Review

An Overview of Microbial Fuel Cells within Constructed Wetland for Simultaneous Nutrient Removal and Power Generation

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Abstract: Water, energy, and food are indispensable for sustainable economic development. Despite nutrients, especially phosphorus and nitrogen, being essential for plant growth and thus food supplies, those present in wastewater are considered an environmental burden. While microbial fuel cells (MFCs) are receiving much interest, combining wastewater treatment with an MFC has emerged as an option for low-cost wastewater treatment. Among others, a constructed wetland (CW) coupled with an MFC (CW-MFC) has the potential to provide a low carbon footprint and low-energy wastewater treatment, as well as nutrient and energy recovery from wastewater. Findings from this review show that the organic and nutrient removal and power generation by the integrated CW-MFC systems are affected by a number of factors including the organic loading rate, hydraulic retention time, system design, plant species, dissolved oxygen, substrate/media type, influent feeding mode, electrode materials and spacing, and external resistance. This review aims to summarize the current state of the CW-MFC and related technologies with particular emphasis on organic and nutrient removal, as well as on the bioenergy recovery from different wastewaters. Despite the benefits that these technologies can offer, the interactive mechanisms between the CW and MFC in the integrated system are still unclear. Further research is needed to fully understand the CW-MFC and related systems. The results of this work provide not only an overview and insight into existing knowledge but also the future direction of the CW-MFC technologies.

Keywords: microbial fuel cell; constructed wetland; hydroponics; wastewater treatment; electricity generation; nutrient removal; low carbon footprint; energy recovery; future direction

1. Introduction

Population growth, urbanization, and industrialization contribute to the rapid depletion of nonrenewable energy, deterioration of water resources [1], growing food demand, and increasing need for wastewater treatment. In past years, wastewater was seen as liquid waste that requires treatment before disposal into the environment. In recent years, however, wastewater has come to be viewed as an untapped steady source of fresh water, nutrients, and renewable energy [2–8]. Unlike other renewable energy sources such as wind and solar, the generation and availability of wastewater are consistent with the human/animal population and their activities and are thus quite predictable. A microbial fuel cell coupled with a constructed wetland is an appealing concept that has the potential to provide highly effective and sustainable resource recovery, bio-electricity generation, and wastewater treatment at the same time.

Wastewater occurs in large quantities, and its treatment with traditional technologies is energy intensive. In conventional municipal wastewater treatment plants (WWTPs), it is estimated that between 950 and 2850 kJ of energy is required to treat 1 m$^3$ of wastewater [9,10]. In addition, municipal wastewater alone accounts for approximately 5% of greenhouse gas, mainly methane, emissions [11]. In the United States, about 30.2 billion kWh of electricity...
is consumed annually in WWTPs, which accounts for 3 to 4% of the nation’s electricity demands [12]. Globally, an estimated 3% of the world’s electricity is consumed for wastewater treatment, and 50% of the wastewater treatment costs are used for sludge treatment and disposal [13,14]. On the other hand, the chemical energy contained in municipal wastewater is estimated to be at least 13 kJ/g-COD, which is approximately nine times more than the current energy demand for its treatment [15,16]. In recent years, technologies capable of recovering energy from wastewater with minimum energy input are receiving increased attention.

Nitrogen (N) and phosphorus (P) are pollutants present in wastewater discharges, which can cause eutrophication in receiving water bodies. Eutrophication is a worldwide environmental issue [17]. The conventional methods for treating large volumes of wastewater are not only costly but also wasteful of the abundant chemical energy and nutrients hidden in wastewater [18]. In chemical terms, 1 m$^3$ of domestic water contains approximately 300–600 g of COD, 40–60 g of N primarily in the form of ammonium and organic compounds, 5–20 g of P mainly in the forms of phosphate and organic compounds, 10–20 g of sulfur (S) in the form of sulfate, and trace amounts of heavy metals [11]. Although P and N can be removed and recovered from wastewater [6,19], conventional wastewater treatment plants (WWTPs) employ costly, energy-intensive processes such as chemical precipitation for the P recovery [19] and biological nitrification-denitrification or Anammox processes to transform various N forms to N$_2$ [20].

Wastewaters from food processing and agricultural activities, in particular, contain high levels of nutrients [21] that are the key constituents of fertilizers [14] and can be exploited as resources. There is an increasing trend of research on the development of technologies to recover nutrients from wastewater in a sustainable manner [6,22]. Such technologies can alleviate future fertilizer demands and contribute to a healthier environment [23–25].

Initial research on microbial fuel cells (MFCs) was mainly targeted toward electricity generation promoting bioelectrochemical systems [26,27]. Later, MFCs emerged as alternative low carbon footprint wastewater-treatment technologies to convert chemical energy contained in wastewater into electrical energy [28–31]. MFCs possess the ability to generate electricity using wastewater as an energy source while simultaneously treating the wastewater with little or no external energy input. In recent years, the application of these technologies has been expanded to nutrient recovery with added benefits such as reduced sludge generation and energy recovery and conservation [6,32].

The basic components of an MFC are an anode electrode that is situated in the anaerobic chamber and a cathode electrode that is either in the cathode chamber or under an aerobic environment. In the anode chamber, electrochemical active bacteria (EAB) convert the chemical energy in wastewater to electrical energy directly through microbial electron transport systems [33–37] with only a small loss of energy, compared to other wastewater treatments such as electricity generation via methane production in an anaerobic digestion process [32]. To prevent short-circuiting, the anode and cathode chambers are separated by an ion-exchange membrane or proton exchange membrane (PEM) in a dual-chamber MFC. The membrane separator allows protons to migrate to the cathode while containing anolyte (substrate) within the anode chamber [38]. The PEM also serves as a barrier to maintaining EAB in the anode chamber [39]. Since the membrane separator is generally expensive and requires maintenance [38], membrane-less single chamber MFCs have also attracted attention.

This review paper aimed to summarize the latest studies for wastewater treatment, nutrient removal, and energy production by the CW-MFC and related systems, considering various factors, including the organic loading rate (OLR), hydraulic retention time (HRT), dissolved oxygen (DO), plant species, media type, system design, and other relevant parameters. To the best of the author’s knowledge, there are no recent review articles mainly emphasizing simultaneous nutrient (P and N) removal and power generation by the CW-MFC. This review paper will benefit researchers exploring the recent advancement.
in nutrient removal and energy recovery (power generation) from various wastewaters by
the CW-MFC systems.

2. Constructed Wetland-Microbial Fuel Cell (CW-MFC) Systems

2.1. CW

A constructed wetland (CW) is an engineered ecological wastewater treatment sys-
ystem that mimics natural water purification processes [40,41]. In CWs, plants, substrate,
soil, and microorganisms play important roles in providing symbiotic physical, chemical,
and biological functions, including filtration, ion exchange, physicochemical adsorption,
chemical precipitation and decomposition, bioabsorption, and microbial reactions such
as ammonification, nitrification, denitrification, and biodegradation [42]. Through the
processes, various organic and inorganic contaminants are removed [43]. Wetland plants
have been shown to increase the density of bacteria by 10 times [44], favoring the growth
of exoelectrogens to improve nutrient removal [45]. The CWs generally use plants with
limited commercial importance (contrary to hydroponics) for the removal of pollutants in
wastewater [46]. The CWs possess the following advantages: (i) low-cost, simple mainte-
nance and operation, (ii) low energy consumption, (iii) eco-friendly, (iv) excellent landscape
integration [17,41,47–49], and (v) suitability to different climatic conditions [50,51]. The
CWs have been employed as a cost-effective viable option for treating low-strength wastew-
aters, and offer reliable, sustainable, and green treatment for developed areas, as well
as economically underserved communities [45]. The performance of the CWs that treat
the high-strength wastewater is prone to be affected by faster substrate clogging [52,53].
As high-strength wastewater poses a high oxygen demand, intermittent aeration can be
an appropriate strategy to achieve satisfactory performance in the removal of organic
pollutants and nitrogen [42]. Additionally, a longer hydraulic retention time is required for
efficient treatment of the high-strength wastewater [54].

2.2. CW-MFC

The CW coupled with an MFC is a relatively new technology for concurrent wastewa-
ter treatment and electricity generation [55]. A typical CW-MFC system has anodic and
cathodic regions, which are separated by the media (e.g., soil, sand, and gravel), fibrous
materials, or proton exchange membranes [45] (Figure 1). The CWs may have one or more
types of media to allow wastewater to flow through and microorganisms to grow on their
surface. The anaerobic and aerobic transformations of chemical substances take place in
the anaerobic and aerobic regions, respectively [37,56]. In general, the anaerobic region is
formed near the bottom of the CW, while the aerobic region is naturally produced adjacent
to the air–water interface. These regions are characterized by a redox gradient.

![Figure 1. Representation of the major components of the CW-MFC system.](image-url)
In the CW-MFC system, the treatment of wastewater and the generation of electricity are simultaneously accomplished by the complex activities of the plants, rhizodeposition, and microorganisms [41]. The vegetation has been shown to improve the bacterial activity to decompose organics [56,57]. The microorganisms in the rhizosphere have been known to break down rhizodeposition products, as well as chemical compounds in wastewater, and thus they play a crucial role in water purification processes [56,57]. The plant stroma furnishes a large specific surface area that can enhance the adsorption of electron-transfer mediators, providing advantages to the CW-MFC systems [45,58].

Both water purification and electricity generation rely on the microbial oxidation of organic and inorganic matter in wastewater [37]. The CW-MFC can significantly improve its performance, compared to the standalone CW or an MFC [40,57,59–63]. Compared to the standalone CW system, the CW-MFC system increased the wastewater treatment efficiency (in terms of COD removal) by 27–49% [64] and yielded a 22% higher NH$_4^+$-N-removal efficiency [65]. The CW-MFC can be an innovative technology that has the potential to become an economical, self-sustaining, and eco-friendly method to accomplish both wastewater treatment and electricity generation at the same time [49,57,61,63].

2.3. Hydroponics

A hydroponic (Hyp) system was included in this literature review, as it is similar to the CW. In a Hyp system, edible and/or ornamental plants can be grown at any time of the year in a protected and soilless environment. Since the Hyp systems are soilless systems, they can be applied in densely populated urban areas where available land is limited or in arid regions for wastewater reuse in agricultural practices. The use of the Hyp system as the tertiary wastewater treatment for nutrient removal and recovery has been explored [66].

In the Hyp system, nutrients contained in wastewater and carbon dioxide released from wastewater can be captured and stored by the plants as biomass, while oxygen is produced via photosynthesis [67]. Consequently, the Hyp system reduces the risk of environmental hazards. The mechanisms of the Hyp system for the removal of pollutants are similar to those of the CW system, involving the combination of the physical, chemical, and biological processes with microorganisms, plants, and media-based interactive reactions [67,68]. The hydroponic plants can improve bacterial activity by releasing rhizodeposition and oxygen into water [48]. The plants are responsible for most of the nutrient removal [69].

The suggested economic benefits of using the Hyp system for wastewater treatment [70,71] are similar to those of the CW systems: i.e., (i) pollutants can be removed, (ii) nutrients (P and N) can be removed and used simultaneously, (iii) maintenance and energy costs can be reduced [67], and (iv) the system can be implemented onsite as less area is required compared to the conventional wastewater treatment [72]. The use of the Hyp technology can result in environmental and economic gain contributing to food supplies and economic opportunities [67].

2.4. Hydroponics-MFC (Hyp-MFC)

The Hyp-MFC systems are similar to the CW-MFC systems. The major difference, however, is that the Hyp-MFC system is a soilless system in which plants can directly grow in water using a floating device with or without media. Only a few studies have been dedicated to the Hyp-MFC system [60,73]. The factors affecting the nutrient removal and electricity generation are similar to those of the CW-MFC: i.e., the type of the systems, plants, and substrates; influent feeding modes; external resistance; electrode materials; and electrode spacing; in addition to the basic operational parameters such as the HRT, OLR, DO, and temperature (Tables 1 and 2). The major advantage of the Hyp-MFC system is that the substrate clogging can be prevented and the contact between roots and dissolved nutrients can be enhanced for nutrient removal. Figure 2 shows a representation of the Hyp-MFC with the major components, in which nonedible plants are grown as the plant roots are in direct contact with wastewater. To grow edible plants, its design needs to be
more innovative to protect public health. Studies have shown that the electrodes facilitate microbial respiration and enhance microbial growth kinetics to break down organics in wastewater [55,63,74]. The Hyp-MFC system can be a potential candidate for simultaneous energy and nutrient recovery and wastewater treatment.

![ Representation of the Hyp-MFC system for nonedible plants with major components.](image)

**Figure 2.** Representation of the Hyp-MFC system for nonedible plants with major components.

3. Energy Generation in the Integrated Hyp-MFC and CW-MFC Systems

3.1. Effect of Organic Loading Rate (OLR) on Electricity Generation

The efficiencies of wastewater treatment and power generation by the CW-MFC are highly dependent on the OLR [63,74], whereas the generation of voltage is related to the concentration of the carbon source and the efficiency of substrate utilization by the microorganisms thriving in the system [14]. Using the CW-MFC, Villaseñor et al. (2013) [74] studied the effects of the OLR (with 13.9 g COD/m²/d) on the COD-removal efficiency. With the low OLR (13.9 g COD/m²/d), the system removed COD up to 100% in the anode region (in the anaerobic environment) and produced the optimal current density of 1.22 mA/m² (normalized to the electrode area) and the maximum power density of roughly 0.15 mW/m². With the higher OLR (61.1 g COD/m²/d), the COD-removal efficiency fell by 80–85%. Their study showed that the CW-MFC system was more efficient in power generation with lower ORLs (around 13.9 g COD/m²/d) [74]. Using the closed-circuit horizontal subsurface CW-MFC (Figure 3), Srivastava et al. (2020) [63] found that their system yielded COD removal efficiencies of ~98% and ~99% with the volumetric OLR of 0.15 and 0.30 kg COD/m³/d, respectively. However, the efficiency dropped to ~33.7% with the higher volumetric OLR (0.52 kg COD/m³/d). The maximum current and power densities were 17.15 mA/m³ and 11.67 mW/m³, respectively.
without a ceramic separator (Figure 4). In their work, synthetic wastewater was fed to the up-flow CW-MFC system with supplemental aeration (Figure 5). Varying the electrogens [40,76]. Without the barrier between the cathode and anode regions, excessive growth of electrogens in the anodic zone [60]. An insufficient DO level in the aerobic zone (cathode region) is explained by the fact that the oxygen diffusion into the anodic region was blocked by the separator, creating a higher anaerobicity (lower redox potential) and favoring the growth of electrogens in the anodic zone [60]. An insufficient DO level in the aerobic zone (cathode region) can result in low power generation [75]. On the other hand, the high DO level in the cathode region enhanced electricity generation and the degradation of organics by electrogens [40,76]. Without the barrier between the cathode and anode regions, excessive DO in the cathode region could disturb the anaerobic environment in the anode region, impeding the metabolic activities of electrogens to reduce the power output [55].

Figure 3. Representation of the closed-circuit horizontal subsurface constructed wetland-MFC (adapted from Srivastava et al., (2020) [63]).

3.2. Effects of Dissolved Oxygen on Electricity Generation

Khuman et al. (2020) [60] investigated the performance of two up-flow hydroponic constructed wetland-microbial fuel cells, namely, hydroponic CW-MFC systems with and without a ceramic separator (Figure 4). In their work, synthetic wastewater was fed to the bottom of the system (anodic zone) and released from the top of the system (cathodic zone) at a rate of 6 L/day. In the system with the separator (Figure 4a), the wastewater stagnated in the anodic zone for 12.8 h before entering the cathodic zone. The COD removal efficiencies with and without the ceramic separator were 86.2 ± 8.1% and 91.5 ± 4.9%, respectively. The volumetric power densities produced by the system with and without the separator were 258.78 mW/m³ and 91.02 mW/m³, respectively. The larger COD removal and smaller power output by the system without the separator indicate that there was an intrusion of oxygen (O₂) into the anodic zone where O₂ was directly consumed by bacteria for the decomposition of organics, instead of accepting electrons at the cathode to generate electric current [34,74]. The larger power output by the system with the separator is explained by the fact that the oxygen diffusion into the anodic region was blocked by the separator, creating a higher anaerobicity (lower redox potential) and favoring the growth of electrogens in the anodic zone [60]. An insufficient DO level in the aerobic zone (cathode region) can result in low power generation [75]. On the other hand, the high DO level in the cathode region enhanced electricity generation and the degradation of organics by electrogens [40,76]. Without the barrier between the cathode and anode regions, excessive DO in the cathode region could disturb the anaerobic environment in the anode region, impeding the metabolic activities of electrogens to reduce the power output [55].

Figure 4. Hydroponic CW-MFC system (a) with ceramic separator and (b) without a ceramic separator (adapted from Khuman et al., (2020) [60]).

3.3. Effects of Vegetation on Electricity Generation

Oon et al., (2017) [55] investigated the effect of the wetland plant, Elodea nuttallii, using the up-flow CW-MFC system with supplemental aeration (Figure 5). Varying the
air-flow rate from 0 to 1900 mL/min, they found that the air-flow rate of 600 mL/min was the optimal flow rate, which yielded the highest Coulombic efficiency of 10.28%. The maximum power density at the optimal air-flow rate was 184.75 ± 7.50 mW/m² of the anode chamber [55]. Yan et al. (2018) [62] investigated two different systems, namely CW-ACMFC and CW-MFC, with *Cannas indica*. The difference between the two systems is that the cathode was placed at the top of the influent level in the CW-ACMFC, while the cathode was submerged in the CW-MFC (Figure 6). Both systems were fed with synthetic wastewater in a fed-batch mode. The voltage produced by the CW-ACMFC and CW-MFC ranged from 0.36 to 0.52 V and from −0.04 to 0.07 V, respectively. The maximum power density produced by the CW-ACMFC (4.21 mW/m² of cathode surface) was considerably larger than that produced by the CW-MFC (0.005 mW/m² of cathode surface) [62]. In the CW-ACMFC system, oxygen released by the plants could have influenced electricity generation as oxygen is an electron acceptor at the cathode. Furthermore, the plants probably played a significant role in power generation as the photosynthetic activity of the plants produces rhizodeposits (e.g., carbohydrates), which are consumed by bacteria and converted to electricity in the system [41,48].

![Figure 5](image1.png)

**Figure 5.** Representation of the up-flow CW-MFC system with supplemental aeration (adapted from Oon et al., 2017 [55]).

![Figure 6](image2.png)

**Figure 6.** Representation of the (a) CW-ACMFC and (b) CW-MFC (adapted from Yan et al., 2018 [62]).
Three different indigenous wetland plants (Cyperus prolifer, Wachendorfia thyrsiflora, and Phragmites australis) were investigated by Oodally et al., (2019) [48]. In their study, the CW-MFC with C. prolifer produced the highest power density (229 ± 52 mW/m³) and voltage (510 mV). The maximum power density was 24% higher with C. prolifer than that of the control (unplanted), whereas the maximum power densities with W. thyrsiflora and P. australis were 42% and 41% less than the control, respectively. Oodally et al., (2019) [48] suspected that W. thyrsiflora and P. australis were not acclimated to the initial environment that had a high concentration of ammonia in the wastewater. Noticing that C. prolifer developed its roots more extensively than the other two species, it can be presumed that the larger root-specific area and biomass are advantageous for power production. The expeditious acclimatization of the plants is an important factor in generating stable voltage [48].

Using the floating treatment wetland-MFC (FTW-MFC), which is similar to the hydroponic system, Colares et al., (2021) [77] treated urban wastewater with five different ornamental plants: (i) Canna generalis, (ii) Chrysopogon zizanioides, (iii) Cyperus papyrus Nanus, (iv) Hymenachne grumosa, and (v) Equisetum hyemale. As is seen in Figure 7, the cathodes were placed below the roots of each plant and the anode was placed on the bottom of the CW. The cathodes and anode were situated in parallel. The water surface was covered with a carpet foam and insect screen. In their setting, the maximum power density of 0.93 mW/m² was produced at the maximum voltage of 108 mV. The larger voltage was produced by the system with the plants that had developed a larger root system than those with a smaller root system. This may be due to the fact that larger amounts of oxygen were released and exudates were excreted from the larger root system. The larger root system provides a larger surface area for the bacteria to grow and forms a biofilm to enhance the generation of electricity [77]. The release of oxygen from the plant’s roots in the rhizosphere (known as radial oxygen loss) and root exudates played significant roles in enhancing nitrogen removal and electricity generation [78].

![Image of the floating treatment wetland-MFC (FTW-MFC)](image_url)

**Figure 7.** Representation of the floating treatment wetland-MFC (FTW-MFC) (adapted from Colares et al., (2021) [77]).

Using an ecological floating bed-MFC (EFB-MFC) (Figure 8), Yang et al., (2021) [78] studied four plants: (i) windmill grass, Cyperus alternifolius Linn. Sbsp. Flabelliformis (Rottb.) Kukenth); (ii) goldfish algae, Ceratophyllum demersum Linn.; (iii) water hyacinth, Eichhornia crassipes (Mart.) Solms Pontederia crassipes Mart.; and (iv) water spinach, Ipomoea aquatic Forsk. In their study, these plants reduced the internal resistance of the system by 21.23–67.66% and increased the voltage by 26.26–62.63%, compared to the control system (with no plants) [78].
Figure 8. Representation of the floating bed-MFC (EFB-MFC) (adapted from Yang et al., 2021 [78]).

3.4. Effects of Electrode Spacing on Electricity Generation

According to Doherty et al., (2015) [79], the availability of oxygen and the distance between the anode and the cathode are two critical factors in maintaining high levels of electricity production in the CW-MFC. Oon et al., (2015) [40] studied the effect of the electrode spacing on the output voltage and power density using the up-flow CW-MFC system with three anodes (Figure 5). Each anode was separated from the cathode at a distance of 15 cm, 30 cm, and 45 cm. In their study, the largest voltage (421.7 mV) and the largest maximum power density (6.12 mW.m⁻²) were produced at the electrode spacing of 15 cm (the smallest distance tested in their study), while it produced ~300 mV and 3.39 mW/m² at 30 cm, and ~250 mV and 3.37 mW/m² at 45 cm. The internal resistance of the system increased with the increasing distance between the anode and cathode; i.e., 820 Ω at 15 cm, 1000 Ω at 30 cm, and 4300 Ω at 45 cm. The variation of the power density was mainly due to the performance of the anode rather than that of the cathode [40]. In a study by Yu et al., (2020) [80], ammonium was found to be the main substrate (for the NH₄⁺ oxidation reaction) that affected electricity generation. Yu et al., (2020) [80] examined the effects of the electrode spacing (at 15 cm and 25 cm) and operating temperature (at 15.9–26.2 °C, 12.8–16.8 °C, and 6.0–10.5 °C) on the electricity generation and NH₄⁺ removal. They observed voltage reversal with the electrode spacing of 25 cm when the temperature dropped below 10 °C. The voltage reversal occurred as the microorganisms’ activity decreased and more electrons were captured by the electrode rather than being used by other microorganisms near the anode [80]. Even though the bacterial activity dropped, the voltage output was increased.

3.5. Effects of the Media Type and Hydraulic Retention Time on Electricity Generation

The media type is another factor that affects electricity generation in the CW-MFC system. Three different media types, namely, sand, zeolite, and volcanic cinder, were investigated by Yakar et al. (2018) [61] using three parallel up-flow CW-MFC systems (UCW-MFC) (Figure 9). At the electrode spacing of 15 cm, they observed voltage ranging from 0.45 to 0.99 V with sand, 0.65 to 1.24 V with zeolite, and 0.5 to 1.19 V with volcanic cinder. The maximum power and current densities were, respectively, 5.09 mW/m² and 7.11 mA/m² for sand, 26.12 mW/m², and 16.1 mA/m² for zeolite, and 8.42 mW/m² and 6.47 mA/m² for volcanic cinder. The larger voltage, power density, and current density produced by zeolite indicate that zeolite can provide an environment more suitable for the growth of electrogenic bacteria, compared to sand and volcanic cinder [61].
Yadav et al., (2020) [73] designed a drip Hyp-MFC system with multiple small reactors that were connected in series and parallel (Figure 10). Their system used graphite for the anode and cathode electrodes, and cocopeat as the medium to support the growth of lemon-grass, *Cymbopogon citratus*. Wetland plants and electrodes were placed in plastic containers filled with cocopeat. In treating domestic wastewater, they found that the maximum power densities and corresponding current densities were, respectively: (i) 31.88 mW/m$^2$ and 35.63 mA/m$^2$ at the external resistance of 20 kΩ in the reactors connected in series; and (ii) 31.58 mW/m$^2$ and 457.8 mA/m$^2$ at the external resistance of 0.120 kΩ in the reactors in a parallel mode [73].

Using the vertical-flow CW-MFC system with up-flow and down-flow chambers (Figure 11), Zhong et al. (2020) [17] examined three different substrates, namely, ceramsite, quartz, and zeolite. Feeding synthetic sewage at three different hydraulic retention times, HRTs (7.6 days, 4 days, and 2.8 days), they found that the average voltage values were the highest with ceramsite in both the up-flow and down-flow chambers at the HRT of...
7.6 days. The average power densities in the up-flow chamber were 120.3 mW/m³ with ceramsite, 11.3 mW/m³ with quartz, and 14.2 mW/m³ with zeolite. When the HRT was decreased to 4 and 2.8 days, the voltage declined to below 0.02 V. At the low HRTs, the influent wastewater with low DO could have flown directly to the cathode region to create the DO deficiency, resulting in low power outputs [17]. On the other hand, the high HRT increased the contact time between the wastewater, microbes, and substrate to give a longer reaction time for the adsorption and degradation of organics [42]. Wang et al., (2019) [76] warned, however, that a large HRT can aggravate the anaerobic conditions in the CWs; thus, the CW-MFC should be operated at an optimal HRT to maintain a high treatment efficiency and bioelectricity generation.

Figure 11. Integrated vertical-flow CW-MFC system with up-flow (right) and down-flow (left) chambers (adapted from Zhong et al., (2020) [17]).

3.6. Effects of System Design on Electricity Generation

Obviously, the design of CW-MFC systems plays an important role in wastewater treatment and electricity generation. Corbella et al., (2015) [57] installed an MFC within the horizontal subsurface CW (Figure 12) to treat real domestic wastewater. Their design consisted of two different systems, i.e., one fed the primary effluent of the conventional settling process (settler line) and the other fed the effluent of the hydrolytic up-flow sludge blanket reactor (HUSB line). The HUSB is a type of anaerobic digester in which, however, methane formation was depressed during hydrolysis of organic matter, providing low HRTs [81]. The HUSB supplies more concentrated fuel (organics) to the MFC, as the effluent of the HUSB contains higher levels of biodegradable organics than that of the conventional settling process [37]. In the work of Corbella et al., (2015) [57], the maximum current density of 219 mA/m² and maximum power density of 36 mW/m² were achieved in the HUSB line. Seasonal variations in the water level affected the cell voltage. When the cathode was exposed to the atmosphere, the power output increased owing to the increased availability of O₂.
Liu et al., (2019a) [43] designed a vertical-flow CW-MFC (Figure 13) to treat slightly polluted source water. The water contained, on average, 15.5 mg/L COD, 0.17 mg/L total phosphorus (TP), 3.3 mg/L total nitrogen (TN), 0.85 mg/L NH$_4^+$-N, 1.6 mg/L NO$_3^-$-N, and 1.11 mg/L NO$_2^-$-N. This system produced the maximum voltage and power density of 777 mV and 8.05 mW/m$^2$, respectively, at an external resistance of 6000 Ω. Denitrification was frequently disturbed. In another study by Liu et al., (2019b) [58], a vertical-flow CW-MFC (Figure 13) was fed swine wastewater after it was treated anaerobically in a biogas tank and an up-flow anaerobic sludge blanket process. An average COD concentration in the influent was 505 mg/L with a range between 324 mg/L and 708 mg/L. To provide a large surface area for the growth of electroactive microbes, a layer of activated carbon was placed at the bottom of the system. This system produced voltage in the range from 598 mV to 713 mV and the largest power densities of 456 mW/m$^2$. The ohmic resistance was mainly rooted in the conductivity of electrolytes, electrodes, and surface biofilms [58]. This process produced a Coulombic efficiency of only 0.386%, which may be due to the fact that a large number of microorganisms from the swine wastewater competed with the electrogenic bacteria for organic food in the reactor, resulting in, at least partially, anaerobic digestion reactions [58].

**Figure 12.** MFC within a horizontal subsurface CW (adapted from Corbella et al., (2015) [57]).

**Figure 13.** Representation of the vertical-flow CW-MFC (adapted from Liu et al., (2019a) [43]).

Ren et al., (2021) [37] treated swine wastewater using the alum sludge-based hybrid CW-MFC with a vertical up-flow system in the first stage and a horizontal subsur-
face flow system in the second stage (Figure 14). Using dewatered alum sludge as the main substrate, their system produced voltage and power density of $0.44 \pm 0.09$ V and $33.3 \pm 13.81$ mW/m$^3$, respectively, in the first stage, and $0.34 \pm 0.09$ V and $9 \pm 2.5$ mW/m$^3$, respectively, in the second stage. Xu et al., (2017) [82] built a stacked CW-MFC (Figure 15), in which the anode electrode was made of stainless-steel mesh sandwiched in graphite gravel, and the cathode electrode was made of stainless-steel mesh with a thin layer of particle-activated carbon. Their system, fed with synthetic wastewater, showed changes in the COD-removal efficiency from 83.2% in the upper part of the system to 88.7% in the outer part of the system. During a 3-month run period, the power density decreased from ~2 mW/m$^2$ (normalized to the cross-section area of the reactor) on the second day of the operation to ~0.3 mW/m$^2$ after 60 days. This downturn was likely due to the deterioration of the air cathode. The cathode electrode (containing particle-activated carbon) could have been contaminated with pollutants in the wastewater, resulting in reduced catalytic functions. The anode performance, however, showed minor changes over the operation period [82]. Gupta et al. (2021) [59] developed the algal-assisted CW-MFC integrated with a sand filter (Figure 16) for the treatment of various pollutants in wastewater and electricity generation. Their system produced the maximum power and current densities of $33.14$ mW/m$^3$ (of the working volume of the anodic region) and $235$ mA/m$^3$, respectively.

Figure 14. Representation of the alum-sludge-based hybrid CW-MFC with a vertical up-flow system in the first stage and a horizontal subsurface flow system in the second stage (adapted from Ren et al., (2021) [37]).

Figure 15. Representation of the stacked CW-MFC (adapted from Xu et al., (2017) [82]).
Figure 16. Representation of the algal-assisted CW-MFC integrated with a sand filter (adapted from Gupta et al., (2015) [59]).
| Wastewater Type | System Type        | Electrodes                                                                                       | Plant Type                  | Coulombic Efficiency | Voltage         | Power Density        | Current Density | Reference |
|-----------------|--------------------|--------------------------------------------------------------------------------------------------|-----------------------------|----------------------|-----------------|----------------------|-----------------|-----------|
| Domestic wastewater | Horizontal subsurface flow CW-MFC               | Anode & Cathode: cylindrical graphite rods wrapped with a stainless-steel mesh.                | Phragmites australis       | NA                  | HUSB line: 164 mV | HUSB line: 36 mW.m⁻² | HUSB line: 219 mA.m⁻² | [57]    |
| Synthetic wastewater | Up-flow CW-MFC                   | Anode & Cathode: carbon felt. Electrode spacing between cathode and anodes (A): A1: 15 cm; A2: 30 cm; A3: 45 cm.  | Typha latifolia           | A1: 8.86%           | A1: 421.7 mV     | A1: 6.12 mW.m⁻²    | A1: ~18 mA.m⁻²             | [40]    |
| Synthetic wastewater | Stacked CW-MFC                  | Anode: stainless-steel mesh. Cathode: stainless-steel mesh net with a thin layer of particle-activated carbon. | NA                          | At 2 days: 1.85%    | At 2 days: ~550 mV | At 2 days: ~2 W.m⁻³  | At 2 days: ~5 A.m⁻³            | [62]    |
| Synthetic wastewater | Up-flow CW-MFC                  | Anode & Cathode: activated carbon. Electrode spacing between cathode and anodes (A): A1: 8 cm; A2: 23 cm; A3: 38 cm.  | Elodea nuttallii          | 0.08-10.28%         | A1: 546 ± 25 mV   | 184.75 ± 7.50 mW.m⁻³ | NA              | [55]    |
| Synthetic wastewater | CW-ACMFC & CW-MFC             | Anode: graphite felt. Cathode: graphite felt.                                                  | Canna indica               | NA                  | CW-ACMFC: 0.36-0.52 V | CW-ACMFC: 4.21 mW.m⁻² | CW-ACMFC: 20.30 mA.m⁻²    | [62]    |
| Synthetic wastewater | Up-flow CW-MFC (three parallel systems) | Anode: graphite. Cathode: magnesium cathode. Electrode spacing between cathode and anodes (A): A1: 35 cm; A2: 15 cm.  | Typha latifolia L.        | Zeolite: 1.64%      | Sand: 0.44-0.93 V | Sand: 5.09 mW.m⁻²    | Sand: 7.11 mA.m⁻²             | [61]    |
| Synthetic wastewater | CW-MFC                          | Anode: graphite-based anode. Cathode: horizontal rectangular magnesium.                        | Typha latifolia (M₁)      | M₁: 6.074 ± 8.42%  | M₁: 0.7-1.24 V    | M₁: 13.4 mW.m⁻²    | M₁: 31.4 mW.m⁻²            | [41]    |
| Synthetic wastewater | Integrated vertical flow CW-MFC | Anode & cathode: graphite panels.                                                             | Canna sp. and Aceras calamus | NA                  | 777 mV          | 8.05 mW.m⁻²        | NA              | [43]    |
| Swine wastewater | Integrated vertical flow CW-MFC | Anode: stainless-steel wire mesh that wraps activated carbon particles. Cathode: carbon felt.  | Canna indica              | 0.386%              | 598-713 mV       | 0.456 W.m⁻³        | 22.5 mAm⁻²          | [58]    |
| Wastewater Type       | System Type                      | Electrodes                                      | Plant Type                                      | Coulombic Efficiency | Voltage | Power Density | Current Density | Reference |
|-----------------------|----------------------------------|-------------------------------------------------|-------------------------------------------------|----------------------|---------|---------------|----------------|-----------|
| Settled sewage        | CW-MFC                           | Anode: an inoculated layer of granular activated carbon. Cathode: platinum-coated carbon. | *Cyperus plicifer* (*N*1); *Wachendorfia thyrsiflora* (*N*2); *Phragmites australis* (*N*3) | NA                   | *N*1: 510 mV | *N*1: 229 ± 52 mWm~−3 | NA             | [48]     |
| Synthetic wastewater  | Integrated CW into a horizontal subsurface CW (HSSF-CW-MFC) | Anode & Cathode: graphite gravel. | *Canna indica* | OLR of 0.15 kg COD/m²/d: 1.86% OLR of 0.30 kg COD/m²/d: 0.6% Closed circuit, OLR of 0.52 kg COD/m²/d: 1.05% | OLR of 0.15 kg COD/m²/d: 1.86% OLR of 0.30 kg COD/m²/d: 0.3% Closed circuit, OLR of 0.52 kg COD/m²/d: 1.05% | 11.67 mW/m³ | 17.15 mA/m³ | [63]   |
| Synthetic wastewater  | Two Upflow hydroponic CW-MFC     | Anode & cathode: carbon felts. | *Canna indica* | NA | With ceramic separator: ~400 mV Without ceramic separator: ~800 mV | With ceramic separator: 258.78 mWm~−3 Without ceramic separator: 91.02 mWm~−3 | With ceramic separator: ~560 mA.m~−2 Without ceramic separator: ~190 mA.m~−2 | [60]     |
| Domestic sewage       | Integrated drip hydropnics-MFC   | Anode & cathode: noncatalyzed disc-shaped graphite. | *Cymbopogon citratus* | NA | In series: 1.49 ± 0.091 V In parallel: 0.158 ± 0.005 V | 31.9 mWm~−2 in series and parallel | In series: ~36 mA.m~−2 In parallel: ~458 mA.m~−2 | [73]     |
| Simulated NH₄⁺ polluted river water | Up-flow CW-MFC               | Anode & cathode: activated carbon-stainless-steel mesh. Electrode spacing between cathode and anodes (A): A1: 25 cm, A2: 15 cm. | *Canna flaccida* | NA | At 15.9–26.2 °C: A1: 9.5 ± 4.9 mV A2: 10.7 ± 4.1 mV At 12.8–16.8 °C: A1: 7.5 ± 7.3 mV A2: 2.2 ± 5.6 mV At 6.0–10.5 °C: A1: 89.9 ± 29.2 mV A2: 89.5 ± 25.3 mV | At 15.9–26.2 °C: A1: 9.4 µWm~−2 A2: 9.0 µWm~−2 At 12.8–16.8 °C: A1: 1.7 µWm~−2 A2: 28 µWm~−2 At 6.0–10.5 °C: A1: 575 µWm~−2 A2: 488.1 µWm~−2 | At 15.9–26.2 °C: A1: 0.3 ± 0.16 mAm~−2; A2: 0.34 ± 0.13 mAm~−2 At 12.8–16.8 °C: A1: 0.24 ± 0.23 mAm~−2; A2: 0.7 ± 0.18 mAm~−2 At 6.0–10.5 °C: A1: 2.86 ± 0.93 mAm~−2; A2: 2.85 ± 0.81 mAm~−2 | [80]     |
| Synthetic sewage wastewater | Integrated vertical-flow CW-MFC (Up-flow and downflow) | Up-flow chamber: anode: granular graphite; cathode: carbon felt. Down-flow chamber: anode & cathode: granular graphite. | *Canna indica* (planted in the up-flow chamber) | NA | HRT of 4 and 2.8 days: all CW-MFCs below 0.02 V. | HRT of 7.6 days: Ceramsite: 120.3 mW.m~−3 Quartz: 11.3 mW.m~−3 Zeolite: 14.2 mW.m~−3 | NA | [17]     |
| Synthetic wastewater  | Algal-assisted CW-MFC integrated with sand filter. | Anode: graphite granules. Cathode: circular carbon felt. | NA | NA | ~0.275 mV | 33.14 mWm~−3 | 235.0 mA.m~−1 | [59]     |
Table 1. Cont.

| Wastewater Type | System Type | Electrodes | Plant Type | Coulombic Efficiency | Voltage | Power Density | Current Density | Reference |
|-----------------|-------------|------------|------------|-----------------------|---------|---------------|----------------|-----------|
| Swine wastewater | Alum sludge-based two stages hybrid CW-MFC system (Vertical flow (1st stage) and horizontal subsurface flow (2nd stage)) | 1st stage: Anode: stainless-steel mesh box filled with charcoal. Cathode: empty stainless-steel mesh box. 2nd stage: Anodes: stainless-steel pads. Cathodes: stainless-steel bridges. | Cyperus | 1st stage: 0.06 ± 0.01% 2nd stage: 0.22 ± 0.04% | 1st stage: 0.44 ± 0.09 V (max. 0.58 V). 2nd stage: 0.34 ± 0.09 V (Max. 0.5 V). | 1st stage: 33.3 ± 13.81 mW.m⁻³ (max. 56.9 mW.m⁻³) 2nd stage: 9 ± 2.5 mW.m⁻³ (max. 15.4 mW.m⁻³) | 1st stage: 0.07 ± 0.02 Am⁻³ 2nd stage: 0.02 ± 0.01 Am⁻³ | [37] |
| Urban wastewater | Floating treatment wetlands-MFC | Cathode: graphite rods. Anode: PVC hose filled with graphite sticks. | *Canna generalis* *Chrysopogon zizanioides* *Cyperus papyrus Nanus* *Hymenachne grumosa* *Equisetum hyemale* | Maximum Voltages in: Open circuit: 225 mV (*C. generalis*) 212 mV (*H. grumosa*) 144 mV (*C. zizanioides*) 137.4 (*C. papyrus Nanus*) 89.6 mV (*E. hyemale*) Closed circuit: 21.0 mV (*C. generalis*) 31.6 mV (*H. grumosa*) 31.7 mV (*C. zizanioides*) 40.4 (*C. papyrus Nanus*) 43.7 mV (*E. hyemale*) | 0.93 mW.m⁻² (When max voltage is 108 mV from all plants in parallel) | NA | NA | [77] |
| Synthetic eutrophication influent | Ecological floating bed-MFC | Anode & Cathode: stainless-steel mesh and carbon felt. | *Cyperus alternifolius* *Linn.*, *subsp. flabelliformis* (Roett.) Kukenth (EFB-MFC1) *Ceratophyllum demersum* Linn (EFB-MFC2) *Eichhornia crassipes* (Mart.) Solms Pinnerea crassipes Mart (EFB-MFC3) *Ipomoea aquatic* Forsk (EFB-MFC4) | NA | Control: 99 mV EFB-MFC1: 125 mV EFB-MFC2: 144 mV EFB-MFC3: 157 mV EFB-MFC4: 161 mV | The maximum power density was EFB-MFC4: 6.03 mW.m⁻² | NA | [78] |

Note: NA (not available).
4. Nutrient Removal in the Integrated Hyp-MFC and CW-MFC System

Nitrogen (N) and phosphorus (P) play a critical role in plant growth and, thus, food supply [83]. Hence, securing a continuous supply of P and N is essential to satisfy the needs for meeting the increasing food demand as the world population is growing [84,85]. The current nitrogen-fixing Haber-Bosh (H-B) process uses fossil energy to fix atmospheric nitrogen into ammonia, which is used in manufacturing fertilizer or more frequently converts N to urea or nitrogen salts for agricultural uses [86]. The H-B process is an energy-intensive process, as it requires high pressure and temperature [14,87], accounts for approximately 2% of the world’s annual energy consumption, and is a major contributor to greenhouse gas emissions into the atmosphere [11,86]. Approximately 95% of the total phosphorus (TP) production is utilized in the agricultural sector, mainly as fertilizers, around the world [84]. In the last decades, researchers have drawn attention to the challenges of global phosphorus scarcity [88–90]. Phosphorus is a nonrenewable resource commonly found in igneous and sedimentary deposits. At the current rates of mining and consumption, it would be exhausted within the next 50 to 100 years [88]. Due to the high cost associated with industrial nitrogen production and diminishing phosphate rock reserves, it is important to recover and reuse nutrients to ensure food security [91] and to reduce energy consumption and carbon emissions [92].

Ammonia is one of the most common forms of N in wastewater, which can be oxidized to nitrate by microorganisms in a CW or transported and accumulated in the cathode region of a CW-MFC system [92]. The major mechanisms of N removal in the CW include plant adsorption, volatilization (as ammonia), and nitrification–denitrification [93,94]. Wetland plants enhance COD and N removal by providing microbes with oxygen for aerobic respiration and rhizosphere secretions [45]. The major P removal processes in the CW are physicochemical processes such as sorption, precipitation, filtration, sedimentation, absorption, ion exchange, complexation reactions [76,95], and biological processes such as plant assimilation [95]. The P removal is mainly controlled by the combined effect of filter media, plants, and microorganisms [96]. Because the specific surface area of the vegetated wetland systems is relatively large, a considerable fraction of the total P is retained in the CW-MFC system [41]. Theoretically, the recovery of phosphate from sewage could potentially supplement between 15 to 20% of the worldwide phosphate demand [97].

4.1. Effects of Dissolved Oxygen on Nutrient Removal

Oon et al., (2017) [55] studied the up-flow CW-MFC with supplemental aeration. Because the anodic region was anaerobic, denitrification was favored by the microorganisms that utilize organic carbon as an electron donor. Molecular oxygen affects nitrogen removal in the CW-MFC system. Oon et al., (2017) [55] reported that the NH$_4^+$ removal decreased from 97% to 81% when the air-flow rate was decreased from 1900 mL/min to 60 mL/min. When the aeration was not provided, the NH$_4^+$ removal further dropped to 47%. In the unplanted CW-MFC system, the NH$_4^+$ removal was only 30% without the aeration [55]. The DO level in the cathodic zone affected the removal of organics and nitrogen in the CW-MFC system [76]. Gupta et al., (2021) [59] treated synthetic wastewater using the algal-assisted CW-MFC integrated with a sand filter. In their study, the removal of phosphate and ammonium was mainly by the algal uptake and nitrification in the photosynthetically oxygenated cathode region. In the anodic region, the concentrations of nitrate and ammonium decreased significantly. Their system effectively removed up to 96.37 ± 2.6% of COD, 85.14 ± 10.73% of NH$_4^+$, and 69.03 ± 10.14% of PO$_4^{3−}$. The sand filter provided an anaerobic environment in the CW-MFC to enhance nitrate removal as high as 68.41 ± 7.63% [59].

4.2. Effects of Plants on Nutrient Removal

Individual plant species have different physiological characteristics and ecological requirements. These properties may directly affect the CW-MFC performance, especially the removal of COD and nutrients, and the generation of electricity [41]. In the study by
Liu et al., (2019a) [43], the main mechanism of the N removal in the CW-MFC system was biological denitrification, which was responsible for the removal of 60% of COD, 51% of NH$_4^+$-N, 81% of NO$_3^-$-N, 29.4% of TP, and 70% of TN.

In a CW, wetland plants supply oxygen to aerobic bacteria to support the oxidation of organics and ammonia and also support the rhizosphere secretions for bacterial growth [45]. In the CW-MFC system, the oxygen generated by photosynthesis can serve as an electron acceptor to produce electric current [41]. Additionally, wetland plants have a substantial capacity to uptake phosphorus from wastewater [95,96]. Furthermore, soils and sediments absorb phosphorus in a wetland [95].

In a study by Saz et al., (2018) [41], the CW-MFC with plants provided higher treatment efficiencies compared to the system without plants. The average removal efficiencies were in a range between 85% and 88% for COD, 88% and 97% for NH$_4^+$, and 72% and 97% for TP. The aerobic environment in the presence of vegetation markedly enhanced nitrification. The system with T. angustifolia provided better NH$_4^+$ removal (97.3 ± 1.57%) than the system with other plant species. This may be due to the greater root system and larger biomass of T. angustifolia, compared to other plant species. The largest nitrate removal (63%) was observed in the unplanted CW-MFC system [41]. The phosphorus-removal efficiency was larger in the CW-MFC systems with the plants than without the plants. The phosphorus uptake by macrophytes was usually highest at the beginning of the growing season [95].

In the study of the CW-MFC with three different plants, Oodally et al., (2019) [48] found that the system with C. prolifer provided a COD-removal efficiency of 97 ± 1%, which was higher than the system with W. thyrsiflora (94 ± 1%), P. australis (94 ± 1%), and the control (unplanted) system (90 ± 1%). The system with C. prolifer provided a higher orthophosphate-removal efficiency (98 ± 0%) than the system with P. australis (81 ± 4%), W. thyrsiflora (58 ± 6%), or the control (72 ± 7%). The removal of ammonia was mainly via nitrification and adsorption [48]. Compared to other wetland plants, C. prolifer produced a higher density of root biomass, which offered a larger surface area for the growth of microorganisms and provided more oxygen for nitrification. However, the control system yielded the highest nitrate-removal efficiency (~93%) compared to the systems with the plants, as the control (unplanted) system produced an anoxic environment that favored denitrification [48]. In another study by Colares et al., (2021) [77] with the floating wetland-MFC with five different ornamental plants, the COD removal was 71.4%, but the removal efficiencies of total N and P were only 8.4% and 11.4%, respectively. The low N removal was likely due to the low oxygen levels, as oxygen plays an important role in nitrification and consequently in N removal [77]. Yang et al. (2021) [78] studied the four plants (i.e., windmill grass, goldfish algae, water hyacinth, and water spinach) in the ecological floating bed-MFC (EFB-MFC). They found that the generation of electricity can stimulate the activity of enzymes associated with N removal and drive the migration of NH$_4^+$-N from the anode to the cathode, enhancing N removal, and a portion of the total N could be removed by the absorption on the plants [78].

4.3. Effects of Media Type on Nutrient Removal

The substrate/medium type is an important factor for designing the CW/Hyp-MFC system, as it supports the growth of plants, as well as of microorganisms. Yadav et al., (2020) [73] examined the drip Hyp-MFC system to treat domestic wastewater (Figure 10). Their study used cocopeat for the medium bed to support Cymbopogon citratus. Due to its structure and water-holding capacity, cocopeat acted as an adsorbent for organics and facilitated the growth of microorganisms that contribute to aerobic treatment. The removal of P can be accomplished by microbial and plant uptake, physical adsorption on the cocopeat matrix, and electrochemical reactions at the electrode. Nitrification and denitrification could occur because of the availability of multiple microenvironments within the cocopeat matrix [73].

Yakar et al., (2018) [61] used three parallel up-flow CW-MFC (UCW-MFC) systems (Figure 9) to examine three different types of media, i.e., sand, zeolite, and volcanic cinder.
Their results showed that zeolite performed better than the other media, providing removal efficiencies of $92.1 \pm 7.27\%$ for COD, $93.2 \pm 7.01\%$ for $\text{NH}_4^+$, and $96.7 \pm 2.9\%$ for TP. The zeolite medium provided the habitats for microorganisms to support the production of enzymes for the efficient biodegradation of organics and to catalyze electron transfer. Zeolite is porous and possesses protuberances and rough morphology, and thus it has a large specific surface area compared to sand or volcanic cinder [61]. Natural zeolite contains aluminosilicate in which aluminum atoms are substituted for silicon. This structure gives zeolite a negative charge that attracts $\text{NH}_4^+$ [98,99]. $\text{NH}_4^+$ is generally adsorbed as an exchangeable ion on clays and is chemisorbed by humic substances or fixed within the clay lattice [95]. The large surface area of zeolite exposes numerous active sites for the chemisorption of organic matter through the ion-exchange process and creates suitable living conditions for microorganisms [45]. In their study with the alum-sludge-based two-stage hybrid CW-MFC system (Figure 14), Ren et al., (2021) [37] achieved removal efficiencies of $72 \pm 7.4\%$ for COD, $88 \pm 8.7\%$ for PO$_4^{3-}$-P, $59 \pm 28.3\%$ for $\text{NH}_4^+$-N, $69 \pm 25.6\%$ for NO$_3^-$-N, $47 \pm 19.7\%$ for TN, and $85 \pm 9.5\%$ for TP. The COD removal was mainly attributed to the biological degradation of organic matter by bacteria grown on the alum sludge. In their study, adsorption and precipitation of the substrates played a critical role in P removal.

4.4. Effects of Hydraulic Retention Time on Nutrient Removal

The hydraulic retention time (HRT) plays an important role in nutrient removal in the CW-MFC system. In the study with the vertical-flow CW-MFC with ceramsite (Figure 11), Zhong et al., (2020) [17] reported that the removal efficiencies of $\text{NH}_4^+$-N and PO$_4^{3-}$-P were 93.8 and 99.6%, respectively, with ceramsite at the HRT of 2.8 days. As the HRT decreased from 7.6 to 2.8 days, the $\text{NH}_4^+$-N level increased at the sampling ports of the up-flow chamber with ceramsite, the down-flow chamber with quartz, and both chambers with zeolite. The relatively high pH values in the up-flow chamber with ceramsite could have contributed to the ammonia loss via volatilization. The difference in the N-removal efficiency with ceramsite, compared to other substrates, can be partly explained by the fact that the higher iron content in milled ceramsite granules contributed to the ferrous ion’s higher oxidation potential and the abundance of denitrifiers for the nitrate reduction. With the ceramsite medium, the changes in the PO$_4^{3-}$-P concentration with varying HRT were similar to the change in the $\text{NH}_4^+$-N concentration, except for the system with quartz. Because quartz is less porous compared to the other two substrates, it could not provide a diverse niche for nitrifying and denitrifying bacteria for the removal of $\text{NH}_4^+$-N and TN. The high P-removal efficiency with ceramsite was mainly due to substrate adsorption and denitrifying phosphorus removal [17]. In addition, the biological P uptake might have contributed to the PO$_4^{3-}$-P removal in the system with ceramsite [17]. In another study by Yadav et al., (2020) [73] with the drip Hyp-MFC system (Figure 10), the HRT also played an important role in treating domestic wastewater. Their study showed that, after 3 h of operation in a batch-recirculation mode, the removal efficiencies of COD, phosphate, and $\text{NH}_4^+$-N were $72 \pm 2.4\%$, $83.2 \pm 1.1\%$, and $35.1 \pm 2.4\%$, respectively. These efficiencies increased considerably to $85.7 \pm 0.6\%$, $85.8 \pm 0.6\%$, and $76.6 \pm 1.1\%$, respectively, after 12 h of operation [73].

4.5. Effects of System Design on Nutrient Removal

The type and design of the CW-MFC system also influence nutrient removal. Xu et al., (2017) [82] treated synthetic wastewater using the up-flow stacked CW-MFC (Figure 15). In their study, wastewater was pumped into the bottom of the CW-MFC system, then it flowed through the stainless-steel mesh net (air-cathode) to the anode region (first stage treatment). The effluent of the first stage was cascaded down to the second stage (the down-flow CW) in which nutrient removal proceeded. Their system provided $83.2\%$ COD removal and $94.5\%$ phosphate removal in the upper part of the system. Their system, however, provided only a small improvement in COD removal (from $83.2\%$ to $88.7\%$) with no additional phosphate
removal in the outer part of the system. On the other hand, the TN-removal efficiency was markedly increased from 53.1% to 75.4% in the outer part of the system. This indicated that the N removal by nitrification and denitrification was significantly improved in the outer down-flow section of the CW because when wastewater flowed through the outer part of the CW the aerobic condition gradually turned anoxic, then anaerobic at the bottom, favoring denitrification [82].

In the study by Oon et al., (2015) [40] with the up-flow CW-MFC system (Figure 5), an aerator was placed 45 cm from the bottom of the CW. Their result showed that the aeration aided the growth of the aerobic microbial community, indicated by the enhanced biodegradation of organics and improved nitrification. However, the aeration inhibited the nitrate removal in the cathode region of the system. The COD removal was 58.6% in the anodic chamber. With the aeration, the COD-removal efficiency was 100%. The reduction in NH$_4^+$ in the aerated region was 91%. The removal of nitrate was 97% in the anaerobic region and 40.17% in the aerated region. The high nitrate-removal efficiency in the anaerobic region is likely attributed to denitrification [40]. In the CW-MFC system studied by Yu et al., (2020) [80], the NH$_4^+$ removal of 87.3 ± 16.3% was achieved in the temperature range between 12.8 and 16.8 °C, with electrode spacing of 15 cm. Their CW-MFC system enhanced the NH$_4^+$ removal by the NH$_4^+$ oxidation augmented by the self-generated electricity [80].

Corbella et al., (2015) [57] used two pilot plant-scale horizontal subsurface-flow CW-MFC (Figure 12). One system was fed with the primary settled wastewater (settler line) and the other with the effluent of the hydrolytic up-flow sludge blanket reactor (HUSB line) (Figure 12). This system was described previously in Section 3.6. The settler line directed the pretreated wastewater to one of the CW-MFC systems and the HUSB line directed the flow to the other CW-MFC. Between the HUSB and settler lines, no significant differences were observed for the removals of NH$_4^+$-N, NO$_3^-$-N, and PO$_4^{3-}$-P. The removal efficiencies for NH$_4^+$-N in the HUSB line and settler line were 58% and 60%, respectively. Since the organic concentration in the settler line was lower than that in the effluent of the HUSB reactor, the better organic removal in the settler line can be attributed to the smaller OLR in the settler line compared to that in the HUSB line. The COD-removal efficiencies in the settler line and the HUSB line were 71% and 61%, respectively [57].

In the study with the integrated vertical-flow CW-MFC system (Figure 13), Liu et al., (2019) [58] reported the average removal efficiencies of 79.65%, 75.13%, and 77.5% for COD, NO$_3^-$-N, and NH$_4^+$-N, respectively. In the aerobic region, the roots of Canna indica encouraged the growth of a special microbial community and the generation of oxygen, which promoted the removal of nitrogen and organic matter. In the cathode region, the organic carbon sources at sufficient levels could encourage the growth of aerobic denitrifiers that can utilize both oxygen and nitrate as terminal electron acceptors to promote denitrification under aerobic conditions [58]. Ammonia-N was removed successfully but very limited denitrification took place in the vertical-flow CW systems [58]. On the other hand, the horizontal-flow CW provided the conditions favorable for denitrification, but the ability of these systems to nitrify ammonia is very limited. Therefore, various types of CWs may be combined to exploit the specific advantages of the individual systems [95].

Overall, the results indicate that CW-MFC systems are capable of removing COD, nitrate, ammonium, and phosphate from various types of wastewater (Table 2). The studies, however, presented a wide range of removal efficiencies, which could be due to the differences in design, operation mode, and other experimental variables. Macrophytes and supplemental aeration in the cathode region could benefit ammonium removal and bioelectricity generation. Since aeration would increase the operational cost, the airflow rate needs to be optimized to obtain cost-effective removals of COD, nitrate, and ammonium, and energy recovery [55].
5. Limitations and Future Scope

To meet the increasing demands for water, energy, and food, it is becoming essential to develop sustainable technologies capable of recovering energy and nutrients from wastewater and treating the wastewater. The CW-MFC systems have attracted attention as they can perform these functions at the same time [79]. Although the CW-MFC systems have been investigated for approximately a decade, the concept is still in a stage of infancy, and several aspects of the systems have not become fully understood [48]. The major drawbacks of these systems are high capital costs, low power output, and various technical limitations such as clogging of the CW [52]. Moreover, the power output largely relies on the effectiveness of the electrode material, type of plant species, and richness of the electroactive microorganisms [45]. Affected by the design and operating conditions, such as the hydraulic loading rate, organic loading rate, substrate porosity, water depth, oxygen supply conditions, and plant growth, the substrate pore size and hydraulic conductivity are reduced over time and, eventually, the CW will be clogged [53]. The clogging significantly reduces the CW-MFC performance.

The usefulness of the CW-MFC system as an electricity generation device has not been extensively examined in real-world scenarios [17]. Currently, its uses are limited to laboratory or small-scale field experiments [45,49,73,74,84]. For the optimal operation of the CW-MFC system to remove organics and nutrients [76] and to generate electric power, it is important to understand the effects of multiple factors. The biological nitrification–denitrification seems to be the major N-removal process in most treatment wetlands. Nitrification occurs in all types of CWs, and their performance is affected by the availability of oxygen [95]. In the up-flow system, the N removal is likely restricted, as nitrification is hindered in the anode region because of a lack of oxygen and denitrification is blocked by the presence of oxygen in the cathode region [17]. In the vertical-flow CWs, ammonia-N can be removed effectively but limited denitrification may take place. On the other hand, the horizontal-flow CWs can provide the conditions for denitrification, but the ability of these systems to nitrify ammonia is limited [95]. The selection of wetland plant species suitable for the CW-MFC is important, as each plant has its characteristic physiology and morphology, which could affect bioelectricity generation and wastewater treatment [48]. Keeping sustainability and field implementation in view, further research is necessary to better understand the operational mechanisms of the CW-MFC systems. The substrate clogging caused by high-strength wastewater, in particular, needs to be addressed. This drawback can be overcome by facilitating a low-cost filtration system. Moreover, detailed studies are needed to understand the degradation kinetics involved in the biochemical processes in the CW/Hyp-MFC systems.

The cost for treating wastewater using an MFC at a flow rate of 318 m³/h was estimated to be around $6.4 million, only 9% of the cost (~$68.2 million) of treating the wastewater by a conventional WWTP [100,101]. In a conventional WWTP, 25–40% of the operating costs are attributed to energy consumption [102]. For developing self-sustaining WWTPs, a high energy efficiency, low cost, and low carbon footprint are consequential requirements in upscaling and implementing treatment facilities. The conventional MFC requires a costly PEM. In contrast, the CW/Hyp-MFC can be operated without the PEM. Therefore, the CW/Hyp-MFC has the potential to make wastewater treatment facilities energy neutral or even turn them into net energy producers.
### Table 2. Nutrient removal in the Hyp-MFC and CW-MFC systems.

| Wastewater Type | System Type | Initial Wastewater Characteristics | Plant | COD Removal | PO₄³⁻ Removal | NH₄⁺ Removal | NO₃⁻ Removal | Reference |
|-----------------|-------------|------------------------------------|-------|-------------|---------------|--------------|--------------|-----------|
| Domestic wastewater | Horizontal subsurface flow CW-MFC | HUSB line: COD: 323 mg/L, NH₄⁺-N: 41 mg/L, NO₃⁻-N: <1 mg/L, PO₄³⁻-P: 9 mg/L; Settler line: COD: 235 mg/L, NH₄⁺-N: 39 mg/L, NO₃⁻-N: <1 mg/L, PO₄³⁻-P: 7 mg/L | Phragmites australis | HUSB line: 61%; Settler line: 71% | NA | HUSB line: 58%; Settler line: 60% | NA | [57] |
| Synthetic wastewater | Up-flow CW-MFC | COD: 314.8 ± 13 mg/L | Typha latifolia | 58.6% in the anode region, 100% with supplementary aeration. | NA | 91% cathode region | 97% anode region, 40.17% cathode region | [40] |
| Synthetic wastewater | Stacked CW-MFC | COD: ~280 mg/L, TN: ~22.5 mg/L, NH₄⁺-N: ~15 mg/L, PO₄³⁻-P: ~11.2 mg/L | NA | 88.7% | 94.5% | 85.1% | NA | [62] |
| Synthetic wastewater | Up-flow CW-MFC | COD: ~650 mg/L, NH₄⁺-N: ~40 mg/L | Elodea nuttallii | 98% | NA | At 1900 mL/min: 97%, At 1200 mL/min: 96%, At 600 mL/min: 84%, At 60 mL/min: 81% | At 1900 mL/min: 97%, At 1200 mL/min: 96%, At 600 mL/min: 44%, At 60 mL/min: 50% | [55] |
| Synthetic wastewater | CW-ACMFC & CW-MFC | COD: 198.4 mg/L, NH₄⁺-N: ~40 mg/L | Cannas indica | - CW-ACMFC: average 69.9% (maximum: 79.13%) - CW-MFC: average 66% | NA | CW-ACMFC: average NH₄⁻N removal 36.2% CW-MFC: average NH₄⁻N removal 32.8% | NA | [62] |
| Synthetic wastewater | Up-flow CW-MFC | COD: 103–554 mg/L, NH₄⁺-N: 19.9–225.5 mg/L, NO₃⁻-N: 12.9–191.5 mg/L, TP: 10–29.8 mg/L | Typha latifolia | Sand: 90.6 ± 4.12%, Zeolite: 92.1 ± 7.27% | Zeolite: 96.7 ± 2.9% | Sand: 92.6 ± 6.05% Zeolite: 93.2 ± 7.01% | Zeolite: 95 ± 5.49% Sand: 81.47 ± 7.3% Zeolite: 82.41 ± 3.45% Volcanic cinder: 77.9 ± 17.5% | [41] |
| Synthetic wastewater | CW-MFC | COD: 344 ± 143 mg/L, NH₄⁺-N: 45.7 ± 32.7 mg/L, NO₃⁻-N: 25.08 ± 16.5 mg/L, TP: 20.7 ± 9.84 mg/L | Typha latifolia (M₁), Typha angustifolia (M₂), Juncus gerardii (M₃), Carex divisa (M₄) Unplanted (M₅) | M₁: 88.25 ± 11.8% Tp: M₁: 96.38 ± 5.64% M₂: 97.05 ± 4.408% M₃: 95 ± 5.49% M₄: 96.9 ± 4.03% M₅: 71.77% | M₁: 95.33 ± 1.93% M₂: 97.15 ± 1.57% M₃: 96.3 ± 2.33% M₄: 95.4 ± 1.54% M₅: 88.1 ± 7.15% | M₁: 95.33 ± 1.93% M₂: 97.15 ± 1.57% M₃: 96.3 ± 2.33% M₄: 95.4 ± 1.54% M₅: 88.1 ± 7.15% | M₁: 52.68 ± 13.81% M₂: 55.52 ± 18.47% M₃: 41.99 ± 24.06% M₄: 38.8 ± 27.79% M₅: 63.6 ± 13.3% | [41] |
| Wastewater Type      | System Type                          | Initial Wastewater Characteristics | Plant                   | COD Removal | PO$_4^{3-}$ Removal | NH$_4^+$ Removal | NO$_3^-$ Removal | Reference |
|----------------------|--------------------------------------|------------------------------------|-------------------------|-------------|---------------------|------------------|------------------|-----------|
| Synthetic wastewater | Integrated vertical-flow CW-MFC      | COD: 10.6–20.4 mg/L; TP: 0.02–0.40 mg/L; TN: 218–6.74 mg/L; NH$_4^+$: 0.123–1.245 mg/L; NO$_3^-$: 1.036–3.326 mg/L | *Canna sp.* and *Acorus calamus* | 60%         | TP: 29.4%           | 51%              | 81%              | [43]      |
| Swine wastewater     | Integrated vertical-flow CW-MFC      | COD: 324–708 mg/L; NH$_4^+$: 138–284 mg/L | *Canna indica*          | 79.65%      | NA                  | 73.5%            | 75.13%           | [58]      |
| Settled sewage       | CW-MFC                               | COD: 730 mg/L; PO$_4^{3-}$: 11.46 mg/L; NH$_4^+$: 106.94 mg/L; NO$_3^-$: 7.53 mg/L | *Cyperus prolifer* (N$_1$) | N$_2$: 97 ± 1% | N$_2$: 98 ± 0%     | N$_2$: 58 ± 6%   | N$_2$: >99%      | [48]      |
| Domestic sewage      | Integrated drip hydroponics-MFC      | COD: 410 ± 16 mg/L; PO$_4^{3-}$: 3.8 ± 0.2 mg/L; NH$_4^+$: 41.5 ± 2.1 mg/L; NO$_3^-$: 0.28 ± 0.13 mg/L | *Cymbopogon citratus*     | 72 ± 2.4% at HRT = 3 h; 85.7 ± 0.6% at HRT = 12 h | 83.2 ± 1.1% at HRT = 3 h; 85.8 ± 0.6% at HRT = 12 h | 35.1 ± 2.4% at HRT = 3 h; 76.6 ± 1% at HRT = 12 h | NA       |
| Simulated NH$_4^+$   | Up-flow CW-MFC                       | NH$_4^+$: 16.3 ± 3.3 mg/L; NH$_3$: 10.5 ± 4.83 mg/L | *Canna flaccida*         | NA          | NA                  | Electrode spacing between cathode and anodes | At 15.9–26.2 °C: A1: 86.1 ± 15.4%; A2: 82.4 ± 16.3% At 12.8–16.8 °C: A1: 42.3 ± 25.6%; A2: 87.3 ± 17.1% At 6.0–10.5 °C: A1: 73.7 ± 13.9%; A2: 66.1 ± 22.5% | NA       |
| polluted river water |                                     |                                    |                         |             |                    |                 |                  |           |
| Synthetic sewage     | Integrated vertical-flow CW-MFC (Up-flow and downflow) | HRT: 7.6 days; COD: 262 ± 69 mg/L; PO$_4^{3-}$: 7 ± 0.55 mg/L; NH$_4^+$: 18.04 ± 2.75 mg/L; HRT: 4 days; COD: 243 ± 22 mg/L; PO$_4^{3-}$: 7.29 ± 0.58 mg/L; NH$_4^+$: 16.36 ± 1.20 mg/L; HRT: 2.8 days; COD: 243 ± 15 mg/L; PO$_4^{3-}$: 2.24 ± 0.19 mg/L; NH$_4^+$: 16.74 ± 1.25 mg/L | *Canna indica*             | Ceramist: HRT = 7.6 d: 96.6%; HRT = 4 d: 62.95%; HRT = 2.8 d: 47.3%; Quartz: HRT = 7.6 d: 95.7%; HRT = 4 d: 92.6%; HRT = 2.8 d: 91.8% | Ceramist: HRT = 7.6 d: 95%; HRT = 4 d: 99.6%; HRT = 2.8 d: 99.6%; Quartz: HRT = 7.6 d: 10.1%; HRT = 4 d: 9.2%; HRT = 2.8 d: ~9.1% | Ceramist: HRT = 7.6 d: 95.8%; HRT = 4 d: 98.3%; HRT = 2.8 d: 93.8%; Quartz: HRT = 7.6 d: 36.1%; HRT = 4 d: 6.7%; HRT = 2.8 d: 0.4%; Zeolite: HRT = 7.6 d: 64.4%; HRT = 4 d: 27%; HRT = 2.8 d: 12.5% | NA       |
| wastewater           |                                     |                                    |                         |             |                    |                 |                  |           |
| Algal-assisted       | CW-MFC integrated with sand filter   | COD: 1505.61 ± 681.54 mg/L; PO$_4^{3-}$: 17.13 ± 5.77 mg/L; NH$_4^+$: 21.72 ± 7.99 mg/L; NO$_3^-$: 40.25 ± 17.38 mg/L | *Canna indica*             | NA          | 96.37 ± 2.6%       | 69.03 ± 10.14%  | 85.14 ± 10.73%  | 68.41 ± 7.63% | [59]      |
Table 2. Cont.

| Wastewater Type          | System Type                          | Initial Wastewater Characteristics | Plant                  | COD Removal | PO\textsubscript{4}− Removal | NH\textsubscript{4}+ Removal | NO\textsubscript{3}− Removal | Reference |
|--------------------------|--------------------------------------|------------------------------------|------------------------|-------------|------------------------------|------------------------------|-----------------------------|-----------|
| Swine wastewater         | Alum sludge-based two stages hybrid CW-MFC system | COD: 526 ± 104.6 mg/L              | Small Cyperus          | 72 ± 7.4%   | 88 ± 8.7%                   | 59 ± 28.3%                   | 69 ± 25.6%                   | [37]      |
| Urban wastewater         | Floating treatment wetlands-MFC       | COD: 525.0 ± 268.5 mg/L TN: 95.2 ± 35.8 mg/L TP: 8.91 ± 2.25 mg/L | Canna generalis Chrysopogon zizanioides Cyperus papyrus Nanus Hymenachne graminoides Equisetum hyemale | 71.4% | TP: 11.4%                   | TN: 8.4%                     | NA                                | [77]      |
| Synthetic eutrophication influent | Ecological floating bed-MFC          | COD: 100 mg/L NH\textsubscript{4}+ N: 30 mg/L | Cyperus alternifolius Limn. subsp. flabelliformis (Rottb.) Kakens (EFB-MFC1) Ceratophyllum demersum Limn (EFB-MFC2) Eichhornia crassipes (Mart.) Solms Ponteria crassipes Mart (EFB-MFC3) Ipomoea aquatica Forsk (EFB-MFC4) | Control: 73.88% | EFB-MFC1: 73% | EFB-MFC2: 76.37% | EFB-MFC3: 78.23% | EFB-MFC4: 82.49% | Control: 38.74% | EFB-MFC1: 34.76% | EFB-MFC2: 41.65% | EFB-MFC3: 51.21% | EFB-MFC4: 55.6% | NA | [78] |

Note: NA (not available).
To the authors’ best knowledge, the Hyp-MFC system is a new technology that needs to be explored further. The Hyp-MFC system can be a potential technology to avoid substrate clogging. Additionally, plants with high economic values may be studied in the Hyp-MFC systems, taking advantage of the nutrients present in wastewater to satisfy the future energy, food, and water demands of the growing population.

6. Conclusions

This paper focused on the integrated CW-MFC systems for energy generation and nutrient removal from various wastewaters. The findings suggested that the performance of the CW-MFC technologies depends on a number of factors including biological, electrochemical, design, and operational factors. The CW-MFC systems discussed in this paper can be alternative sustainable technologies to alleviate the escalating water, energy, and food demands, coinciding with the diminishing nonrenewable natural resources. Despite the significant advantages of integrated CW-MFC systems, these systems are facing many challenges. Hydroponics as a standalone system has been widely explored; however, little work on the Hyp-MFC system has been found. There is a need for research on the Hyp-MFC systems. This literature review revealed that further research is necessary to better understand the CW/Hyp-MFC systems for the implementation of these technologies in real-world settings.

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