Electrolysis-Driven Bioremediation to Enhance Nitrogen and Phosphorus Removal From Polluted River Sediment

Sanshan Li
  Nanjing University

Tangming Ma
  Nanjing University

Chaoqun Zheng
  Nanjing University

Zhaofang He
  Nanjing University

Liuyan Yang
  Nanjing University

Yan Gao (✉ gaoyan@nju.edu.cn)
  Nanjing University

Abstract

In order to testify the effect of electrolysis and microbial remediation technology in polluted river sediment. Here, we explored the possibility of electrochemically removing ammoniacal nitrogen-nitrogen (NH_3-N), nitrate-nitrogen (NO_3^--N) and phosphate ions-phosphorous (PO_4^{3-}-P) by using a titanium (Ti) mesh cathode, a Ti/Ti dioxide (TiO_2)/Ruthenium (IV) oxide (RuO_2) (RuO_2-IrO_2/Ti) mesh, and a magnesium-aluminum (Mg−Al) alloy anode placed within the sediment and overlying water. Results showed that approximately 151.82 ± 21.69 mg TN was removed which was five times more effective than the non-electrolytic controls (30.21 ± 13.73 mg), NH_3-N concentration in the sediment was substantially reduced (up to 2.9 times) compared to the non-electrolytic controls. Its efficiency lies in the electrolysis process, which may directly remove NH_3-N through electrochemical oxidation and simultaneously produce oxygen which helps nitrifying bacteria to convert NH_3-N into NO_3^--N by the role of anode; and electrolysis may directly remove NO_3^--N in the overlying water through electrochemical reduction while simultaneously producing hydrogen electron donor for hydrogen autotrophic microorganism as *Hydrogenophohaga*, to be the dominant species in sediment to enhance the removal of NO_3^--N by the role of cathode. Electrolysis also reduced the PO_4^{3-}-P through electro-coagulation since Mg^{2+} ions could also produce since sacrificial Mg−Al alloy anode was used and electro-deposition on Ti mesh cathode both to increase PO_4^{3-}-P removal in overlying water and sediment. This study verifies the benefits of electrolysis-driven bioremediation as a sustainable technology for the bioremediation of N and P polluted river sediments.

1. Introduction

The pollution of river sediment by high concentration of nitrogen (N) and phosphorus (P) is a widespread environmental problem especially when the TN (total nitrogen) concentration was more than 1000 mg/kg and TP (total phosphorus) concentration are more than 420 mg/kg defined in US Environmental Protection Agency (EPA 2001). N mobilizing processes in sediments include the mineralization of organic matter to produce ammonium nitrogen (NH_4^+-N) (ammonification) and chemoautotrophic oxidation, then NH_4^+-N transform to nitrite/nitrate nitrogen (NOx-N) under aerobic conditions (nitrification) (Kuypers et al. 2018). As for P, which was mostly accumulated by sorption onto the sediments of dissolved phosphate ions-phosphorous (PO_4^{3-}-P) ions and the in-stream mineralization of sewage organic matter, the recycling of allochthonous particulate-P by aquatic flora or by microorganisms in the sediment can also generate autochthonous pools of phosphate ions (PO_4^{3-}) (Pistocchi et al. 2017). The active PO_4^{3-}-P remobilized from the sediments mostly are NH_4Cl-P (loosely-bound P plus labile organic P) which transferred from NaOH-P (Fe-/Al-bound P plus moderately resistant organic P) under anaerobic condition which increase the bioavailability of P and increased PO_4^{3-}-P concentration in the overlying water (Wu et al. 2019). The continuous release of N and P from sediment is the main reason for the continuous deterioration of water bodies worldwide.

Biological methods for the cleanup of sediment ecosystems contaminated by eutrophic river represent a sustainable option, due to their relatively low costs, low environmental impacts, and wide applicability to a range of sediment contamination scenarios (Zhao et al. 2019). Bioremediation takes advantage of the metabolism of N multifunction micro-organisms, which are widespread in nature, particularly in environments where microbes are naturally present, i.e., in polluted river sediments. Moreover, the lack of oxygen needed to serve as electron acceptors results in a decrease in the nitrification rate and the accumulation of extra ammoniacal nitrogen-nitrogen (NH_3-N) during the N transforming process in sediment. Meanwhile, this lack of oxygen is beneficial to the release of NaOH-P to NH_4Cl-P in the sediment and increased the PO_4^{3-}-P concentration in the overlying water. Therefore, increasing oxygen availability has long been regarded as a way to reduce NH_3-N and PO_4^{3-}-P in anaerobic sediment environments (Wang et al. 2020). The use of oxygen-releasing compounds (ORC) based on calcium or magnesium peroxide has been proposed as a strategy to ensure a long-lasting release of oxygen for NH_3-N removal in contaminated subsurface anaerobic sediment and released Mg^{2+} and Ca^{2+} ions for the PO_4^{3-}-P control (Huang et al. 2015; Yang et al. 2015; Seifan et al. 2017; Li et al. 2020). However, a number of drawbacks have prevented the use of ORC for the remediation process, including: (i) the rate of oxygen release and the resulting oxygen availability is difficult to control; (ii) repeated injections may be required due to the oxygen consumption/scavenging by biotic and abiotic side reactions, (iii) ORC may have secondary harmful effects on microbe and aquatic biota such as higher pH environment was caused as hydroxide will produced during process of ORC release oxygen.

Recently, electrochemical and microbial methods have been successfully applied to accelerate the bioremediation of phenanthrene, poly (allylamine hydrochloride) (PAH), and oil-contaminated marine sediments (Cecconet et al. 2020; Li et al. 2020). The electrochemical process can provide virtually inexhaustible source of electron donors/acceptors for the reductive/oxidative degradation of contaminants (Bellagamba et al. 2017). The electrochemical oxidation of NH_3-N to dinitrogen (N_2) and the electrochemical reduction of NO_3^--N to N_2 were researched for the removal of inorganic N (Tang et al. 2013; Yao et al. 2016; Song et al. 2019; Cecconet et al. 2020; Zhai et al. 2020). As for P removal, the electrolysis sacrificial iron anode was also used in biofilters (Gao et al. 2016) and in a modified biological aerated filter system configured for Fe-C micro electrolysis (Zhang et al. 2018) to enhance PO_4^{3-}-P removal. It can be concluded that the electrochemical method is a flexible technology which is conducive to the removal of N and P in polluted river sediment.

In this research, we explored the possibility of an electrolysis-driven bioremediation system to manipulate the mechanism of N and P transformation process of polluted river sediment by low voltage electrolysis which using a constant 40 mA current and voltage was about 5 V–6 V. In theory, the bioremediation of N in polluted river sediment could promoted transformation directly through reductive/oxidative degradation by electrolysis process and indirectly through the oxygen acceptor produced by Ti/Ti dioxide (TiO_2)/Ruthenium (IV) oxide (RuO_2) (RuO_2-IrO_2/Ti) mesh anode which was promoted the nitrification of NH_3-N and NO_3^--N could remove by hydrogen autotrophic microbes since the hydrogen donor was

produced by Ti mesh cathode; and PO_4^{3-}-P could remove through electro-coagulation by using a sacrificial magnesium-aluminum (Mg−Al) alloy anode which released mostly Mg^{2+} ions and electro-deposition in a titanium (Ti) mesh cathode. The objectives of this study were (a) to evaluate the enhancement of electrolysis-driven bioremediation of N and P removal in polluted river sediment and overlying water; (b) to determine the variations of the N transformation microbial community structure under electrolysis in polluted river sediment; (c) to explore the degradation mechanisms of P in this system; and (d) study the N and P secondary release from sediment after electrolysis.

2. Methods And Material

2.1 The experimental sediment and simulated overlying water

The polluted river sediment was collected from the Jiuxiang River near Nanjing in the Jiangsu Province of China (32°N and 118°E). A Peterson grab sampler was used to collect sediment at a height of about 15 cm from the surface of the riverbed. The collected sediment was immediately sealed, shaded and transported to the laboratory. After the preliminary removal of the residual leaves, stones, and debris, the sediment was screened through 20 mesh screens (Diameter of each mesh was 32.26 mm), and evenly mixed for use. The overlying water was prepared by dissolving ammonium chloride (NH_4Cl), potassium nitrate (KNO_3) and potassium dihydrogen phosphate (KH_2PO_4) in tap water to maintain concentrations of 12 mg/L NH_3-N, 2.50 mg/L NO_3^--N, and 0.20 mg/L PO_4^{3-}-P, with a pH ranging between 7.5 and 7.7 and a dissolved oxygen (DO) concentration of 4.10 mg/L−4.35 mg/L, which was simulated as overlying water in polluted river sediment throughout the entire experiment.

2.2 Experimental apparatus and operation conditions

Figure 1 was a schematic drawing of the experimental device used for electrolyzing sediment and overlying water which was placed in a 1 L beaker. In each beaker, 400 g of polluted river sediment (about 300 mL) was added, and 500 mL of overlying water was slowly injected into the beaker along the wall of the beaker by using a siphon and static device for one day. The electrolysis was conducted by two pairs of electrodes, one was a Mg−Al alloy bar anode, 180 mm long, 10 mm wide and 3 mm thick; the other was a Ti mesh cathode, 140 mm long, 50 mm wide and 1.5 mm thick. After four days of electrolysis reaction, another group of electrolytic pairs were added for common electrolysis, i.e., the RuO_2-IrO_2/Ti mesh anode and the Ti mesh cathode, which were the same size (Fig. 1). And the distance between each of the two pairs of electrodes was 7 cm. The electrodes were connected using wiring alligator clips with a diameter of 1 mm to a Model KXN-3020D DC regulated power supply (Zhaoxin Electronic Instruments and Equipment Co., Ltd., Shenzhen, China). The voltage ranged between 0 V and 30 V and the amperage was between 0 A and 5 A, while the DC regulated power provided a constant low current of 40 mA for electrolysis with a voltage of 5 V−6 V. In the non-electrolytic control group, the same weight of polluted river sediment and overlying water were added without electrolysis. Both the electrolysis group and

control group were replicated in three groups. The experimental apparatuses were maintained within a temperature range of 25 °C to 30 °C. During the experiment period, the water loss from evaporation and transpiration was replaced by adding deionized water to the original volume every day.

2.3 Sampling of sediment and overlying water

The samples of sediment and overlying water were collected after 0 d, 2 d, 3 d, 4 d, 5 d, 7 d, 9 d, 11 d, 13 d, 15 d, 17 d, 20 d, 24 d, 27 d and 32 d to determine the physical and chemical indicators of water quality, and at each sampling time point, there were three sets of repeats for the electrolyzed sediment and the non-electrolytic control group. 20 mL of overlying water were collected from the electrolysis group and the non-electrolytic control group with a syringe and put into a 50 mL Corning centrifuge tube for analysis; then, removed all the overlying water, extracted 50 mL mixed sediment into a 50 mL Corning centrifuge tube. For overlying water samples, the pH and DO were immediately measured by using a portable Hach HQ30d multi-parameter analyzer, a PHC101-30 pH electrode, and an LDO101-03 DO electrode (Hach Company, Loveland, CO, USA). After the corresponding standard pretreatment and reagent additions were completed, the NO_3^--N was determined by ultraviolet spectrophotometry, NH_3-N was used as the Nessler's reagent spectrometry, TN was determined by using the alkaline potassium persulfate digestion method, PO_4^{3-}-P was determined by the ascorbic acid method, and TP was measured by the ammonium molybdate spectrophotometry method. All the testing procedures were according to Standard Methods for the Examination of Water and Wastewater by the Ministry of Environmental Protection (MEP of China 2002). For sediment samples, the pH and Eh were immediately measured using a portable Hach HQ30d multi-parameter analyzer, a PHC101-30 pH electrode, and an MTC101 Eh electrode (all from the Hach Company, Loveland, CO, USA). After the corresponding standard pretreatment and reagent additions were completed, the NO_3^--N, NO_2^--N, and NH_3-N were measured by extraction with potassium chloride solution as per the Chinese Solution-Spectrophotometric Method of the HJ 634−2012 (MEPPRC 2012). TN was measured using persulfate digestion, and the TP was analyzed using the digestion-Mo-Sb anti-spectrophotometric method and measured with ultraviolet and visible V1800 spectrophotometers (Shimazu Corp., Kyoto, Japan). The SMT continuous step extraction method was used for the determination of NH_4Cl-P and NaOH-P (Psenner et al. 1988).

2.4 Characterization of the microbial community and diversity analysis

The bacterial community was examined in sediment electrolytic groups and non-electrolytic controls, using DNA-based molecular techniques. About 15 mL of sediment were taken from each electrolytic sediment sample when the experiment was ended. The sediments were extracted according to the instruction of an OMEGA E.Z.N.ATM-Bind Soil DNA Kit. High-throughput sequencing (HTS) was used to identify and estimate changes in the relative abundances of bacteria. DNA concentration and purity were determined by a NanoDrop ND-1000 microspectrophotometer (NanoDrop Technologies, Wilmington, DE, USA). Three replicate DNA extractions were combined into one sample for Illumina HTS after the genomic

DNA was extracted. HTS was performed externally (Sangon Biotech, Shanghai, China) using standard protocols on a MiSeq platform (Illumina, Inc., San Diego, CA, USA). The resulting sequencing data was processed using Quantitative Insights into Microbial Ecology (QIIME™) software (Caporaso et al. 2010b). Archaeal, bacterial, and eukaryotic reads were distinguished based on differences in their PCR primers and then analyzed separately. The reads were then clustered into operational taxonomic units (OTUs) based on 97% similarity with UCLUST43 (Edgar 2010). Representative sequences from each OTU were assigned a taxonomy using an RDP Classifier44 (Wang et al. 2007) with a minimum support threshold of 80% and aligned using the Greengenes reference database (version 13_8) 45 by PyNAST42 (DeSantis et al. 2006; Caporaso et al. 2010a). Raw sequence data (FASTQ files) generated for this study have been deposited in NCBI's Sequence Read Archive under BioProject number 16S190521. Microbial diversity was measured by analyzing the alpha and beta diversities, as determined using the QIIME™ pipeline and based on the OTUs. For alpha diversity, Chao1 richness and the Shannon and Simpson diversity indices were calculated using QIIME™ (Caporaso et al. 2010b).

2.5 Morphology and composition analysis of precipitates

At the end of the experiment, the precipitates deposited on the Ti mesh cathodes and the precipitates between the sediment and overlying water were both collected and put into refrigerator set at -80 °C for freezing before treatment; they were then freeze dried for 48 hours using an LGJ-18S vacuum freeze dryer (Beijing Songyuan Huaxing Technology Development Co., China). The scanning electron microscope (SEM) and energy dispersive spectrometer (EDS) were used to observe the surface morphology and composition of the precipitates. The dried precipitates were also tested by a Fourier transform infrared (FTIR) spectrometer with a scanning range of $4000-400$ cm^{-1}. In addition, the TP concentration in the precipitates were also determined by the soil-determination of TP by alkali fusion$-$Mo-Sb anti spectrophotometric method according to the Chinese standard, HJ 632$-$2011 issued by MEPPRC.

2.6 Nitrogen and phosphorus secondary release from sediment after electrolysis

In order to explore the secondary release amount of NH_3-N and PO_4^{3-}-P into the overlying water. Taken the electrolysis group of 13-days and 32-days sediment and removed all the overlying water, and then the sediment was fully stirred and mixed then adding 500 mL deionized water as initial overlying water which was slowly added along the beaker wall by siphon, and then it was keep completely still, waiting for N and P secondary released from sediment into the overlying water. The overlying water was sampled at 0 d, 1 d, 3 d, 5 d, 7 d and 10 d, respectively, and the concentrations of NH_3-N and PO_4^{3-}-P in the overlying water were determined.

2.7 Data analysis and drawing

All data statistics were performed with SPSS 23.0 (IBM Corp., 2015) and all figures were drawn by using Origin 2018 (OriginLab 2018). Significant differences among treatments were identified through one-way

analysis of variance (ANOVA). Typically, homogeneity of variance for the obtained data was tested, and data of the test values $p > 0.05$ were adopted for the ANOVA analysis.

3. Results And Discussion

3.1 Nitrogen removal by electrolysis in sediment and overlying water

To further explore the migration and transformation of N in the electrolytic sediment and overlying water, concentrations of TN, NO_3^--N and NH_3-N in both the sediment and overlying water were shown at different times in Fig. 2. At initial four days the electrolysis was carried out by a Mg−Al alloy anode and Ti mesh cathode, the NH_3-N concentration in sediment increased from 71.56 ± 3.24 mg/kg to 104.03 ± 20.34 mg/kg, as the concentration of NO_3^--N also increased from 30.29 ± 0.28 mg/kg to 52.90 ± 2.42 mg/kg after 2 days electrolysis. After 4 days electrolysis, another electrode pair of RuO_2-IrO_2/Ti mesh anode and Ti mesh cathode were inserted in the overlying water and sediment to enhance the NH_3-N and NO_3^--N removal. Then, the NH_3-N concentration in sediment decreased from 83.24 ± 5.24 mg/kg on the 4th day to 28.94 ± 2.52 mg/kg on the 32nd day and the concentration in overlying water decreased from 32.64 ± 1 mg/L on 4th day to 1.66 ± 0.15 mg/L on the 32nd day. TN concentration in sediment decreased from 942.60 ± 81.70 mg/kg on 4th day to 512.45 ± 54.22 mg/kg, respectively, TN concentration in overlying water decreased from 36.05 ± 1.20 mg/L on 4th day to 2.02 ± 0.15 mg/L at the end of the experiment. Electrolysis promoted the transformation of NH_3-N to NO_3^--N by nitrifying bacteria since the oxygen production was enhanced during the electrolysis process (Fig. S1) and the concentration of NO_3^--N in sediment increased from 30.29 ± 0.28 mg/kg to 80.82 ± 1.79 mg/kg on the 9th day and then decreased to 45.06 ± 0.76 mg/kg on 32nd day. And the concentration in the electrolysis group was lower than 0.5 mg/L in the overlying water, which was lower than the control group which was increased from 0.04 ± 0.001 mg/L on 1st day to 1.47 ± 0.12 mg/L on 32nd day.

These results shown that a combined electrode Mg−Al anode paired with a Ti mesh cathode and an RuO_2-IrO_2/Ti anode paired with a Ti mesh cathode can effectively reduce TN released from sediment to overlying water with a total removal amount of 151.82 ± 21.69 mg, which was approximately 3 times higher than control group (30.21 ± 13.73 mg) ($p = 0.005$) (Figs. 3a and 3b). There are two main reasons for the decrease of N in the sediment and overlying water are: 1) the added RuO_2-IrO_2/Ti anode in the sediment could directly or indirectly oxidize NH_3-N to N_2 in the sediment and overlying water; Direct electro-oxidation involves NH_3-N adsorption onto the anode surface, as well as its direct electron transfer, and ultimately, its conversion into N_2 (Sun et al. 2020). Indirect electro-oxidation of NH_3-N when performed via anodic reaction forms an intermediate oxidant (e.g., Cl_2, HOCl, and ClO^-) as the 2.17 V was the theoretical voltage which could produce Cl_2, which would react with NO_2^- to NO_3^- and under the mole basic of 1.5 the N_2 could be produced and N_2H_2 and NH_2OH would be produced lower the mole basic of

1.5 (Snoeyink & Jenkins 1980; Kapałka et al. 2010). Moreover, the results were consistent with the adsorption and degradation of NH_3-N and organic nitrogen by the IrO_2 anode as reported by Kim et al (2005) and Ihara et al (2006); 2) when the voltage was higher than 1.23 V, the hydrogen electron acceptor produced by Ti mesh cathode was used for hydrogen autotrophic microorganisms to remove NO_3^--N in the sediment (Xing et al. 2020); and 3) the added Ti mesh cathode contributed to the electro-reduction of NO_3^--N to N_2; and 4) the nitrification of NH_3-N by nitrifying bacteria was promoted in a higher DO and pH environment (Fig. S2).

3.2 Phosphorus removal by electrolysis in sediment and overlying water

P in the polluted river sediment can release to the overlying water as bioavailable P mainly was PO_4^{3-}-P in overlaying water under anaerobic conditions which leads to the continuous eutrophication of a water body (Wu et al. 2019). To further explore the effects of P removal and migration in polluted river sediment and overlying water resulting from electrochemical reaction, the concentrations of NH_4Cl-P (loosely-bound P plus labile organic P) and NaOH-P (Fe-/Al-bound P plus moderately resistant organic P) in polluted river sediment, TP and PO_4^{3-}-P concentrations in overlying water were measured and the results were shown in Fig. 4, after 20 days electrolysis the concentration of NH_4Cl-P increased from 23.71 ± 0.32 mg/kg to 63.83 ± 3.32 mg/kg, and then gradually decreased to 15.91 ± 0.20 mg/kg on 32nd day which was lower than that of the control group (22.02 ± 1.03 mg/kg). The concentration of NaOH-P in the sediment decreased from 64.23 ± 3.85 mg/kg on 1st day to 23.64 ± 1.35 mg/kg on 32nd day (Figs. 4a and 4b). Thus, the electrolysis enhanced the decrease of NaOH-P, and initial promoted the concentration of NH_4Cl-P which was weakly bound P easily absorbed by plants and microbes, and then decreased in the sediment. The increase of weakly adsorbed P of NH_4Cl-P in the initial may was caused by the increased pH value which increase the release rate of NaOH-P as OH^- ions can exchange with HPO_4^- in Fe-/Al-bound P (Fig. S2) (Temporetti et al. 2019).

The changes of P concentration in the overlying water of electrolysis system were shown in Figs. 4c and 4d. The concentrations of TP and PO_4^{3-}-P decreased rapidly within the first five days of electrolysis, the TP concentration decreased from 0.34 ± 0.009 mg/L to about 0.076 ± 0.01 mg/L and the PO_4^{3-}-P concentration in the overlying water decreased from 0.14 ± 0.005 mg/L to nearly 0 mg/L. the TP removal amount was 0.16 ± 0.005 mg on the end of the experiment (Fig. 5). As for control group, In the first nine days the was a sustained P released from the sediment to the overlying water and the TP concentration increased from 0.19 ± 0.05 mg/L to 0.44 ± 0.023 mg/L, and the PO_4^{3-}-P increased from 0.079 ± 0.026 mg/L to 0.45 ± 0.035 mg/L on 9th day which mean that the sediment was a P sink, and after the release period of P, the concentrations of TP and PO_4^{3-}-P in overlying water gradually decreased and there was only about 0.06 ± 0.003 mg PO_4^{3-}-P deposited from overlying water to the sediment at the end

of the experiment (Fig. 5). It can be seen that use electrolysis method to remove PO_4^{3-}-P is reasonable when the P release from sediment is high.

The combined electrode pair of Mg−Al alloy anode and Ti mesh cathode electrode pair and the other electrode pair of RuO_2-IrO_2/Ti mesh anode and Ti mesh cathode can effectively remove TP and PO_4^{3-}-P. During the electrolysis, the anode of Mg−Al alloy could produce Mg^{2+} and Al^{3+} ions which might be fixed by the formation of magnesium phosphate, calcium phosphate, hydroxyapatite (HAP, $Ca_{10}(PO_4)_6(OH)_2$), and other precipitates in the sediment-overlying water interface during the electro-coagulation (Fig. S4) (Jiang et al. 2019). Simultaneously, partial PO_4^{3-}-P was deposited on the Ti mesh cathode through the electro-deposition the he product of P may be HAP, Mg (NH_4) $PO_4 \cdot 6H_2O$ (MAP) and other compounds as another PO_4^{3-}-P removal way (Table S1) (Lei et al. 2018; Feijoo et al. 2020). After the electrolysis reaction, the concentration of PO_4^{3-}-P in the sediment and overlying water interface precipitates was 129.36 mg/kg, and concentration of precipitates on the Ti mesh cathode surface was 433.37 mg/kg. Thus, electrolysis proved to be an easy method to undertake for P removal via electro-coagulation and electro-deposition, which had higher removal rates.

3.3 Effect of electrolysis on bacterial community in sediment

To further explore the bioremediation mechanism of polluted river sediment driven by electrolysis, we used HTS to investigate the structural changes in the bacterial community's polluted river sediment after electrolysis. There were 6143 OTUs in the control group and 5474 OTUs in the electrolysis group and the same number of OTUs shared by the two groups was 2009 which accounted for 26.44% of the total OTU number in these two groups (Table S2). This indicated that the sediment-based microorganisms underwent some obvious changes after electrolysis. Meanwhile, the OTU number and the Shannon value of the bacterial community in the electrolysis sediment was lower while the Simpson value was higher, indicating that after electrolysis, the diversity and abundance of microorganisms decreased obviously comparing with the control system (Table S2). After the 32 day electrolysis treatment, the highest genera of bacteria in the electrolytic sediment were *Hydrogenophohaga* (23.75%), *Sulfuricurvum* (4.89%), *Proteiniclasticum* (3.29%), *Silanimonas* (2.3%) and *Gemmobacter* (2.29%); the highest genera of bacteria in the sediment of the control group were *Thiobacillus* (4.46%), *Dehloromonas* (2.63%), *Povalibacter* (2.21%), *Ornatilinea* (2.12%) and *GP6* (2.12%) (Fig. 6). There were about 15 genera of bacteria that were different from that in the control group with a *p* value lower than 0.001 (Fig. 6a).

It is obvious that after electrolysis, the community structure of the polluted river sediment's bacteria had changed significantly. Additionally, *Hydrogenophohaga* was the dominant species which was the hydrogen autotrophic denitrifying bacteria after electrolysis when the current voltage was 5 V−6 V. This voltage is higher than the theoretical voltage value of water (1.23 V), so the hydrogen gas was produced by Ti mesh cathode which served as the electronic donor of hydrogen autotrophic denitrifying bacteria, which was conducive to the further growth of hydrogen autotrophic denitrifying bacteria. This not only

enhanced the denitrification process but can also expedited NO_3^--N removal (Gao et al. 2017). In addition, compared with the control group, the abundances of denitrifying bacteria such as *Proteiniclasticum* and *Gemmobacter* in the electrolyzed sediment were also increased after electrolysis treatment, which was one of the reasons for the NO_3^--N concentration decreased in the electrolytic sediment. While the abundances of *Thiobacillus* decreased, which was the predominance bacteria in different sulfur-based denitrification systems after treating nitrate-contaminated wastewater (Yang et al. 2018). The sulfur-oxidizing bacteria *Sulfuricurvum* increased after electrolysis and proved capable of reducing the concentration of SO_4^{3-} in the polluted sediment. Thus, the electrolysis of the Mg−Al alloy anode paired with a Ti mesh cathode and the RuO_2-IrO_2/Ti anode paired with a Ti mesh cathode could reduce the diversity of the bacterial community in the sediment and greatly changed the community structure of the sediment (Fig. 6b).

Electrolysis had an enhanced effect on denitrifying bacteria such as hydrogen autotrophic denitrifying bacteria which could reduce the NO_3^--N in polluted river sediment. Simultaneously, the increased concentration of sulfur-oxidizing bacteria meant that electrolysis was producing more oxygen which heighted the oxygen content of sediment thereby promoting the transformation of highly toxic reduced sulfur to harmless oxidized sulfur. This transformation inferred that the electrolysis process reduced the toxicity of sulfur and, therefore, may be a superior way to remove S which needs further study.

3.4 Analysis of secondary release of nitrogen and phosphorus from sediment after electrolysis

To further verify the secondary release of NH_3-N and PO_4^{3-}-P from the electrolysis sediment, the electrolyzed 13-day and 32-d sediments were used for the secondary release experiment. As shown in Figs. 7a and 7b, the concentration of NH_3-N in the overlying water of the electrolysis group and the control group initially increased and then stabilized. On the 5th day, the concentration of NH_3-N secondary release from the sediment was 6.20 ± 1.02 mg/L in the 13-day electrolyzed sediment, 4.31 ± 0.92 mg/L in the electrolyzed 32-d sediment, and 2.84 ± 0.69 mg/L in the control group's non-electrolytic sediment. The total released NH_3-N was 22.6 ± 0.54 mg/L from the 13-day electrolyzed sediment, 12.1 ± 0.36 mg/L from the 32-day electrolyzed sediment, and 17.0 ± 0.41 mg/L from the non-electrolytic control group. When the electrolysis time was 32 days, the release of NH_3-N was the lowest. However, no significant difference was reported in the NH_3-N release between the three groups.

As the PO_4^{3-}-P released from the 32-day electrolyzed sediment, the secondary release amount was lower than that of 13-day electrolyzed sediment. The total secondary released PO_4^{3-}-P amounts were 1.43 ± 0.02 mg/L from the 13-day electrolyzed sediment, 0.80 ± 0.03 mg/L from the 32-day electrolyzed sediment, and 0.83 ± 0.03 mg/L from the non-electrolytic control group. The short period electrolyzed sediment easily released PO_4^{3-}-P to the overlying water mainly due to the strong alkaline reduction and electric field effect caused by electrolysis that promoted the transformation of NaOH-P and NH_4Cl-P to

PO_4^{3-}-P, which lead to more PO_4^{3-}-P entering to the overlying water. Thus, enhanced the electrolysis time could reduce the secondary release of NH_3-N and PO_4^{3-}-P to the overlying water.

4. Conclusions

This work elucidated the advantages of electrolysis for removing N and P removal. The device's low-voltage (5 V–6 V) and current 40 mA electrolysis proved efficient to remove N and P. Electrolysis can remove part of N through direct oxidation-reduction on the electrode surface, while indirectly changing the denitrifying bacteria, sulfur-oxidizing bacteria and sulfide bacteria community structure in the sediment by producing oxygen and hydrogen. NH_3-N was effectively removed through the electrochemical oxidation process by the paired RuO_2-IrO_2/Ti and Ti mesh anode. NO_3^--N was directly removed through electrochemical reduction by a Ti mesh cathode and indirectly through the role of hydrogen denitrifying bacteria in the sediment. Approximately 151.82 ± 21.69 mg TN were removed by electrolysis reaction compared to 30.21 ± 13.73 mg TN removed in control group; thus, the electrolysis proved five times more effective than the control group. In the HTS analysis, *Hydrogenophohaga* was the dominant species along with the increased concentration of sulfur-oxidizing bacteria and the decreased concentration of sulfide bacteria, which means electrolysis promoted the transformation of highly toxic reduced sulfur to harmless oxidized sulfur thereby reducing the toxicity of sulfur. Test results showed that electrolysis promotes the removal of PO_4^{3-}-P and TP into the overlying water via Mg^{2+} and Al^{3+} ions from a Mg–Al alloy anode; moreover, some PO_4^{3-}-P was deposited on the Ti mesh cathode. Through the analysis of the secondary release of N and P from sediment after electrolysis, the electrolysis interval of polluted river sediment was five days after the initial electrolysis which provides more economic viability for the proposed electrolysis-driven bioremediation approach.

Declarations

Not applicable

Authors' contributions

Yan Gao, Liuyan Yang provided the original proposal and designed the experiment. Sanshan Li and Chaoqun Zheng conducted the experiment, water quality tests and characterization of the microbial community, diversity, and statistical analysis. Yan Gao wrote the manuscript with the help of Liuyan Yang and Tangming Ma. All of the authors have reviewed the manuscript, and provided valuable suggestions.

Funding

Conflicts of interest/Competing interests

The authors declare no competing interests.

References

Bellagamba M, Viggi C C, Ademollo N et al (2017) Electrolysis-driven bioremediation of crude oil-contaminated marine sediments. New Biotechnol 38: 84–90

Caporaso J G, Bittinger K, Bushman F D et al (2010a) PyNAST: a flexible tool for aligning sequences to a template alignment. Bioinformatics 26: 266–267

Caporaso J G, Kuczynski J, Stombaugh J et al (2010b) QIIME allows analysis of high-throughput community sequencing data. Nat Methods 7: 335–336

Cecconet D, Sabba F, Devecseri M et al (2020) In situ groundwater remediation with bioelectrochemical (BES) systems: A critical review and future perspectives. Environ Int 137: doi:10.1016/j.envint.2020.105550

DeSantis T Z, Hugenholtz P, Larsen N et al (2006) Greengenes, a chimera-checked 16S rRNA gene database and workbench compatible with ARB. Appl Environ Microbiol 72: 5069–5072

Edgar R C (2010) Search and clustering orders of magnitude faster than BLAST. Bioinformatics 26: 2460–2461

EPA (2001) Methods for collection, storage and manipulation of sediments for chemical and toxicological analyses: technical manual. United States environmental protection office of water 4305: EPA-823-F-01-023

Feijoo J, Fort R, Gomez-Villalba L S et al (2020) Electroprecipitation of Magnesium and Calcium Compounds for Weathering Protection of Ornamental Rocks. Cryst Growth Des 20: 2337–2355

Gao Y, Xie Y, Zhang Q et al (2017) Intensified nitrate and phosphorus removal in an electrolysis-integrated horizontal subsurface-flow constructed wetland. Water. Res 108: 39–45

Gao Y, Xie Y, Zhang Q et al (2016) High performance of nitrogen and phosphorus removal in an electrolysis-integrated biofilter. Water Sci Technol 74: 714–21

Huang G, Liu F, Yang Y et al (2015) Ammonium-nitrogen-contaminated groundwater remediation by a sequential three-zone permeable reactive barrier (multibarrier) with oxygen-releasing compound (orc)/clinoptilolite/spongy iron: column studies. Environ Sci Pollut R 22(5): 3705–3714

IBM Corp, Released (2015) IBM SPSS Statistic for Windows. Version 23.0 Armonk NY IBM Corp

Ihara I, Umetsu K, Kanamura K et al (2006) Electrochemical oxidation of the effluent from anaerobic digestion of dairy manure. Bioresour Technol 97(12): 1360−1364

Jiang X, Yan B, Chen J et al (2019) Transport and retention of phosphorus in soil with addition of Mg-Al layered double hydroxides: Effects of material dosage, flow velocity and pH. Chem Eng J 378:122154

Kapałka A, Katsaounis A, Michels N L et al (2010) Ammonia oxidation to nitrogen mediated by electrogenerated active chlorine on Ti/PtOx-IrO$_2$. Electrochem Commun 12(9): 1203−1205

Kim K W, Kim Y J, Kim I T et al (2005) The electrolytic decomposition mechanism of ammonia to nitrogen at an IrO$_2$ anode. Electrochim Acta 50 (22): 4356−4364

Kuypers M M M, Marchant H K, & Kartal B (2018) The microbial nitrogen-cycling network. Nat Rev Microbiol 16 (5): 263−276

Lei Y, Remmers J C, Saakes M et al (2018) Is There a Precipitation Sequence in Municipal Wastewater Induced by Electrolysis? Environ Sci Technol 52: 8399−8407

Li Z, Sun P, Zhang R et al (2020) A novel magnesium-based oxygen releasing compound for eutrophic water remediation. Chemosphere 251: 126326

Ministry of Environmental Protection, People's Republic of China (MEPPRC) (2011) Soil-determination of total phosphorus by alkali fusion−Mo-Sb anti spectrophotometric method. Chinese Standard HJ 632-2011

OriginLab. Origin Version 2018. OriginLab Corporation Northampton MA USA https://www.originlab.com/2018

Pistocchi C, Tamburini F, Gruau G et al (2017) Tracing the sources and cycling of phosphorus in river sediments using oxygen isotopes: Methodological adaptations and first results from a case study in France. Water Res 111: 346−356

Psenner R, Bostrom B, Dinka M et al (1988) Fractionation of phosphorus in suspended matter and sediment. Arch Hydrobiol Beih Ergeb Limnol 30: 98−103

Seifan M, Samani A K, & Berenjian A (2017) A novel approach to accelerate bacterially induced calcium carbonate precipitation using oxygen releasing compounds (ORCs). Biocatal Agric Biotechnol 12: 299−307

Snoeyink V L, Jenkins D (1980) Water chemistry, 1st edn. Wiley. New York, pp 392—393.

Song Q, Li M, Wang L et al (2019) Mechanism and optimization of electrochemical system for simultaneous removal of nitrate and ammonia. J Hazard Mater 363: 119−126

Sun D, Hong X, Wu K et al (2020) Simultaneous removal of ammonia and phosphate by electro-oxidation and electrocoagulation using RuO_2-IrO_2/Ti and microscale zero-valent iron composite electrode. Water Res 169: 115239

Tang J, Guo J, Fang F et al (2013) Oxidation behavior of ammonium in a 3-dimensional biofilm-electrode reactor. J Environ Sci 25(12): 2403–2409

Temporetti P, Beamud G, Nichela D et al (2019) The effect of pH on phosphorus sorbed from sediments in a river with a natural pH gradient. Chemosphere 228: 287–299

The Ministry of Environmental Protection (MEP of China) (2002) Water and wastewater monitoring and analysis method, 4th edn. China Environmental Science Press, Beijing, pp243–285

Wang J, Chen J, Yu P et al (2020) Oxygenation and synchronous control of nitrogen and phosphorus release at the sediment-water interface using oxygen nano-bubble modified material. Sci Total Environ 725: 138258

Wang Q, Garrity G M, Tiedje J M et al (2007) Naive Bayesian classifier for rapid assignment of rRNA sequences into the new bacterial taxonomy. Appl Environ Microbiol 73: 5261–5267

Wu Z, Wang S, & Ji N (2019) Phosphorus (P) release risk in lake sediment evaluated by DIFS odel and sediment properties: A new sediment P release risk index (SPRRI). Environ Pollut 255: 113279

Xing W, Wang Y, Hao T et al (2020) pH control and microbial community analysis with HCl or CO_2 addition in H_2-based autotrophic denitrification. Water Res 168: 115200

Yang J, Lin F, Yang L et al (2015) A bench-scale assessment for phosphorus release control of sediment by an oxygen-releasing compound (ORC). J Environ Sci Heal A 50: 49–59

Yang Y, Gerrity S, Collins G et al (2018) Enrichment and characterization of autotrophic *Thiobacillus* denitrifiers from anaerobic sludge for nitrate removal. Process Biochem 68: 165–170

Yao J, Zhou M, Wen D et al (2016) Electrochemical conversion of ammonia to nitrogen in non-chlorinated aqueous solution by controlling pH value. J Electroanal Chem 776: 53–58

Zhai S, Ji M, Zhao Y et al (2020) Shift of bacterial community and denitrification functional genes in biofilm electrode reactor in response to high salinity. Environ Res 184: 109007

Zhang L, Yue Q, Yang K et al (2018) Analysis of extracellular polymeric substances (EPS) and ciprofloxacin-degrading microbial community in the combined Fe-C micro-electrolysis-UBAF process for the elimination of high-level ciprofloxacin. Chemosphere 193: 645–654

Zhao Y, Bai Y, Guo Q et al (2019) Bioremediation of contaminated urban river sediment with methanol stimulation: Metabolic processes accompanied with microbial community changes. Sci Total Environ

653: 649–657