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Chapter

Expedited Biodegradation of Organic Pollutants and Refractory Compounds Using Bio-Electrochemical Systems

Eustace Fernando, Godfrey Kyazze, Ahmed Ahsan and Pavithra Fernando

Abstract

Biodegradation of xenobiotics is often considered to be a slow process. This is especially true if the xenobiotic in question is polymeric in nature, contains many chemical substituent groups or generally exhibits high level of toxicity to environmental microbiota. Due to this observed slow kinetics of degradation, removal of many xenobiotics from contaminated environments using conventional bioremediation technologies is a difficult problem. To alleviate this, alternative technologies showing improved kinetics of biodegradation are sought by the scientific community. One such promising approach is the usage of the novel technology of bio-electrochemical systems for improved degradation of xenobiotics. Due to the newness of this technology and affiliated methods, not much information about its usage for biodegradation of xenobiotics is available in literature. Therefore, this chapter aims to address that gap and bring about a comprehensive analysis on the usage of bio-electrochemical systems for rapid removal of xenobiotic contaminants from the environment.

Keywords: bio-electrochemical systems, bioremediation, biodegradation, pollutants, xenobiotics

1. Introduction

Bioelectrochemical systems (BES) are devices that drive electrochemical reactions using biological agents as catalysts. Primarily, there are two types of BES. They are microbial fuel cells (MFCs) and Microbial electrolysis cells (MECs). Both depend on two electrodes where anodic and cathodic electrochemical reactions occur and both types are driven by microorganisms. In MFC systems, organic substrates, most often organic pollutants are microbially broken down at the anodic compartment. The electrons released are harnessed by the anode electrode and are driven towards an external circuit where it can produce a usable current. These electrons are then accepted by a cathode electrode where it is coupled with protons and an electron acceptor such as molecular oxygen to produce water (**Figure 1a**). MFCs contain a proton exchange membrane to exchange protons between anode

Figure 1.

Schematic diagrams showing the operating principles of (a) MFC and (b) MEC systems (modified figure adapted from [1], with permission).

and cathode chambers. On the contrary, MEC systems rely on a small exogenous supply of electrons to the cathode or anode electrode to produce combustible gases hydrogen or methane. It can therefore be termed as an MFC system run in reverse to produce gases with fuel value or other value added products. Both types of BES come in many different designs, shapes and sizes [2–4]. Out of the two BES types, MFCs are the most ubiquitously used type for laboratory and pilot scale studies and full-scale operation.

The primary utility of MFCs is to be used as a wastewater treatment technology and simultaneous recovery of electrical energy as a fringe advantage. However, the amount of recoverable electricity still remains low for large-scale practical applications of MFCs as electricity generation units. Until a research breakthrough in MFC electrical power generation is made, the focus therefore, firmly lies within wastewater and waste treatment capabilities of MFCs. One of the main benefits of MFCs in this regard is that they are demonstrated to possess better degradation kinetics for biodegradation of many recalcitrant organic pollutant types over their conventional counterpart systems such as activated sludge systems, anaerobic sludge blankets and constructed wetland systems [5–7]. Some of the refractory organic pollutants such as azo dyes and nitrophenolic compounds are known to be problematic due to their poor or non-degradable nature in conventional wastewater treatment systems such as activated sludge systems. These compounds are therefore known as highly recalcitrant organic pollutants. Earlier studies that use of MFCs however, have demonstrated that some of these compounds can be effectively degraded, transformed or mineralized into simpler intermediates with the use of BES. This chapter examines the utility of such BES systems for enhanced degradation and removal of such recalcitrant environmental pollutants.

2. Removal of azo dyes in BES

Azo dyes by far, are the most widely studied class of environmental pollutant remediated by electro active microorganisms. Azo dyes are characterized by one or more of the azo moieties, which are of oxidative nature. Therefore, the most obvious conversion mechanism of azo dyes by electro active microorganisms is by the azo pollutant acting as an electron acceptor and undergoing reduction into their constituent amines. Azo moieties are flanked by R groups which may contain various electron-rich or electron-poor substituent groups, which would influence the redox potential of the dye [8]. In recent years, Microbial Fuel Cell (MFC) technology has been explored extensively for their innovative features and environmental benefits [9]. At the anode, organic co-substrate is oxidized by electrochemically active microorganisms. Subsequently, the microorganisms transfer the electrons

Table 1.

BES studies involving azo dyes and the bioelectrochemical characteristics of the BES systems during azo dye remediation.

resulting from this oxidation to the anode via extracellular electron transfer which then passes through an external circuit to the cathode, thus producing current. Protons migrate through an ion exchange membrane to the cathode where they combine with azo dye and electrons and lead to the degradation of azo bond. Reduction of azo bond results in the formation of colorless and biodegradable aromatic amines [10]. It has been demonstrated in earlier studies that BES can be

BES type	Matrix	Pollutant	Microorganisms	or cathodic surface area (measured in $mW/m2$) normalized by anodic Increase in maximal Power density	or cathodic surface area normalized by anodic (measured in $mA/m2$) Increase in Maximal Current density	normalized by anodic or cathodic surface area (measured in Maximal Voltage mV	References
MFC	Synthetic Medium	phenanthrene Benzene,	P. aeruginosa, S. oneidensis and Undefined cultures	1.25	$\frac{8}{2}$	65	$[28]$
MFC	waste water Domestic	Petroleum Sludge Real Field	Bacteria in Anaerobic sludge	20.6			$[26]$
SMFC	Saline Soil	TPHs, n-alkanes & PAHs	Polluted soil containing a microbial consortium		299		$[29]$
SMFC	Saline Soil	C8-C 40 n-alkanes, 16 PAHs	Petroleum contaminated soil containing Geobacteraceae sp. and Escherichia sp.	37	102	366	$[30]$
MFC	Soil	C8-C40 n-alkanes TPHs, 16 PAHs,	Microorganisms in polluted soil				$[31]$
MFC/MFC + anoxic cathode	wastewater	wastewater (BOD Petrochemical 5/COD: 0.36).	consortium from effluent treatment aerobic consortium & anaerobic plants		14.3 mW/m ² (aerobic) 17.1 mW/m ² (anoxic),	OCV: 248 and 280 mV	$[32]$
SMFC	Sediment	PAHs	Caldisericum sp. & other sediment bacteria			$0.4 - 0.6$ V	$[33]$
SMFC	Sediment	Benzo(a)pyrene	Aerobic genera (Vogesella, Pseudomonas, anaerobic genera (Longilinea, Bellilinea, Flavobacterium and Rhizobium) and Desulfobacca and Anaeromyxobacter)		$\overline{17}$	$61 - 65$ mV,	$[34]$

Table 2.
BES studies involving PAHs and the bioelectrochemical characteristics of the BES systems during PAH remediation. *BES studies involving PAHs and the bioelectrochemical characteristics of the BES systems during PAH remediation.*

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successfully coupled to other wastewater treatment technologies such as activated sludge systems and up-flow anaerobic sludge blanket reactors (UASBs) in order to fully mineralize azo dye pollutants [6, 11]. The use of BES has shown higher kinetic rates of azo dye biotransformation rates compared to other conventional treatment methods [12, 13]. Many other studies have hitherto demonstrated this ability of BES to effectively biodegrade many different azo dyes (**Table 1**).

3. Removal of polycyclic aromatic hydrocarbons (PAHs) in BES

Incomplete combustion of organic matter and fuels leads to creation of PAHs. Additionally, various industrial processes use large amounts of PAH compounds such and naphthalene, phenanthrene and chrysene. These PAHs are degraded into compounds which are less toxic in nature. This degradation takes place while these PAHs are being used as a source of carbon and energy in solid or liquid media. However, there is no evidence for the existence of a single microorganism species which can work on PAHs of several types. This is the reason why using microbial consortia can be very useful. Usage of microbial consortium is advantageous because, they are cheap, show higher applicability and produce lower negative environmental impacts [23].

Degradation activities of PAHs are extremely long, taking up to years or at least months [24]. All PAHs are "fat-loving" or "water-hating chemicals [25]. They become less available for biodegradation in water, as they have a tendency to adsorb to organic molecules. Even though various bioremediation techniques are used to treat PAHs in polluted environments, they have limitations due to the negative effect caused by activity and diversity of indigenous hydrocarbon degrading bacteria, low abundance, slow growth rates, less availability in aqueous solutions among others. Further problems can also be created due to addition of co-substrates and nutrients to improve indigenous microbial activity due to high cost & and the fact that the added chemicals having the ability to act upon anything other than the target compounds. Hence, using Microbial fuel cells remains as a viable solution circumvent some of these problems [24].

Less complex compounds can be formed from recalcitrant PAHs, if bio electrochemical activity can increase the metabolism of PAH degraders closer to the electrodes [26]. When a microbial fuel cell with a single chambered air-cathode working upon a Winogradsky solution was used, detoxification activity by microflora of a specialized variety growing on PAHs in that Winogradsky solution was found. Microbial Fuel Cells not only shows improved degradation activities, but it also show higher amount of microbial detoxification activity [23]. Inocula extracted from complex microbial communities such as soil or sediments to drive electrochemical reactions in BES and to degrade xenobiotic pollutants may confer other benefits such as simultaneous nitrogen and phosphorus removal from wastewater [27]. Many previous studies have demonstrated that various BES types employing many different electrochemically active microorganisms are effective in biodegradation of a range of PAH pollutants (**Table 2**).

4. Removal of pesticides and their residues using BES

Pesticides are defined as substances or mixtures designed for destroying and mitigating any group of pests such as insects and plants. During the past decades, different types of pesticides had widely been used in agriculture for high yield productions. Applications of pesticides have secured almost one-third of crop

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production in the world. Pesticides have led to the improvement of food production to secure the demands of an ever increasing human population [35]. Majority of pesticides and their residues can cause unintended splash damage to other organisms than the target pest. The widespread use of pesticides not only brought adverse influence on agro ecosystems but also caused alteration in physiological processes of non-target organisms [36].

The over and misuse of pesticide has precedence to immense health problems, economic loss and various environmental problems. The resultant health problems of pesticides include cancer, birth defects, reproductive problems, liver, kidney, and neural problems among others. In many developing countries majority of pesticides are associated with adverse effect on human health and environment due to the overuse of pesticide. On the other hand the overuse of pesticide also results in the environmental pollution such as water and soil pollution and cause imbalance of ecosystem [37].

Presently, the soil bioelectrochemical remediation system (BERS) can effectively remove pesticides, e.g., hexachlorobenzene [38], isoproturon [39] and Metalachlor [40] from soils by means of native functional microorganisms, with the advantage of the production of bioelectricity. This new technology has been paid more attention by the researchers and engineers in the field of environmental management. In the soil BERS, microorganisms is in charge of system performance since degraders and electroactive bacteria were responsible for pollutant degradation and electricity generation. Thus, the insights to the structure and evolution of microbial community in the remediation process of soil BERS is the most important for revealing the mechanism of microbial electrochemical degradation [41]. **Table 3** summarizes a range of different studies that have successfully utilized BES to bio-transform or biodegrade a variety of pesticides.

5. Removal of polychlorinated biphenyls (PCBs) using BES

Polychlorinated biphenyls (PCBs) are among the persistent organic pollutants which mainly accumulate in soils and sediments [46]. These lipophilic compounds are of major concern due to their low biodegradation, high bio-concentration and persistence in environment [47]. Polychlorinated biphenyls (PCBs) is a kind of light yellow or deep yellow oily liquid with the properties of insulating ability, thermal stability, resistance to acids, oxidation, hydrolysis, and flame resistance. Due to these unique physical and chemical properties, PCBs were widely used in many products especially in transformers and power capacitors [48]. Polychlorinated biphenyls are a subset of the synthetic organic chemicals known as polychlorinated hydrocarbons. They are composed of two attached benzene rings and multiple bonded chlorine atoms. PCBs consist of 209 isomers and congeners, with a 10,000 fold range in n-octanol/water partition coefficient (Kow, indicator of lipophilicity and then, of potential bioaccumulation) values between the mono-substituted chlorobiphenyl and the fully-substituted decachlorobiphenyl [49].

It is known that PCBs induce a wide variety of toxic responses in human, wildlife, plants and laboratory animals. They can penetrate the human body through skin contact, by inhalation of PCBs contaminated vapors and by consuming food contaminated with PCB residues [50]. Toxicological studies of PCBs in human were found to increase the rate of melanoma, gall bladder cancer, brain cancer, liver cancer, biliary tract cancer, gastrointestinal tract cancer and possibly connected to breast cancer. The people exposed to high levels of PCBs through skin contact or by consumption experience skin irritations like severe acne and rashes, nose and lungs infections and eye issues [51]. The uptake of PCBs and their negative

effects on plants has also been examined. For instance, PCBs firmly bind to the soil organic matter and provides low bioavailability for plants and microorganisms. But accumulation of PCBs in soil is perilous to all kinds of organisms including plants. The higher PCB concentrations affect the biosynthesis, ultrastructure of plant cell, membrane stability and plant DNA. Also, the plants that are highly exposed to PCBs result in reduction of photosynthesis, water/nutrients uptake and show visible symptoms of growth inhibition, browning of root tips and even death [52].

Anaerobic biodegradation is a major process for PCBs removal in sediments. However, a lack of or deficiency in terminal electron acceptors such as sulfate and nitrate can result in a decreased removal rate of bioremediation or no biodegradation at all [53]. Therefore, bio-stimulation by introducing electron acceptors could improve the microbiological activity in sediments [54].

Sediment microbial fuel cells (SMFCs), as a bio-electrochemical system, have been demonstrated to enhance the biodegradation of organic compounds, i.e. PCB. Xu et al. found that the combined application of a sediment microbial fuel cell (SMFC) and surfactant led to the highest removal efficiencies (43.26% of PCBs) after 60 days of operation, and produced the maximum power output (0.821 V of voltage and 18.30 W m−3 of power density) [55]. The degradation of polychlorinated biphenyls (PCBs) by sediment microbial fuel cell (SMFC) with/without nano-scale zero-valent iron (NZVI) addition was investigated by [56]. It was found that the combined application led to the highest removal efficiencies of PCBs $(37.55 \pm 1.11\%)$ and TOC $(49.72 \pm 1.54\%)$ in all circumstances and produced a higher power density (108.89 mW/m²) [56]

6. Removal of cellulose using BES

Cellulose is a β-1,4-linkaged polymer of Glucose. Cellulosic biomass, usually obtained from agricultural and industrial activities, can be considered as one of the highly available sources of renewable energy on earth. Cellulosic materials can be used in a process of sustainable energy production by transforming them to H_2 , ethanol, and methane. But due to economic and technical problems, it remains difficult to produce such products [57].

One of the main major downsides of Cellulose is that it's a moderately recalcitrant compound. It can be converted to different types of fuels such as bioethanol, methane and Hydrogen, but the energy recovery and treatment in waste water is difficult because of its low density in terms of energy and highly recalcitrant nature [58]. Another problem is that, cellulose is adhered with lignin or/and Hemicellulose. Lignin may cause problems by not allowing cellulases to digest cellulose material due to this tight adherence. However microbial fuel cells (MFCs) can be used to convert cellulosic materials to Electrical energy and it has been found to be an effective technology which can be used as an alternative. The microorganism or microorganisms must be able to breakdown cellulose anaerobically to sugar monomers and they should also be electrochemically active in order to directly convert liberated sugars to electricity in a Microbial Fuel Cell. Here, they should use an anode as an alternative electron acceptor. Metabolites of cellulose hydrolysis are oxidized in this manner to produce direct electrical energy [57].

Anaerobic microorganisms of facultative varieties can be found in the rumen. Through fermentation or anaerobic respiration, they hydrolyze cellulose while conserving energy significantly. Hence, Rumen microbial consortium can be used in Microbial Fuel Cells to generate electricity from cellulose [57]. Cellulose is different from other organic materials because, it needs this consortium to metabolize it [58]. When the Rumen consortia was used in Microbial Fuel cells, it showed the

Table 4.

BES studies involving cellulose and the bioelectrochemical characteristics of the BES systems during cellulose remediation.

presence of respiratory anaerobes & hydrolytic enzymes which involved in cellulose hydrolysis. Phylogenetic analysis was also used to understand this relationship [57].

Clostridium cellulolyticum and *Geobacter sulfurreducens* were used in one study to demonstrate the production of electricity in Microbial Fuel cells which used particulate MN301 Cellulose. Fluorescent *in situ* hybridization and quantitative PCR showed that most *Clostridium* cells were attached to cellulose which was there as a suspension. In the meantime, *Geobacter* cells were attached to the electrode. However, in contrast, use of soluble carboxymethyl cellulose caused bacteria to remain as suspension and biofilm. This explains that it is possible to improve electricity conversion from solution by decanting supernatant and settling Cellulose, as Cellulose hydrolysis can be improved by optimizing the reactor operations. Cellulose transformation performance can be increased with further research. The performance of the reactor with increasing biocatalyst density should be further analyzed. Hence, the effectiveness of decant and settling at cellulolytic density of higher concentration is one part which needs further focus to identify cellulose conversion extent [58]. Several previous studies have demonstrated the effectiveness of various BES types to biodegrade and convert the energy content of cellulose and its derivatives into electrical energy (**Table 4**).

7. Removal of lignin its derivatives in BES

Lignin is a phenolic polymer which is heterogenous in nature. It is made of renewable source of aromatic chemicals. Main components of Lignin are three alcohols namely phydroxyphenyl guaiacyl and syringyl. Different types of lignin in different plant components like grass, softwood and hardwood contain different amounts of methoxy groups depending on the degree of these three types of alcohols present in them. Microbial Fuel Cell (MFC) is a type of biological decomposition method which can be used to degrade lignin efficiently [59]. Microbial

fuel cells are devices which produce electricity from catalysis of microorganisms by using chemical energy as their energy source [60]. They contain a cathode chamber which is aerobic and an anode chamber which is anaerobic. Both of these chambers are connected either by a salt bridge or a membrane where the protons can be exchanged. In the anode chamber, microorganisms like fermenting bacteria produce smaller fermentation products from larger organic molecules. $CO₂$, protons, and electrons are produced by oxidation using anaerobic bacteria afterwards. The circuit is completed by electrons passing to the anode interface which are then transferred to the cathode through an external wire. The cathode reduces the oxygen molecules to either form water or H_2O_2 . Here, H_2O_2 is produced by a two-step reaction consisting of a two-electron reaction [59]. The lignin undergoes hydrogen peroxide mediated oxidative depolymerization when introduced to the cathode. In aerobic, low pH conditions this oxidative reaction will proceed better as oxygen is required for H_2O_2 production. An effective method for H_2O_2 production has already been observed recently via a microbial electrochemical cell [61].

8. Removal of micropollutants using BES

Micropollutants are natural or anthropogenic residue organic refractory compounds such as pharmaceutical compounds and their metabolic intermediates, antibiotics, personal care products, pesticides and industrial chemicals. These micropollutant compounds are present in polluted water in sub-microgram per liter levels [62]. In addition to their refractory nature, their presence such low concentrations in affected environments pose additional challenges for conventional biodegradation processes. Furthermore, some micropollutants such as endocrine disrupting chemicals (i.e. estradiols and their derivatives, antibiotic residues) can exert their undesirable effects on the receiving water bodies even at sub-microgram per liter concentrations [62]. Presence of antibiotics and their residues in natural waterways also promotes the development of undesired antibiotic resistance mechanisms

Table 5.

The utility of BES and affiliated hybrid systems for removal of micropollutants.

within bacteria found in natural aquatic ecosystems. It has been established in multiple studies that such micropollutants can easily pass through conventional wastewater treatment systems such as activated sludge systems, anaerobic digester systems and membrane bioreactors completely unchanged, back into the consumers of recycled water [63]. It is therefore clear that novel technologies are required to tackle such emerging pollutants such as various types of micropollutants.

The removal of some micropollutants has been demonstrated using BES in several previous studies. In a study conducted by [64], it was demonstrated that constructed wetland-MEC hybrid system operated at pilot-scale could successfully remove several micropollutants individually and their mixtures. The removal of four pharmaceutical compounds carbamazepine (CBZ), diclofenac (DCF), ibuprofen (IBU) and naproxen (NPX) were monitored in a constructed wetland – BES hybrid environment and the removal of all of these micropollutant compounds was observed (**Table 5**) [64].

Other studies conducted in this area also indicate similar results where micropollutant compounds such as chloramphenicol [65], β-lactam antibiotics removal from pharmaceutical wastewater [66] and 17β-Estradiol removal [67] were reported in earlier studies.

9. Conclusion

From the above analysis, it is evident that BES excels at removing certain types of refractory xenobiotic compounds more efficiently than other conventional treatment technologies. Certain types of xenobiotic pollutants such as azo dyes that resist biotransformation and subsequent degradation in conventional treatment systems can be dealt with relatively easily using BES systems such as MFCs and MEC systems or with hybrid systems combining BES with conventional treatment systems such as activated sludge systems and UASB reactors. The current level of scientific knowledge on the utility of this technology in biodegradation processes is insufficient but it is currently evolving into a better level of understanding where it could potentially be applied in pilot or industrial scales in order to deal with these refractory organic waste types in a more meaningful manner.

Conflict of interest

All authors declare no conflict of interest.

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