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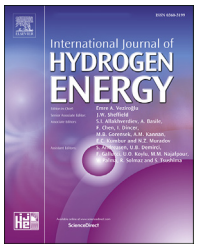

# Cellulose – Carrageenan coated carbon felt as potential anode structure for yeast microbial fuel cell

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## HIGHLIGHTS

- Yeast biocatalyst and two different cellulose treated felts were investigated.
- κ-carrageenan was used as entrapping polymer.
- MPD ranged from 48.38 $mW \cdot m^{-2}$ with CF to 70.98 $mW \cdot m^{-2}$ with CF/[KC/CMF].
- Addition of κ-carrageenan made a better contact between anolyte — and carbon felt.

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## ABSTRACT

Anode material is an important part of Microbial Fuel Cells (MFCs). Carrageenan and cellulose are strong candidates for modifying anode due to their many advantages, especially their biocompatibility. Cellulose microfibrils and microcrystalline were trapped on the surface of carbon felt (CF) using carrageenan (KC). The MFC adopted with CF/[KC/CMF] as anodes structures produced a power density of 70.98 $mW \cdot m^{-2}$, higher than MFC that used plain CF. The presence of KC changed the CF properties from hydrophobic to hydrophilic. This can be seen from the weight of biofilms formed in CF/KC, CF/[KC/CMC], and CF/[KC/CMF] being 60, 80, and 90 mg, respectively, higher than plain CF (60 mg). Carrageenan was also successful in entrapping cellulose. Cellulose donated hydrogen ions to form oxycellulose, which has a carboxyl group, wherein can increase Direct Electron Transfer (DET) between yeast and the anode. CF/[KC/CMF] anode structure showed excellent performance and has the potential to be developed in the future.



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## Introduction

Microbial fuel cells are one of the developed alternative technologies which utilize reactions carried out by microorganisms in converting the substrate to electrical energy [1]. In addition to generating energy, technology with the MFCs approach can also be used and has good prospects in a broad field, including waste treatment, desalination, hydrogen production, and environmental biosensors [2–4]. MFCs are considered as one of the environmentally friendly technologies because it uses biomass in its application. Moreover, the reaction from MFCs are not harmful to the environment, and it can even be used for other applications.

Although it is a promising technology, several challenges must be faced in developing MFCs due to its low power output compared to conventional fuel cells. The high cost of material and core parts causes existing MFCs to become difficult to be up-scaled or commercialized [5]. Costs and performance play an essential role in the MFC technology development sector. The lack of biofilms due to poor microbial adhesion performance with the anode surface and not facilitating the extracellular electron transfer activity will also limit the performance of MFC.

The anode is the most important part of the MFCs performance, and it is one of the keys to the MFC technology development. An anode is a place where biofilms are formed from microorganisms which are cultured in an anode chamber to produce electricity. In addition to capturing electrons produced by bacteria through an exoelectrogenic process, the anode also serves as a support medium for electrochemically active bacteria. On the other hand, the material from the anode also affects the capital costs of MFC commercialization. High conductivity, good electrochemical reversibility, good biocompatibility, excellent microbial adhesion performance, good stability, commercial availability, and low prices are the criteria of the anode material sought and are being developed in the MFCs [6–10]. Various carbon types such as bulk and particulate porous carbon, powdery carbon, and fibrous carbon are often widely used as a material from the MFC anode. These materials are usually modified with other materials to enhance MFC performance. Carbon-based electrodes are chosen because of their low price compared to nanoparticles and noble metals [11].

Carbon Felt (CF) is one of the carbon-based support materials considered for use in MFCs' anodes because it has many advantages, for instance: high conductivity, low cost, high surface area and porosity, excellent electrolytic efficiency, and excellent mechanical strength [12–15]. CF was created to support the 3D structure that is very useful in MFC as a place for the biofilms formation to support electron transfer processes [16,17]. Despite having advantages, the use of CF directly (plain CF) is not recommended because of its insufficient wettability and electrochemical activity in aqueous solutions due to their hydrophobic surface nature [18]. Moreover, they have poor kinetics for reduction and oxidation reactions [19]. Therefore, it is compulsory to modify the surface of the CF to improve the excellent properties in the CF. Electrode modifications using natural polymer materials such as chitosan, alginate, carrageenan were believed to be able to enhance the performance of electrochemical devices [20–22]. However, synergistic performance between CF and natural polymer for microbial fuel cells are rarely studied. Carrageenan is a gel-forming and viscosifying polysaccharide which is obtained by extraction from Rhodophyceae and known as an acidic hydrophilic polyanion [23]. Commonly, Carrageenan is used in food preparation for its gelling, thickening, and emulsifying properties, as well as in pharmaceutical applications and experimental medicine [24–26].

Cellulose is a carbohydrate polymer that has hydrophilic and insoluble water properties [27], so it is suitable to be used as a promising biomaterial to support the modification of the MFC anode. Cellulose is disintegrated chemically or mechanically into micro- and nano-sizes, namely Cellulose Micro Fibril (CMFs), Cellulose Nano Fibril (CNFs), Cellulose Micro Crystalline (CMC), and Cellulose Nano Crystalline (CNC) [28–30]. They are commonly used to modify electrochemical devices. The need for micro-scale (and nano-scale) cellulose in composite materials has increased due to their high strength and stiffness, low weight, biodegradability, and renewability [27].

Several researchers have investigated the carrageenan-cellulose composites. Kassab et al., fabricated composites from κ-carrageenan with nanocrystal cellulose (CNC), where the addition of cellulose to the carrageenan matrix can strengthen the mechanical characteristics of the composite [31]. Composite between cellulose and carrageenan can also be used for enzyme immobilization as an entrapping agent [32]. Prasad et al. also fabricated the cellulose-carrageenan composite using an ionic liquid to become gel material [33].

In this study, we exploited the benefits provided by CF, κ-Carrageenan (KC), CMC, and CMF to improve the performance of yeast MFCs, which adopting *Saccharomyces cerevisiae* as a biocatalyst. Yeast *Saccharomyces cerevisiae* based MFCs have significant potential to be developed, and it is interesting to be studied [34]. To enhance the performance of MFC anodes, cellulose is mixed with KC solution to form a mixture of KC-cellulose. Then the mixture solution is coated onto the surface of the CF to make a promising anode structure; CF/[KC/CMC] or CF/[KC/CMF]. Plain CF was used as a control sample, while CF coated with KC only, without cellulose, was also fabricated to determine the role of cellulose in the bioelectrochemical activity. All samples were characterized physiochemically and electrochemically. Three Dimension Optical Microscopy (OM-3D) was used to characterize the morphology of electrodes, while the electrochemical characteristics were done by determining the polarization curve, Closed Circuit Voltage (CCV), and internal resistance of MFCs. Approach through collaboration between CF-KC-cellulose as anode material for MFC has never been done by previous researchers, make it become our academic novelty. Moreover, this study aimed to promote and apply the advantages of low-cost carrageenan combined with cellulose and CF-based material as an anode material of yeast MFC. Thus, making a significant and beneficial contribution to the commercialization of MFC in the future.

## Materials and methods

### Electrode fabrication

The electrode fabrication method in this study was inspired by Ref. [34]. CF was produced by KWK Steel Co., Ltd (Zhejiang, China), while KC was obtained from IndoGum (Jakarta, Indonesia). CMC was produced by Merck (Darmstadt, Germany), and CMF was obtained from PT. Polytech (South Tangerang, Indonesia). Separately, 5 mg/mL of CMC and CMF were fed in the 0.1% KC solution (in 1% w/v NaCl) to form a stable dispersion, namely KC/CMC and KC/CMF. Then CF was prepared as the backbone of the electrode, while stainless steel wire acted as a current collector. In a unique fabrication process, firstly, CF was treated in de-ionized (DI) for hours to remove impurities on the CF surface. After that, CF was dipped in a KC/CMC and KC/CMF dispersion solution for 30 min. Later, composite electrodes were washed using DI water to remove the unbound component and then dried at room temperature overnight. Finally, the CF/[KC/CMC] and CF/[KC/CMF] composite electrodes were successfully fabricated. The CF/KC electrode, as a control, was also made by the same method, by dipping CF in a KC solution and drying it.

### Fuel cell configuration

A single cubic reactor made from polyacrylic (Phychemi Co. Ltd., Beijing, China) was used in this study with a total reactor volume of 28 mL and a geometric surface area of 7 $cm^2$. Modified CF and plain CF were used as anodes and cathodes, respectively. Nafion 117 was used as a separator between the anode chamber and the cathode. Previously, Nafion 117 was treated with 3% wt. $H_2O_2$, 0.5 M $H_2SO_4$, and DI water.

An anolyte solution containing yeast and glucose acted as an anolyte. The composition consisted of 13 mg/mL yeast from *Saccharomyces cerevisiae* (Societe Industrielle Lesaffre, Marcq-en-Baroeul, France) and 13 mg/mL glucose powder (Beihai Jinzhi Natural Health Products Co. Ltd., Guangxi, China). The glucose/yeast ratio was 1:1, which was obtained from an optimization result from previous studies [35]. The MFC process was carried out in semi-aerobic conditions [36], where there was a small hole in the upper side of the anode chamber, which was left open as the place where $CO_2$ gas was released, and air entered. The stirring process was carried out in the anode chamber to keep the anolyte solution homogeneous. The glucose consumption by yeast measured by using commercial glucose sensor. The experiment conducted at room temperature around 25–28 °C with relative humidity (RH) in the range of 73–75%, with duplicate repetition. The configuration of yeast microbial fuel cell can be shown in Fig. 1.

### Fuel cell characterization

Yeast MFC was operated in four cycles with a total time of 288 h (12 days) or 72 h (3 days) each cycle. 1000 Ω was used as external resistance [37]. In the first three cycles, fresh anolyte solution (yeast + glucose) was injected into the anode chamber to replace the old anolyte solution. In those cycles, the yeast biofilm was allowed to grow on the anode surface. In the last cycle (fourth cycle), only glucose solution was injected, to prove the performance of the biofilm in converting glucose to electricity. Instead of the YPD medium, glucose was only used as a growth medium of yeast because later, this system will be applied to rural areas. So that low-cost media is applied here. Fuel cell performance was monitored using a digital multimeter Krisbow KW06-267 (Jakarta, Indonesia), which was monitored every 6 h, with a recording interval of 10 min for 30 min. Polarization curves were measured to calculate the maximum power density at the end of every cycle by applying external resistance stepwise from 1 MΩ to 50 Ω at an interval time of 15 min.

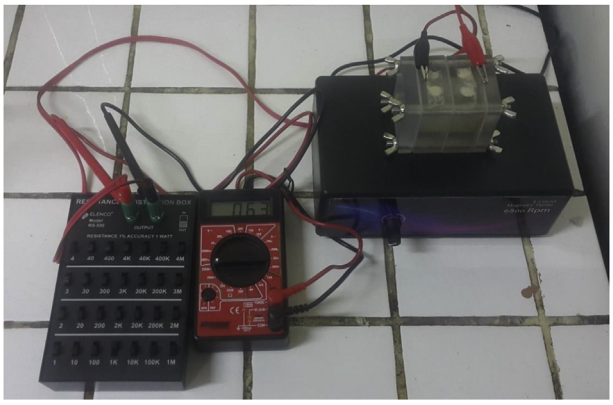

**Fig. 1 – Yeast microbial fuel cell Configuration.**

### Morphology characterization

The surface morphology of modified CF before and after completion of the MFC cell test was examined by 3D digital optical microscopy (OM-3D) to verify the yeast growth on the modified CF surface. Keyence VHX-600 (Itasca, Illinois, USA) was used for this purpose. The OM-3D operation is generally similar to commercial microscopes. The sample is placed on the preparation table near the objective lens. Then the ocular part is connected directly to the computer. Sample magnification can be set on the user interface seen on the computer display.

### Coulombic efficiency and energy conversion efficiency measurement

Coulomb efficiency (CE) is the efficiency of electron transfer in a system for conducting electrochemical reactions [38], calculated using Eq. (1):

$$CE = \frac{M_S \times I \times t_b}{F \times b_{es} \times V_{an} \times \Delta C} \times 100\% \quad (1)$$

where $M_S$ is the molecular weight of the substrate, I is the average result in the current (A), $t_b$ is processing time in one cycle (s), $V_{an}$ is the anode chamber volume ($m^3$), $b_{es}$ is the number of moles of electrons produced by substrate oxidation (24 mol $e^-$ per mol glucose), F is the Faraday's constant (96,500 s A/mol), and ΔC is the concentration of the substrate oxidized during the time.

Energy conversion efficiency (EE) is the ratio between the total energy produced by MFC and the maximum energy that can be obtained from biomass [38]. This maximum energy can be attributed to biomass standard molar enthalpy. The efficiency of energy conversion can be calculated using the equation:

$$EE = \frac{P \times t_b}{V_{an} \times \Delta C \times \Delta H_f} \quad (2)$$

where P is the resulted average energy or output power, tb is time for substrate consumption (s), $V_{an}$ is the anode chamber volume ($m^3$), C is the concentration of substrate in the anode space (C = 0.72 mol per $m^3$, glucose), and $\Delta H_f$ is the standard formation molar enthalpy (2801 kJ/mol for glucose) [39].

## Results and discussion

### *Possible chemical bonding and morphology of anode structure*

The effect of adding carrageenan and cellulose to carbon felt as an MFC anode was initially investigated. Fig. 2 shows the possible bonding that can occur in CF, CF/KC, CF/[KC/CMC], and CF/[KC/CMF]. In CF/KC, carrageenan might wrap the carbon fiber surface on carbon felt. Carrageenan is a hydrophilic polymer which absorbs water for the gelling process [40]. KC formed a thin layer on the surface of carbon fiber and made the carbon surface that was originally hydrophobic turn into hydrophilic. This hydrophilic nature was highly desirable in MFC because electrolyte solutions (which contain microbes) entered deeper inside the carbon felt to form biofilms, not just on the upper surface [41]. In CF/[KC/CMC] and CF/[KC/CMF], KC had a role as an entrapping polymer where it trapped the cellulose (CMC or CMF) so that it stuck and was distributed to carbon felt.

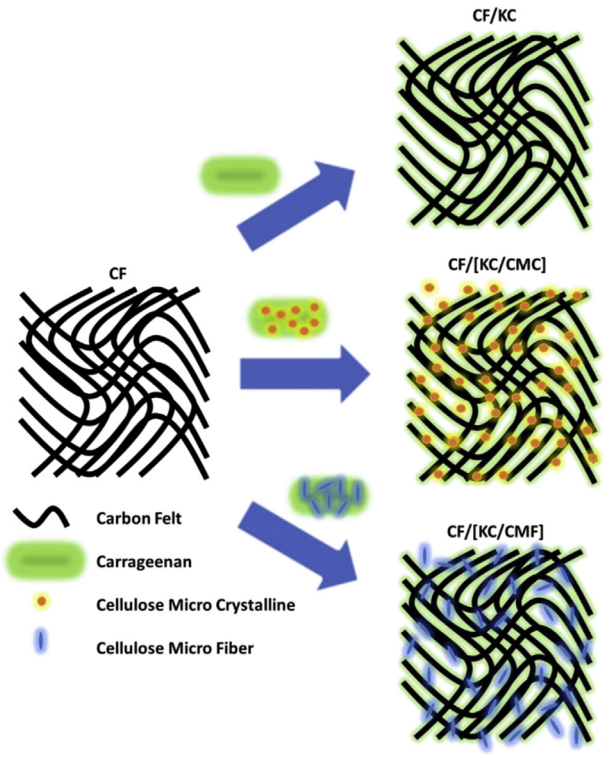


**Fig. 2 – Possibility bonding mechanism of CF, CF/KC, CF/[KC/CMC], CF/[KC/CMF] anode structure.**

The morphology of the anode structure after the fabrication process is essential to be investigated. Fig. 3 shows the morphology of CF, CF/KC, CF/[KC/CMC], and CF/[KC/CMF] anode structures via OM-3D. There were no significant differences in the morphology of CF and CF/KC, as shown in Fig. 3a and b. KC that covered carbon felt had a very thin and transparent size, so it was rather difficult to identify. While the morphology of CF/[KC/CMC] anode structure shown in Fig. 3c, shows there were some glowing points. It was identified as the cellulose crystals from CMC. CMC has a very small size close to the size of nano so that CMC crystals could enter inside the felt carbon during the fabrication process. Fig. 3d shows the morphology of CF/[KC/CMF]. Crystals from CMF were found in the figure, and the condition was similar to crystals in CMC. The crystals in CMF come from very fine fibers, which could penetrate inside the carbon felt, even though coarse fibers stuck to the upper surface. By its nature, the degree of crystallinity of CMC is higher than CMF [42]. The non-uniform size of the CMF made them only to be concentrated on the surface of the CF.

### *Bioelectrochemical analysis*

MFC's performance with various anode structures was tested for its current generation for four cycles with external resistance of 1000 Ω. Fig. 4a shows the current density generated from MFC that adopts CF, CF/KC, CF/[KC/CMC], and CF/[KC/CMF] as the catalyst structure of the anode. There are some remarkable things found in these phenomena. A plain CF anode produced a current density as low as 86.81 ± 0.88 $mA \cdot m^{-2}$. After refilling with fresh anolyte consisting of yeast and glucose solution, in each MFC cycle (after the first and second cycles), current density increased rapidly. Then the current density decreased gradually and started to rise after a while to 111.49 ± 2.01 $mA \cdot m^{-2}$. This trend is related to the adaptation process of yeast to the new environment in the anode chamber [36]. The current density decreased again near the end of the cycle due to the depletion of the substrate because the yeast consumed it. When adding fresh substrate (without yeast) in the fourth cycle, the current density increased but lower than the previous cycles. These phenomena prove that biofilms formed on the surface of the CF, although the total microbes were lower than others.

The current output of MFC shows a substantial impact by coating the CF anodes using KC, CMC, and CMF. MFC adopting CF/[KC/CMF] anode structure produced the highest current density (148.37 ± 4.41 $mA \cdot m^{-2}$), although it was not significantly higher compared to other anode structures. MFC adopting anode structure of CF/KC and CF/[KC/CMC] resulted in a current density of around 118.41 ± 5.12 and 127.81 ± 0.85 $mA \cdot m^{-2}$, respectively. Coating the anode CF with a carrageenan-cellulose composite can increase the production of bioelectricity. Cellulose, which was in the carrageenan matrix is likely to oxidize to oxycellulose because it is in an acidic condition for a long time [43]. The acid comes from

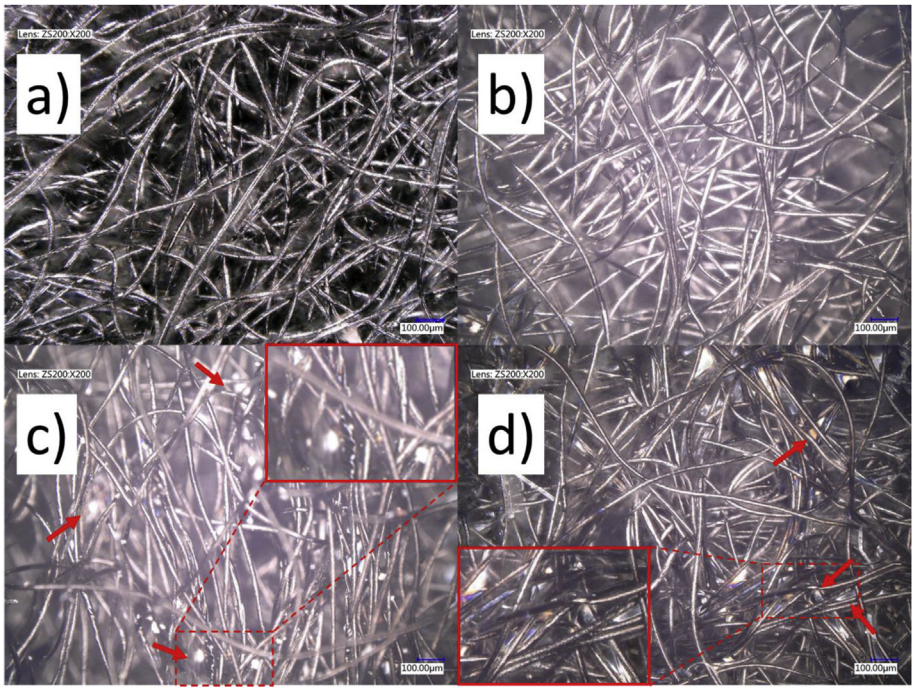


**Fig. 3 – Morphology of a) CF, b) CF/KC, c) CF/[KC/CMC], d) CF/[KC/CMF] anode structure before MFC incubation process. Arrows and insets show the presence of cellulose crystal from CMC or CMF.**

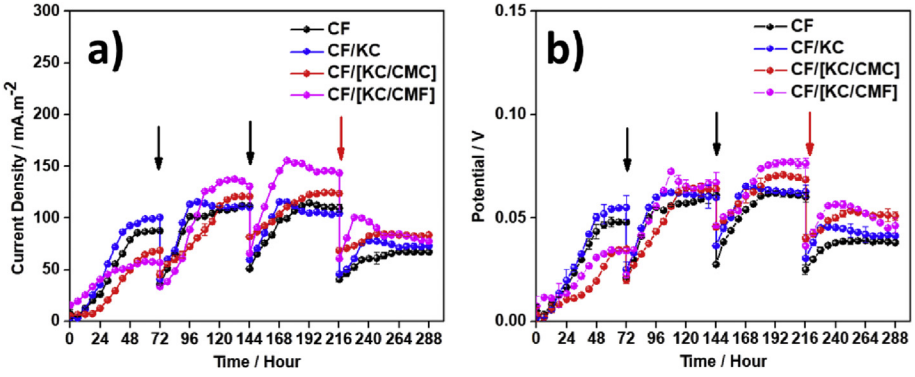


**Fig. 4 – a) Potential and b) current density of MFC adopting CF, CF/KC, CF/[KC/CMC], and CF/[KC/CMF] as anode structures. Applied external resistance was 1000 Ω. Black arrows show yeast and glucose replacement, while red arrows show only glucose replacement. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)**

acetic acid, which was produced from the semi-aerobic process of glucose by the yeast. The $-CH_2OH$ group in cellulose was oxidized to –COOH (carboxyl) by releasing hydrogen ions (protons) and gaining oxygen from acids [42]. The carboxyl group on oxycellulose helps to increase electron mobility [44].

Fig. 4b shows the potential observed from MFCs adopting CF, CF/KC, CF/[KC/CMC], and CF/[KC/CMF] as anode structures. In all samples, the MFC potential slowly decreased slightly because the lag phase occurred where the yeast adapted to the new environment. After a while, the potential exponentially increased, followed by the stationary phase until the potential decreased before the end of the cycle due to the depletion of glucose as substrate and the formation of acid and alcohol in the anolyte. This made the characteristic of anolyte changed and affect to anode potential. The average potential obtained for MFC with plain CF anode was 61.23 ± 0.75 mV after the third cycle. Likewise, the potential obtained for MFC with the CF/KC anode was 69.77 ± 3.33 mV after the third cycle. Even though the MFC potential with CF/[KC/CMC] anode increased around 2-folds from 32.19 ± 3.94 mV in the first cycle to 67.47 ± 3.75 mV in the third cycle. The highest potential occurred with MFC with the CF/[KC/CMF] as anode structure that was 129% from 33.16 ± 1.48 mV in the first cycle to 76.07 ± 1.11 mV in the third cycle. These results clearly indicate that biofilms grow well with CF anodes coated with KC, CMC, or CMF than with plain CF without any coating.

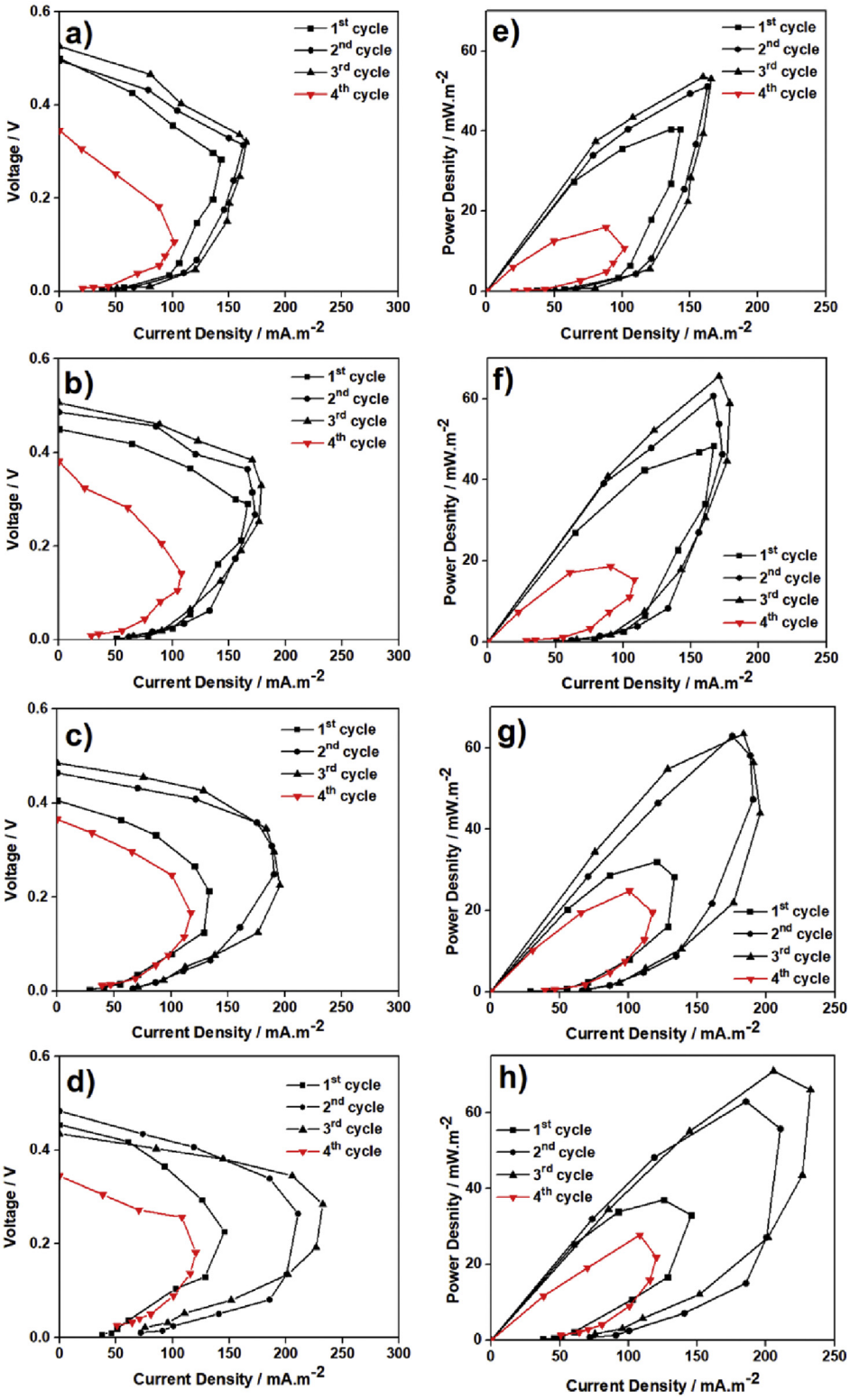


**Fig. 5 – Polarization curves of a) CF, b) CF/KC c) CF/[KC/CMC], and d) CF/[KC/CMF]. While (e–h) is their power curves, respectively.**

Fig. 5a–h shows the polarization and power curves taken every MFC cycle. There are several observable phenomena. First, MFC with CF/[KC/CMF] anode had a maximum power density of 70.98 $mW \cdot m^{-2}$. These results are slightly higher than other samples, namely MFC with CF/[KC/CMC] anode (62.43 $mW \cdot m^{-2}$), MFC with CF/KC anode (58.19 $mW \cdot m^{-2}$), and MFC with plain CF anode (48.38 $mW \cdot m^{-2}$). In this condition, the presence of oxycellulose plays an important role in

electron transfer activity and slightly changes the environmental condition in the anode system. Although coating the CF with carrageenan made the electrode become hydrophilic and preferably by yeast to perform colonization, the gelling properties of carrageenan allowed the distance between the surface of CF and yeast to be slightly far. This presents a small problem in electron transfer activity. On the other hand, plain CF has poor biocompatibility due to its hydrophobic nature, which can interfere with yeast activity in producing and transferring electrons.

Second, it can be seen that the maximum power density in all samples in the first to third cycles increased. This is because biofilms grow well on the surface of all samples. Carrageenan coating of the CF surface allowed a better colonization process than plain CF surface so that the yeast biofilm was well-formed. Moreover, the presence of oxycellulose from CMF or CMC was also probable to facilitate increasing direct electron transfer (DET) between yeast biofilm and the electrode surface. In the fourth cycle, the MPD values were lower than previous cycles due to the limitation of total microbes, which resulted in electrons.

Third, all the power curves for all MFC samples underwent doubling back or power D-overshoot when the low-value resistances were applied to MFC. According to some literature, power overshoot occurred because MFC experiences anodic electron depletion from exceeded cathodic overpotential, and this was related to a kinetic limitation at the anode part [45,46].

Other parameters which could be determined were CE and EE information of the MFC systems. As an abovementioned explanation, CE means the recovered electrons as resulted current to the total coulombs in the substrate during the MFC process, while energy efficiency is the percentage between resulted power and the maximum power that can be obtained during MFC process. As can be seen, the CEs of CF, CF/KC, CF/[KC/CMC], and CF/[KC/CMF] are 5.21, 6.07, 6.55, and 7.61% respectively. While their EEs are 4.82, 5.90, 6.33, and 7.20%, respectively. It explains that the addition of carrageenan and cellulose increased the CEs and EEs slightly. The complete data about CE and EE can be shown in Table 1. However, the CE and EE value of this MFC system is generally still very low. It can also be seen that its CEs in this study are not too far different from the result Pt catalyst by Esmaeili et al., which resulted in CE of 18.4% [47].

### *Biofilm and internal resistance relationship*

Ohmic loss and overpotential are involved in energy loss in MFCs. Ohmic loss has a strong relationship to internal resistance or areal resistivity, where the value of internal resistance can be determined using the ohmic region slope in polarization curves [48]. While the total overpotential is cumulative of overpotentials at both electrode, anode and cathode. Overpotential is encouraged by the presence of a pH inconsistency at the anode and cathode chambers, as well as membrane separator [49]. Areal resistivity is credited to the resistance of electron generation by microbes metabolism and electron transfer activity from microbes to anode surface [48]. The areal resistivity of CF, CF/KC, CF/[KC/CMC], and CF/[KC/CMF] were 13.03 ± 2.23, 8.04 ± 1.36, 6.39 ± 2.00, and 6.08 ± 2.42 $k\Omega \cdot cm^2$ respectively, where areal resistivity decreased as an increase in power density and biofilm mass. The formed biofilm of CF, CF/KC, CF/[KC/CMC], and CF/[KC/CMF] anode structure were 60, 82, 91, and 112 mg respectively. Biofilm has conductive nature where their presence in the surface of the anode structure will decrease the areal resistivity and increase the electron transfer activity between microbes and anodes [50–52]. The biofilm activity obtained by dividing the weight of biofilm per result in MPD is as shown in Table 1. This number gives inspiration to how effective the biofilm community resulted in electricity through the electrogenic process. Using this approach, the advantage effect of KC and cellulose addition on CF is clear that the presence of KC and cellulose increased the electrogenic activity. The full dataset of areal resistivity, the weight of biofilm, and biofilm activity can be seen in Table 2.

OM-3D images were used to verify the number of yeast electroactive biofilm, which affected the power generation of MFC. Fig. 6a–d shows clear evidence that the interaction between electrodes, yeast, and substrate formed a biofilm, in the form of a thin layer, between single CF fibers. In plain CF (Fig. 6a), the biofilms formed were not distributed well, was not homogeneous, and only formed between single fibers. Whereas in the CF/KC anode (Fig. 6b), more biofilm layers were formed, although biofilm formation was not well distributed. The increase in the number of biofilms is believed to be the effect of changing the CF nature after being coated with carrageenan, from initially hydrophobic, to being hydrophilic (or semi-hydrophilic). This was following previous studies where the hydrophobicity or hydrophilicity of CF had some influences on the number of biofilms formed. Surprisingly, the yeast biofilm layer that grew in the CF/[KC/CMC] anode had more quantity than the previous two samples, as shown in Fig. 6c. It is possible that the weak bond or interaction between cellulose and yeast, such as van der Waals bond or hydrogen bond, results in a yeast biofilm that can accumulate well on the anode surface. Moreover, biofilms that grew on the surface of CF/[KC/CMF] anodes had the highest number of biofilms compared to all samples, and they were well distributed (Fig. 6d). It was previously explained that the hydrophilicity of anodes affects the distribution of anolyte (including glucose and yeast). It made the interactions between the three components (anodes, glucose, yeast) grow well and produced many biofilms. Consequently, this enables better electron transfer between the yeast and the anode.

**Table 1 – Coulombic Efficiency and Energy Efficiency of MFCs adopted CF, CF/KC, CF/[KC/CMC], and CF/[KC/CMF] as anode catalyst structure.**

| Sample | Coulombic Efficiency (%) | Energy Efficiency (%) |
|---|---|---|
| CF | 5.21 | 4.82 |
| CF/KC | 6.07 | 5.90 |
| CF/[KC/CMC] | 6.55 | 6.33 |
| CF/[KC/CMF] | 7.61 | 7.20 |

### *Glucose oxidation*

Decreased glucose concentration was observed by analyzing anolytes taken from the anode chamber periodically every

**Table 2 – Dry weight and specific activity of yeast biofilm after full-cell tests.**

| Sample | Initial Dry Weight (mg) | Final Dry Weight (mg) | Dry Biofilm Weight (mg) | MPD ($mW \cdot m^{-2}$) | Specific Biofilm Activity ($m^{-2} \cdot mg \cdot mW^{-1}$) | Internal Resistant (kΩ) |
|---|---|---|---|---|---|---|
| CF | 990 | 1050 | 60 | 48.38 | 1.24 | 13.03 |
| CF/KC | 870 | 950 | 80 | 58.19 | 1.37 | 8.04 |
| CF/[KC/CMC] | 940 | 1030 | 90 | 62.43 | 1.44 | 6.39 |
| CF/[KC/CMF] | 990 | 1110 | 120 | 70.98 | 1.69 | 6.08 |

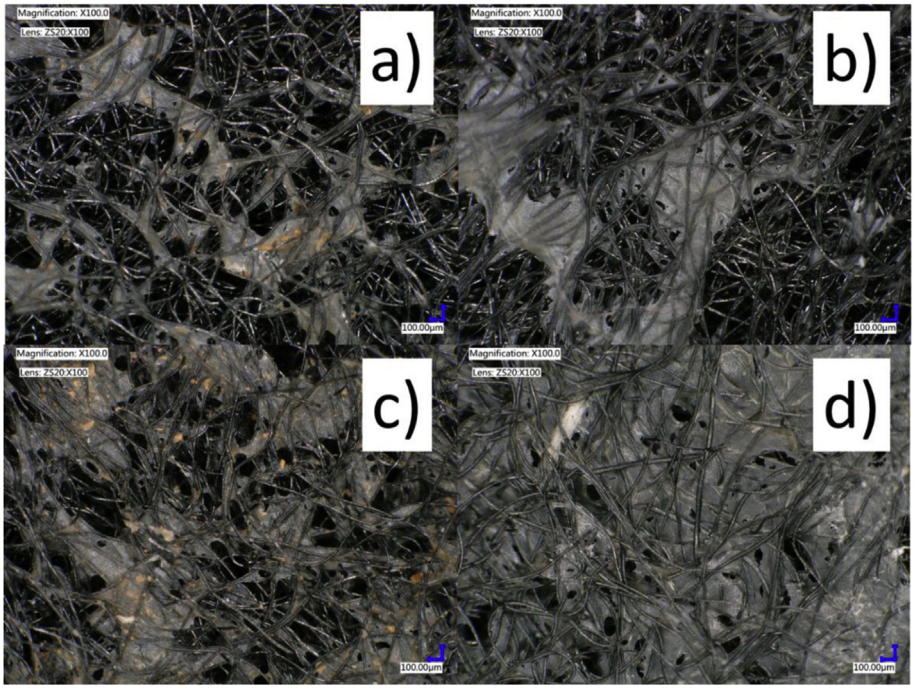


**Fig. 6 – Biofilm formation in the a) CF, b) CF/KC, c) CF/[KC/CMC], d) CF/[KC/CMF] as anode structure after MFC incubation process.**

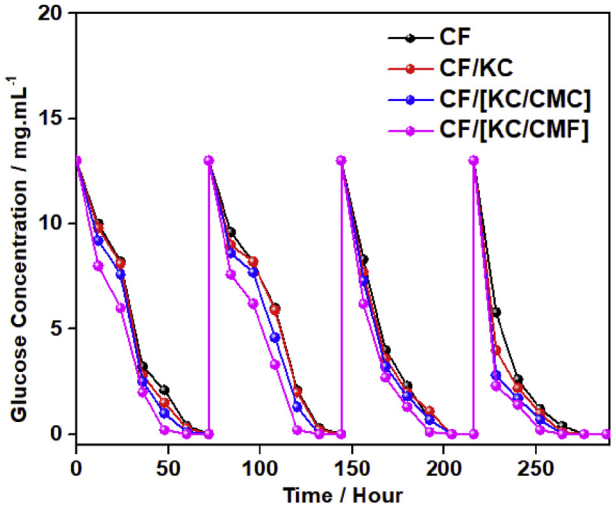


**Fig. 7 – Glucose consumption during MFC incubation which adopting CF, CF/KC, CF/[KC/CMC], and CF/[KC/CMF] as anode structures.**

12 h from the beginning of the first cycle to the end of the fourth cycle. Glucose will be oxidized to alcohol and acetic acid as the product of fermentation process [53,54]. At the beginning of the first, second, and third cycle, glucose was fed to the reactor along with yeast extract and peptone as the substrate. While only glucose was fed to the MFC reactor as a substrate to generate electricity in the beginning of fourth cycle. In Fig. 7, it can be seen that the glucose concentration was almost depleted after 48 h of the incubation process in all MFCs with CF, CF/KC, CF/[KC/CMC], and CF/[KC/CMF] anode structure. If related to Fig. 4, this phase was occurred at the end of the logarithmic phase, and the glucose depletion including other subsrates caused the stagnant voltage or even decrease. Moreover, glucose concentration closed to zero as it approaches the 204$^{th}$ hour of incubation time in the third cycle. While glucose ran out faster in the fourth cycle because there were no other substrates besides glucose. Yeast biofilms were focused on consuming glucose to metabolize and generate electricity.

## Conclusion

Entrapping cellulose, in the form of crystalline or fibril, on the surface of CF using KC changed the characteristics of the anode so that it could improve the performance of yeast MFC system. When CF was modified with KC, biofilms distributed in almost all parts of the CF, not only on the upper surface. The formation of colonies from yeast in forming biofilms was driven by changes in the physical property of felt carbon from hydrophobic to hydrophilic. The presence of cellulose did not affect the ability of yeast to form biofilms in modified CF by KC and cellulose.

The fuel cell test showed that current densities and voltage had decreased due to the yeast adaptation process in the new environment. Then the phenomenon was followed by an increase in current density and voltage in the second and third cycles caused by the increase in the number of biofilms on the anode surface. The current density and voltage at the fourth cycle were not as large as the previous cycles because fresh yeast was not injected, so the current and voltage generated were purely from glucose conversion by biofilm yeast. Yeast MFC that adopted a CF/[KC/CMF] structure anode had a higher current density, voltage, and power density than other structures. During the process, cellulose from CMF changed to oxycellulose, which has a –COOH function group which could increase the DET between the yeast and the electrode.

The construction of a biofilm on the electrode surface was proven by micrograph inspection using 3D digital microscopy. Quantitatively, the number of biofilms attached to the anode was obtained from the dry weight of the biofilm, while the effectiveness of biofilms was attained from the dry weight of biofilms divided by MPD.

Finally, modification of CF using KC and cellulose was considered to be one way to increase the DET between the yeast and the anode. Several characteristics were obtained and showed that KC played a role in changing CF properties, while cellulose that turns into oxycellulose increased electron transfer using their –COOH function groups. Research on the use of KC and/or cellulose still needs to be explored deeper to produce a yeast MFC system that can produce high power.

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