Effects of Different Biological Carriers in Microbial Fuel Cells

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ABSTRACT: In this study, to investigate their effects on battery power generation performance and wastewater treatment capacity, coal semicoke granular-activated carbon, granular graphite, and walnut shell-activated carbon were added to the anode compartment of a microbial fuel cell. As revealed from the experimental results, adding activated carbon and graphite can significantly decrease the startup time of microbial fuel cells as well as provide the shortest startup time of coal semicoke-activated carbon fluidized bed microbial fuel cells (MGAC-MFCs). The activated carbon particle diameter did not increase from 0.275 to 0.55 mm, and the voltage changed the chemical oxygen demand (COD) degradation efficiency. However, the 0.275 mm activated carbon exhibited a maximum open-circuit voltage of 935 mV as well as a COD degradation efficiency of 95%, and the operation cycle was shortened. After running a cycle, the COD removals of different systems were 85, 93, and 89%, and the maximum value was obtained by the MGAC-MFC system. After the activated carbon and graphite particles were added, the electrical performance and production capacity of the sewage treatment microbial fuel cells were significantly enhanced.

1. INTRODUCTION

Microbial fuel cells (MFCs) have emerged as an innovative power generation technology and have recently aroused substantial research interest. The selection of an appropriate reactor and biocarrier is of high significance. Specifically, the fluidized bed reactor acts as the anode chamber, and an interactive collision takes place between the sewage as the liquid phase and the biocarrier as the solid phase. Accordingly, microorganisms grow continuously on the biocarrier surface and are oxidized to decompose the organic substances in the sewage.1–6 In the MFC reaction of a fluidized bed reactor, activated carbon is added as a biological attachment material, promoting microorganisms to decrease the adaptive growth time6 and rapidly enriching the anode with microorganisms. Moreover, adhesion materials (e.g., nanowires and chitosan) are generated during microbial growth that can be tightly attached to the surface of the activated carbon and resist being washed away by water during the fluidization process.7 Various anode materials exhibit different conductivity and enrichment properties, so selecting suitable anode materials can enhance the electrical performance of MFC systems.

Thus far, numerous studies on graphite particles as biocarriers have been conducted. Compared with activated carbon, graphite particles have a relatively small pore size and a small specific surface area. In addition, their ability to adhere to microbes and adsorb organic matter is inferior to those of activated carbon. Their chemical oxygen demand (COD) removal rate is slightly lower than that of activated carbon microbial fuel cells. Biofilms on activated carbon are more developed than those on a graphite surface, primarily because activated carbon exhibits a porous structure and a rough surface, suggesting that microorganisms can be easily adsorbed on the membrane. Although graphite exhibits a layered structure, its specific surface area is relatively smaller than that of activated carbon, creating a narrower adhesion area for microbes, and it cannot easily form developed biofilms. However, compared with activated carbon, graphite particles exhibit better conductivity, which effectively reduces the internal resistance of biofuel cells and increases the open-circuit voltage and polarization power. In this study, a quartz tube fluidized bed reactor was taken as the anode chamber in the MFC system; in addition, activated carbon and graphite particles exhibiting different particle sizes were introduced as the solid phase fluidization medium and biocarriers and were compared with the battery system in the absence of biocarriers. The cultivated mixed bacteria were adopted to replace the conventional activated sludge. The effects of different electrode
2. MATERIALS AND METHODS

2.1. Experimental Materials. The novel mixed bacterial strains (e.g., bacillus, nitrifying bacteria, and photosynthetic bacteria) were added to the microbial fuel cell system as active substances for degradation and electricity generation.

2.2. Experimental Apparatus. In the present study, a microbial fuel cell acted as the experimental apparatus with a height of 800 mm and a diameter of 40 mm; its anode chamber was made of a cylindrical quartz material. A funnel-shaped end with a hole was connected at the bottom as well as a water intake device of 80 mm in diameter on its top. The schematic diagram is presented in Figure 1. Such an MFC reactor was filled with various activated carbon particles, and a carbon rod was inserted into the anode chamber as a working anode in the design. The air cathode was made of a hydrophobic carbon fiber cloth material exhibiting a diameter of approximately 2 mm in which the precious metal platinum content was 0.35 mg m\(^{-2}\).

The waste liquid storage tank was arranged in a thermostatic water bath. Under a peristaltic pump, the sewage in the storage tank was introduced to the MFC reaction system via a porous distribution plate. After a reaction in the reactor, the waste liquid was returned to the storage tank via a return pipe at the top of the anode chamber. The mentioned process was repeated in the reaction. The pollutants in the matrix in the reactor were metabolized and decomposed by the microorganisms distributed on the biocarrier as well as on the surface and inside of the anode. Additionally, the reaction generated numerous protons and electrons. The protons arrived at the cathode through the fluidization of the system, and electrons arrived at the cathode via the external circuit of the anode, thereby generating a stable current and output voltage. The prepared COD in a 2000 mg L\(^{-1}\) simulated sewage acted as the degradation substrate.

2.3. Preparation of Activated Carbon. First, the coal semicoke and walnut shells were repeatedly cleaned with deionized water and placed into a muffle furnace to be dried at 80 °C for 2 h. The dried raw materials were crushed and sieved in a 300 mesh and were adopted as pretreatment raw materials. The pretreated material was introduced to a fluidized bed reactor (Figure 2) and heated continuously in a N\(_2\) atmosphere as inert gas. As the temperature increased to 200 °C, the reaction started. The initial stage referred to the decomposition of a part of the organic matter to produce tar, etc. When the temperature rose to over 300 °C, carbonization took place, and the carbonization temperature was in the relatively wide range of 300–600 °C. The carbonization period ranged from 2 to 3 h. With the continuous increase of the temperature, the temperature required for activation was relatively high, generally at 800–1200 °C. At the beginning of activation, the gas was switched from inert N\(_2\) to active CO\(_2\), and the activation time was set to approximately 4–6 h. The schematic diagram is presented in Figure 3. When the experiments were achieved, the active gas was switched to N\(_2\), and the activated carbon samples were harvested and then selected to be characterized and analyzed.

2.4. Sewage Quality Evaluation in the Experiment and Inoculation Process. To investigate the microbial reaction and advanced microorganisms required for electricity generation and degradation, simulated sewage with simple components acted as the degradation substrate in this study, and the COD content was regulated by the added amount of sucrose. The simulated sewage primarily consisted of sucrose, inorganic salt, and other trace elements. It was maintained at a pH value of nearly 7.0 and a COD of approximately 2000 mg:
L. Initially, the separated microorganisms were acclimated. Afterward, all the microbial colonies applied for inoculation were cultivated to the logarithmic phase in the medium. The cultivation conditions were set to a strain ratio of 1:1. Mixtures with a blending ratio between bacterial solution and an acclimated sewage of 1:100 were inoculated into the simulated sewage with a COD of 200 mg·L⁻¹, then, they were cultivated on an oscillating table at 30 °C, acclimated for 8 h, and added to the MFC reaction system.

2.5. Evaluation of the Wear Resistance of Activated Carbon. In this part, the wear resistance ability of the activated carbon was determined. After the carrier M1 was weighed in 50 mL of water, it was magnetically stirred for 8 h to remove the residue. Subsequently, it was put into the drying oven and dried to a constant weight at M2. The equation for calculating the wear rate of the carrier is

\[ \delta = \frac{(M1 - M2)}{M1} \times 100\% \]

3. RESULTS AND DISCUSSION

3.1. Industrial Analysis and Elemental Analysis of Raw Materials. The representative walnut shell and coal semicoke samples were selected as the raw material for activated carbon preparation, and their ultimate and proximate analyses are listed in Table 1. The volatiles and ash involved in the coal semicoke reached 32.81 and 4.07%, respectively, while the fixed carbon content was only 59.31%. In the activation, volatiles were released from the semicoke, and partially closed pores were opened, so more pores were generated. For the walnut shells, the volatiles accounted for only 1.71%, while the fixed carbon content reached 68.45%, indicating that the formation of the developed pore structure was limited, whereas a high yield could be obtained. In general, both raw materials were acceptable for preparing the activated carbon.

3.2. The Physico-Chemical Property Analysis of the Activated Carbon. The wear resistances of the different biocarriers are listed in Table 2, showing that the wear rate was 2.7% when coal semicoke was used as the biocarrier, exhibiting high hardness and good wear resistance. In addition, the wear resistance of graphite was the worst with the lowest hardness and a wear rate of 4.3%. Thus, a comparison for the wear rates showed that coal semicoke-activated carbon was the most suitable for biocarriers in the fluidized bed reactor in the MFC system.

Table 1. Proximate and Ultimate Analysis of the Walnut Shell and Coal Semicoke

| material          | proximate analysis (%) | elemental analysis (% dry basis) |
|-------------------|------------------------|----------------------------------|
|                   | M  | A  | FC | V  | C  | H  | O  | N  |
| walnut shell      | 7.48| 1.71| 68.45| 22.36| 57.05| 9.03| 32.02| 0.19|
| coal semicoke     | 3.81| 4.07| 59.31| 32.81| 62.01| 1.36| 21.86| 1.44|

Table 2. Wear Rates of the Three Organism Carriers

| biocarrier     | M1 (g) | M2 (g) | wear rate (%) |
|----------------|--------|--------|---------------|
| walnut shell   | 30     | 28.92  | 3.6           |
| coal semicoke  | 30     | 29.19  | 2.7           |
| graphite       | 30     | 28.71  | 4.3           |

Table 3. Activated Carbon and Commercially Activated Carbon for Refinement of Injection

| activated carbon | pore volume (cm³·g⁻¹) | mesopore volume (cm³·g⁻¹) | surface area (m²·g⁻¹) | average pore size (nm) |
|------------------|------------------------|---------------------------|------------------------|------------------------|
| HGAC             | 0.43                   | 0.37                      | 1069.67                | 1.91                   |
| MGAC             | 0.45                   | 0.47                      | 1104.15                | 3.29                   |

Figure 4. N₂ adsorption–desorption isotherms of the walnut shell and coal semicoke-activated carbon.

Figure 5. Pore size distribution for the walnut shell and coal semicoke-activated carbon.

Figure 6. FT-IR spectrum of the (a) walnut shell and (b) walnut shell-based-activated carbon.
3.3. Brunauer–Emmett–Teller (BET) Analysis for Different Activated Carbons. As shown in Figure 4, the nitrogen absorption–desorption isotherm curves of the walnut shell-activated carbon (HGAC) could not completely comply with those of coal semicoke-activated carbon (MGAC), and an obvious retention space formed by the macropores and mesopores was contained in the activated carbon. Although the activation time was relatively long when carbon dioxide acted as an active agent, a diverse pore structure (e.g., micropores, macropores, and mesopores) was beneficial for the photocatalysis load and microbial attachment. The isotherm adsorption curve of the coal semicoke-activated carbon was

Figure 7. FT-IR spectrum of coal char and coal char-activated carbon. (a) Coal-based semicoke and (b) coal-based semicoke-activated carbon.

Figure 8. Voltage curve of the MGAC-MFC, GG-MFC, HGAC-MFC, and MFC at the startup period.

Figure 9. Voltage generation of the MGAC-MFC, GG-MFC, HGAC-MFC, and MFC.

Figure 10. Variations of the COD removal rate with operation time in different systems.

Figure 11. Curve of COD versus operation time with different particle sizes of activated carbon as the biocarrier.

Figure 12. Polarization curve and power density curve of MGAC-MFC with different particle sizes of activated carbon as the biocarrier.
basically a type IV curve, and an H3-type hysteresis loop appeared between $P/P_0 = 0.6−0.9$. For the walnut shell-activated carbon, as $P/P_0$ increased from 0 to 0.1, the adsorption quantity rapidly increased, reaching the saturated adsorption value and fitting a type I adsorption isotherm for a $P/P_0$ of up to 0.3. Activated carbon exhibiting a developed pore structure and a high specific surface area was formed from the coal semicoke and walnut shells, which is a beneficial supporting skeleton structure for microbial carriers and catalysts (Figure 5). The pore size and surface area data of different activated carbons are shown in Table 3. The two activated carbon parameters meet the requirements.

3.4. FT-IR Analysis for Activated Carbon. The prepared walnut shells and coal semicoke were harvested for Fourier transform infrared (FT-IR) analysis, and the results are shown in Figures 6 and 7. It is suggested that the surface functional group of coal semicoke was similar to that of the walnut shell, and a strong adsorption peak appeared at 3500−3000 cm$^{-1}$, which was attributed to the stretching vibration of alcohols, phenols, some organic acids, and O−H bonds or N−H bonds. The stretching vibration of saturated C−H bonds might cause an adsorption peak at 3000−2500 cm$^{-1}$. However, compared with the walnut shells, the peak intensity decreased at this location, which could be explained by considerable volatiles being released in this period. The adsorption peak of the C=O stretching vibration at 200−1000 cm$^{-1}$ and the C−O stretching vibration at 1000 cm$^{-1}$ indicated that carboxyl and ester functional groups can exist in activated carbon with walnut shells and coal semicoke as biocarriers. As suggested from the figure, a weak adsorption peak was observed at 1000 cm$^{-1}$ for the walnut shells, while for the coal semicoke, an apparent peak appeared. It can be concluded that C−O bonds existed in the coal semicoke, whereas C−O bonds hardly existed in the walnut shells.

3.5. Effect of Biocarriers on Microbial Fuel Cell (MFC) Startup Time. The startup time of the microbial fuel cells was the growth period of the microorganisms. As the microorganisms adapted to the growth environment, they started to reproduce and load on the surface as well as inside the pores of the different biocarriers. For the various surfaces and structures of the different activated carbon materials, the effects of the different attachment and reproduction mechanisms for the microbes should be assessed. The cultivated mixed strain and activated carbon were added to different MFC fluidized bed devices. To study the effect of different biocarriers on the startup time, simulated sewage with a COD of 2000 mg·L$^{-1}$ was fed into the system on a day-to-day basis. At the startup stage of the battery, the temperature was fixed at 30°C, and the relation between the time and output voltage is presented in Figure 8.

Figure 13. SEM image of activated carbon. (a, b) Coal semicoke-activated carbon. (c, d) Walnut shell-activated carbon.

Figure 14. SEM image of microorganisms on a graphite biocarrier. (a) Granular graphite. (b) Biological granular graphite.
As suggested from the results, the startup time of the MFC without activated carbon was 108 h. The voltage variation curve represents the inoculation of the mixed strains in GG-MFC and MGAC-MFC. The maximum values were reached for the first time at 30, 35, and 40 h. In addition, the second maximum voltage values occurred at 75, 70, and 81 h. Obviously, the startup times of the different MFC systems were shortened significantly in the presence of activated carbon. In contrast to those of the activated carbon with graphite and walnut shells as bio-carriers, the increase in the startup time of the coal semicoke was more significant, and a transitory stable period was presented at both peak times. It could be concluded that a more developed pore structure and various pores existed in the coal semicoke, facilitating the attachment and growth of microorganisms. Because of the relatively good conductivity of graphite, the output voltage of the graphite system was relatively high for a period. However, since the generation of the voltage was primarily dependent on the microorganism metabolism to produce electrons in the later period, the voltage in the coal semicoke active system was higher than that of the graphite particle system.

3.6. The Effect of Biocarriers on the MFC Output Voltage. As bio-carriers, the graphite particles and two different activated carbons provided attachment sites for microbial growth. The attachment and enrichment of microorganisms as well as the formation of biofilms were directly impacted by the specific surface area. Moreover, the specific surface area was directly affected in the reaction and electron transfer. The types of bio-carriers significantly impacted the performance of the MFCs. The power generation performance of the fuel cell in one cycle is shown in Figure 9.

As revealed from the results, the output voltage of the coal-based semicoke system was the highest, reaching 535 mV. In the operation of the MGAC-MFC system, the microorganisms were rapidly enriched at the anode, and the electric energy was directly increased by the metabolism of the microorganisms. Moreover, the increase in the electrical energy in the MFC system was not directly proportional to the specific surface area of the biocarrier; it was related to the microbial species, metabolic capacity, growth rate, and the performance of the electrode material itself. As suggested from the figure, domestication can shorten the startup time of the MFC and enhance the electrical performance. Graphite is a good conductor, and adding graphite as a biocarrier can reduce the internal resistance of the system and increase the output current. The voltage increased rapidly in the initial stage, reaching a maximum of 875 mV at 30 h, which was higher than that of the other systems in the identical period; however, since the graphite particles were not suitable bio-carriers, the voltage increased slowly in the later period, and the overall voltage curve was lower than that of the coal semicoke system. A transitory period took place after the logarithmic period during microbial growth, the voltage decreased, and the voltage curve presented a descending trend, primarily caused by the inhabitation effects of the nitrifying bacteria on the growth of the photosynthetic bacteria.

3.7. The Effect of Biocarriers on COD Removal of Simulated Sewage. The COD removal rate of different bio-carriers in system operation is illustrated in Figure 10. After one cycle, the COD removal rate reached 85.7, 31.8, and 24.4%, marking increases of 29.9, 31.8, and 24.4%, respectively, compared with the condition without bio-carriers. The removal rate curve of the coal semicoke was higher than those of the other two carriers, primarily due to the synergetic effect between the adsorption of coal semicoke-activated carbon and the metabolic decomposition of microorganisms. The mixed microbial community introduced to the reaction was attached onto the surface and interior pore of the activated carbon to form the biofilm-activated carbon. The complex pore structure of activated carbon created active sites for the growth of microorganisms, which were tightly attached onto the pores of activated carbon and would not be washed away by water flow during the reaction in the fluidized bed reactor. Biofilm-activated carbon was integrated with organics in sewage to complete sewage degradation via metabolic processes. Meanwhile, under the synergism of activated carbon adsorption and degradation of microorganisms, the sewage absorbed in the pores of activated carbon could fully react with the attached microorganisms. Furthermore, the pore structure of activated carbon after the degradation was regenerated and reduced, and it continued to absorb the substances in the sewage, thereby further elevating the degradation rate and electron transfer rate of organics.

Moreover, as indicated from the results, all three biocarrier particles were conductive materials. The electrons generated in the microbial catalytic organic degradation process can be transferred via the biocarrier in the fluidization process in the fluidized bed, thereby further enhancing the electron transfer and gradually enhancing the reaction performance in the MFC system. Compared with activated carbon, the layered structure of graphite was not suitable for the attachment growth of microorganisms, and only a small number of microorganisms could be attached on the graphite surface. Thus, the COD degradation efficiency of the granular graphite system was lower than that of the activated carbon system.

3.8. The Effect of Coal Semicoke Particles on COD Removal of Simulated Sewage. To investigate the effect of particles on the COD removal in the system, coal semicoke-activated carbon particles with average particle sizes of 0.275, 0.375, and 0.55 mm were added to the system. As a result, after operation for one cycle, the COD removal rate reached 91.2 and 90.4%. Figure 11 suggests that there were no obvious differences in the COD degradation capacities of the three systems with different particle sizes, whereas the COD degradation rate of the 0.275 mm system stabilized in approximately 40 h, while the other two groups took 47 and 53 h. Because of the porous structure of the biocarriers, in a certain range, the smaller particles exhibited a larger specific surface area, which was more conducive to the attachment and growth of microorganisms. The biocarriers with small particle sizes could promote the rapid propagation of microorganisms and rapidly elevate the quantity of microorganisms; as a result, the degradation efficiency of COD was enhanced.

3.9. The Effect of Biocarrier Particle Size on the MGAC-MFC Electricity Production Performance. The particle sizes of the activated carbon critically affected the stability of the MFCs. Thus, to investigate the effect of the particle size on MGAC-MFC for generating power, coal semicoke-activated carbon with various particle sizes of 0.275, 0.375, and 0.55 mm were added to the system. The power curve obtained by measuring the output voltage and current is exhibited in Figure 12. It is suggested that the power reached a maximum of 373.1 mW·m⁻² in the MFC system when the 0.275 mm coal semicoke particles acted as the biocarrier. As shown in the figure, as the particles increased in size, the
output power decreased, and the maximum value of the power was 357.5 W·m⁻² for a particle size of 0.55 mm. This could be explained by the fact that the small particle size semicoke has a higher surface area and a more developed pore structure, facilitating the growth and reproduction of microorganisms. This material had obvious advantages, significantly helping the anode material to achieve microbial enrichment. Moreover, it can promote microbial metabolism and electron transfer, leading to enhanced electrical performance of the MGAC-MFC system.

3.10. SEM Analysis of Biofilm-Activated Carbon. A biofilm was generated on the surface of the biocarrier in the reaction stage. Small amounts of coal semicoke-activated carbon, walnut shell-activated carbon, and granular graphite were collected and then fixed for the scanning electron microscopy (SEM) test. The SEM images are shown in Figure 13. The mixed bacteria were primarily Bacillus and Staphylococcus aureus, and in the growth of the biofilm on the surface of the activated carbon, a single colony was attached. The coal semicoke-activated carbon had a larger pore size, which facilitated the growth of the microorganisms. Additionally, numerous microorganisms adhered to the surface and interior of the activated carbon, thereby forming biofilms.

A comparison of Figures 13 and 14 shows that the surface and interior of the activated carbon were filled with microorganisms, and abundant microorganisms existed in regular holes, while no microorganisms attached to the graphite layers. As revealed in the results, the original layer structure maintained development, whereas the graphite surface was covered with microorganisms. The activated carbon acted as a porous material with a variety of internal large holes and mesoporous structures. The surface was rough, and its pore structure was more regular, which facilitated microbial growth and reproduction. However, the internal structure of the graphite was layered with a small space between the layers and a relatively small specific surface area, so it was not prone to forming more developed biofilms for microorganisms to attach to on the graphite surface.⁹,¹⁷,22 Almost no microorganisms could be identified between the graphite internal layers.

4. CONCLUSIONS

In the present study, the effects of different biocarriers on MFCs regarding generating power and degrading COD were investigated. In addition, activated carbon and graphite particles prepared from coal semicoke and walnut shells were employed, and the conclusions obtained from this study are listed as below.

The startup time of the MFC was obviously shortened in the absence of a biocarrier, and the activated carbon MFC prepared from coal semicoke yielded the shortest startup time. The output voltage and power of the MFC were significantly improved in the presence of activated carbon, and graphite particles were added, which decreased the internal resistance and increased power generation. A favorable sewage treatment result could be observed in both the MGAC-MFC and GG-MFC systems, and the COD removal rates reached 86 and 79% after a cycle, respectively. As the particle sizes of the semicoke-activated carbon increased from 0.275 to 0.550 mm, the output power decreased. However, different particle sizes exerted limited effects on the output voltage and COD removal rate, and a stable run time and shorter removal degradation time were achieved in the 0.275 mm system.

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