Assessing the efficacy of bio-composite starch/biochar based microbial fuel cell Pb(II) removal from contaminated seawater

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Abstract: *The efficient removal of toxic chemicals from wastewater using effective sorbents has gained significant attention in recent years. In this study, a starch/biochar composite was synthesized from rice straw to achieve the removal of Pb(II) from wastewater. The highest performing starch/biochar composite was then utilized as a microbial fuel cell electrode for generating electricity in synthetic seawater contaminated with Pb(II) and petroleum hydrocarbons. Results showed that the maximal power density and current density were 2.18±0.20 W/m^3 and 8.33±0.50 A/m^3, respectively, and that the composite achieved removal rates of 95.10±1.50% for Pb(II) and 55.10±2.20% for petroleum hydrocarbons. These findings suggest that the starch/biochar composite can serve as an effective sorbent for remediating environmental contamination, while also having potential for use in sustainable energy generation.*

Keywords: Biochar, Heavy metal, Lead, Low-cost electrode, Marine pollution

1 Introduction

The synthetic polymer industry commonly employs fillers such as silica, carbon black, and calcium carbonate, despite their drawbacks such as high cost, compromised mechanical properties, and adverse environmental effects during disposal [1]. Consequently, extensive research has been conducted to identify a cost-effective, biodegradable substitute that can enhance the mechanical properties of rubbers. Various bio-based materials including cellulose [2-4], protein, and starch have been examined for this purpose, with starch proving to be the most promising and extensively studied alternative [5].

Starch possesses several notable benefits such as its affordability, renewability, biodegradability, and wide-ranging applications in diverse industries. To serve as a filler, starch has been processed into starch

bio-composites [6], starch nanocrystals [7], chemically modified starch [8], or thermoplastic starch [9]. Although these techniques have demonstrated some success in enhancing specific mechanical properties, they are often intricate, protracted, or unsuitable for industrial-scale production [10].

Biochar can be obtained from rice straw via several thermo-chemical conversion methods, such as pyrolysis [11], torrefaction or carbonization [12], hydrothermal liquefaction [13], and gasification [14]. The quality of the biochar produced is heavily influenced by process variables, such as temperature, pressure, reactor configurations, and catalysts employed. Pristine rice straws generally exhibit high ash content (ranging from 8.5% to 20.4%), which is greater than that of other straw types, such as wheat straw (5.0–8.5%), barley straw (7.4%), corn straw (5.1–7.9%), and sugarcane straw (4.1%) (Wang et al., 2020). The primary component of the ash is silica oxide, accompanied by other oxides such as aluminium oxide and calcium oxide [15]. Rice straw also contains approximately 40% carbon, 30% oxygen, 5-6% hydrogen, 1% nitrogen, and less than 0.2% sulphur [16]. These straw-type biomasses typically have similar elemental composition (C, H, N, S, O) and a low moisture content of less than 10%, making them ideal pyrolysis feedstocks for biochar production. Furthermore, the moderate fixed carbon content of rice straw, as opposed to other straw-type biomasses like sugarcane straw and wheat straw, which contain less than 10% fixed carbon, is a desirable feature for the synthesis of carbon-based adsorbents [17].

The pollution of marine systems by heavy metals has emerged as a significant global environmental issue, and is currently one of the most important concerns being monitored in marine environments [18]. Heavy metals possess high levels of toxicity and persistence, and have the potential for biomagnification, bioaccumulation, and incorporation into the food chain once they reach a certain threshold in the ocean [19]. The contamination caused by Pb(II) is particularly worrisome as these metals are increasingly contributing to environmental deterioration, and have detrimental effects on marine life [20].

To address this problem, various approaches have been developed by related departments, including stringent environmental regulations that require the removal of heavy metal compounds from seawater and sediments. Traditional techniques and technologies utilized for the removal of heavy metals include precipitation and membrane filtration [21]. However, adsorption has been proven to be the most effective method for heavy metal removal [22].

Benthic Microbial Fuel Cells (MFCs) were first introduced in 2001, and played a critical role in generating interest in the broader field of MFCs. These fuel cells are typically composed of a graphite anode embedded in anaerobic marine sediments. They are connected via an electrical circuit, such as a marine scientific instrument or capacitor, to a cathode located in the aerobic seawater above [23, 24]. A key characteristic of marine MFCs is their sustainability, which is attributed to the constant supply of fuel and oxidant by environmental processes. Typically, this is derived from the settling of dead phytoplankton and/or plant debris, along with the continuous regeneration of microbial electrode catalysts.

A novel type of marine Microbial Fuel Cell (MFC) has been developed to mitigate the ohmic loss resulting from the anode-cathode spacing. This design is based on the concept of a floating MFC previously described by An et al. [25] and Huang et al. [26]. The new marine floating MFC features a constant inter-electrode spacing of 15 cm, making it particularly suitable for areas that experience variations in water column height due to tidal phenomena. Furthermore, the use of a floating system offers several advantages, including the ability to benefit from the natural agitation caused by wave motion. This agitation can promote mass transfer within the anodic compartment. Overall, this new design of marine floating MFC shows considerable promise for enhancing MFC performance in marine environments [27].

Petroleum hydrocarbons are the most commonly found environmental pollutants, and their introduction into a previously uncontaminated environment alters its nature and reduces its ecosystem functionality. The principal biological process responsible for the removal of these hydrocarbons from the environment is natural attenuation. This process involves the degradation of the organic pollutants by microorganisms already present at the site, without the need for external bioremediation enhancers such as electron donors, electron acceptors, other microorganisms, or nutrients [28].

The impact of hydrocarbon contamination on biota can vary greatly and may be influenced by several factors, including the presence of co-contaminants such as heavy metals. Studies have shown that the toxicity of petroleum hydrocarbons to microorganisms can be significantly increased when heavy metals are present as co-contaminants, as compared to when hydrocarbons are the sole contaminant [29]. Research by Khudur et al. (2019) has demonstrated that hydrocarbon-contaminated sites are more resistant to microbial bioremediation when co-contaminated with lead (Pb). This is because lead has the ability to inhibit numerous metabolic pathways, including enzymatic and respiratory processes of various bacteria, thereby creating additional stress on hydrocarbon-degrading species [30].

The primary objective of this study is to develop a composite material consisting of biochar and starch, which can be used as an anodic electrode for the removal of Pb(II) and petroleum hydrocarbons. Additionally, this composite material will also generate electricity from contaminated synthetic seawater through the use of a floating Microbial Fuel Cell (MFC) setup.

2 Methodology

2.1 Microbial Consortium

The petroleum hydrocarbon-degrading marine bacterial consortium MB 11, primarily consisted of *Enterococcus* sp. as depicted in Figure 1. This consortium was obtained from the Faculty of Science at Thaksin University in Thailand and was preserved in modified nutrient broth, which comprised 3 g/L beef extract, 5 g/L peptone, and 5 g/L NaCl until its utilization in subsequent experiments.

2.2 Synthetic Seawater

In the study, a modified method developed by Shi et al. [31] was employed to prepare synthetic seawater contaminated with Pb(II). The synthetic seawater was composed of various salts and minerals, including 5.00 g/L NaCl, 0.06 g/L Na_2HPO_4, 0.05 g/L KH_2PO_4, 0.02 g/L $CaCl_2$, 0.04 g/L $FeSO_4$, 0.14 g/L $MgSO_4$, and 200 mg/L $PbNO_3$. To ensure that the synthetic seawater was free of any microorganisms, it was sterilized at 121 °C for 15 min. Aseptic technique was employed to add 0.1% (v/v) diesel to the synthetic seawater before use.

2.3 Biochar Pyrolysis

The rice straw used in the experiment was purchased from the local market. To prepare it for use, the rice straw was dried at 60 ºC for 48 hr. It was then ground and sieved through a 40-mesh sieve before being subjected to pyrolysis. The pyrolysis process was carried out at a temperature of 300 ºC for 2 hr under an oxygen-limited atmosphere. To study the morphology of the resulting rice straw biochar surface, a scanning electron microscope (Quanta 450, FEI) was employed. This was done to gain a better understanding of the structure and characteristics of the biochar material. The biochar was activated using 30% (w/w) H_2O_2 solution before use.

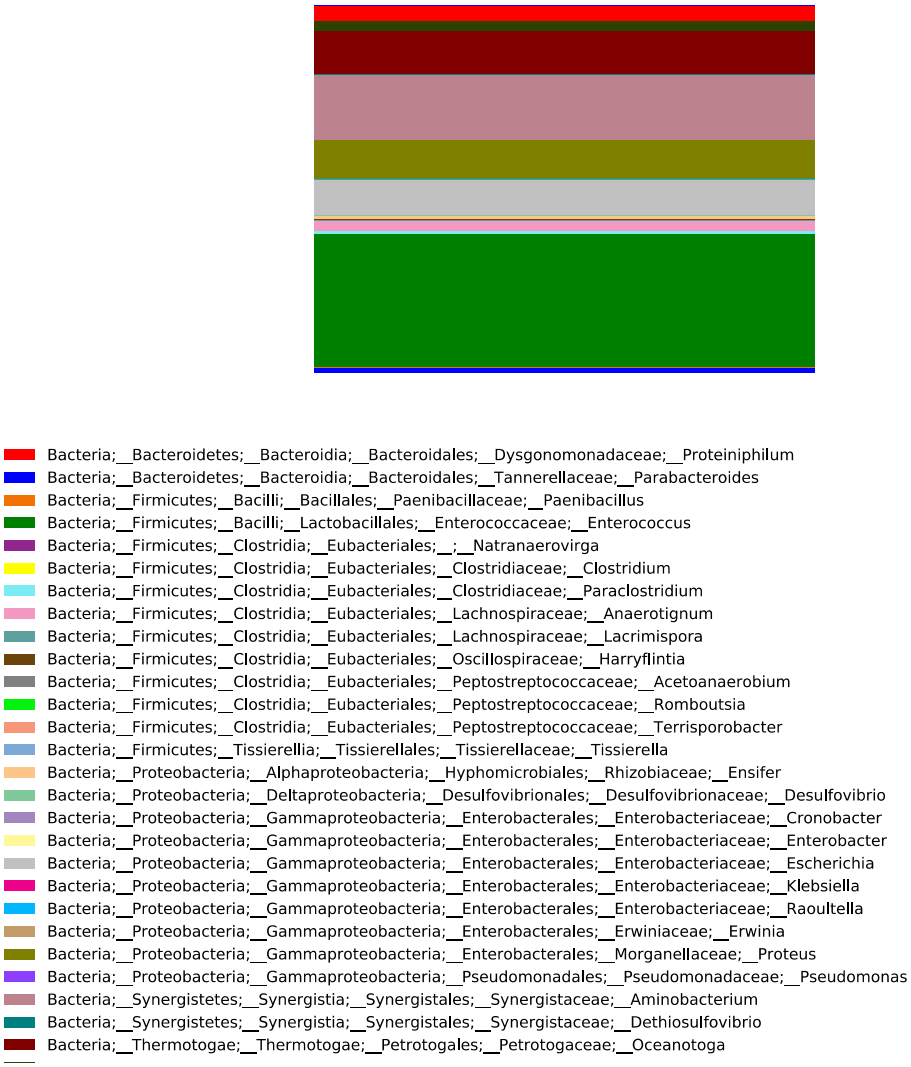

Figure 1: The petroleum degraded – marine bacterial consortium MB11 community.

2.4 Starch/Biochar Composite

In accordance with the method described by Kim et al. [32], heat-treated cassava starch was prepared at various temperatures ranging from 50 ºC to 180 ºC for 1 hr in the heating block. A 20% (w/v) concentration of cassava starch was used for the preparation. After the heating process, the cassava starch was allowed to cool.

To prepare the composite material, 30 g of rice straw biochar was mixed with 94 g of the heat-treated cassava starch for a duration of 30 min. The mixture was then placed into a circle-shaped mold and dried

for 48 hr at 60 ºC. Finally, the material was heated at a temperature of 130 ºC for 2 hr to produce the final product.

2.5 Pb(II) Adsorption

To investigate the adsorption of Pb(II), starch/biochar composites weighing 5 g were introduced into a solution consisting of 100 mL of synthetic seawater contaminated with 200 mg/L of Pb(II). The mixture was subjected to agitation at a speed of 150 rpm for 60 min.

The removal efficiency of Pb(II) was determined using the 2,5-dimercapto-1,3,4-thiadiazole (DMTD) method. Specifically, 1 mL of the sample was combined with 1 mL of DMTD solution and 2 mL of 0.005 M HCl. The resulting solution was allowed to incubate for 1 min and subsequently measured at a wavelength of 375 nm using a UV-Vis spectrophotometer. The adsorption capacity of the starch/biochar composites was calculated based on the obtained results and the specific formula used for this purpose.

$$\text{Pb(II) removal (\%)} = [(CA - CB) / CA] \times 100 \quad (1)$$

$$Qe\ (mg/g) = (CA - CE) \times (V/M) \quad (2)$$

Where Qe is the adsorption capacity (mg/g), CA is the initial Pb(II) concentration (mg/L), CB is the final Pb(II) concentration (mg/L), CE is the equilibrium Pb(II) concentration (mg/L), V is the volume of Pb(II) solution (mL), and M is the amount of biochar (g).

2.6 Floating MFC Design

In this experiment, a floating microbial fuel cell (MFC) was utilized as shown in Figure 2. The MFC comprised a 10 g starch/biochar composite and a stainless steel net as the anodic electrode, a ceramic plate as the proton exchange membrane [33], and an air-cathode constructed from Pt-coated carbon cloth. The floating portion of the MFC was made from polyethylene foam.

To immobilize the bacterial consortium, 10 mL of the active consortium in modified nutrient broth was added to the starch/biochar composite. The mixture was allowed to adsorb and immobilize for 24 hr at room temperature [34].

2.7 MFC Operation

During the experimental operation, the floating MFC containing the immobilized bacterial consortium was placed in a glass chamber filled with 100 mL of contaminated seawater. The open circuit voltage (OCV) was monitored every 60 minutes over a 24-hr period. To generate the polarization curve, the close circuit voltage (CCV) was measured at resistance values ranging from 300 to 5,000 Ω. The electrochemical properties were subsequently calculated using Ohm's law. The Pb(II) and petroleum hydrocarbon removal were measured.

$$I = V / R \quad (3)$$

$$P = I \times V \quad (4)$$

$$CD = I / D \quad (5)$$

$$PD = P / D \quad (6)$$

Where I is the current (A), V is the close-circuit voltage (V), R is the resistance (Ω), P is the power (W), D is the working volume (m^3) CD is the current density (A/m^3) and PD is the power density (W/m^3).

The quantification of petroleum removal was carried out based on the methodology described in a previous study. Specifically, a 10 mL volume of n-hexane was added to 100 mL of the sample and the resulting mixture was subjected to shaking for a period of 20 min. Subsequently, the solution was transferred to a separating funnel and the supernatant was collected. The absorbance of the supernatant was then measured at a wavelength of 225 nm using UV-Vis spectrophotometry, as outlined in reference [35].

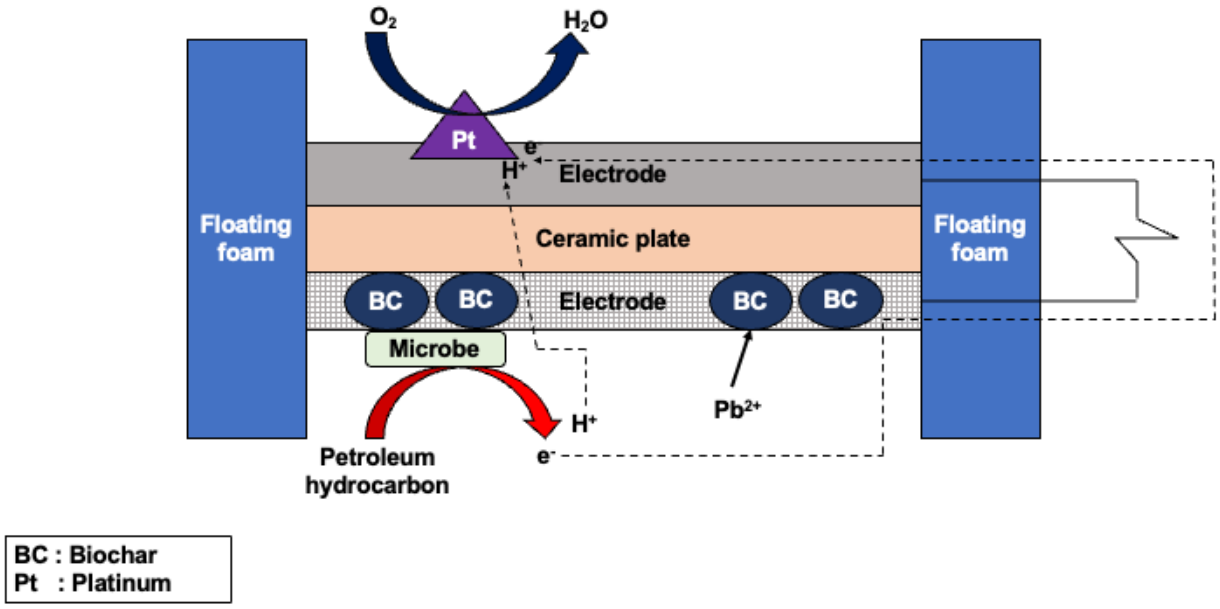

Figure 2: The floating MFC used in this experiment.

3 Results and Discussions

3.1 Biochar Morphology

Biochars are increasingly being acknowledged as environmentally friendly and cost-effective remediation agents for addressing pollution issues. In the case of rice straw biomass that contains amorphous carbon and amorphous silicon. The process of dehydration (below 250 °C) leads to the polymerization of silicic acid, resulting in a closer integration of carbon and silicon. When subjected to moderate pyrolysis temperatures (250–350 °C), a significant cracking of carbon components occurs which exposes the silicon present within the inner tissue. Upon exposure to high pyrolysis temperatures (500–700 °C), the biochar undergoes condensation due to the aromatization of carbon and the crystallization of silicon [36]. The present study involved the preparation of rice straw biochar through a low-temperature pyrolysis process conducted at a temperature of 300 °C. To analyze the surface morphology of the biochar, a scanning electron microscope was employed, and the corresponding results have been presented in Figure 3.

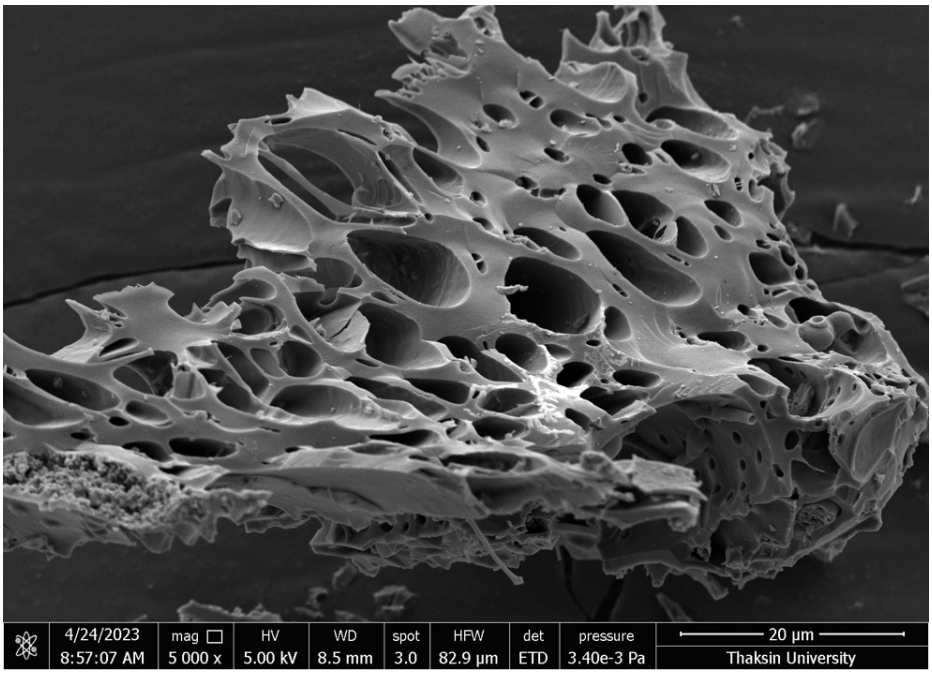

Figure 3: The surface morphology of pyrolyzed biochar.

3.2 Pb(II) Adsorption

Lead (Pb), a highly toxic heavy metal, is known to exist extensively in the soil, air, and water, posing a significant threat to the environment and human health. This non-biodegradable metal easily accumulates in living organisms, leading to a range of diseases and disorders, including carcinogenic, genotoxic, anemia, reproductive, and neurological effects, with children being particularly vulnerable. Given its widespread use in mining, metallurgical engineering, battery manufacturing processes, and traditional gasoline, the urgent need to develop effective methods to separate lead from the environment has garnered the attention of researchers and scientists worldwide [37].

In the present investigation, a starch/biochar bio-composite was synthesized by subjecting it to different heat-treating temperatures of 50, 80, 120, 150, and 180 ºC. The bio-composite was then tested for its ability to remove Pb(II) from an initial concentration of 200 mg/L. The percentage of Pb(II) removal was subsequently determined and presented graphically in Figure 4. The maximal Pb(II) removal of 93.10±1.50% was gained from the 150 ºC prepared starch/biochar composite with an adsorption capacity of 276±2 mg/g.

In the study conducted by Khandanlou et al., rice straw/magnetic nanocomposites were synthesized using the co-precipitation method to investigate their potential for removing Pb(II) from aqueous solutions. The researchers determined the optimum conditions for the adsorption of Pb(II) to be an initial ion concentration of 100 mg/L and an adsorbent dosage of 0.13 g. Under these conditions, the maximum removal efficiency of Pb(II) was found to be 96.25% [38]. Pyrolysis of rice straw can be used to produce biochar, which serves as a cost-effective and efficient adsorbent for removing metal contaminants from water. Biochar was created at different pyrolysis temperatures ranging from 300 to 500 °C, and its physicochemical properties were analyzed. Increasing the pyrolysis temperature enhanced the pH (from 8.23 to 10.25), surface area (from 12.78 to 64.56 m^2/g), and surface morphology of the biochar. The biochar produced at 500 °C demonstrated the highest adsorption capacity for Pb (II) with a value of 17.93 mg/g

[39]. Table 1 provides a review of the utilization of rice mill by-product biochar for the removal of Pb(II). The table presents a summary of the relevant information regarding the application of this adsorbent for the purpose of removing this metal ion.

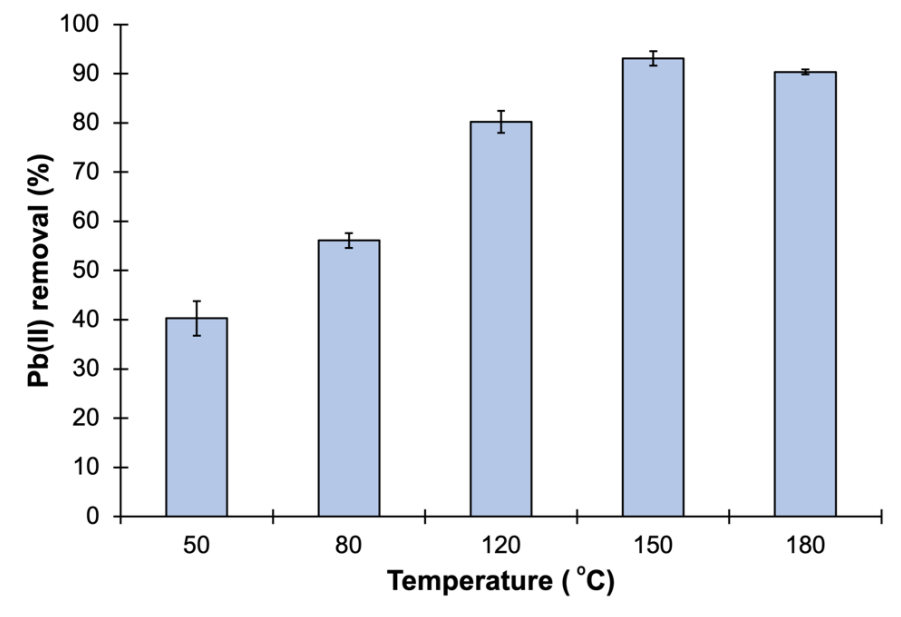

Figure 4: The Pb(II) removal of starch/biochar composites from the synthetic seawater.

Table 1. The utilization of rice mill by-product biochar for the removal of Pb(II).

Substrate	Pyrolysis temperature (°C)	Removal (%)	Adsorption capacity (mg/g)	Reference
Rice straw	300	93.10±1.50%	276±2	This study
Rice straw	700	99.00	-	[40]
Rice straw	400	-	131.24	[41]
Rice husk	600	90.00	-	[42]
Rice husk	300	-	240.13	[43]

3.2 Electrochemical Properties and Bioremediation

Wastewater generated by the petroleum industry is composed of complex constituents that contain carbon which is slow to decompose, also known as recalcitrant. The major contaminants commonly found in wastewater from petroleum refinery industries include volatile phenols, sulphides, benzene, ammonia, dissolved solids, suspended solids, cyanides, and nitrogen compounds [44, 45]. The hydrocarbons present in petroleum-contaminated water are collectively known as total petroleum hydrocarbons (TPHs), which comprise both aliphatic and aromatic hydrocarbons. Due to the recalcitrant nature of these compounds, they

can pose significant challenges to the effective treatment and remediation of petroleum-contaminated wastewater [46, 47].

Microbial fuel cells (MFCs) have been shown to be effective in treating wastewater that has low biodegradability, including petroleum-contaminated wastewater [48]. Several studies have reported treatment efficiencies for such wastewater in the range of 30-60% using MFCs. This suggests that MFCs are a promising technology for the treatment of recalcitrant wastewater, such as petroleum-contaminated wastewater. Further research is needed to optimize the performance of MFCs in treating such wastewater and to develop strategies for scaling up the technology for practical applications [44, 49].

In this study, a starch/biochar composite was utilized as the anodic electrode in a floating microbial fuel cell (MFC) for the treatment of seawater contaminated with Pb(II) and petroleum hydrocarbons, as well as for electricity generation. Results indicated that the maximum open circuit voltage (OCV) achieved was 851.08±5.76 mV, as shown in Figure 5. Additionally, the maximal power density (PD) and current density (CD) generated were 2.18±0.20 W/m^3 and 8.33±0.50 A/m^3, respectively as shown in Figure 6. In terms of contaminated water treatment, the maximum removal rates of 95.10±1.50% for Pb(II) and 55.10±2.20% for petroleum hydrocarbons were achieved in this experiment. These findings suggest that the starch/biochar composite has potential for use as an effective electrode material in MFCs for the treatment of contaminated seawater while also generating electricity.

In Abu-Reesh et al., The highest cell voltage of 762 mV and volumetric power density of 0.328 W/m^3 were achieved in a continuously operated dual-chamber microbial fuel cell (MFC) with a hydraulic retention time (HRT) of 16 hours. These results suggest that the MFC system has potential for generating electricity from wastewater while also providing effective treatment [50].

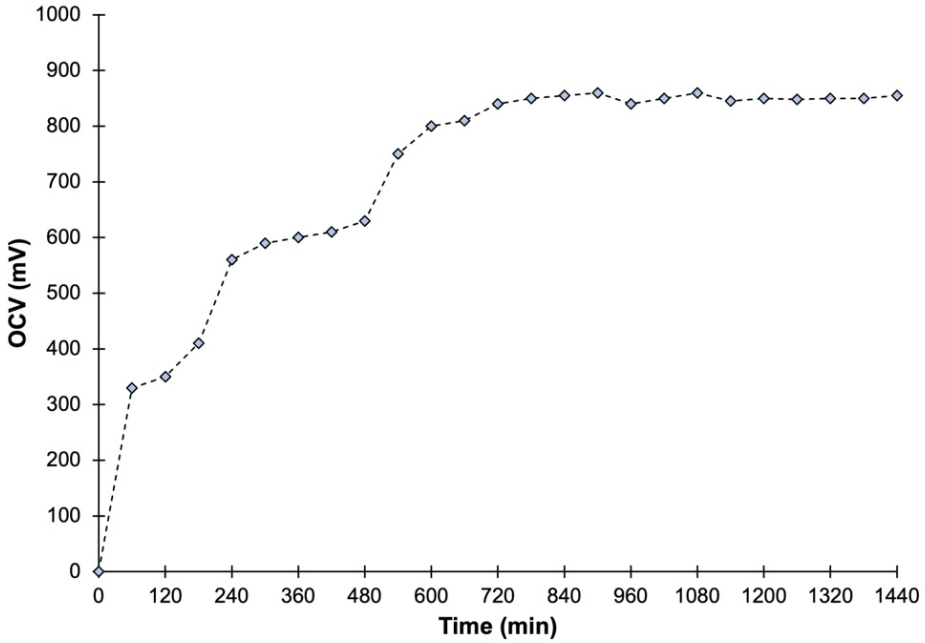

Figure 5: The open circuit voltage (OCV) of floating MFC with starch/biochar composite.

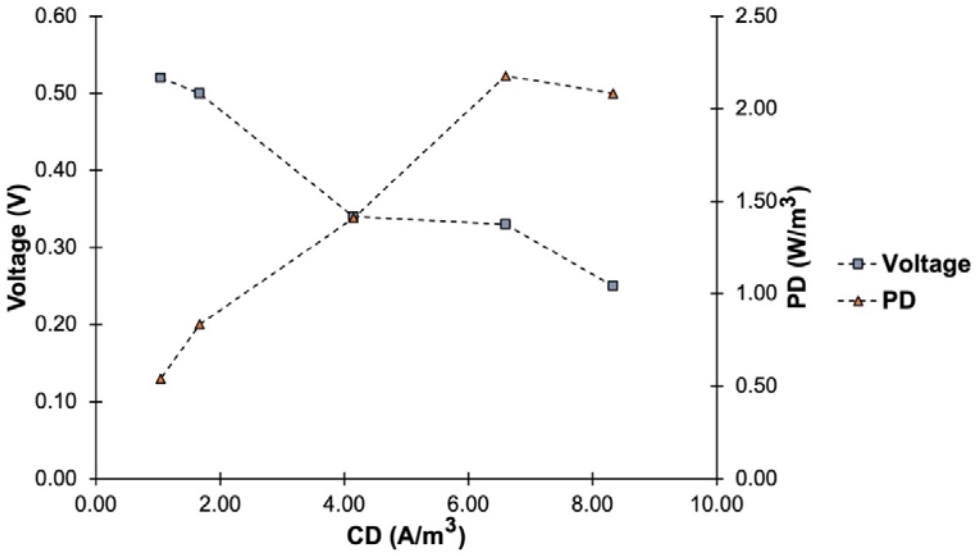

Figure 6: The polarization curve of floating MFC with starch/biochar composite

4 Conclusion

The results of this study demonstrate the potential of a starch/biochar composite synthesized from rice straw as an effective sorbent for the removal of Pb(II) from wastewater. Moreover, the composite exhibited promising performance as a microbial fuel cell electrode for generating electricity in the presence of Pb(II) and petroleum hydrocarbon contaminants. The maximal power density and current density achieved were 2.18±0.20 W/m^3 and 8.33±0.50 A/m^3 respectively, while the composite achieved removal rates of 95.10±1.50% for Pb(II) and 55.10±2.20% for petroleum hydrocarbons. These findings suggest that the starch/biochar composite has significant potential for use in environmental remediation and sustainable energy generation. Further research is warranted to optimize the composite's performance and explore its potential for real-world applications.

Authors' Contribution: PC all participated in idea generation, data analysis and drafted manuscript. PM, AK, and JT do experiment and data collection.

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Conflicts of Interest: The authors declare no conflict of interest.

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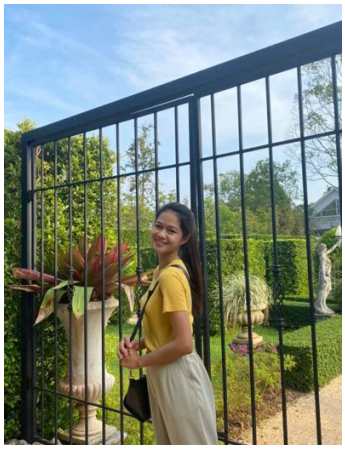

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