8 Engineering Perspectives on the Application of Photosynthetic Algal Microbial Fuel Cells for Simultaneous Wastewater Remediation and Bioelectricity Generation

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CONTENTS

8.1 INTRODUCTION

Renewable bioresources like solar, water, and wind hold immense potential, but their inefficient utilization along with high operational and installation costs is a significant drawback in harnessing them at full capacity. To overcome the above-mentioned drawbacks and improve the overall process yields, low-cost microbial fuel cells (MFCs) and photosynthetic microbial fuel cells (PMFC) were sought after. However, MFCs are often crippled with problems like CO_2 discharge to the environment and energy intensity due to mechanical aeration for oxygen supply, constituting 50% additional cost. Integrating this technology with algae can tackle the bottlenecks, as they consume

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CO_2 during photosynthesis (reducing carbon footprint) and release oxygen (acting as an electron acceptor) which can help increase the power output. Figure 8.1 represents the working of a typical photosynthetic algal microbial fuel cell (PAMFC). Such algae-assisted MFCs were also reported to generate more power density compared to stand-alone MFCs (Zhang et al., 2019).

The fundamental mechanism of a PAMFC process is that the bacteria present in the anodic chamber utilize the organic matter of wastewater in an anoxic environment and release CO_2, electrons, and hydrogen ions (Kannan & Donnellan, 2021). The electrons pass through an external circuit to reach the cathodic chamber, where the algae take up sunlight and CO_2 (from anodic chamber) to photosynthesize and release oxygen, while generating biomass (Arun et al., 2020). This released oxygen acts as an electron acceptor for the generation of bioelectricity. Specifically, microalgae have great potential to be employed for PAMFC as they can utilize nitrogen and phosphate from wastewater, assisting in pollutant removal and thereby eutrophication (Arun et al., 2022). In addition, the algae can be harvested after each cycle to extract pigments and lipid for biofuel production. The biomass can also be used as substrate in the anode for subsequent processes to stimulate sustainability (Shukla & Kumar, 2018). Various factors like pH, temperature variations, substrate type, CO_2 and oxygen concentration, light intensity, and illumination devices affect the working of a PAMFC, thus affecting the power output. These factors need to be optimized for sustainable and efficient bioelectricity generation (Reddy et al., 2019).

Equations 8.1–8.3 are the biochemical reactions that take place in an electrochemical cell (He et al., 2014):

Anodic chamber:

$$C_6H_{12}O_6 + 6\,H_2O \rightarrow 6\,CO_2 + 24\,H^+ + 24\,e^- \tag{8.1}$$

Cathodic chamber:

$$n\,CO_2 + n\,H_2O \rightarrow \text{Algae+ light}(CH_2O)\,(\text{Biomass}) + n\,O_2 \tag{8.2}$$

$$O_2 + 4\,H^+ + 4\,e^- \rightarrow 2\,H_2O \tag{8.3}$$

Researchers have conducted lab-scale studies using microalgae as a carbon source in the fuel cells, which solely focused on the metabolic role of algae in different compartments and the production of bioelectricity (Enamala et al., 2020). Addition of substrate to the photosynthetic and bacterial

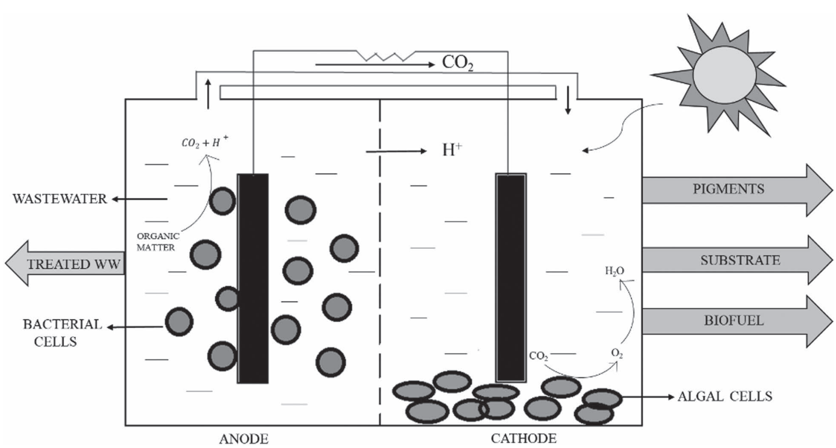

FIGURE 8.1 Schematic representation of a typical PAMFC setup.

components like glucose, acetate, and wastewater from different industries have been observed to enhance bioelectricity production (Arun et al., 2020; Eom et al., 2020). Especially, acetate and domestic wastewater have shown prominent results as substrate source in this aspect (Bhande et al., 2019; Firdous et al., 2018; Pant et al., 2010). However, wastewater treatment domain is not completely explored due to various limitations concerning efficiency, cost, etc., which has further prevented the scale-up of the technology. The knowledge gaps in simultaneous wastewater technology and production of value-added products have also limited the real-time use of this technology. The applications and various value-added products that can be obtained from an PAMFC system is showed in Figure 8.2.

The aim of this chapter is to analyze the research interest and trends of MFCs and its advancements since the very commencement of research in this area. Second, the chapter reviews the research activity conducted in the last 5 years to develop an understanding of the significant factors affecting the power density output in an PAMFC. Third, it throws light on the cost-affecting factors and the application of this integrated system in developing a zero-carbon output, sustainable wastewater treatment technology. In addition, the chapter deals with the efficiency of the PAMFC to produce electricity along with other benefits using algae in such bioeletrochemical systems. Lastly, the chapter addresses the setbacks in scaling up and commercialization of the technology and the future scope in developing into an efficient system.

8.2 OVERVIEW OF RESEARCH ACTIVITY ON MICROBIAL AND PHOTOSYNTHETIC MICROBIAL FUEL CELLS

The generation of electricity using bacteria was reported in the early twentieth century (Apollon et al., 2021). However, the number of publications on using algae as a biocathode material for simultaneous wastewater management and bioelectricity generation has increased only in the last 10 years. In order to get a comparative preview, the number of hits obtained from different online databases like Google Scholar, Web of Science (WOS), and Scopus, from 2001 to 2022, using various keyword combinations like "Microbial Fuel Cell," "Photosynthetic Microbial Fuel Cell," "Algal Microbial Fuel Cell," and "Bacterio-Algal Fuel Cell" are shown in Table 8.1. The result showed that "microbial fuel cell" got the highest number of hits from all the three databases, whereas photosynthetic/algal fuel cell was relatively a less explored area. To get a more detailed view of the research work conducted on the microbial fuel cells containing algae, all the bibliographical data collected using the previously mentioned keywords were fed to a scientometric analysis software, called Citespace 5.8.R3. In Figure 8.3, the number of published articles on microbial fuel cell was

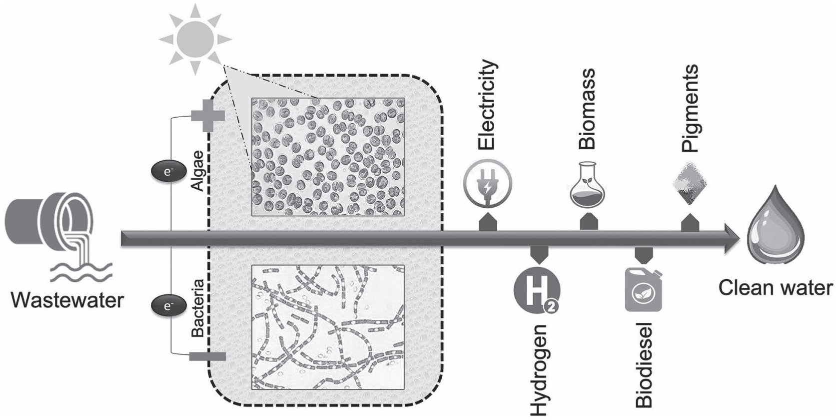

FIGURE 8.2 Various value-added products that can be obtained from an PAMFC system.

TABLE 8.1
Number of Publication Found with Different Keyword Combinations from Different Online Repositories from 2001 to 2022

Keywords Used	**Number of Hits Obtained from Different Online Repositories**		
Google Scholar		**Scopus**	**Web of Science**
Microbial fuel cell	21,000	12,268	12,216
Photosynthetic microbial fuel cell	5,380	242	323
Algal microbial fuel cell	4,800	177	229
Algal fuel cell	37	688	659
Photosynthetic algal microbial fuel cell	283	51	53
Bacterio-algal fuel cell	4	1	1

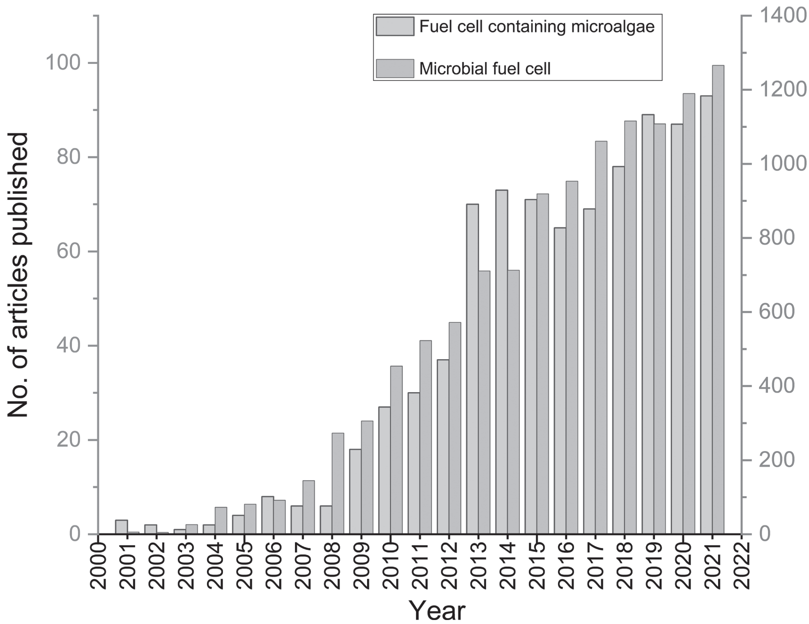

FIGURE 8.3 Number of articles published in the area of microbial fuel cell and microbial fuel cell containing microalgae in the last two decades.

compared with the number of articles on the microalgal fuel cell published over the last two decades. The result depicts that even though articles focusing on the microalgal fuel cell were less, it showed a steady growth over the years. Here it is worth mentioning that especially in the last decade, the rapid growth in the fuel cell containing photosynthetic microorganisms has gained more focus due to the realization of the advantages offered over the bacterial ones. Further, bibliographical analysis on the microbial fuel cell containing algae showed that 68% of the published documents are scientific research articles and 17% are review papers (Figure 8.4a). The subject distribution of the data set showed that 21% of the published documents fall under the area of "Environmental science," whereas other areas like "Energy," "Chemical Engineering," and "Biochemistry, Genetics

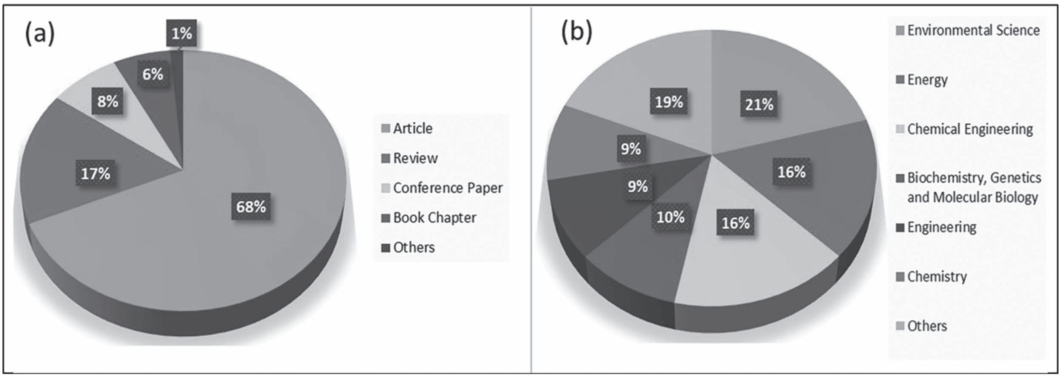

FIGURE 8.4 Distribution of the different types of published documents (a) in different areas and (b) in the field of algal microbial fuel cell.

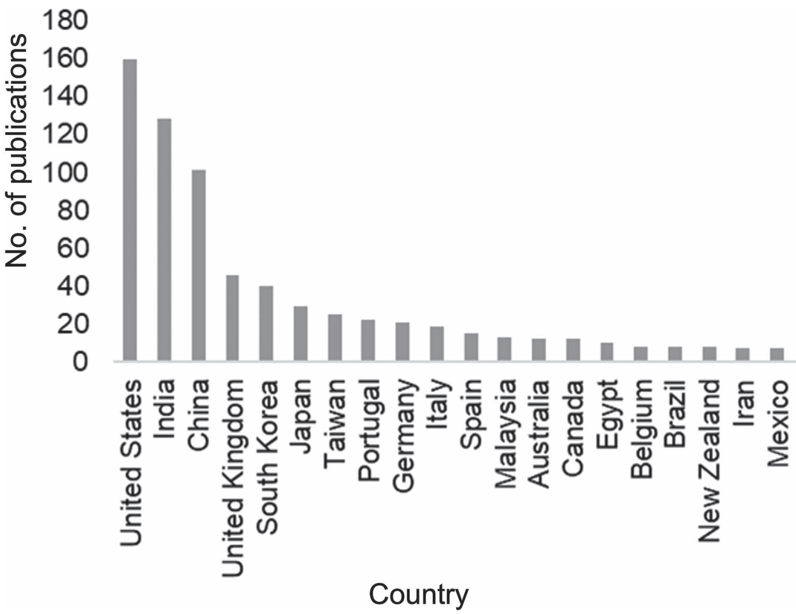

FIGURE 8.5 Top 20 contributing countries/territories in the field of algal fuel cell.

and Molecular Biology" contain 16%, 16%, and 10% of the total published documents, respectively (Figure 8.4b). This means that the latter aspects of the systems are yet to be explored. Among the different contributing countries in this field, the United States holds the first position followed by India and China (Figure 8.5). Apart from that, other Asian countries like South Korea, Japan, Taiwan, and Malaysia also showed significant contribution to this field.

8.3 IMPORTANT FACTORS INFLUENCING THE FUNCTIONING OF A PAMFC

For the efficient working of a PAMFC and maximum power output, various factors like pH, temperature, light intensity and artificial illumination devices, carbon dioxide and oxygen concentration, substrate type, and design configuration need to be studied closely.

8.3.1 pH

pH plays a crucial role in regulating the metabolic pathways taking place inside algae and bacteria, which aids in bioelectricity generation (Zhang et al., 2019). Microbes tend to perform optimally at low ion concentration and neutral pH. Thus, pH at anode should ideally vary between 6 and 7 and the pH at cathode can be neutral or slightly alkaline to be favorable for algal growth. Also, this condition is inevitable due to oxygen reduction (Arun et al., 2020). Even in single-chambered MFCs or with air-cathodes, both the rise and fall in pH affects the efficiency. Low pH affects the membrane efficiency (Saba et al., 2017), but a pH of 9.5 showed to generate a maximum power density of 0.66 W/m^3 (Kusmayadi et al., 2020). CO_2 generated by bacterial cells in an anode compartment can interact with water and form H_2CO_3, which can lower the pH. This issue can be prevented by inoculating algae at a high initial concentration for simultaneous CO_2 assimilation (Zhang et al., 2014). Another reason for the acidification of the anodic chamber may be due to the production of fatty acids by non-electrogenic organisms under an anaerobic environment (Zhang et al., 2019). Maintenance of pH can be brought about by buffers like MES (2[*N*-morpholino]ethane sulfonate), HEPES(4(2-hydroxyethyl)1-piperazine ethane sulfonic acid), PIPES (piperazine-*N*,*N′*-bis[2-ethane sulfonate]), caustic soda, carbonate, and zwitterionic buffers. For large-scale applications, carbonate buffers are most economical (Shukla & Kumar, 2018), but they can also accelerate the growth of methanogens and are non-biodegradable, resulting in eutrophication when released into waterbodies (Wang et al., 2018). However, zwitterionic buffers are stable, non-toxic, and do not adversely affect the metabolic pathways of the microorganisms (Kusmayadi et al., 2020). Ideally, algae in the cathode should utilize protons and electrons and generate water and oxygen. However, when non-noble carbon electrodes are used for being more economical, oxygen reduction in the cathode results in the production of hydroxyl ions. This observation was confirmed by Wang et al. (2018) that anode acidification can be balanced by the production of hydroxyl ions in the cathode area without the addition of buffers.

8.3.2 Temperature

Shukla and Kumar (2018) reported 35 °C as the optimum working temperature, which may vary with the organism used. Temperature and dissolved oxygen (DO) concentration are directly interdependent; as a result, an increase in temperature results in an increase in DO. Seasonal variations in temperature also affect the algal efficiency due to variation in photosynthetic activity (Saba et al., 2017). Higher operating temperature resulted in improved reaction kinetics and higher power density along with better nutrient removal due to faster rates of biochemical reaction. Higher reaction rates are because of rapid substrate absorption (Kusmayadi et al., 2020) and faster movement of electrons (Inglesby et al., 2013). Increased membrane conductivity was observed at higher temperatures ranging between 20 and 60 °C with proton-exchange membranes (Pérez-Page & Pérez-Herranz, 2011). However, extensively high temperature decreases the membrane conductivity due to water deficit in the membrane, as membrane hydration is an important factor in determining its durability and performance. Dehydrated membranes show higher ionic resistance and can even be damaged irreversibly (Shukla & Kumar, 2018). It was observed that with the temperature increase from 25 °C to 30 °C, the maximum power density increased somewhat from 1811.9 mW/m^3 to 2072.1 mW/m^3, but higher temperatures (38 °C) also lead to cell death (He et al., 2014).

8.3.3 Light Intensity and Artificial Illumination

In a photobioreactor, the most influencing factor affecting the growth of algal biomass is found to be light intensity and spectral composition (Greenman et al., 2019). Major emphasis has been laid on the light source and intensity as it impacts the chlorophyll content, stomatal opening, and photosynthesis. When light intensity varies between 3500 and 10,000 lux, algal biomass concentration increases gradually, but beyond this range, the PAMFC becomes saturated (Arun et al., 2020). The cultivation of biomass in the presence of substrate and light can be seen in Equation 8.4:

$$\text{Organics} + \text{light} \rightarrow \text{Biomass} + \text{Oxygen} \tag{8.4}$$

Biomass and oxygen are produced during the light period and the algae consume the oxygen produced and reduce the organic matter during the dark period. Further, it was found that red light with high intensity (900 lux) gave better power output as compared to blue light with low intensity (100–600 lux) (Jaiswal et al., 2020; Mekuto et al., 2020). This occurs due to the high absorption of light energy by the photosynthetic system of algae at this wavelength. Jaiswal et al. (2020) further reported that continuous supply of light intensity is better than batch-mode operation. On the downside, the light intensity is a crucial parameter that needs to be monitored closely as low light can limit the growth of algae adversely affecting the power output. Also, high algal density can limit the penetration of light at deeper levels. High light intensity can slow down the photosynthesis rate, a phenomenon called photoinhibition. Thus reactor, designing needs to be done strategically to provide a maximum surface area (Lee et al., 2015). They have further cited that light intensity has an indirect effect on resistance in the fuel cell as well. DO increases with an increased rate of photosynthesis, which resulted in an increase of cathodic resistance.

In some cases, the anode department is covered to prevent the growth of algae along the anode (Mekuto et al., 2020). This system had a higher voltage and power density compared to uncovered anodic chamber (Jaiswal et al., 2020). The performance and efficiency of MFCs were enhanced when LED was used as a source of light as compared to conventional light sources such as compact fluorescent light (Arun et al., 2020). When an artificial illumination device was used, lower power (6–12 W compared to 12–18 W) generated higher power output. It is further reported that cathodic resistance could be decreased, and power density can be increased by optimizing the duration of the light and dark cycle (Lee et al., 2015).

8.3.4 Carbon Dioxide Concentration

As shown in Equation 8.1, the anaerobic breakdown of organic substances by anoxic bacteria leads to the formation of CO_2. In a typical microbial fuel cell, this CO_2 gets released into the environment, which leaves a significant carbon footprint. But in a photosynthetic microbial fuel cell, this CO_2 can be directed to the cathode chamber, using an external pipe (Liu et al., 2015), where the algae can utilize it during photosynthesis and help create a zero-carbon footprint sustainable system. As a consequence, this in situ carbon sequestration by algae acts as a carbon sink and aids in cleaning up the environment (Arun et al., 2020). Sparging CO_2 consumes extra energy and bears additional costs (Liu et al., 2015). It was also found by a group of researchers that in a double-chambered PAMFC, some concentration of the CO_2 passes to the cathodic side through the Nafion membrane present in-between. The result of growing heterotrophic microorganisms alongside autotrophic algae is the 100% assimilation of CO_2 from the anode (Saba et al., 2017). Thus, CO_2 is also an essential influencing parameter in the generation of bioelectricity and any changes in its rate or concentration can affect oxygen generation, which will ultimately affect the power output. In the initial stages of the wastewater treatment and bioelectricity generation cycle, low algal biomass concentration can lead to the dissolution of CO_2 into the water to produce H_2CO_3 which can decrease the pH. To prevent this from occurring, the initial algal biomass inoculum concentration should be high (Saba et al., 2017). At higher CO_2 concentrations, greater biomass density is achieved and algae produce 6% more lipid content, leading to a better quantity of biodiesel (Mehrabadi et al., 2016).

8.3.5 Oxygen Concentration

The anodic chamber needs to maintain an anaerobic environment for the anoxic breakdown of the substrate, whereas the cathodic chamber needs to be oxygenated as oxygen acts as the electron acceptor, necessary for bioelectricity generation (Shukla & Kumar, 2018). Oxygen can be supplied either through mechanical aeration by sparging air or pure oxygen or with the help of algae at the cathode. By applying a catalyst at the cathode, oxygen reduction can be enhanced. Other methods include adding potassium ferricyanide, rotating the electrode, or using an air-cathode (Saba et al., 2017). Thus, one of the major drawbacks of single-chambered algal MFCs is that during the process, the back diffusion of oxygen from the cathodic to the anodic side occurs. This situation can be fatal for obligate anaerobes or can lead to change in metabolic pathways in facultative anaerobes or can change the mixed microbial consortium (Saba et al., 2017). For example, Arun et al. (2020) cited an observation where a 53.4% decrease in power density was observed when the concentration of dissolved oxygen was increased from 7.8 mg/L to 9.5 mg/L in the cathodic chamber. During the beginning of each cycle, the power density falls as the initial concentration of oxygen released by the algal biomass is low, leading to a shortage of electron acceptor. But as the algal biomass density increases, power output improves due to an increase in oxygen concentration (Zhang et al., 2019). However, thick biofilm formation on the reactor surface can limit the movement of oxygen (Saba et al., 2017).

8.3.6 Substrate Type

Microorganisms breakdown the organic substrates in an anoxic environment in the anodic chamber to release electrons, protons, and CO_2 and reduce the overall chemical oxygen demand (COD) (Saba et al., 2017). In the cathode compartment, algae can be grown either in BG11 medium or in a wastewater source containing a high concentration of nitrogen and phosphorus or in a combination of both. PAMFCs can use a variety of substrates ranging from pure to complex ones, including wastewater. The easily digestible substrates, such as glucose, starch, molasses, volatile fatty acids, etc., have been extensively used in MFCs, but they are quite expensive (Saba et al., 2017). Moreover, easily degradable substances cannot sustain long-term bioelectricity generation and hence long-release substrates should be explored that will help in self-sustained bioelectricity production (Qi et al., 2018). Besides supplying oxygen in the cathode, algae can also be used as substrate at anode in the form of either live cells or powdered form or as dead biomass or pretreated biomass or after extraction of lipids (Shukla & Kumar, 2018), which will reduce the cost sufficiently and create a closed-loop system (Mekuto et al., 2020). As this setup is being tested for wastewater remediation with simultaneous bioelectricity generation, various wastewater sources have also been used as substrates like swine wastewater (Zhang et al., 2019), kitchen wastewater (Naina Mohamed et al., 2020), anaerobic sludge (Ma et al., 2017), paper recycling wastewater (Radha & Kanmani, 2017), food-based wastewater (Saba et al., 2017), dye wastewater (Enamala et al., 2020), brewery wastewater (Harewood et al., 2017), landfill leachate (Hernández-Flores et al., 2017), fermentation effluents (Dai et al., 2021), dairy wastewater (Choudhury et al., 2021), oil refinery wastewater (Ng et al., 2021), pharmaceutical wastewater (Nayak & Ghosh, 2019) etc. Before being used as substrates, sometimes these wastewater sources might need to be pretreated with methods like ultrasonication, heat treatment, alkali treatment, and microwave treatment in order to break down the more complex substances present for easier hydrolysis by bacteria (Shukla & Kumar, 2018). Some researchers are also trying to explore the potential of *Arthrospira maxima* as a substrate (Qi et al., 2018). Substrate feeding rate or the growth-limiting substance can act as the rate-limiting step (Greenman et al., 2019) as these factors determine the morphology of biofilm and bacterial growth (Kusmayadi et al., 2020). It was also reported by Nam et al. (2010) that an increase in substrate loading rate increased electricity production. Sometimes a mixture of substrates or wastewater sources with carbon sources like glucose (Enamala et al., 2020; Li et al., 2019) have also been tested for better power output. Also, the microbial population's assembly and activity are determined by the type of substrate used (Mekuto et al., 2020).

8.3.7 Design Configuration

PAMFCs resemble MFCs in many ways and can be single-chambered or double-chambered. In single-chambered PAMFCs (Figure 8.6), both electrodes are present in the same compartment, with

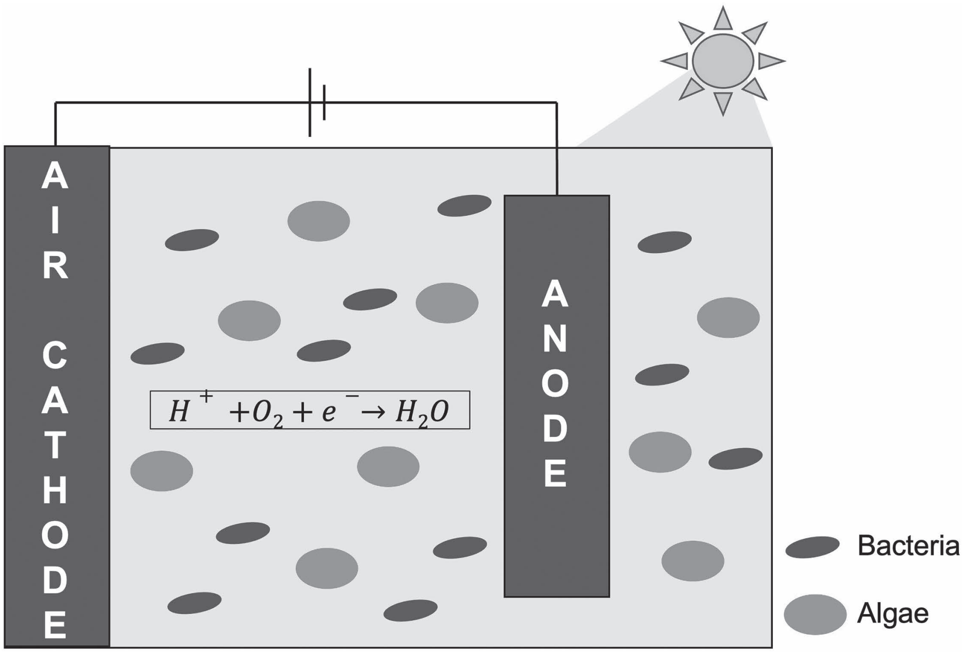

FIGURE 8.6 Diagrammatic representation of a typical single-chambered photosynthetic microbial fuel cell.

an air-cathode and no added oxygen supply unit. Algae and bacteria reside in the same chamber and grow synergistically (Saba et al., 2017). Thus, they can be easily scaled up due to simple design, low operational costs (no membrane required), and maintenance. But one major drawback is that pH changes occur simultaneously in both the cathode and anode regions and back diffusion of oxygen from cathodic to anodic region can affect power output. In a dual-chambered PAMFC, the electrodes are present in two separate chambers connected with an external circuit and separated with a membrane, which is most commonly a proton-exchange membrane (PEM) like Nafion. The light source is usually kept on the algal side and in some cases, the anode side maybe covered to prevent algal growth and avoid disturbance to the anoxic bacteria (Saba et al., 2017). Sometimes a photobioreactor might be connected to the cathode for easier maintenance and supply of algae. H-shaped reactors have also been designed, but narrow membrane area and low ion-exchange resulted in low power outputs. The major drawbacks of dual-chambered cells are membrane crossover, high internal resistance, and membrane fouling (Saba et al., 2017). The construction material of these fuel cells varies from plastic to acrylic to glass for easier penetration of light, promoting algal growth.

Electrode material plays an important role in the designing of a PMAFC. The material selection is based on cost, surface area, biocompatibility, anticorrosiveness, stability, and conductivity (Shukla & Kumar, 2018). It can be of two types: bio-electrodes, and chemical electrodes. Most common electrode materials are carbon cloth (Wang et al., 2018), stainless steel mesh (Bazdar et al., 2018), platinum, copper oxide (Liu et al., 2015), titanium (Kusmayadi et al., 2020), copper, aluminum, zinc, Ti-TiO_2 electrode (Taskan et al., 2014), aluminum–graphite (AL-C), iron–graphite (Fe-C), and copper–graphite (Cu-C) (Hou et al., 2016). But carbon electrodes find their usage in most large-scale applications due to their non-toxicity and low cost (Shukla & Kumar, 2018). They also help in forming stable bacterial biofilms (Saba et al., 2017). To prevent thick biofilm formation on the electrode, it is advised to make the electrode surface smooth rather than rough. In most studies, anode and cathode materials are the same, but in some studies, the electrodes are made of different materials to support biofilm formation in one and not the other. The distance between two electrodes needs to be spaced with careful consideration as it is crucial for power output. Placing the electrodes too close to each other results in decreased power density (Arun et al., 2020). Due to high tensile strength, ductility, and conductivity, copper wire is more frequently used than titanium.

The ion-exchange membrane can be of three types, namely, cation-exchange membrane (CEM), anion-exchange membrane (AEM), and bipolar membrane (BPM), based on the ionic groups attached. This, in turn, affects the passage of molecules from one chamber to another allowing protons, electrons, and CO_2 to pass through while restricting oxygen and substrate. The membrane should be featured to overcome internal resistance, pH splitting, and oxygen permeability to reduce the overall cost of MFC. Nafion is the most used in MFCs because it renders more specific conductivity for protons, increased columbic efficiency of MFC, and the increased microorganism lifetime. However, it has been reported that it does not function efficiently in alkaline conditions (Hernández-Flores et al., 2017). Clayware membrane, ceramic membrane, and natural rubber membrane are a few examples of low-cost membranes (Xu et al., 2015).

With the advancement in material science and technology, we look forward to new non-toxic buffers, cheaper membranes, and efficient electrode materials.

8.4 MOST COMMONLY USED ALGAL AND BACTERIAL CULTURE

In a typical microbial cell, anoxic electrochemically active bacteria (EAB) were initially used to generate electricity using an external circuit. To step up from the setbacks faced in the functioning of these cells, algae were used as biocathode, with or without immobilization, for CO_2 assimilation and in situ oxygen generation. Compared to other autotrophs, microalgae are fast-growing with tremendous photosynthetic efficiency and require lesser water than terrestrial plants (Angioni et al., 2018). Singular or mixed algal species can be utilized for power generation. The most frequently used algal species reported in the literature have been listed in Table 8.2. As discussed earlier, algae can also be used at the anode as a nutrient source for bacteria. It can be used in various forms like live algal biomass, dry powder form, pretreated or lipid-extracted, and in different concentrations. A pretreated powdered form of *Scenedesmus* was utilized by Liu et al. (2015) at the anode to obtain a maximum power density of 514.2 ± 19.4 mW/m^2. *Bacillus cereus*, *Pseudomonas aeruginosa*,

TABLE 8.2
Algal Species Used as Biocathode in a PAMFC and Their Corresponding Power Outputs

Algal Species	Power Density (W/m^3)	References
Chlorella vulgaris	126 mW/m^3	(Bazdar et al., 2018)
Scenedismus obliquus	153 mW/m^2	(Kakarla & Min, 2014)
S. platensis	10 mW/m^2	(Lin et al., 2013)
Fresh pond water	128 μW	(Gajda et al., 2015)
Ulva lactuca	0.98 W/m^2	(Arun et al., 2020)
Chlamydomonas reinhardtii and *Pseudokirchneriella subcapitata*	2.2	(Xiao et al., 2012)
Chlorella and *Phormidium*	26 W	(Juang et al., 2012)
Microcystis aeruginosa and *Chlorella vulgaris*	4.14	(Wang et al., 2012)
Microcystis aeruginosa	0.058	(Cai et al., 2013)
Desmodesmus sp. *AS*	0.82	(Wu et al., 2014)

Cyanobacteria, *Geobacter sulfurreducens*, *Shewanella oneidensis* MR-1, *Clostridia*, *Geobacter metallireducens*, *Shewanella putrefaciens*, *Aeromonas hydrophila*, *Rhodoferax ferrireducens*, *Enterococcus faecium*, *Pseudomonas aeruginosa*, and *Betaproteobacteria* are a few examples of commonly used bacterial consortium. Using genetic engineering, new strains can be engineered with better electrogenic properties for optimum output.

Protons travel through the membranes to reduce oxygen and the electrons are taken up by the anode which then travel through an external circuit to cathode without the help of any mediators. Electron transfer can be either a direct or an indirect mode of transfer, depending on the type of bacterial species. The most common form of extracellular electron transfer occurs with the help of bacterial cytochromes (Shukla & Kumar, 2018) or redox molecules. In some cases, bacteria have appendages like nanopili or nanowire that aid in electron transfer (Lesnik & Liu, 2014). This direct form of electron transfer can be seen in *Shewanella oneidensis* and *Geobacter sulfurreducens*. Another system of indirect electron transfer is cyclic diffusion, where bacterial cells (like *Escherichia coli*, *Bacillus* sp., and *Clostridia* sp.) donate electrons to anode and in turn take up soluble compounds from solution via polymeric redox mediators or primary metabolites like hydrogen (Patil et al., 2012). Addition of external mediators like potassium ferrocyanide, platinum catalysts, methyl blue, neutral red, thionine, methyl viologen, and humic acid have also been reported. Algal cells from suspension form a biofilm with time and directly take up the electrons from the circuit, which penetrate the algal body.

8.5 HARVESTING OF ALGAE

Efficient harvesting of algae from cathode or anode for further extraction of value-added products like biofuels, pigments, etc. can help in offsetting the additional costs endured during the functioning of the fuel cells. Recently, production of biochar from microalgae has been gaining interest for application as adsorbent, fertilizer, etc. (Pathy et al., 2022). Harvesting of algae comprises 30% of downstream costs (Shukla & Kumar, 2018). One of the methods to reduce harvesting costs is the immobilization of algae into beads. Immobilization of algae not only reduces costs but also adds benefits like high algal density, stable operation, and faster reaction speed (He et al., 2014). Sand filtration and coagulation have also been proposed for harvesting algae, but they have not been proved to be efficient in removing algae due to their small size and low density. Direct electrolysis could be a potential option for effective harvesting if not for the enormous energy requirement (Monasterio et al., 2017). Modification of this electrochemical system by incorporating flocculation, combined with either flotation or sedimentation, can make it energy-efficient and cost-effective (Shin et al., 2017; Behera et al., 2020). These methods can be broadly classified as electrolytic coagulation, electrolytic flotation, and electrolytic flocculation (Richardson et al., 2014; Krishnamoorthy et al., 2021).

In the electrolytic coagulation method, sacrificial electrodes are used to release positively charged ions which form aggregates with negatively charged algal cells due to charge neutralization. The coagulates settle down and can be separated with the age-old method of sedimentation (Richardson et al., 2014; Nageshwari et al., 2021). During this process, oxygen and hydrogen bubbles maybe released from the cathode, which can help the already formed flocs to rise to the surface. This process is called electro-floatation (Bardone et al., 2018). The addition of flocculants like chitosan or modified starch can lead to the formation of algal flocs, called flocculation (Behera et al., 2019). It can also be induced with the help of electrolysis, ultrasound waves, or magnetic waves (Pirwitz et al., 2015). To maximize efficiency, these methods are used in combination with one another as electrocoagulation–flocculation (Fayad et al., 2017) and electrocoagulation–flotation (Parmentier et al., 2020; Shi et al., 2017).

An innovative method was suggested by Monasterio et al. (2017), in which microbial fuel cell was integrated with electrochemical removal of algae. MFCs were operated on wastewater for the generation of electricity and simultaneously, an electrolysis unit connected in series with four to five MFCs was operated for electrochemical removal of algae.

8.6 APPLICATION OF THE INTEGRATED SYSTEMS

Microbial fuel cells were the first electrochemical devices to utilize live organisms for wastewater treatment and to produce bioelectricity. But it faced a few bottlenecks such as non-eco-friendliness due to release of CO_2 produced by the bacteria into the atmosphere, high operational cost due to supplying of CO_2, pH alteration due to its dissolution into the water, and aeration costs up to 50% because of oxygen. Thus, adding algae into the cathode not only solved the bottlenecks but also gave added products that could offset the additional costs. A comparative study between MFC and PAMFC has been presented in Figure 8.7. The following are the advantages of using algae at cathode as reported in the literature:

(i) Value-added pigments production (chlorophyll, ß-carotene, canthaxanthin, lutein, zeaxanthin) (Arun et al., 2020)
(ii) Production of algal biomass (Shukla & Kumar, 2018)
(iii) Use as industrial filters (Mekuto et al., 2020)
(iv) Has a lesser water footprint compared to other plant-based fuel industries (Ma et al., 2017)
(v) Pollution control as discharge of biomass into waterbodies can cause eutrophication (Liu et al., 2015)
(vi) Wastewater treatment (COD, nitrogen, and phosphorus removal) (Ma et al., 2017)
(vii) Carbon fixation in turn reducing global warming (Li et al., 2019)
(viii) Biodiesel production (lipid extraction), biogas production (anaerobic digestion of algae), hydrocarbon fuel (hydrothermal liquefaction) (Shukla & Kumar, 2018)

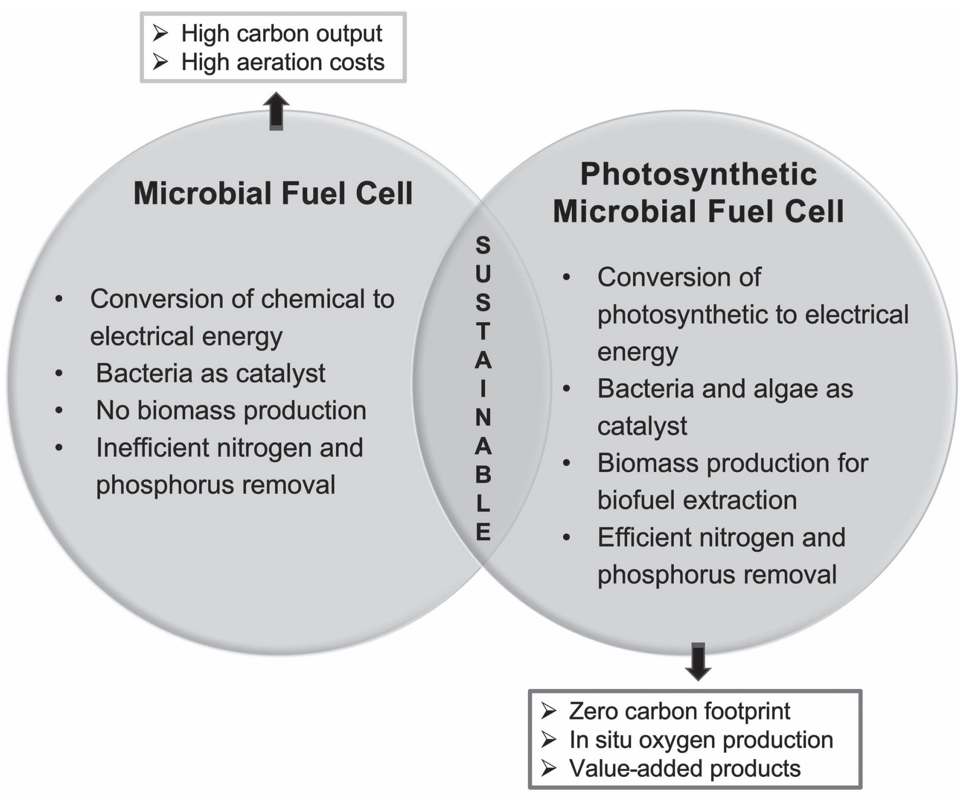

FIGURE 8.7 Comparison of characteristics between MFC and PAMFC.

(ix) Algal biomass can be also used as substrate in various forms:
  (a) Fresh algal biomass (Xu et al., 2015)
  (b) Algal powder (Shukla & Kumar, 2018)
  (c) Lipid extracted (Khandelwal et al., 2018)
  (d) Pretreated (sonication, autoclave, microwave, thermal, acid/alkali) (Arun et al., 2020)
(x) Algae grow in adverse ecological conditions as well as remove nutrients and toxic metals (Naina Mohamed et al., 2020)

8.7 RESEARCH GAP

1. Though articles have mentioned the direct use of algal biomass, some articles have also pointed out the need for pretreating the algal biomass as the cellulose present in the cell wall is not easy for the bacteria to hydrolyze (Xu et al., 2015). Requirement of pretreatments like thermal, sonication, alkaline, and acid treatment can increase the production costs. In addition, lipid-extracted algae were reported to contain harmful solvents and toxic chemicals, which can affect the growth of bacteria at the anode.
2. When the algal biomass density increases exponentially, they tend to form biofilms on reactor walls and the electrode surface. The thickness of cathodic biofilm influences oxygen diffusion, CO_2 transfer rate, and light penetration, which will ultimately affect electricity production. Studying biofilm formation in detail will help us develop efficient algal harvesting systems.
3. Buffer-less PAMFC systems need to be studied more in detail so that the pH can be regulated without the use of a buffer to limit the usage of chemicals and to prevent additional costs.
4. Algae being photosynthetic organisms require sunlight for biomass production and oxygen generation. Thus, the reactor setup should be such that it can receive the required amount of sunlight. Also, light penetration should not be affected even when the biomass becomes very dense at deeper levels of the reactor. In a year, the sunlight intensity varies according to season and to prevent it from affecting power generation, artificial illumination devices need to be set up to provide uninterrupted and consistent light throughout the year.
5. Membranes are expensive in nature. Thus, finding a cheaper alternative that will also prevent the unwanted diffusion of oxygen and substrate is required.
6. Stability, long-term performance, efficiency, and scaling up process from lab- to full-scale are the future challenges to be faced by many researchers.

8.8 SCOPE FOR FUTURE RESEARCH

The need for the development of robust technologies is critical because conventional systems consume a lot of energy. Reduction of CO_2 and greenhouse gases emission by these technologies is necessary. In addition, accomplishment of the permissible limits of nitrogen and phosphorus in the recycled wastewater needs to be achieved to prevent eutrophication and algal blooms. They also need to be low-cost to be installed in big to small cities, without the need of external supervision and excessive human resources. The development of an integrated system for the treatment of wastewater and bioelectricity generation using live organisms is still in its early phases and a lot of drawbacks need to be overcome before it can be utilized at its full potential at a commercial scale. Algal strains which can be fit for use at both electrodes need to be determined. The synergistic relationship between the bacteria and algae and the electron transfer mechanisms in addition to CO_2 uptake needs to be studied in detail for better control of growth and maximizing power output. In terms of bioelectricity generation, despite all the combinations of wastewater sources with bacterio-algal strains and electrodes being tried, the best combination is yet to be found for maximum power output. Various reactor configurations have also been tested over the years, but the optimum design

TABLE 8.3
Research Gaps and the Proposed Solution to Improve the Overall Efficiency and Commercialization of PAMFC

Research Gap	Proposed Solution
Use of buffers for pH regulation increases overall cost	Search for novel buffer-less systems
Low availability of reactor configurations in literature	Selection of optimum reactor design for maximum power output
Use of single algal species or in consortia	Selection of algal strains that can be used at both cathode and anode
Expensive membranes	Development of an efficient and cheaper membrane alternatives
Biofilm formation adversely affects power generation	Biofilm formation needs to be studied in detail to prevent occurrence
Lack of commercial scale plants	Optimization of parameters and standardization of influential factors to develop a sustainable technology

is yet to be determined. Ways to prevent thick biofilm formation needs to be discovered, so that power output does not get affected. Table 8.3 summarizes the research gaps along with their proposed solutions to develop an understanding of the need of this research area. Due to variations in wastewater or nutrient sources, microorganism strains, reactor environment, and operational setup, there have been discrepancies in data. This issue needs to be resolved for getting a consistent output and to develop field-based criteria. Also, the influencing parameters need to be optimized for keeping these factors in play.

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