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ENHANCED ELECTROCHEMICAL AND BIOLOGICAL PHOSPHORUS REMOVAL IN A SOLE REACTOR

Abdelmajeed, Adam.[1, 2], Maria, Elektorowicz [1] and Sharif, Ibeid[1].

[1] Department of Building, Civil & Environmental Engineering (BCEE), Concordia University, Canada
[2] ab_ada@encs.concordia.ca

Abstract: Excessive phosphorus levels in aquatic environments create toxic algal bloom resulting and eutrophication, which ultimately lead to poor water quality. To control this undesirable phenomenon, the phosphorus removal from municipal wastewater treatment plants (WWTPs) is essentially required. The removal of nutrients requires constructing various operation units to provide different conditions (i.e. anoxic and aerobic) intended to promote the removal efficiency of each single nutrient compound. Such designs have high energy consumption and a big footprint. In this work, the possibility of simultaneous electrochemical and biological phosphorus removal in a single reactor was investigated. Bench scale electro-bioreactor was fed with synthetic wastewater containing phosphorus in the range of 14−18 mg $PO_4 - P$/L. At bench scale steady state conditions, the removal efficiency of phosphorus (as $PO_4 - P$) reached over 99%. It was concluded that such high percentage of phosphorus removal was due to both electrocoagulation process and polyphosphate-accumulating organisms (PAOs) growth in the reactor since a single submerged membrane electro-bioreactor (SMEBR) was capable of creating anoxic and aerobic conditions suitable for PAO growth. Furthermore, the electrocoagulation generated Al^{3+} due to the electrolytic dissolution of aluminum anode led to higher electrochemical phosphorus removal in the system.

Keywords: Advanced wastewater treatment processes, Phosphorus removal, Electrochemical treatment, Submerged Membrane Electro-Bioreactor (SMEBR), Polyphosphate-accumulating organisms (PAOs), Sustainable design.

1 INTRODUCTION

Over the recent years, the providing of a fresh water supply and safely treated effluent has become an urgent global issue. The discharge of insufficiently treated wastewater effluent can potentially create a significant squeeze to fresh water resources and environmental milieus. Among of these impurities that discharged effluent usually containing are nutrients, in particular nitrogen (N) and phosphorus (P), which are constantly challenging and expensive to remove (Bodini et al. 2015). The accumulation of these nutrients in aquatic environments such as lakes and artificial reservoirs can pose its deterioration. Discharged nutrients have numerous effects including toxic algal growth and eutrophication leading to oxygen depletion in surface waters. Phosphorus is considered as the key component of municipal sewage, as human waste from the toilet is a substantial source (Nedjah and Laskri 2015), and thus the removal of phosphorus during treating processes is of great importance.

Several techniques have been established for the removal of phosphorus (i.e. biological treatment (EBPR), chemical treatment (precipitation, sometimes supported with membrane filtration) (Nassef 2012). Each

process does exhibit drawbacks when they operate separately. For instance, membrane filtration process (microfiltration and ultrafiltration) is not capable of an adequate removal of phosphorus from wastewater to an acceptable level. Reversible and irreversible fouling during membrane processes is also considered as a major obstacle facing this technique since it results in a flux decline and an increase in the overall resistance to the filtration process, and hence, increasing the energy requirement and decreasing the life-span of the membrane (Wilbey 2013, Basile et al. 2015). Likewise, biological means to enhance phosphate accumulation by biomass requires more than one biological reactor and pump systems to ensure recirculation of wastewater. Such processes can drop the reactive phosphorus concentration as low as 0.2 mg $PO_4 - P$/L, but it requires space and energy that not all wastewater treatment plants and municipalities can accommodate and handle. In the areas where population density is low and climate conditions are extreme, the above techniques would be difficult to apply. Chemical treatment, which uses coagulation agents to co-precipitate phosphorous, leads to the increase of the operation costs and generate huge sludge as by-products. Thus, these conventional processes have economic and sustainability limitations as a result of excess sludge disposal, high energy inputs, poor effluent quality, and limited phosphorus removal (Posadas et al. 2013). An alternative strategy is to use electrokinetic processes in combination with biological and membrane processes to eliminate phosphorous at low cost, while the requirement for space is minimal.

Electrocoagulation as a result of electrokinetic processes is a promising technology, which has been proved to be an efficient and inexpensive technique for wastewater treatment. Electrocoagulation (EC) for wastewater treatment provides a set of unique advantages over chemical coagulation processes. Electrocoagulation process as an example can operate with no chemical additives while, alkalinity is not consumed through the processes so that pH adjustment is not necessary (Al-Shannag et al. 2013). EC process does not require coagulation agents such as alum and ferric sulfate, and hence, reduces the cost of operation and produces less sludge as an end-product (Zhu et al. 2005). Moreover, EC has a potential in an efficient removal of metals, colloids, and soluble inorganic pollutants, resulting in higher quality of the effluent (Hasan et al. 2014).

The Submerged Membrane Electro-Bioreactor (SMEBR), presented in this paper, implements EC to combine it with conventional membrane bioreactor at a low direct current in a single vessel (Elektorowicz et al. 2009, Bani-Melhem and Elektorowicz 2010). The novelty of this technology bases on allowing the electrokinetic phenomena to create alternated reduction – oxidation conditions (e.g. between -150 mV to +150 mV) in the same vessel to provide an adequate environment for the growth of all diverse types of microorganisms including PAO (Ibeid et al. 2011). The process takes fully advantage of phenomena initiated by direct current applied intermittently into the vessel (Eq. 1– 4) (Ibeid et al. 2011). Such phenomena control physicochemical as well as biological processes in the reactor (Ibeid et al. 2013). The operation mode (DC current system ON and OFF) is a state-of-art and can be adjusted depending on treatment objectives (e.g. carbon removal, phosphorous or all impurities simultaneously, etc.) (Hasan et al. 2012, Ibeid 2011). Since physicochemical, and biological and filtration processes take place simultaneously in the same tank, the SMEBR leads to the possible minimizing of space requirements and footprint (Ibeid et al. 2016). Additionally, the system also requires less aeration supply and no additional of chemicals comparing to a conventional MBR system where intensive aeration and chemicals usage are inevitable. This system has been recognized as an efficient, operational cost-effective and environmentally friendly (Ibeid et al. 2016). Furthermore, SMEBR is capable of removing metallic impurities (Hasan et al. 2014). Major electrochemical reactions taking place around the anode and cathode area in SMBER system are presented in the equations (1– 4).

Anode area (oxidation):

[1] $\mathrm{Al} \rightarrow \mathrm{Al}^{+3} + 3\mathrm{e}^{-}$

[2] $2\mathrm{H_2O} \rightarrow \mathrm{O_2\,(gas)} + 4\mathrm{H}^{+}\mathrm{(aq)} + 4\mathrm{e}^{-}$

Cathode area (reduction):

[3] $3\mathrm{H_2O} + 3\mathrm{e}^{-} \rightarrow \frac{3}{2}\mathrm{H_2\,(gas)} + 3\mathrm{OH}^{-}$

[4] $0.5\ O_2 + 2e^- + H_2O \rightarrow 2OH^-$

This article displays an outstanding performance of integrated processes comprising electrokinetic, biological and membrane filtration in one vessel for treating wastewater with respect to phosphorous. Moreover, a potential of the simultaneous biological and electrochemical phosphorus removal in this single reactor was discussed.

2 MATERIALS AND METHODS

2.1 Material and feed media

The tested systems were supplied with a synthetic wastewater, where phosphorous was provided by potassium phosphate (KH_2PO_4) and potassium chloride (KCl). Such composition was selected to simulate real municipal wastewater in terms of organic compounds, nitrogen, phosphorus, and soluble salts content. Phosphorus in the form of potassium phosphate (KH_2PO_4) changed between 14 to 18 mg $PO_4 - P/L$ in feed wastewater over the experimental period. The organic portion of the feeding consisted of both glucose ($C_6H_{12}O_6$) and sodium acetate ($CH_3COONa.3H_2O$). Ammonium sulfate $(NH_4)_2SO_4$ was the major source of nitrogen. Other trace elements were also added to the synthetic wastewater. Table 1 shows the characteristics of influent ("feed wastewater").

Table 1: Initial physicochemical characteristics of synthetic wastewater

Parameter	Abbreviate	Average Value/or Range
Chemical oxygen demand [mg/L]	COD	280
Ammonia-nitrogen [mg/L]	$NH_4^+ - N$	23.3
Nitrate-nitrogen [mg/L]	$NO_3^- - N$	0.148
Nitrite -nitrogen [mg/L]	$NO_2^- - N$	≤ 0.05
Total nitrogen [mg/L]	T-N	24.6
Phosphorus [mg/L]	$PO_4 - P$	14−18
Potential of hydrogen	pH	6.62
Conductivity [µS/cm]	EC, κ	97.4
Oxidation-reduction potential [mV]	ORP, E_h	6−8
Total alkalinity (as CaCO3 mg/L)	TALK	98.5
Temperature [ºC]	T	15.8−17.2

2.2 Experimental setup

Two bench scale continuous flow reactors: one submerged membrane electro-bioreactor– SMEBR (Figure 1) and one conventional MBR system without DC electrical field (reference reactor) were run in parallel to assess the phosphorus removal. Both reactors were continuously fed with the same influent composition and a constant flow rate of 7 L/d. The corresponding hydraulic residence time (HRT) and sludge residence time (SRT) were 24h and 20 days respectively in both reactors (Table 2). To obtain comparative conditions between the processes of SMBER and MBR, alum (as aluminum sulfate, $Al_2(SO_4)_3 \cdot 18H_2O$) was added to the supernatant of MBR system. The configuration of the electro-bioreactor was identical to patented SMEBR design (Ibeid et al. 2011, Elektorowicz et al. 2009). SMEBR consisted of a cylindrical polyethylene container with a total effective working volume of (7 L), a hollow fiber ultrafiltration membrane module (Zeeweed-1, GE, Canada), two perforated electrodes, and 8 fine bubble air diffusers. The air diffusers supplied oxygen for biological activities, achieving a proper mixing, and restraining the membrane fouling. The membrane, with a pore size of 0.04 µm and surface area of 0.047 m^2, was located

centrically and surrounded by electrodes (Figure 1). A DC power supply connected to a timer was used to provide intermittent current (at a current density of 12.5 A/m^2). The system was inoculated with fresh activated sludge from a local WWTP. The activated sludge was diluted (0.5 L of activated sludge and 6.5 L water) to provide the initial mixed liquor suspended solid (MLSS) concentration of 3000 mg/L. About of 350 mL of sludge was wasted on a daily basis to maintain target SRT of 20 days. The temperature of 22±2 ˚C was kept at during the experiment while neutral pH was also preserved. Table 2 summarizes the major operational factors of SMEBR and MBR systems.

Table 2: Operation condition of SMEBR and MBR

Parameter	SMEBR	MBR
DO (mg/L)	0.5-3.5	5-6
HRT (h)	24	24
SRT (d)	20	20
Wasted sludge (mL)	350	350
Effluent flowrate (L/d)	7	7
Effective working volume (L)	7	7
MLSS (mg/L)	3000	3000
Current density (A/m^2)	12.5	N/A*

*N/A= not applicable

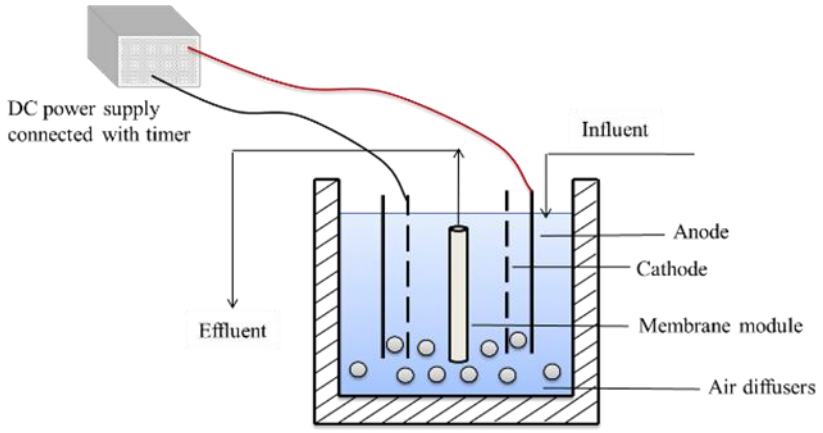


Figure 1: Schematic diagram of submerged membrane electro-bioreactor, SMEBR

2.3 Analytical methods

The concentrations of phosphorus in influent and effluent were determined as orthophosphate-phosphorus ($PO_4 - P$) using Hach procedure (Ascorbic Acid Method 10209, TNTplus™ 844, Hach, DR 2800, USA). The values of pH and temperature were measured by using a pH probe (HQ30d, multi- parameters meter, Hach, SA). Measurements of dissolved oxygen (DO) was conducted using a digital DO meter (YSI, Model 52, USA), while oxidation-reduction potential (ORP) was measured by a digital IntelliCAL™ MTC101 Standard Gel-Filled ORP electrode (Hach). These parameters are interrelated and are of importance in controlling the reactor performance. Besides, the measurement of ORP assures the accuracy of DO distribution. Calibration of DO and pH meters were performed on a weekly basis. The removal efficiency of phosphorus (% Removal) was calculated as per Eq. 5:

[5] $(\%\ \text{Removal}) = \frac{C_i - C_f}{C_i} \times 100$

Where; C_f and C_i are the final and initial phosphorus concentration in "mg/ L," respectively.

To assess the biomass activity, mixed liquor suspended solids (MLSS) and mixed liquor volatile suspended solids (MLVSS) were measured regularly according to a procedure described by the Standard Methods 2540 D and 2540 E, respectively (APHA 2012). Such techniques were mainly based on weight lost, at 105 °C and 550 °C for TSS and VSS, respectively. Calculations were done based on the given equations 6 and 7:

[6] mg TSS/L = ([103–105 °C] dry weight - weight of filter)*1000, mg/volume of sample, mL.

[7] mg VSS/L= (550 °C dry weight – [103–105 °C] dry weight)*1000, mg/volume of sample, mL.

Furthermore, the viability of bacteria in the electrical field were counted using the plate count agar (PCA) method. Such method is used to provide a rough estimation of the number of aerobic and facultatively anaerobic bacteria present in water sample. In this practice, dilution of tested samples "serial dilution technique" between 10^1 to 10^5 were performed as described in Standard Methods for Microbiological Examination of Dairy Products, (APHA Method, 7157) (Marshall 1993). Diluted samples that yielded a plate with an average of 30 ~ 300 colonies were chosen. 0.1 mL volume of the diluted specimen was utilized to inoculate or to quantify bacterial counts in the sample coming from the electro-bioreactor. Samples were incubated at 32±1°C and examined for growth after 48 hours. Duplicated plates were used to test bacteria count, and the value was recorded as an average. The number of bacteria in the representative samples were calculated using this formula: CFU/mL= CFU * dilution factor * 1/aliquot, where magnifying colony counter was used when needed.

Applying a DC electrical field in a biological reactor is most likely to impair the bacteria's metabolism, shape, movement, and membrane integrity. Thus, the available commercially LIVE/DEAD *Bac*Light bacterial viability kit (L13152) provided Molecular Probes, Inc. (Eugene, OR, USA) was applied to test the bacterial viability in view of assessing the impact of electrokinetics processes on microbial activity. The *Bac*Light viability stain package provides an indication of the fraction of physiologically active and dead "inactive" cells based on membrane integrity. In this protocol, cells are treated with two different nucleic acid-binding dyes: SYTO®9 and propidium iodide (PI). SYTO®9 is a small green-fluorescent nucleic acid that penetrates all bacterial membranes (both those with intact and damaged membranes), and hence it is used for assessing total cell counts (live and dead). In contrast, propidium iodide is a large red-fluorescent nucleic acid that penetrates only cells with a compromised membrane and consequently visualizing dead microbes. PI is, as a result, causing a reducing in the SYTO®9 stain fluorescence intensity when both dyes are present. The simultaneous use of SYTO9 and PI, therefore, enables differentiation and measurement of the relative ratio of active cells "green" with an intact membrane and dead cells "red" with a damaged cytoplasmic membrane.

Two fluorescent nucleic acid stain were either used alone or in combination: SYTO9 (Invitrogen AG, Basel, Switzerland) and propidium iodide (Invitrogen). The working solution of these dyes was prepared as recommended by the supplier. All stock solutions were stored at -20°C. Staining was done by mixing equal volumes of SYTO®9 and PI, dissolved in 5 mL deionized water. 1.5 mL of two staining mixture was added directly to 1.5 mL of the appropriate bacterial suspension. Incubation was accomplished in the dark at room temperature for 15 minutes. Calibration was done according to manufacturer's instructions except for *E. coli* suspensions which were replaced by the MBR activated sludge. Five samples were prepared to achieve effective concentrations ranging from 0 to 100% to create a standard curve (all mixtures were five specimens * 1.5 mL = 7.5 mL total). A standard curve was then prepared for each microplate. Stained bacterial suspensions were observed under a confocal laser-scanning microscopy (CLSM) with 1000X total magnification. The excitation/emission was 488/535 nm for viable cells, and 488/580 nm for dead cells. The cells were then visualized and quantitated using Image processing and analysis in Java.

3 RESULTS AND DISCUSSION

3.1 Phosphorus removal

The phosphorus concentration in effluent and removal efficiency by SMEBR and MBR systems are shown in Table 3 and Figure 2. The results have displayed that the ability of SMEBR system to remove phosphorus to an appreciable extent. Treatment by SMEBR process reduced orthophosphate with an average of 94.86% compared with 48.99% obtained by MBR. The maximum phosphorous removal in SMEBR was 99.28%, while in conventional MBR was only 64.44%. Yet, this removal percentage in MBR system was only achieved by adding of coagulant agents to help phosphorus removal. The same observations regarding to hinder the P-removal efficiency of the MBR system was also reported by (Hasan et al. 2012, Wei et al. 2012, Wei et al. 2009, Zuthi et al. 2013). Zuthi et al. (2013) indicated that the application of MBR treatment targets for P-removal and effluent quality requirements is feasible if the biological processes relevant to P-removal could be fully understood and linked to other process parameters.

Such results may suggest the necessity of an additional operation unit to MBR system to deal with the persisting phosphorus compounds. Contrary, the SMEBR system was enhanced with the electrokinetic process in the same operation unit which also is able to remove other impurities. For example, Hasan et al. (2014) reported that the introducing of electrical filed to the reactor contributed in electro-oxidation of organic impurities and making them possibly much more bioavailable due to the production of hydroxyl radicals ($OH^{\bullet}$) at the anode area. Hydroxyl radical is highly reactive aqueous and a strong oxidizing agent that may interact with organic impurities (Apaydin et al. 2009, Dohare and Sisodia 2014). Electrokinetic also controlled the reaction of dissolved ions with phosphorous and its fate within the reactor (e.g. deposition on electrodes) (Hasan et al. 2014). *In situ* generation of hydroxyl radicals for the treatment of non-biodegradable and refractory phosphorus fractions is displayed in the equation 8:

[8] $\mathrm{M} + \mathrm{H_2O_2} \rightarrow \mathrm{M^{n+}} + \mathrm{OH^-} + \cdot\mathrm{OH}$

Where; (M) is the metal which is oxidized to its cation ($\mathrm{M^{n+}}$)

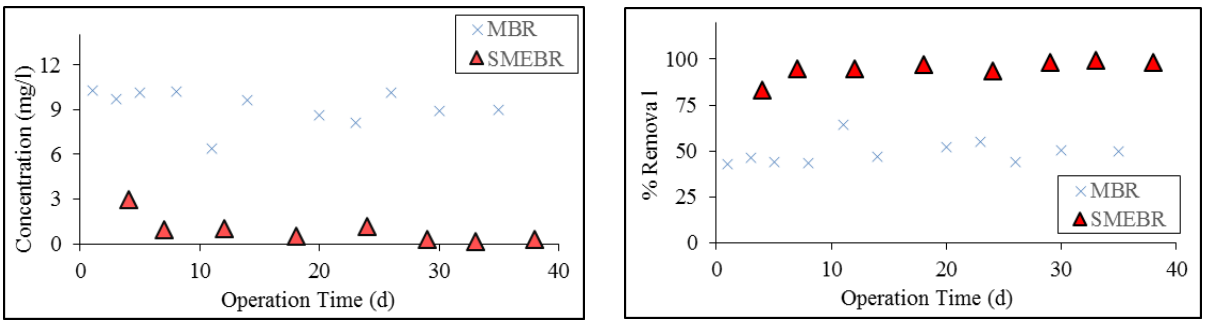
In situ aluminum ions (Al^{3+}) generated due to the electrolytic dissolution of anode led to removal and precipitation of phosphorus (Eq. 9 and 10). Phosphorus was incorporated into a long chain of a complex compound that became a part of the suspended solids and removed due to the liquid-solid separation using membranes (Elektorowicz et al. 2014, Ibeid et al. 2011).

[9] $\mathrm{Al^{+3}} + 3\mathrm{H_2O} \rightarrow \mathrm{AL(OH)_3} + 3\mathrm{H^+}$

[10] $\mathrm{Al^{+3}} + \mathrm{H_nPO_4^{3-n}} \leftrightarrow \mathrm{ALPO_4} + \mathrm{nH^+}$

Table 3: Statistical parameters for the phosphorus concentration in effluent and its removal efficiency in SMEBR and MBR.

Parameter	Conventional MBR		SMEBR	
	Effluent (mg/L)	% Removal	Effluent (mg/L)	% Removal
Mean	9.18	48.99	0.93	94.86
Standard Deviation	1.17	6.52	0.92	5.13
Variance	1.38	42.52	0.85	26.30
Median	9.60	46.67	0.73	95.96
Mode	10.10	43.89	-	-
Minimum	6.40	42.78	0.13	83.33
Maximum	10.30	64.44	3	99.28



(a) **(b)**

Figure 2 : Performance of SMEBR and MBR processes with respect to phosphorus: (a) effluent concentration and (b) removal efficiency

It was concluded that excellent results in phosphorous removal by SMEBR system were due to both electrocoagulation and PAOs (bio-flocs) growth. It is speculated that the SMEBR system provided adequate (anoxic/aerobic) for the growth of polyphosphate-accumulating organisms due to the ability of the system to work in a wide range of redox potentials. In theory, biological phosphorus removal (Bio-P) requires the retention of biomass in sequential anaerobic and aerobic phases. During this process, P-release and organic matters uptake happen under anaerobic phase, while P-removal or uptake occurs under the following aerobic conditions (Sathasivan 2008). In SMEBR, the alternating anoxic/aerobic conditions were provided by periodic switching on/off the DC power supply. Thus, it is assumed that the electro-bioreactor was able to fluctuate oxidation-reduction potential (ORP), providing a suitable environmental for the activity of PAO organisms, and subsequently allowing to support the removal of large amounts of phosphorus. Such fluctuating ORP levels was ensured by the application of electrokinetic processes (Eq. 1– 4) and adequate air supply in the system where the measured DO in the reactor fluctuated between 0.5 and 3.5 mg/L. Ibeid et al. (2011) reported that the anaerobic condition was gradually supported when the electrical field was activated, while the aerobic condition was maintained by disconnecting the electrical current. In contrary, the MBR as all conventional systems preserved aerobic conditions without affecting phosphorous removal.

Previous studies have reported that an intermittent aeration membrane bioreactor (MBR) can achieve simultaneous carbon, nitrogen and phosphorous removal (Wang et al. 2015, Gnirss et al. 2003). However, while these studies have indicated the development of polyphosphate-accumulating organisms (PAOs) within the processes, the removal efficiency of phosphorous did not exceed 60%. The removal efficiency of phosphorus was also affected by the influent phosphorus concentration. It is speculated that in the present investigations, a high phosphorous removal was caused by both PAOs biomass growth and the electrolysis using a sacrificial aluminum anode.

3.2 Biomass activity assay

In order to support above conclusion with respect to biological removal of phosphorus, the biomass activity measurements for both the SMEBR and MBR systems were conducted. To assess the impact of electrokinetics processes on microbial activity, the LIVE/DEAD *Bac*Light bacterial viability kit was used to determine the ratio of viable bacteria in both reactors. The application of low current density with intermittent exposure regime to the bioreactor did not reduce significantly the percentage of live cells comparing to the control reactor. The average of live cells in both the SMEBR and MBR systems were 90% and 96%, respectively. Such test is based on the membrane integrity, while some bacteria having compromised membranes may be able to recover from the electrical field impact and resume their activity. According to Alshawabkeh et al. (2004), anaerobic cultures were able to recover their activities when the DC supply was switched off. The findings of our study suggest that intermittent electrical field of CD =12.5 A/m^2 applied into SMEBR were not negatively affecting the microbial biomass viability. These results were not in agreement with Wei et al. (2011) who reported 15% reduction in the live cell under CD =12.3 A/m^2. This

slight discrepancy may be attributed to different electro-bioreactor configurations applied in both studies. In the current study, the continuous flow electro-bioreactor provided sufficient mixing and well-aerated condition that reduced the potential adverse effect of the DC on bacterial cells, while the batch electro-bioreactor mode used in the previous study may not provide a proper mixing and aeration conditions.

To further assess the activity of the microbial biomass, the bacterial plate count agar (PCA) procedure was undertaken to quantify the number of viable bacteria in both reactors. Remarkably, the applied current density of 12.5 A/m^2 to the bioreactor resulted in electro-stimulation of microbial cells and increasing in bacterial counts comparing to the reference reactor " without current." The highest bacterial counts were observed at the SMEBR system with an average of counted cells of 46 x 10^4 CFU/mL, whereas the MBR system had an average of bacterial counts of 31 x 10^4 CFU/mL. Such results support the hypothesis of that a low-level DC electric field enhances the bacterial growth, which was also confirmed in this study (Berg 1995, Fiedler et al. 1995, Alshawabkeh et al. 2004). Low current density stimulated biomass activity as a result of DNA changes, protein and biopolymer synthesis, enzyme reactions, cell membrane permeability and transport, and cell growth (Nakanishi et al. 1998, Berg 1995). Moreover, an intermittent current density below 25A/m^2 for the MLSS concentration of 6,000 mg/L was found in the literature to stimulate microbial biomass activity (Bayar and Karagunduz 2014). Wei et al. (2011) had studied the viability of the bacteria in the electrical system by which biological processes were not significantly affected by adequate interrupted electrical current density leading to great nutrient removal efficiencies. Furthermore, Yin et al. (2015) investigated the enhancement of the activity of anammox bacteria using electric current through which the total nitrogen removal rate (NRR) was significantly increased by 38.7 % over the control reactor. Chen et al. (2016) found that the electrical field could speed up the processes of hydrolysis and acidification. The elevated abundance of *Pseudomonas* (break down proteins) and *Methanoregula* (generate methane) were typically recorded at an electric voltage of 0.6 V/cm, with species richnesses of 19.1% and 53.3%, respectively. Such results support the fact of that the installation of electrodes on the SMEBR system enhanced the biomass activities. However, as the total number of polyphosphate-accumulating organisms was not directly investigated, it is uncertain to what extent the activity of PAOs have contributed in the removal of phosphorus.

Likewise, the investigation of mixed liquor suspended solids (MLSS) illustrated the high microbial growth and activity in the SMEBR comparing to conventional MBR, which was increased from 3000 mg/L to 4900 mg/L and hence played a key role removal efficiency of organics. Whereas, MLSS concentration in MBR system increased from 3000 mg/L to 3700 mg/L after 40 days' operation period. A significant increase in MLSS was also due to the dissolution of the aluminum anode at 12.5 A/m^2 experiment. MLVSS/MLSS ratio had shown high microorganism content of the suspended material. The investigation of MLVSS fraction illustrated the microbial growth and activity in the SMEBR, which was increased from 2520 mg/L to 3480 mg/L and hence played a key role in removal efficiency of phosphorous. Besides, the concentration of total phosphorus in activated sludge indicated that the phosphorous was likely stored it in biomass. The concentration of orthophosphate in the supernatant was higher than the concentration of phosphorous in the influent wastewater (date not shown). Sophisticated microbial and molecular techniques are continuing to be used in our upcoming research work. Even if we are not able to directly prove the presence of PAOs, it is evident from this research that the system had substantial phosphorous removal and electrode processes stimulated the biological activity in the system. The processes of electron transfer (ET) through microbial redox and their mechanisms have been previously reported by many studies (Watanabe et al. 2009, Summers et al. 2010, Ailijiang et al. 2016, and Liu et al. 2015). These studies have shown that some bacteria species are capable of performing anodic and cathodic half reactions via electrodes as electron donors and acceptors, respectively. Ailijiang et al. (2016) assumed that the conductive minerals would help electron transfer between microbial biomass and electrodes, and hence promote the processes of electrostimulation. Another study reported that electrically conductive magnetite nanoparticles permit the interspecies electron transfer (IET) through bacteria community such as *Geobacter Sulfurreducens* to *Thiobacillus Denitrificans*, leading to enhance simultaneous acetate oxidation and nitrate reduction (Zeyoudi et al. 2015). Such evidence support above conclusion with respect of POA stimulation by the electrical field and adequate redox conditions.

4 CONCLUSION

The potential of phosphorus removal from synthetic wastewater using a hybrid electro-bioreactor has been investigated. The system has apparently shown its capability to combine electrochemical and biological phosphorus removal in a single operation unit by providing an adequate direct current density (CD) and dissolved oxygen (DO) concertation. A substantial phosphorus removal of nearly 99% was attained in the SMEBR process, whereas the average of observed removal in conventional MBR was 47.84%. The SMEBR system distinguished by a compact hybrid unit, where electrokinetic, biological (biodegradation) and membrane processes (ultrafiltration) take place simultaneously. It was also assumed that due to alternative anoxic/aerobic conditions, PAOs are also participating in phosphorous removal. Such promising technique for wastewater treatment can improve treatment efficiency, reduce phosphorus to an acceptable level and decrease fouling rate while minimizing footprint and overall energy costs. The developed system may have greatest potential application to decentralized wastewater treatment facilities.

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