

Recent advances in soil microbial fuel cells based self-powered biosensor

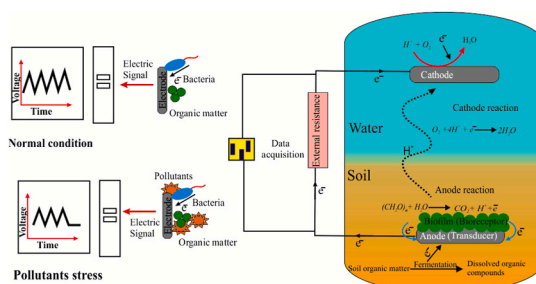
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HIGHLIGHTS

- Soil microbial fuel cell-based biosensor is an innovative technology.
- SMFC could monitor pollutants and DO in the different depths of soil.
- The SMFC based biosensors could be used as an early-alert program in polluted soil.
- Fundamentals, optimization, and future challenges of SMFC-based biosensors.

GRAPHICAL ABSTRACT



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ABSTRACT

The soil microbial fuel cell (SMFC) is a new device that was originally designed to generate electricity from organic matter in soil using microorganisms. Currently, SMFC based biosensors are emerging as a new and promising research direction for real-time and rapid monitoring of soil quality or soil pollution. Compared to conventional biosensors, SMFC based biosensors exhibit advantages such as low-cost, simple design, in-situ, and long-term self-powering monitoring, which makes it become attractive devices for in-situ long-term soil quality or soil pollution monitoring. Thus, this review aims to provide a comprehensive overview of SMFC based biosensors. In this review, different prototypes of SMFC based biosensors developed in recent years are introduced, the biosensing mechanisms and the roles of SMFC are highlighted, and the emerging applications of these SMFC based biosensors are discussed. Since the SMFC based biosensors are applied in open-air conditions, the effects of different environmental factors on the biosensing response are also summarized. Finally, to further expand the understanding and boost the practical application of the SMFC based biosensors, future perspectives including fundamental mechanism exploration and investigation of the full-scale application are proposed.

1. Introduction

Soil is one of the most fundamental resource for human survival on the earth. Soil pollution not only affects ecological security but also seriously threatens human life. How to protect the soil is a severe

challenge for all mankind. Globally, China is one of the countries with serious soil pollution problems (Yuxin, 2020). The State Council 21353 of China proposed the “Soil Pollution Prevention and Control Action Plan” in 2016, pointing out that soil environmental pollution has become a short-board for building a well-off society in an all-round way,

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and clearly emphasized that soil pollution prevention and control are key components of ecological civilization and ecological security construction (Zhang and Li, 2016). Therefore, the prevention of soil pollution is of great significance.

To control the soil pollution, the quality of soil environment should be fully grasped. Therefore, soil environmental monitoring is listed as the “Top Ten Tasks” of the Soil Pollution Prevention and Control Action Plan (Li et al., 2019). At present, soil environmental monitoring mainly involves on-site sampling and offline laboratory analysis. Although, these method could achieve accurate quantitative detection of different pollutants but often encounters time-consuming and high-cost. These methods are difficult to be used for on-site rapid detection and long-term in-situ monitoring of soil quality. However, in-situ rapid detection is essential for real-time monitoring and rapid warning of soil pollution. Biological toxicity is the most important indicator of environmental pollution and has a direct correlation with human health and ecological safety. Environmental pollution warning is the most critical part of early detection, prevention, and reduction of safety risks. Therefore, there is urgent need to develop rapid, in-situ monitoring methods for soil toxicity.

Soil Microbial Fuel Cells (SMFC) is a new technology that utilizes the abundant electroactive microbial flora in the soil to convert organic matter into a current signal. SMFC relies primarily on electroactive microorganisms and their interaction with non-electroactive microbial populations to achieve efficient conversion of organic chemical energy to electricity (Abbas and Rafatullah, 2021). Electroactive microorganisms are capable of extracellular electron transportability (Moradian et al., 2021). SMFC can be used not only to provide continuous power for small sensors but also reduce soil heavy metal toxicity. According to the literature, the first membrane-less laboratory-based SMFC was developed by Dunaj et al. (2012). They used the homogenized soil with deionized water, analysed the water for pollutants detection and identified the *Geobacter* spp. Later on, Li et al. (2014) used SMFC to bioremediate petroleum hydrocarbons from saline soil. First time, SMFC with multi-anode system was designed and *Escherichia* sp. and *Geobacteraceae* sp. were reported. Recently, SMFC has attracted the attention of many researchers due to simultaneously sustainable power generation and soil pollutants remediation. SMFC can provide inexhaustible source of electron donors or acceptors and can be easily controlled by adjusting the electrochemical parameters (Abbas et al., 2022). Furthermore, SMFC could be employed to monitor soil toxicity, microbial activity and methane emission in paddy soil by generating electrical signals (Jiang et al., 2016).

Therefore, there is urgent need to construct an efficient SMFC to monitor different toxic pollutants. There are some advantages of SMFC based biosensor as follow: (1) its inhibit the inoculation process because SMFC based sensor use soil native exoelectrogens to produce voltage baseline (2) the sub-surface exoelectrogens prevent from pollutants shock because surface soil adsorb most of the pollutants and cathode senses the pollutants instead of exoelectrogens (Sonoda et al., 2019) (3) the sensor's components are bunched into an integrated system. Thus, monitoring can be easily started by putting the sensor into the flooded soil (Patil et al., 2010; Liu et al., 2020). At present, the mechanism of electron transport between soil and electrode is still unclear. An in-depth clarification of the electronic interaction mechanism between the soil complex microbial flora and electrodes should be investigated to understand extracellular electron transport (EET). Therefore, there is urgent need for in-depth scientific exploration of cutting-edge.

This review not only covers the different prototype of SMFC based biosensors but also clarifies the electronic interaction mechanism between bacteria and the complex soil environment. On the other hand, this review can summarize not only the effect of different parameters on SMFC performance but also rapid in-situ monitoring and early warning of soil pollution toxicity. At the same time, it can supply the power to in-situ biosensors which will greatly reduce the input cost of in-situ monitoring. More importantly, it would provide the potential for in-

situ, real-time and long-term monitoring of soil pollution in remote areas, and has applications and prospects.

2. Different prototypes of SMFC based biosensor and biosensing mechanisms

The SMFC based biosensor consists of an anode embedded in the soil and a cathode placed in the overlying water. The electrogens produce electrons that transfer to the anode and then finally complete the circuit by oxygen reduction at the cathode which ultimately generate quantitatively electrical signals. These electrical signals are quantitatively used to monitor the concentration of analytes in the soil. Recently, some SMFC based biosensors have been designed and categorized following: based on the analytes (soil pollution, such as heavy metal, PAH, NO₃, etc.; soil quality, such as dissolved oxygen (DO), biological oxygen demand (BOD), chemical oxygen demand (COD), pH, temperature, etc.) as shown in Table 1. Mostly, single chamber SMFC are designed but until low power density is obtained as compared to conventional MFC. The performance of SMFC in terms of earlier detection response depends highly on the soil's BOD, COD, pH, temperature and nitrogen.

2.1. Different prototypes of SMFC based biosensor

As shown in the Fig. 1, biosensor consists of biological material with transducers which have been developed for the quantitatively or semi-quantitatively monitoring of contaminants like heavy metals (Cu²⁺, Cd²⁺, and Pb²⁺), volatile fatty acid, analytes, BOD and COD (Zhao et al., 2018; Raj and Maiti, 2020). SMFC used the microbes as biocatalysts for power generation and the pollutants concentrations are monitored by electrical signals. The metabolic activities of electroactive bacteria (EAB) inhibited by toxicants, which leads to low electron transfer rate from exoelectrogens to electrodes.

However, there is a directly proportional relationship between voltage generation and substrate concentration. Different electron shuttles like riboflavin enhances the electrons transfer to electrodes. However, low cell membrane permeability restricts the electrons transfer from microbes to electrodes (Khan et al., 2020). Furthermore, pollutants quantity in the soil can be directly transferred into digital signals which is easier way of in-situ monitoring. Liu et al. (2020) designed SMFC based sensors for in-situ and online Cu²⁺ detection in the paddy soil, as shown in Fig. 2(a). These SMFC based sensors were operated for eight months in the field, and different concentrations of CuCl₂ were applied at different seasons. They applied 4 SMFC based sensors in different positions to detect Cu²⁺ in the paddy soil. They reported that these SMFC based sensors could be operated for long-time in different weather seasons and cathode could also detect Cu²⁺ shock. The voltage or power density generated in these SMFC were limited by soil particles, high internal resistance, less exoelectrogens, low freshwater conductivity, low organic matter, and low O₂ reduction at the cathode. Previously designed SMFC based sensors could not detect repeated pollutants concentrations until exoelectrogens were isolated and re-inoculated. This SMFC based sensor was a breakthrough in detecting the repeated Cu²⁺ concentrations in different weather seasons. Wu et al. (2018) were also constructed paddy soil-based SMFC sensors to detect the Cu²⁺ shock. Each sensor was consisted of stainless steel tube-shaped anode and platinum grid cathode in plastic pipe. They reported that baseline voltage was stable before Cu²⁺ addition. The peak voltage is not the optimum signal of Cu²⁺ concentrations because it is largely influenced by baseline voltage.

Olias et al. (2020a) designed ceramic SMFC (CSMFC) to detect the DO in the saturated soil as shown in Fig. 2(b). The parafilm sealed anode was buried in the soil inside the terracotta vessel, the cathode was wrapped outside the ceramic vessel with non-conductive wire, and titanium wire was attached for electrons collection. The stable signals were obtained after one month without water and nutrients addition. They reported that higher anode area could enhance the cathode

Table 1
Studies on the performance of different prototypes of SMFC based biosensors with different parameters.

Reactor	Lowest concentration detected	Pollutants	Power generation (mW/m ²)	BOD (g/kg)	COD (g/kg)	Temperature (°C)	pH	Total Nitrogen (g/kg)	Comment	Reference
Single chamber	0.22 mg/kg	Cu ²⁺	0.22 ± 0.03	73.58	5.47	35	–	2.63	Stainless anode and platinum based cathode in the SMFC biosensor inserted in the paddy soil to monitor Cu ²⁺	Liu et al., (2020)
Single chamber	10 mg/L	Cu ²⁺	0.1	0.11	35.22	15	7.35	2.87	Tube shaped anode and grid shaped cathode placed in the paddy soil to monitor Cu ²⁺ by giving Cu ²⁺ shock	Wu et al., (2018)
Single chamber	–	Petroleum hydrocarbons	0.2	–	142	–	8.26	46	Graphite anode with activated carbon cathode used to monitor the petroleum hydrocarbon in the soil	Li et al., (2016)
Single chamber	0.2 mg/L	Cr (VI)	48.88	12.65	–	40	6.4	–	Carbon fiber based electrodes filled with sediment and sterile water used to monitor Cr (VI)	Zhao et al., (2018)
Double chamber	<50 mA/m	U (VI)	–	20–100	–	–	–	–	Monitor the U (VI) in the soil sub-surfaces in the open and enclosed SMFC	Bereza-Malcolm and Franks, (2015)
Single chamber	0.1 µg/L	Diuron Glyphosate Atrazine	0.06 ± 0.34	4.5	–	20	1.65	–	Algal based biocathode inside ceramic vessel used to monitor pesticide	Olias and Di Lorenzo, (2020)
Single chamber	0.003 mg/L	Atrazine	64 ± 21	17.4	–	–	6.5	0.001	SMFC fitted in the polypropylene boxes. Graphite felt electrodes used to monitor the Atrazine in the contaminated soil	Nandy et al., (2022)
Single chamber	10 mg/L	–	2.79 ± 1.87	2.0	200	28	5.4	9.1	Single chamber SMFC inoculated with Andean soil used to monitor BOD	Logroño et al., (2016)
Single chamber	–	–	6	2.9	–	20	–	–	Single chamber PMFC was designed to monitor water soil contents	Tapia et al., (2018)
Single chamber	–	–	28.9	–	300	64	7.2	1.3	Single chamber SMFC was designed for in-situ monitoring of water quality	Velasquez-Orta et al., (2017)
Single chamber	–	Urea	9	3.3	–	–	7.0	0.2	Single chamber SMFC was designed to employ in the field to monitor the urea in the soil	Barbato et al., (2021)
Single chamber	–	–	0.5	–	33,420	30	7.3	–	A portable SMFC was designed to monitor the water contents in the soil	Nguyen and Taguchi, (2021)
Stacked	–	Urea	6.54	–	17.4	30	7.0	10	Stacked SMFC were constructed to monitor the urea in the soil and also power the wastewater treatment chamber	Dziegielowski et al. (2020)

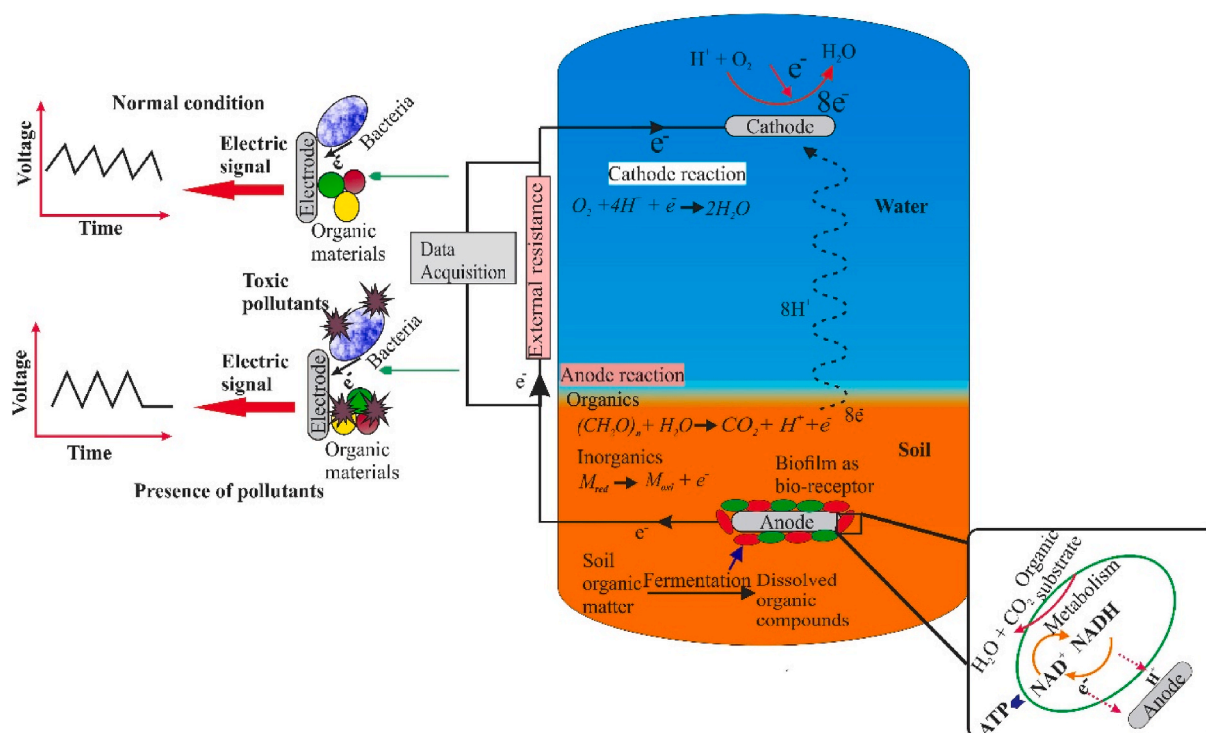


Fig. 1. The prototype of SMFC based sensor and pollutants detection mechanism in SMFC.

diffusion layers, leads to inaccurate DO detection. Water is the main source of ions and nutrients transportation in the soil. So, in this study, maximum power density was obtained after nutrients additions and moisture was kept in the anode chamber to obtain stable signals because signal stability highly depends upon soil hydration. Brunelli et al. (2016) designed simple soil based MFCs in which nutrients enriched soil filled in the cylindrical plastic container, as shown in Fig. 2(c). This SMFC could run for a month, if the soil is highly enriched with nutrients, even with low output voltage. To boost the voltage output, a plant was introduced in the SMFC. So, a continuous nutrients supply to exoelectrogens was established by photosynthesis. This plant MFC (PMFC) operated under photosynthesis and the continuous supply of organic compounds to anodic exoelectrogens. The voltage output could be influenced by parasite attacks or irregular growth of the plant. This PMFC could be employed to monitor plants health and supply power to wireless electronic systems.

Olias et al. (2020b) also designed CSMFC to detect earlier eutrophication by measuring different photosynthetic DO patterns in water, as shown in Fig. 2(d). The increased power output on days and lower at night were directly proportional to algal activity. Conclusively, this was low-cost CSMFC with minimal maintenance in which a large anode/cathode surface area measured DO in the form of a stable signal.

2.2. Biosensing mechanisms

Normally, SMFC based biosensors do not generate high power density as they produce different electrical signals under stress of various pollutants. The biofilm of exoelectrogens on the anode acts as bio-receptor and anode acts as transducing element. In the anodic chamber, different pollutants concentrations affect the rate of electrons transfer by influencing oxidation rate of pollutants in the form of signal generation (Chung et al., 2021). Recently, enzymatic recognition and whole-cell layer have been developed in the bio-electrochemical systems for the recognition of heavy metals ions. The EAB interact with the anode act as bioreceptor which could monitor heavy metal ions by fluctuating the current signals. The EAB on the electrode transfer electrons to heavy

metal ions via their respiratory cycle (Noori et al., 2021). The voltage pattern highly depends upon the heavy metals concentrations in the soil. Under heavy metal stress, some EAB show resistance by expelling heavy metals ions within the body by using ATP. As the EAB use more ATPs, their life cycle move to the death phase (Baldiris et al., 2018). Still, no study was carried out to find the effect of electrode materials on organics detection in SMFC because the electrode material is essential as an electron acceptor. For field applications, response time and detection range are important factors. These factors may be influenced by SMFC volume. Some researchers reported that maximum detection range (MDR) and sensing time (ST) are not influenced by SMFC volume, but some researchers proposed a relationship between normalized MDR (NMDR) and normalized ST (NST) to chamber volume (CV) as follows (Xu et al., 2021);

$$\text{NMDR} = \text{MDR}/\text{CV} \quad (1)$$

$$\text{NST} = \text{ST}/\text{CV} \quad (2)$$

Nevertheless, the voltage produced from soil can act as a signal as baseline voltage varies between rainy days. Previously, MFCs based biosensors were used for pollutants detection but these sensors could not detect the repeated pollutants before exoelectrogens were re-inoculated (Asai et al., 2017; Wang et al., 2020). These exoelectrogens are suitable only for the detection of non-toxic pollutants like BOD and COD (Schrader et al., 2016; Liu et al., 2020). The development of SMFC based biosensor for detecting repeated pollutants would be a breakthrough in the future. The sensing mechanisms based on bioanode, biocathode, and abiotic cathode in the SMFC are summarized below.

Bioanodic sensor: Bioanodic sensor element is highly studied due to the outward EET mechanism. As shown in Fig. 3(b), the metal ions enter the cell through specific transporters like extracellular polymeric substances for Pb^{2+} adsorption on the cell wall, sulphate transporters for Cr^{6+} , and phosphate-specific transporters for As^{5+} . Once the heavy metal ions intrude inside the cell their toxicity is reduced by different mechanisms following; (1) pumping out of metal ions inside the cell body by using reserve ATP (2) by conjugate formation (3) moving

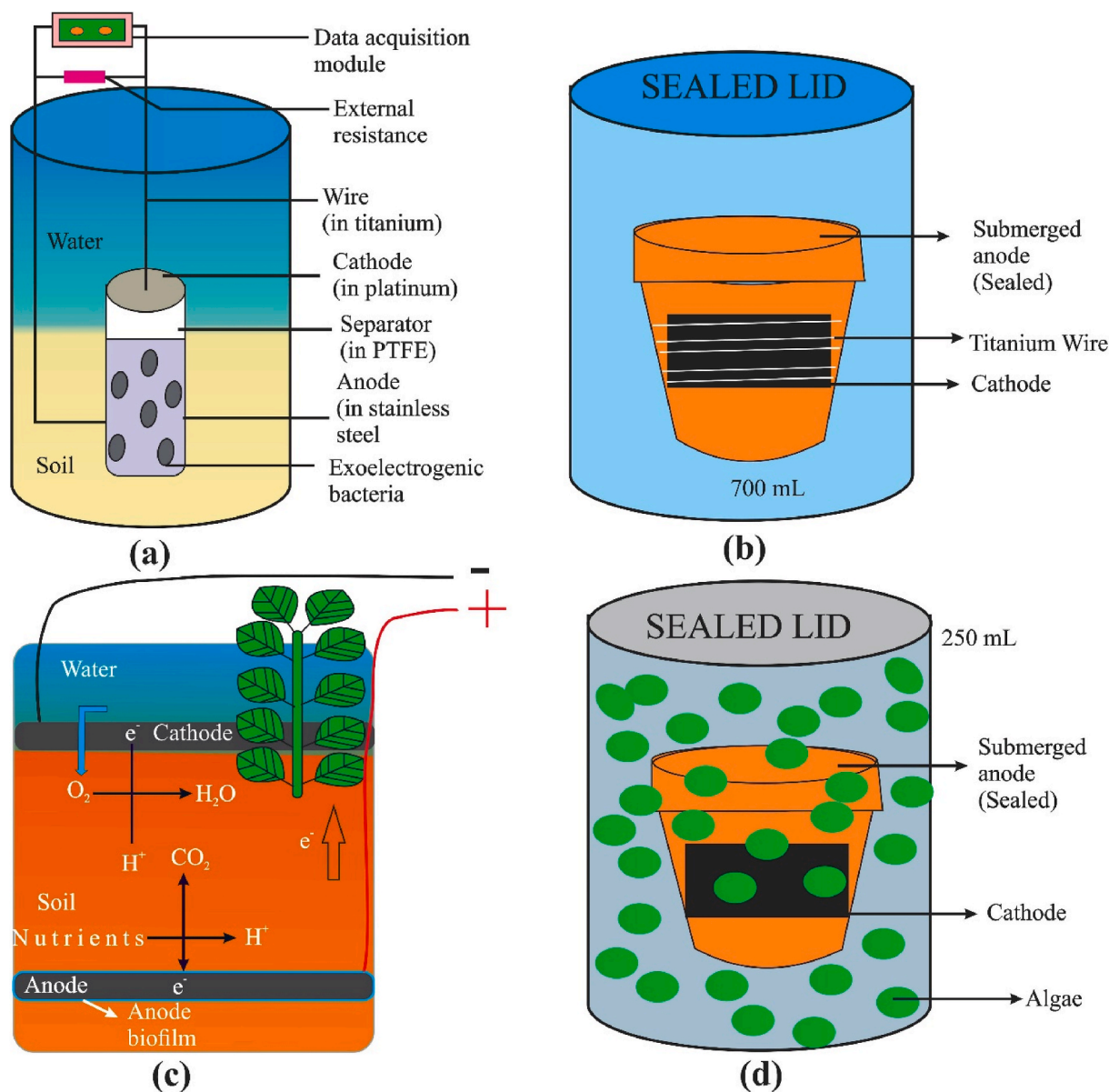


Fig. 2. Different prototypes of SMFC based biosensors. (a): sediment microbial fuel cell. Reprinted with permission from (Liu et al., 2020), Copyright (2020) Elsevier; (b): ceramic soil microbial fuel cell. Reprinted with permission from (Olias et al., 2020a), Copyright (2020) Elsevier; (c): plant-microbial fuel cell. Reprinted with permission from (Brunelli et al., 2016), Copyright (2016) University of Trento, Trento, Italy; (d): ceramic soil microbial fuel cell. Reprinted with permission from (Olias et al., 2020b), Copyright (2020) MDPI.

electrons flow toward specific reductase enzymes (4) metallothionein sequestration (Noori et al., 2021). Many researchers used bioanode to monitor different pollutants like antibiotics, heavy metals, and pesticides. Enriching EAB on the anode is very crucial, as some EAB require syntrophic partner to produce detectable electric signal from soil aromatic compounds. Some researchers reported, heterotrophic EAB primarily enrich in soil organic matter. So, the electrical signal produced by bioanode is directly proportional to organic matter concentration. The herbicide near bioanode enriched with non-specific enzymes like polyphenol oxidase, dehydrogenase, peroxidase, laccase, and urease. The synthesis of these enzymes needs energy which leads to non-detectable signals generation (Li et al., 2019). The soil humification also affects bioanode sensing ability (Li et al., 2018). The microbial biodiversity on bioanode is usually selective due to the enrichment of EAB. The main phyla at bioanode are *Chloroflexi* (12.83%), *Proteobacteria* (46.65%), and *Bacteroidetes* (12.48%), but *Proteobacteria* (46.65%) and *Bacteroidetes* are also present on bioanode under electrochemical condition

(Liang et al., 2021).

Qi et al. (2019) reported that detection time and bioanode sensitivity could be enhanced by enriched luminescent *Vibrio fischeri* for Cu(II) detection. This luminescent *Vibrio fischeri* could not generate the synchronous and optical signals in the absence of oxygen and organic matter. During soil respiration, the quantification of released electrons has been used to monitor soil microbial activity (Jin et al., 2020). SMFC multi-bioanode could be used to detect methane emission efflux (Wu et al., 2019) in sediment bulking (Wang and Jiang, 2019). The sensitivity of bioanode sensor is poor as compared to biocathode due to thickness of electroactive biofilm on bioanode increased with time which leads to less mass transfer. The sensing performance also varies with distance from the bioanode as the pollutants near bioanode are easily biodegraded through in-situ anaerobic process.

The bioanode employment for soil biotoxicity monitoring is normally restricted by low accuracy. To optimize the accuracy, bioanode and biocathode could be constructed together in the SMFC with a

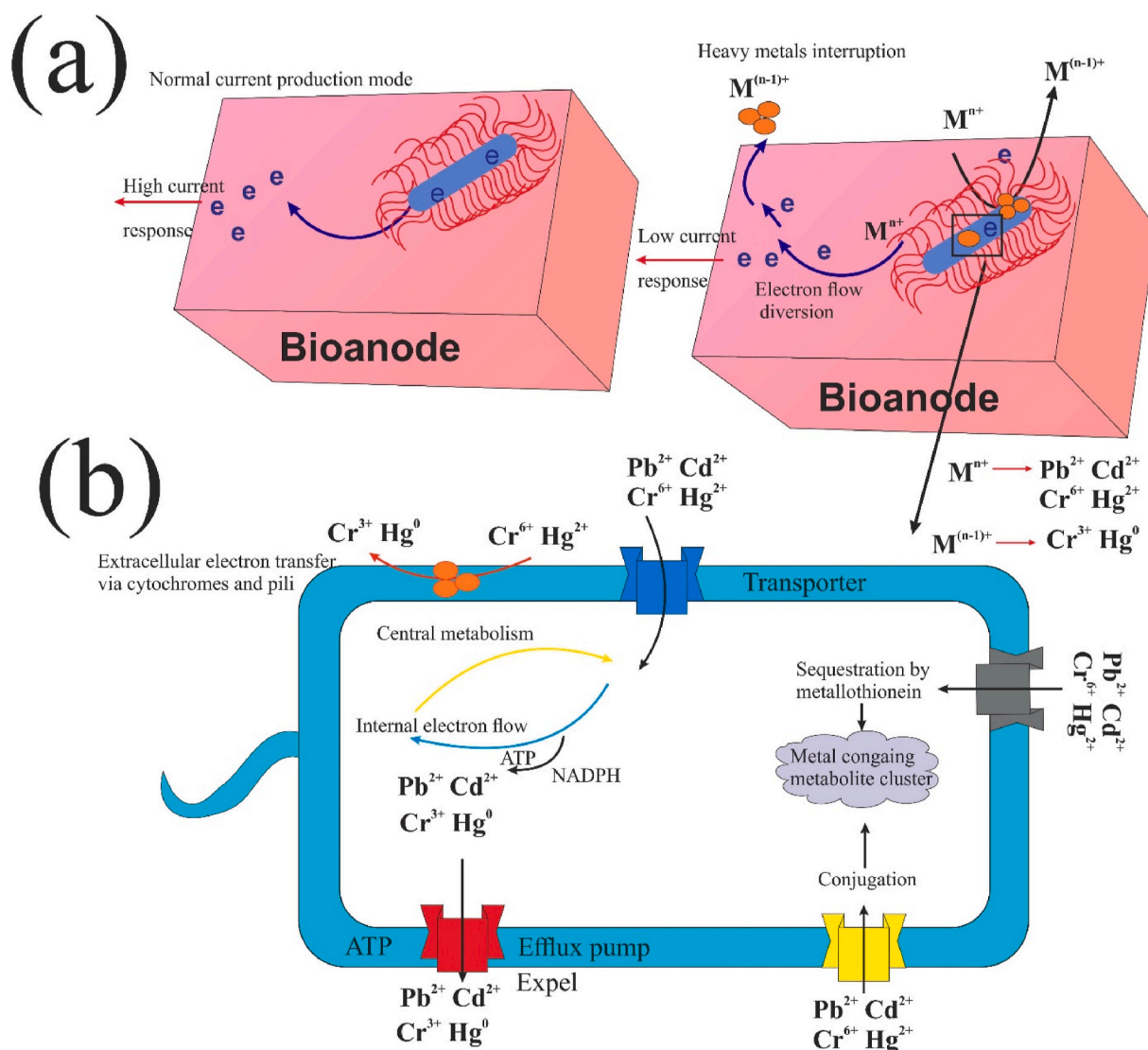


Fig. 3. (a) Extracellular electron transfer mechanism among electrogen and bioanode (b) interaction of metal ion with electrogens and their reduction mechanism. Reprinted with permission from (Noori et al., 2021), Copyright (2021) Elsevier.

bilateral passive oxygen supply. Bioanode could only detect the pollutants exposed to biofilm. However, the rest of the pollutants go undetected. In the future, research should be focused to detect all pollutants near bioanode. Furthermore, investigating the whole reaction mechanisms near bioanode will be helpful for the development of a portable SMFC bioanode.

Biocathodic sensor: biocathode is a self-renewable and organic electrofuels producer. Sometimes, cathode reactions limited by low mass and ohmic losses. So, biocathode with oxidizing bacteria is an excellent option to monitor the soil pollutants. The working mechanism of biocathode sensor based on autotrophic oxygen reduction reaction (ORR). So, the voltage produced by this biocathode could be used to monitor soil quality. Chen et al. (2012) designed a novel SMFC biocathode in the rice rhizosphere to deliver electrons to the root. In this study, the voltage was initially excelled but after one month decreased. Furthermore, root maturity was highly affected by oxygen efflux rate from rice roots which leads to development of potent biocathode from excreted oxygen from wetland plant roots. The soil pollutants primarily inhibit the inward EET in the biocathode attached biofilms. So, problems of errors and low sensitivity could be solved by using the biocathode sensor as an alternative to conventional bioanode sensors. The biocathode sensors are suitable for earlier detection of trace toxicants in the

soil and potentiostat-poised half cells as shown in Fig. 4. To reduce the competition between Cu (II) and O_2 as terminal electron acceptor (TEA) on the cathode, the biocathode is normally installed in SMFC. Different materials are used for biocathode to enhance the electro-migration of Cu (II) in the SMFC. The biocathodes are classified based on the TEA. The electrons mediators like iron and manganese are first abiotically reduced and then re-oxidized by microbe in the case of aerobic biocathode. While, the TEA in the case of anaerobic biocathodes, like sulphate and nitrate are directly reduced by accepting electrons from the cathode (He and Angenent, 2006). The warning signals of the biocathode sensor are highly affected by cathode potential and configuration. As earlier reported, the positive potential will boost the cathode current, and the negative potential will facilitate the biocathode start-up (Xia et al., 2012). The enrichment of electroactive autotrophs on the cathode is time-consuming because these autotrophs are slow growing. A biocathode sensor in the CSMFC was formed by algae to detect earlier eutrophication (Olias and Di Lorenzo, 2020).

These studies showed that biocathode sensors could provide new ways to monitor soil biotoxicity. However, some problems need to be resolved before scaling-up. For example, the parameters which affect the electroactive adhesion to the cathode need to be optimized. The interaction between cathodic electroactive microbes and pollutants should

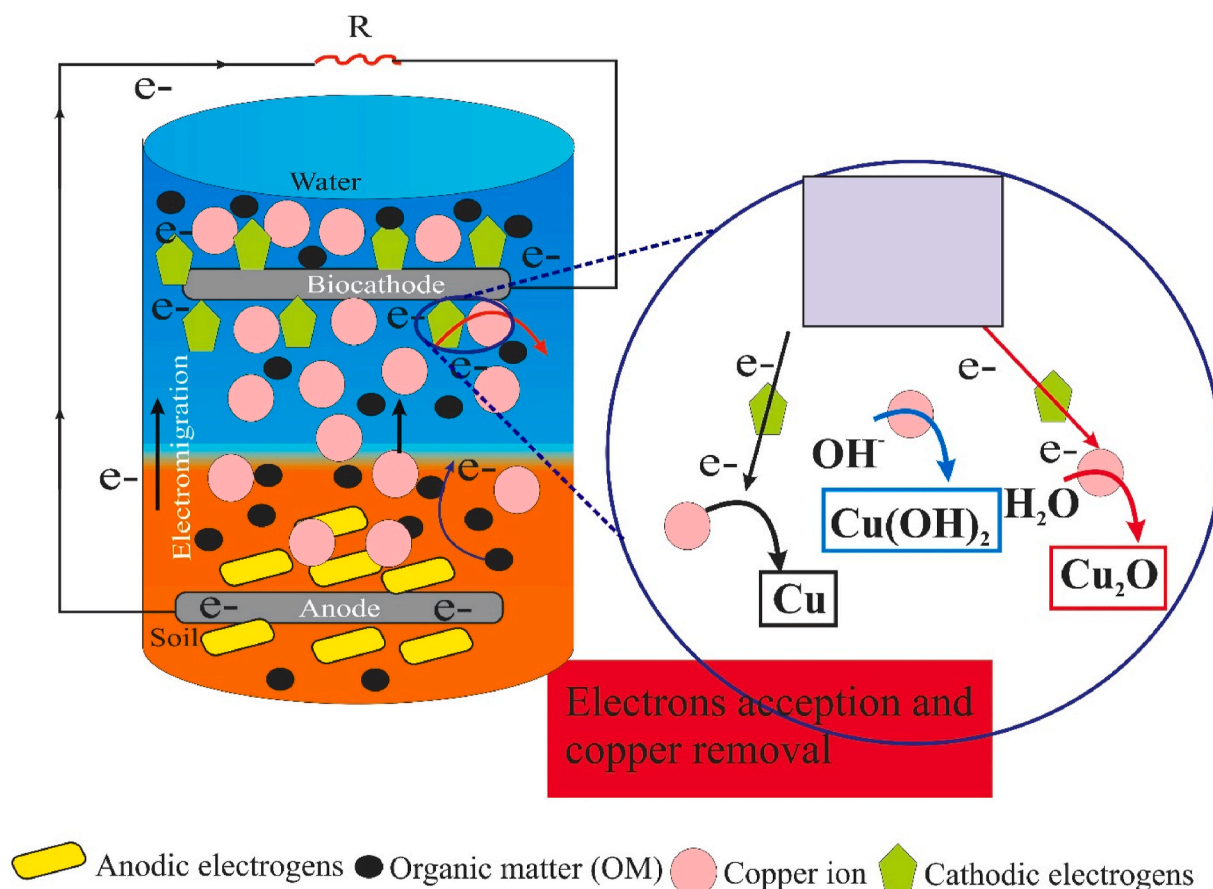


Fig. 4. The electrons transfer and copper detection mechanisms in the biocathode of SMFC. Reprinted with permission from (Liu et al., 2021), Copyright (2021) Elsevier.

be investigated for the construction of microbial electrochemical biosensors. The researchers should focus on the modification and exploration of a particular enzyme for biocathode configuration that can address limitations of low stability and short lifetime of enzymatic biofuels cells. The physico-chemical responses of cathodic bacteria under pollutants stress should be more investigated for the earlier detection of soil pollutants. In the future, laccase and bilirubin oxidase have been more attractive options for researchers to develop enzymatic biocathodes.

Abiotic cathode sensor: the abiotic cathode sensors have been employed in some SMFC as shown in Fig. 5. Its working principle mainly depends upon inhibiting the electrochemical reduction or ORR of the target cathodic analyte (Chu et al., 2021). The abiotic cathode normally lacks the SMFC potential difference caused by the organic matter-degrading bacteria at the cathode and the anode.

Recently, the performance of abiotic cathode sensor has been compared with biocathode sensor in terms of copper detection. The detection range of abiotic cathode is lower than biocathode because higher concentration of organic matter inhibits final electron acceptor due to more microbial oxygen consumption (Liu et al., 2021). Furthermore, some researchers reported, abiotic cathodes used iron, cobalt, and platinum as electron mediators for ORR. The multi-abiotic cathodes were also used to detect different heavy metals (Cr) and pollutants like H_2O_2 (Zhao et al., 2018; Liu et al., 2019). The disadvantage of abiotic cathode sensing element is that expensive catalysts like platinum or cobalt are coated on the abiotic cathode. These catalysts are also continuously replaced that ultimately increase maintenance cost. The advantage of an abiotic cathode sensor is that large cathodic surface area enhances the rate of the cathodic reactions. According to the previous literature review, abiotic cathode sensors are mostly used to detect

water quality. However, abiotic cathode has been rarely used to monitor the soil quality. The biocathode sensor is still vulnerable to toxic shock. This problem can be solved by using an abiotic cathodic sensing element as some heavy metals have high redox potential and could directly accept electrons from the cathode and produce considerable power signals (Gustave et al., 2019).

In the future, it will be challenging to use appropriate method for the monitoring of the target analyte by using an abiotic cathode sensing element. In the abiotic cathode, microbes are not participated, so more attention should be paid to enhance the electron transfer efficiency. The electrons mediators like platinum and cobalt are used in the abiotic cathode which are too expensive. So, in the future, more investigations should be focused on the development of low-cost biocatalyst materials like nanoparticles. Furthermore, an optimal SMFC design should be constructed that could easily run with newly developed abiotic cathode sensing element. The novel electrochemical methods could be used to enhance the sensitivity and selectivity of abiotic cathode sensing elements. The long-term running of abiotic cathode sensors is also challenging in the future because coated catalysts need to replace over time. So, more research should be directed to reduce the maintenance cost of the abiotic cathode sensor.

3. Effect of environmental factors on SMFC based biosensing

Many environmental factors could affect the performance of SMFC based biosensors like temperature, pH, and soil conductivity. Abbas et al. (2017) reported higher power density (129.6 mW/m^2) and toxic metals (cadmium, arsenic and lead) removal rate (79.03%, 66.22% and 71.17%) at 45°C in the aerated SMFC. They recorded the lower power density at thermophilic conditions and lower temperature. At 45°C ,

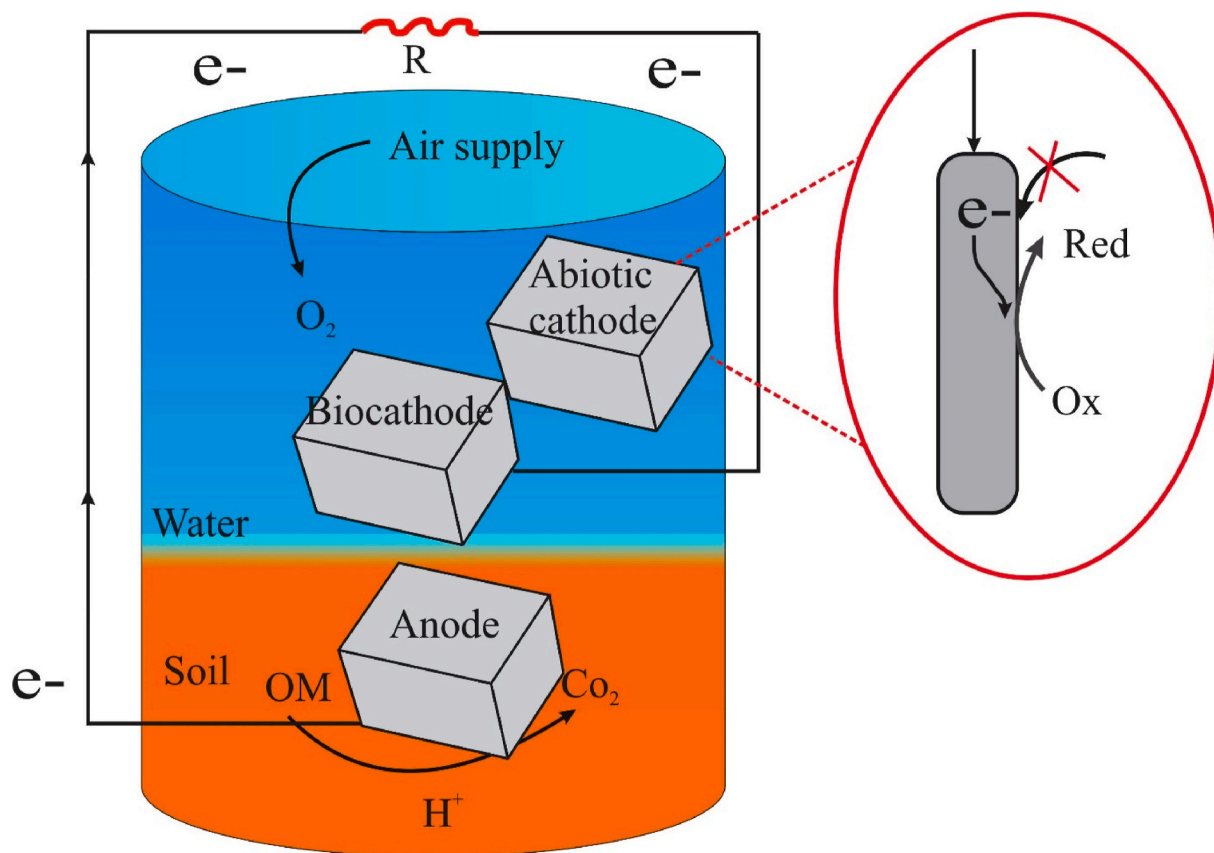


Fig. 5. Abiotic cathode sensing element for monitoring of soil biotoxicity.

higher toxic metal removal rate was observed due to higher conductivity of overlying water, better metabolic pathways, and ohmic reduction. As the temperature increase, the metabolic activities of exoelectrogens are affected by the denaturing of enzymes, except for some thermophilic bacteria which are still rarely reported as exoelectrogens. Some studies also reported 30–45°C temperature is required for better detection of toxic metals in the SMFC based biosensor (Yang and Chen, 2021). The current density varies from soil layers. So, if the anode could adjust in the deeper soil layer, a better response could be detected.

Regarding the influence of pH on the SMFC based biosensor, some researchers reported less power density at low pH due to the production of acidic products by fermentation and H^+ ions diffusion from cathode to anode. The pH difference in the cathode and anode chamber is negligible in aerated SMFC but important in non-aerated SMFC (Abbas et al., 2017). The alkaline pH is also reported as biofilm adhesion on the anode in the SMFC (Yuan et al., 2011).

Olias et al. (2020a) reported the higher voltage and DO at low pH due to favourable ORR and fewer hydroxyl ions (OH^-) formation. The pH was not evident in the non-catalyzed carbon due to the pre-dominant 2-electron reduction pathway. The soil buffering capacity may cause the delayed response and data uncertainty. Some exoelectrogens can tolerate acidic pH, but due to releasing e^- and H^+ ions affect substrate metabolism. The exoelectrogens susceptibility to methanogenic metabolism is due to methane formation by the combination of e^- and H^+ ions with CO_2 . Some studies also reported higher power density and biofilm formation at neutral pH due to suppression of methanogenic activity (Jin et al., 2003; García-Muñoz et al., 2011; Yuan et al., 2011). Regardless of salinity, SMFC biosensors perform well at mid-point salinity. However, at higher salinity, ohmic losses and conductivity increase, but osmotic imbalance may affects the anodic exoelectrogens (Miyahara et al., 2015).

4. Application of SMFC based biosensor

SMFC biosensor to monitor soil pollutants: The SMFC based biosensor could be employed to detect repeated heavy metals shock, monitor plants health and early detection of algal blooms (Brunelli et al., 2016; Olias et al., 2020b). These sensors are used to detect different volatile fatty acids species as concentrations of these fatty acids is directly proportional to power density generation. SMFC based biosensors could also be employed to differentiate fermentable substrates (starch and glucose) and non-fermentable substrates (butyrate and acetate) by investigating CV peaks (Yang et al., 2015).

SMFC biosensors could also be used to monitor oil spills in the natural water as shown in Fig. 6. The vertically arranged cathode in the SMFC could be used to monitor oil on surface water as oil on the cathode will inhibit the power generation. These sensors could monitor the excessive amount of organic matter and other secondary pollutants like NO_3^- , S^{2-} , Fe^{3+} and other substrates with higher redox potentials (Xu et al., 2021).

The effect of toxic pollutants on microbial activities could be monitored by these sensors. In MFC based sensors, the toxic pollutants inhibit the microbial activities around anode, which ultimately produce low power density but in the SMFC based sensor the effect of toxic pollutants on microbial activities is different as shown in Fig. 7. Many fast degrading organic pollutants are present in the natural sediments and overlying water. The anode in the SMFC is used as an alternative electron acceptor for the degradation of organic matter. Under anaerobic degradation, organic pollutants like carbon dioxide, sulphate, nitrate, and Fe(III) coupled with methane emission, sulphide, nitrite, and Fe(II). The organic matter degradation around the anode is normally enhanced by electroactive microbes like *Geobacter*, *Pseudomonas*, and *Desulfohalobus* (Yang and Chen, 2021). Wu et al. (2018) reported Cu^{2+} shock only inhibited the exoelectrogenic activity in the surface soil (0–3 cm)

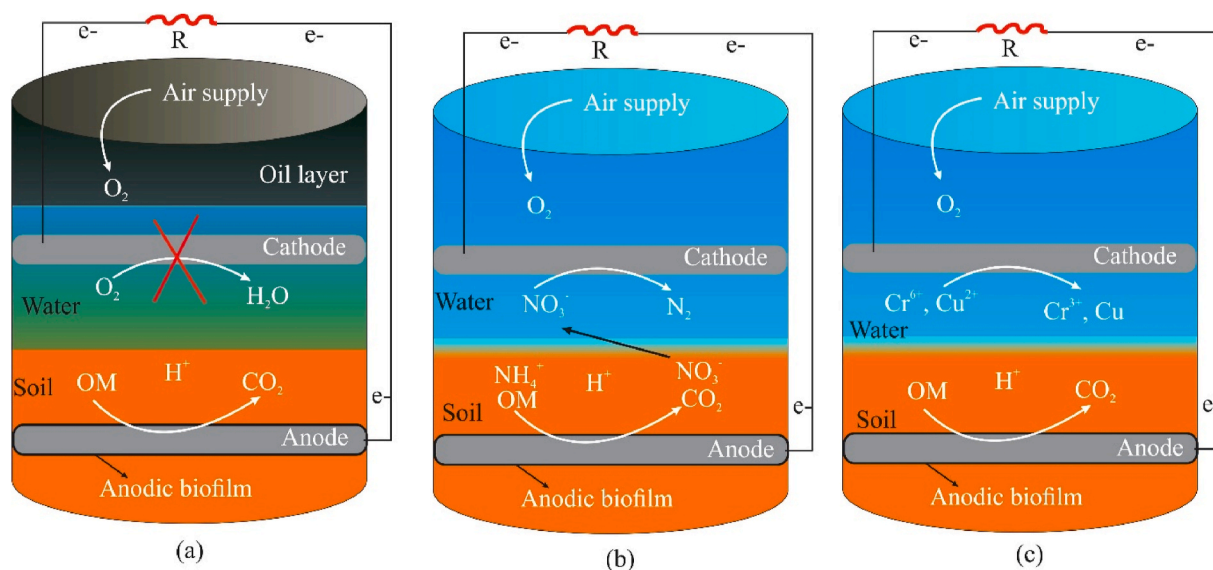


Fig. 6. Monitoring pathways of different pollutants by SMFC based biosensors; (a) oil (b) nitrate (c) copper and chromium.

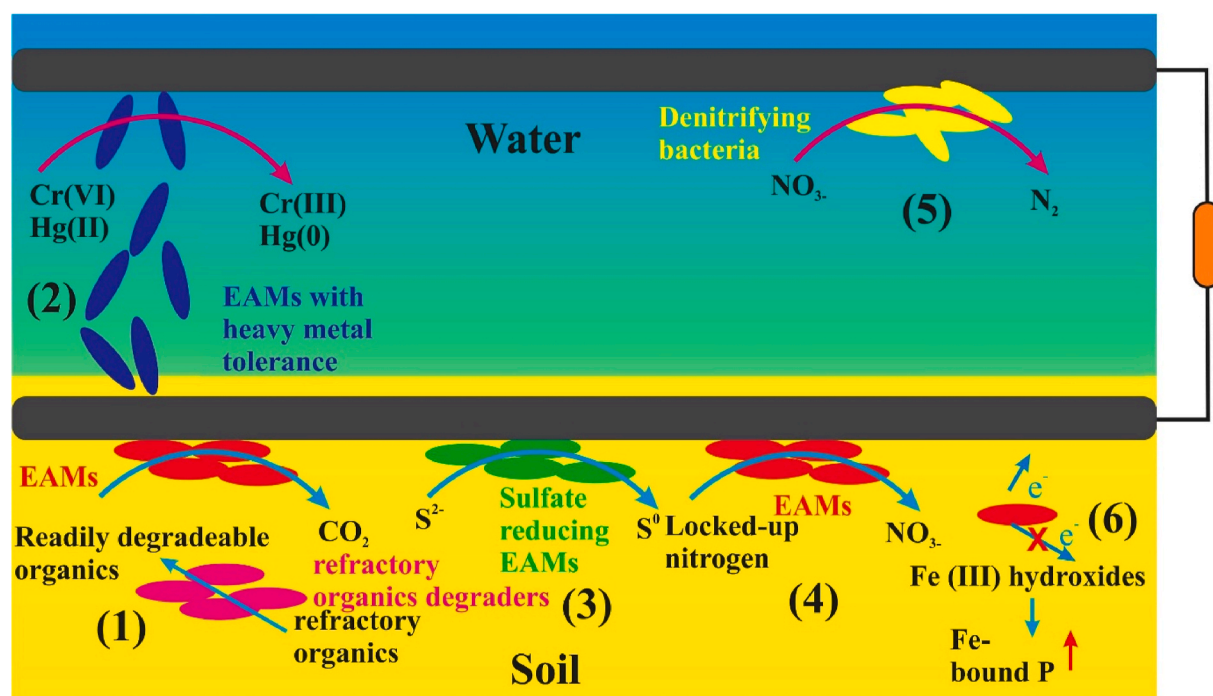


Fig. 7. The monitoring of microbial activities in the SMFC during pollutants degradation. Reprinted with permission from (Yang and Chen, 2021), Copyright (2021) Elsevier.

rather than in sub-surface soil (3–6 cm). The surface soil adsorbed sufficient Cu^{2+} and prevented leaking to the subsurface, so exoelectrogenic activity in the subsurface was not inhibited as compared to surface soil exoelectrogens. The Cu^{2+} concentration is directly proportional to voltage because the Cu^{2+} acted as the electron acceptor and also precipitated to $\text{Cu}(\text{OH})_2$ in the cathode chamber. The voltage baseline is normally stable in all seasons due to the less inhibitive effect of Cu^{2+} on the subsurface anaerobic exoelectrogens than surface exoelectrogens (Liu et al., 2020). Normally, voltage is measured in the SMFC based biosensor as digital signals to monitor the toxicants. The employment of coulombic efficiency, integral power, and colorimetric monitoring could be more suitable to enhance the range of toxicants detection. These parameters could be easily monitored by cell phone but are limited by

low sensitivity. The micro spacing electrodes would also be more suitable option to monitor the toxicants. The overall bottlenecks to implementing the SMFC at the pilot scale are providing a passive and steady supply of organic matter to the anode, decoupling of signalling with toxicants shocks, and stabilizing the baseline concerning environmental conditions. These issues could be resolved by providing a solid analyte for slow release and long-lasting source of electrons donors.

Soil quality monitoring: These cost-effective biosensors could be used for the online monitoring of DO in the different depths of the lake without extra servicing like substrate supplementation or cleaning the electrodes. The SMFC based biosensors quickly respond to DO concentration in the lake so they could also be used to alarm the water pollution like dead zones. These biosensors could easily integrated with

wastewater and paddy soil bio-sensing and real-time monitoring because they quickly adapt to different environments (Song et al., 2019). This integrated technology could also be used for DO monitoring in the different soil depths after analysing the effect of different factors like short start-up time and electrode distance. SMFC based biosensors selectivity could be enhanced by synchronizing the SMFC sensor in a multisensory platform with conductivity probes, DO, pH, temperature, and using biocathodes/bioanodes as bioreceptors. Furthermore, more research should be focused on the shortening of stable voltage generation which ultimately will lead to the field application.

SMFC as power station: SMFC based biosensors were used to charge the high capacity ultracapacitor, which powered wireless temperature sensors. This highcapacity ultracapacitor could record three times more data than power management systems (PMS) (Zhang et al., 2011). The SMFC based biosensor could efficiently supply the power (10 mW–2.5 mW) to the underwater sensors like electro-chemical, optical, barometric, acoustic, thermocouple, and piezoelectric sensors which are used for different purposes like oceanography, seismic monitoring, military surveillance, habitat and environmental monitoring as shown in Fig. 8. These SMFC could supply the power to the underwater ultrasonic sensors and underground PMS without reserve charged batteries (Donovan et al., 2013). The PMS could track the time monitoring by using a real-time clock. The longdistance data transmission in the PMS needs to be long-term stability. The long-term stability under the real environmental conditions could be further investigated. The stacked SMFC is also used to power the water treatment reactor. This field experiment was carried out in Brazil. These stacked SMFCs showed stable performance over many months and maintained soil contents up to 50%. This integrated SMFC system could be employed in remote areas and could be an effective way for practical employment and scalability

of stacked SMFC for power production and self-powered water purification (Dziegielowski et al., 2020). A portable SMFC could be used to monitor water contents in the soil (Nguyen and Taguchi, 2021). This portable SMFC with low-cost materials implemented in the natural agricultural soil to monitor the humidity for many crops in Japan as compared to conventional methods. To minimize the errors in early warning signals, decision algorithms with signal treatments should be improved. Equally, a holistic pathway to calibrate the sensor (i.e. DoE) is recommended to monitor the influence of different environmental factors on the signal output.

5. Future perspectives

Output signal stability is one of the biggest challenges for long-term monitoring with SMFC based biosensors. The output signal of the SMFC would be largely affected by the electrode fouling and environmental factors fluctuation. Therefore, development of inexpensive electrode materials that resist fouling could be very crucial for future applications to meet long-term (month/years) monitoring requirements. Exploration and design of new methods and strategies to construct SMFC that could be robust for operating under different environments are also urgent tasks for the future development of SMFC based biosensors. Improvement of the analytical performance is always a key topic for SMFC based biosensors. Currently, the detection limit of SMFC based biosensors still can't meet requirement for the soil quality monitoring. The detection limit of SMFC based biosensors is normally limited by the mass-transfer barrier in soil and the sensitivity of the biofilm. To enhance SMFC based biosensor detection limit and reliability, systematic investigations to improve the analyte availability in soil and enhance the bio-sensitivity of the biofilm are indispensable. Meanwhile, the electrode material,

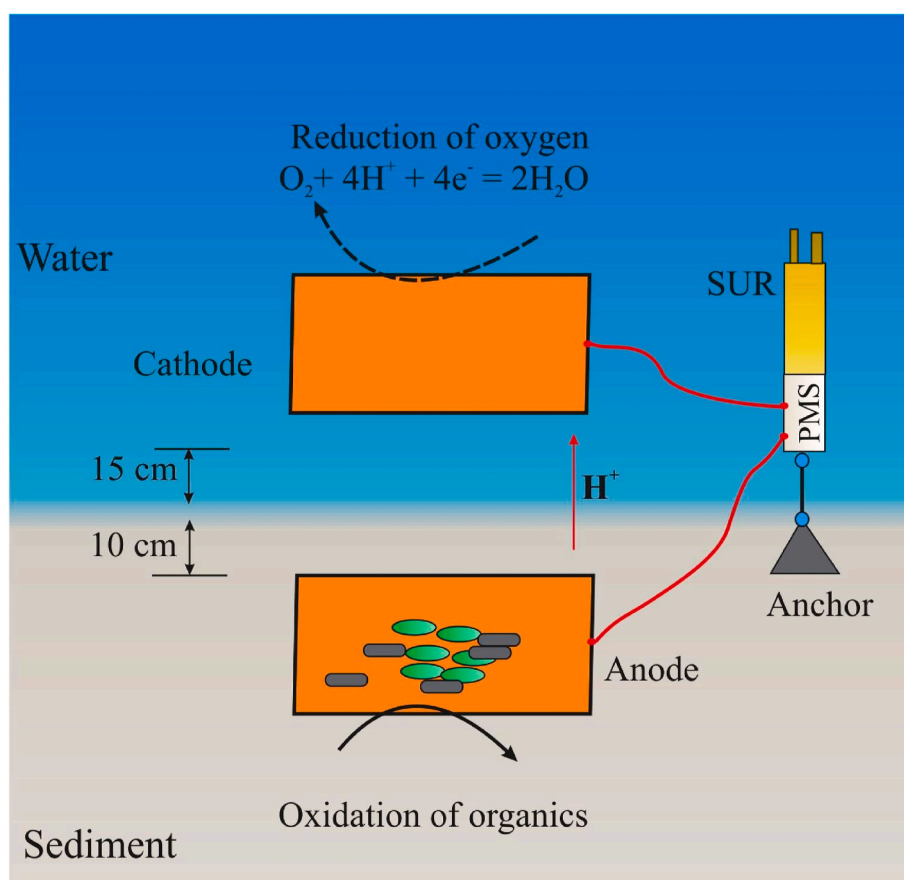


Fig. 8. Schematic diagram of the sediment microbial fuel cell connected to the power management systems (PMS) and the submersible ultrasonic receiver (SUR). Reprinted with permission from (Donovan et al., 2013), Copyright (2013) Elsevier.

configuration, catalysts and sensing methods should also be taken into account to optimize the analytical performance of the SMFC based biosensors. In addition, to meet the requirement of the real applications, more analysis targets should be considered and multiplexed biosensors are expected to be established in the near future.

Furthermore, in-field test and scaling-up are also urgent assignments for practical application of SMFC. Currently, most of SMFC based biosensors operate under laboratory conditions. But, in the field application, many parameters like pH, temperature, oxygen concentrations, ionic strength etc. affect their performance. So, SMFC based biosensors should be further tested under real conditions and in-field evaluation is required before practical application. In addition, scaling-up is another big challenge to SMFC. Development and operating the SMFC with real application scale should be also considered in the future. Overall, practical application oriented designing and optimizing the SMFC based biosensors should be the right way for the next step to move forward to the commercial applications.

6. Conclusions

The development of SMFC biosensors contributes to the field of biosensors and attracts the attention of researchers from all over the world. The SMFC biosensors are cost-effective and could be used for in-situ monitoring of BOD, COD, and toxic metals in the soil. The SMFC based biosensors could detect different pollutants in the soil, but their selectivity, sensitivity, repeatability, and stability should be improved. Future research should focus on the cost-effective and eco-friendly development of fabricated electrode material employed in the SMFC based biosensors for better performance for industrial applications. Furthermore, more efforts should be directed toward the development of self-sustainable and wireless SMFC for distant areas with poor infrastructure.

Author contributions statement

Syed Zaghum Abbas: Literature survey, analysis and drafting the paper. **Jia-Yi Wang:** Conceptualization and methodology. **Hongcheng Wang:** Validation, resources and project administration. **Jing-Xian Wang:** Formal analysis, resources and project administration. **Yi-Ting Wang:** Visualization and software investigation. **Yang-Chun Yong:** Supervision, conceptualization, revising and editing the paper.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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