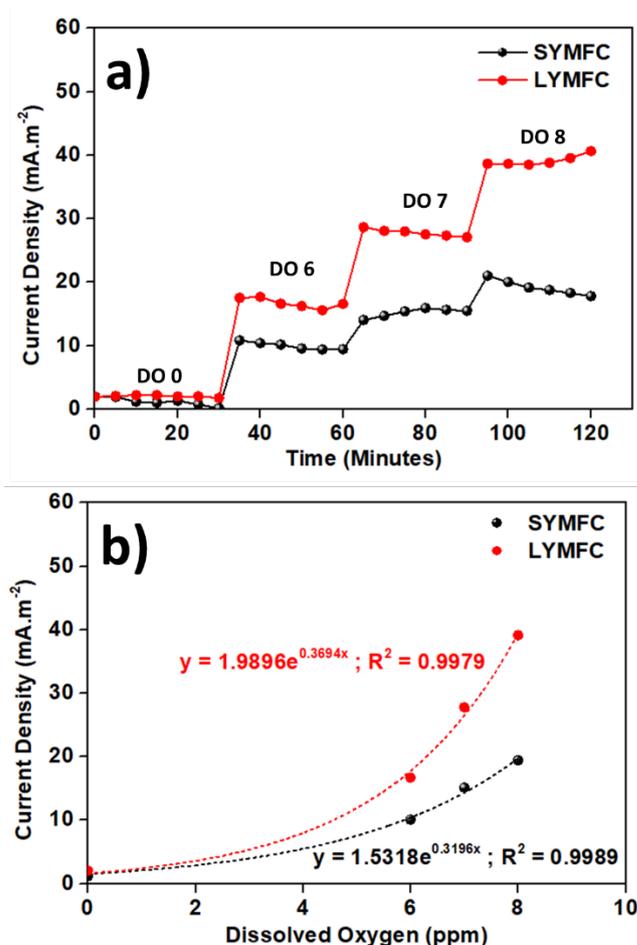


Fig. 2 a) Current density of Yeast MFC based biosensor in various DO concentration and b) correlation between DO level vs. current density

Figure 2b shows the correlation between DO concentration and current density in SYMFC and LYMFC. Several things can be studied. First, the correlation between DO levels and current density did not indicate a linear increase, but rather an exponential. The values of R^2 were 0.9989 and 0.9979 for the exponential equation in SYMFC and LYMFC, respectively. This is because the low DO concentration limited the cathode ORR activity in MFCs (Rismani-Yazdi *et al.* 2008; Rago *et al.*, 2017). While higher DO, concentration can significantly increase current density production due to increased Oxygen Reduction Reaction (ORR) activity. Second, the current density produced in LYMFC was higher than SYMFC, which affected by two things, namely the surface area of the separator membrane and the cathode. The higher the surface area, the more protons diffused from the anode to the cathode. This is consistent with experiments carried out by Oh and Logan (2006) and Hadiyanto *et al.* (2019), which stated that the area of the membrane affects the produced current density. The area of the cathode also affects the generation of current density. The ORR will occur more when the surface area of the cathode gets bigger. The ORR rate influences the rate of proton transfer from the anode to the cathode, where the rate of proton diffusion is proportional to ORR activity.

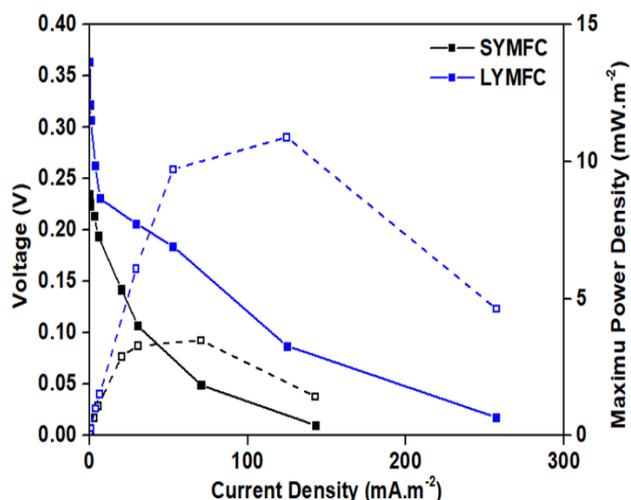


Fig. 3 Maximum Power Density of Yeast MFC as a DO biosensor. For the test, distilled water with average DO concentration was 6 used as the sample

3.2 Maximum Power Density of Yeast MFC

The performance of SYMFC and LYMFC as DO biosensors was investigated by measuring and making the polarization curve in DW without the influence of external oxygen. Figure 3 shows the MPD from SYMFC, where the result was 3.48 mW/m^2 , while the MPD on LYMFC has a value of 10.89 mW/m^2 or higher 3.13 times higher. These results prove the truth that the energy produced by MFCs is influenced by several factors, including the surface area of the separator membrane and the surface area of the cathode. Current density is directly proportional to power density, $P = V \times I$, so the increase in current density affects the increase in the value of power density.

3.3 Application of Yeast MFC as DO Biosensor

SYMFC and LYMFC as DO biosensors were applied to the irrigation canal, which has a flow rate of 0.125 m/s . The placement position of SYMFC and LYMFC in the river flow can be seen in Figure 4. The value of the current density of SYMFC and LYMFC was measured using multimeter, as well as the DO value of the river flow measured using DO meter for 30 minutes, as seen in Figure 5a-b. The average current density value in SYMFC was $14.45 \pm 0.55 \text{ mA/m}^2$, while in LYMFC was $21.91 \pm 1.55 \text{ mA/m}^2$. The average DO value at that time was $6.79 \pm 0.09 \text{ ppm}$. The current density value obtained is then entered in the equation obtained on the DO vs. current density curve in Figure 2b. From these equations, the DO values obtained were 7.02 and 6.49 for SYMFC and LYMFC, respectively. DO values obtained in SYMFC had a percent error of 3.39%, smaller than LYMFC, which had a % error of 4.42%. From this, we know that in addition to power density, the % error in DO measurements is influenced by the architecture and size of the MFC. This is consistent with references stating that the size and architecture of MFCs affect the surface area of electrodes and membranes, and also the volume of electrolytes present in the anode and/or cathode chamber (Lanas *et al* 2014; Fan *et al* 2012; Cheng and Logan, 2011).

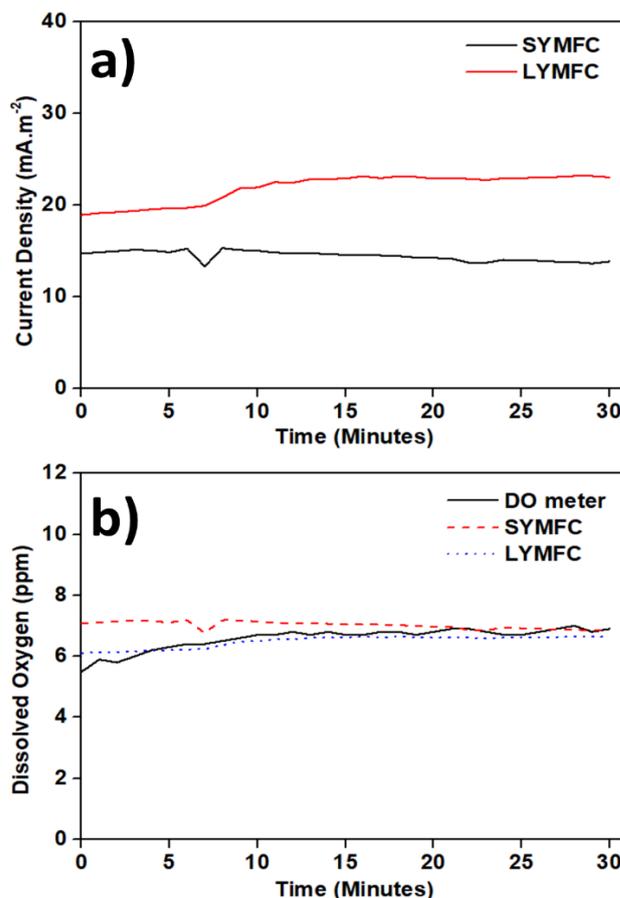


Fig. 4 a) Current density of SYMFC and LYMFC as DO biosensor applied in an irrigation canal and b) DO level of water in irrigation canal measured using DO meter, SYMFC, and LYMFC

3.4 Techno-economic Analysis of Yeast MFC as DO Biosensor

Our results show a comparable correlation between power output and size of MFC-based biosensors, suggesting that larger MFC sizes can have relatively higher power output when compared to MFCs of smaller size, despite having a reactor, electrode, and membrane type the same one. According to Li *et al* (2014), the cost is one of the main obstacles to implementing MFC on a larger scale, so that focusing on reducing the capital costs of MFC-based DO biosensor systems becomes an alternative strategy rather than increasing power output.

The next step is to test the economic feasibility of the SYMFC and LYMFC system assemblies by considering each of its parts, including the reactor, anode, cathode, current collector, separating membrane, biocatalyst, and microorganism growth media, which are installed directly into the system. With this, the capital costs of a larger or smaller scale electrode assembly in future generations are estimated, and the cost-effectiveness can be evaluated.

In short, we assume that SYMFC requires 28 mL of anolyte consisting of media and yeast culture to generate electricity as a DO biosensor. While LYMFC requires a larger anolyte volume of 154 mL or 5.5 times more than SYMFC. Single-chamber acrylic MFC reactors with air-cathode systems used in both the SYMFC and LYMFC systems, have an electrode active surface area of 7 cm^2 for SYMFC and 38.48 cm^2 for LYMFC with spaces between the electrodes being fixed (4 cm).

Table 1

Cost of currently available materials for a small and large MFC system

Component	Material	Quantity	Cost (\$)	SYMFC		LYMFC		Manufacturer
				Required	Cost/Cell	Required	Cost/Cell	
Cell reactor	Cubic acrylic	1 set	53	1 set	53	1 set	80.16	Small: Phyhemi Large: home-made
Electrode	Carbon felt	600 cm ²	21.18	7 cm ² x 2	0.4942	38.48 cm ² x 2	2.7166	KWS
Membrane	Nafion 117	516 cm ²	777.91	25 cm ²	37.69	100 cm ²	150.76	Sigma Aldrich
Current collector	Stainless steel 304 – 0.05 mm	3000 cm	3.85	2.5 cm ² x 2	0.0064	7.5 cm ² x 2	0.0192	Saky Steel
Biocatalyst	Yeast <i>Saccharomyces cerevisiae</i>	11 gr	0.35	0.392 gr	0.0124	2.156 gr	0.0686	S.I.L. France
Media	<ul style="list-style-type: none"> • yeast extract • peptone • D-glucose 	<ul style="list-style-type: none"> • 600 gr • 500 gr • 250 gr 	<ul style="list-style-type: none"> • 128.14 • 55.74 • 33 	<ul style="list-style-type: none"> • 0.140 gr • 0.070 gr • 0.392 gr 	<ul style="list-style-type: none"> • 0.0299 • 0.0078 • 0.0517 	<ul style="list-style-type: none"> • 0.770 gr • 0.385 gr • 2.156 gr 	<ul style="list-style-type: none"> • 0.1644 • 0.0429 • 0.2846 	<ul style="list-style-type: none"> • Merck • Himedia • Merck
Total Cost of power			1073.17		91.2924		234.2163	
					26.23		21.51	

MFC-based DO biosensor assembly costs were calculated based on the current material prices of several producers, as shown in Table 2. Because price variability depends on location, taxes and shipping costs can be excluded. Our analysis shows that SYMFC will be cheaper at \$ 91.2924, but more expensive at \$ 26.23 of capital costs per mW/m² generated. In contrast, the LYMFC capital cost will be 2.6 times higher, but 1.2 times the lower cost per mW/m² (\$ 234.22 and \$ 21.51 per mW/m²). We find the fact that increasing the power in the biosensor can be directly proportional to the economics of the device; the cost reduction must also be considered. We have shown that the compactness and complexity of MFC size materials cannot guarantee cost savings on an MFC system.

4. Conclusion

In this study, we compared the performance of yeast MFC-based DO biosensors from different sizes, small and large, and also study their techno-economic analysis. The correlation between DO and current density had a non-linear (exponential) tendency rather than linear, with R² values of 0.9989 and 0.9979 for SYMFC and LYMFC, respectively. SYMFC and LYMFC as DO biosensors produced maximum power densities of 3.48 and 10.89 mW/m². When applied to the irrigation canal, DO measurements showed errors of 3.39 and 4.42% for SYMFC and LYMFC, respectively, when compared to DO measurements using DO meters. This shows DO measurements using Yeast MFC with various sizes are still acceptable.

The size of the MFC reactor affects the components used as DO biosensors, so the techno-economic analysis needs to be carried out to assess the feasibility of the MFC-based DO biosensors in various sizes. Simple economic analysis showed that for SYMFC, the capital cost of a set of systems equipped with the reactor, electrodes, membrane, biocatalyst, and media would be \$ 91.29, or \$ 26.23 per mW/m² of power generated. While for LYMFC, the capital cost of a set of systems was \$ 234.22, or \$ 21.51 per mW/m² of power generated. This shows that SYMFC is preferred than LYMFC because it only requires fewer

capital costs and by considering cost per mW/m², which is more or less similar.

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