Denitrification in an integrated bioelectro-photocatalytic system

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Abstract

Since nitrate causes severe ecological and health risks, nitrate contamination of drinking water sources has become one of the most important water quality concerns all over the world. Photocatalytic reduction of nitrate to molecular nitrogen presents a promising approach to remove nitrate from drinking water sources. However, harmful intermediates like NO_2^- , NO , NO_2 and N_2O are usually formed, and metal loading or hole scavengers are generally needed to reduce the recombination of photo-generated electrons and holes, which will cause secondary pollution to drinking water. In this work, an efficient, selective and sustainable bioelectro-photocatalytic nitrate-reducing system by utilizing commercial TiO_2 nanoparticles P25 as the photocatalyst and bio-electrons from microbial metabolism as the hole scavenger is reported. In this system, bio-electrons extracted from organic substrates in bioanode are transferred to the photocathode through an external circuit for hole quenching. With the utilization of the residual photogenerated electrons, nitrate is completely reduced to nitrogen without accumulation of harmful nitrite or ammonium. The experimental results and the mechanistic analysis using the first-principles density functional theory calculations demonstrate that toxic by-products like nitrite or ammonium will not be accumulated in this system. Thus, this approach has a great potential for sustainable remediation of nitrate-contaminated drinking water sources.

Keywords: Bio-electrons; Denitrification; Photocatalysis; Remediation; Titanium dioxide

1. Introduction

Nitrate (NO_3^-) is one of the most widespread contaminants in surface water and subsurface aqueous environments, resulting from wide fertilizer use, sewage/livestock wastewater discharge, and landfill leachates, etc (Jensen et al., 2014; Liu et al., 2007; Senn and Hemond, 2002). Nitrate accumulation in water bodies leads to a series of ecological and health problems. For example, algae bloom frequently occurs in freshwaters or estuarines at high nitrate concentrations (Galloway et al., 2008; Lewis et al., 2011). Especially, high contents of nitrate and its reduction intermediates, e.g., nitrite (NO_2^-), in drinking water sources have been demonstrated to cause methemoglobinemia and cancer (Addiscott and Benjamin, 2004; Lundberg et al., 2004). Thus, the World Health Organization has recommended the concentration limit of 50 mg/l NO_3^- and 3 mg/l NO_2^- in drinking water. However, their actual nitrate concentrations in drinking water sources, especially in many developing countries like China, mostly far exceed this limit. In the north China plain region where groundwater is a main drinking water source, the groundwater NO_3^- content reaches up to 160 mg/l, severely threatening drinking water security. Thus, remediation technologies for efficient nitrate elimination without accumulation of harmful intermediates are greatly needed (Foley et al., 2010; Wunderlin et al., 2012).

Currently, physicochemical methods such as ion exchange, reverse osmosis, and electrodialysis have been used to remove nitrate from water, but they generally require further treatment of the concentrated solution, cause the losses of other

useful elements (Chaplin et al., 2012; Li et al., 2016; Zhang et al., 2014), and usually consume high energy. Biological approaches are useful for nitrate removal from wastewater, though their application for water environmental remediation is limited, due to the challenges of performance maintenance and secondary pollution associated with carbon source supply and the pathogen concerns (Cardoso et al., 2016; Cheng et al., 2012; Pereira et al., 2014). Performance of a biological treatment system is affected by many factors, especially microbial communities involved in the system. The complexity and variations of microbial communities cause the instability and difficulty in performance maintenance of biological treatment systems (Barwal et al., 2014). In addition, such bioconversion is usually incomplete, resulting in the formation of even more harmful by-products such as NO_2^- , NO, NO_2 and N_2O (Peng et al., 2016; Upadhyaya et al., 2012).

In comparison, photocatalytic method presents a more environmentally-benign option, which enables efficient reduction of nitrate to N_2 without introducing any chemicals or bacteria and requiring any post-treatment (Choe et al., 2013; Hoffmann et al., 1995; Sa et al., 2009; Zhang et al., 2005). In particular, titanium dioxide (TiO_2), attributed to its excellent optical and electronic properties, chemical stability, nontoxicity, and low cost, has been extensively investigated for such purposes (Hoffmann et al., 1995). However, TiO_2 has a relative poor photocatalytic reduction activity because of the rapid recombination of the photogenerated electrons and holes. Nano-structured TiO_2 can even catalyze nitrate production from nitrogen and oxygen (Yuan et al., 2013). To reduce the recombination of electron-hole pairs, metal doping

or hole scavengers are usually applied. However, this incurs extra costs and arouses environmental concerns on the dosed chemicals (Chaudhuri and Lovley, 2003). In addition, due to a low reaction selectivity, harmful by-products such as nitrite or ammonium are usually generated (Chaudhuri and Lovley, 2003; Cheng et al., 2012). These limitations highlight the needs for more selective, cost-effective and sustainable photocatalytic processes to reduce nitrate to N_2 completely.

In this work, we report a novel bioelectro-photocatalytic denitrification system by utilizing commercial TiO_2 nanoparticles P25 as a nitrate-reducing photocatalyst and using bio-electrons derived from microbial metabolism as a cost-effective and sustainable hole scavenger (Chaudhuri and Lovley, 2003; Rabaey and Rozendal, 2010). The geometry structures of the reactants, intermediates, and products adsorbed on the abundant (101) surface of TiO_2 nanoparticles in the reaction process in water solution were energy-minimized, and the mechanisms of the highly-selective nitrate reduction were elucidated. This bioelectro-photocatalytic system synergically utilizes both bioenergy and photoenergy for efficient and complete nitrate reduction, and has a potential for nitrate removal from drinking water sources and water environmental remediation.

2. Materials and methods

2.1. Reactor construction and operation

The system contained a carbon paper bioanode (3×7 cm, GEFC Co., China) and a TiO₂ photocathode, separated by an ion-exchange membrane and electrically connected through a 10 Ω external resistance. The bioanode was pre-cultivated in a microbial fuel cell for 2 months with anaerobic sludge as inoculums and acetate as substrate, thus a consortium of electrochemically active microorganisms was well enriched in the anode chamber. The photocathode was prepared by painting TiO₂ (P25, Degussa Co., Germany) on a carbon paper with 0.1 g of TiO₂ loaded (Yuan et al., 2013). The anode chamber was filled with 80 mL of autoclaved medium containing (in 1 L of phosphate buffer, pH 7.0): NaAc, 100 mg; NH₄Cl, 310 mg; KCl, 130 mg; CaCl₂, 10 mg; MgCl₂ · 6H₂O, 20 mg; NaCl, 2 mg; FeCl₂, 5 mg; CoCl₂ · 2H₂O, 1 mg; MnCl₂ · 4H₂O, 1 mg; AlCl₃, 0.5 mg; (NH₄)₆Mo₇O₂₄, 3 mg; H₃BO₃, 1 mg; NiCl₂ · 6H₂O, 0.1 mg; CuSO₄ · 5H₂O, 1 mg; ZnCl₂, 1 mg (Sun et al., 2008), whereas the cathode chamber was equipped with a quartz glass window (2×4 cm) and filled with 0.25 mM nitrate solution (80 mL). Nitrite and ammonia nitrogen were not added in the initial media.

A 30-W low-pressure mercury lamp was used as the light source. The lamp was fixed outside the cathode chamber, which had a distance of 5 cm from the photoelectrode surface. Na₂SO₄ solution (0.01 M) was used as the catholyte and constantly stirred by a magnetic stirrer at a speed of 200 rpm. In all the tests, a mixed catholyte of 0.25 mM nitrate solution and 0.01 M Na₂SO₄ was used. All batch tests were carried out in duplicate at ambient temperatures of around 20 °C.

The current was continuously recorded directly with a potentiostat (CHI660C,

Chenhua Inc., China) connected to a computer. The current density was calculated relative to the projected surface area of the bioanode from the current data. The photo-electrochemical response of the system was also analyzed using the potentiostat. The morphology of the prepared TiO₂-loaded photoelectrodes was imaged using scanning electron microscopy (SEM) (FEI Co., the Netherlands), and its crystal structure was characterized by X-ray diffraction (XRD, X' Pert PRO, Philips Co., the Netherlands). Both nitrate and nitrite concentrations were analyzed using ion chromatography (ICS-1000, Dionex Co., USA) with a conductometric detector set at 43 mA. A solution containing 8 mM Na₂CO₃ and 1 mM NaHCO₃ was used as eluent. Ammonia concentration after the Nessler reaction was determined by spectrophotometry (UV-2401, Shimadzu Co., Japan) at a wavelength of 420 nm. Gaseous products (N₂ and NO) was measured by a gas-chromatograph (6890, Agilent Co., USA) equipped with an electron-capture detector, and an FTIR (TENSOR 27, Bruker Co., Germany) equipped with a mercury-cadmium-telluride detector for analysis of the potential NO_x intermediates (Yuan et al., 2013). All reagents used were of analytical grade unless otherwise specified. All solutions were prepared using deionized water (Millipore Co., USA).

TiO₂ P25 contains anatase and rutile phases in a ratio of about 3: 1 (Ohno et al., 2001). The mechanisms of intermediates formation on the (101) surface of TiO₂ P25 were explored by the first-principles DFT calculations.

2.2 Quantum chemical calculations

The anatase TiO₂ (101) surface was modeled as a periodic slab with layers of oxide, the vacuum region is the space between slabs and has a width of about 10 Å. In this model, the vacuum region was usually used to represent the (101) surface of TiO₂. Each slab contains 18 Ti atoms and 36 O atoms. The (101) surface consists of Ti atoms with 5- or 6-fold coordination (Ti-5c, Ti-6c) and O atoms with 2- or 3-fold coordination (O-2c, O-3c) (He et al., 2009). Nitrate, hydrated proton, water and product molecules were adsorbed on one side of the slab only (Gong et al., 2006; He et al., 2009).

First-principles DFT calculations were performed with the generalized gradient approximation (GGA) using plane-wave basis sets and ultrasoft pseudopotentials, as implemented in the CASTEP module (Segall et al., 2002) of Materials Studio. The exchange-correlation energy and potential were described self-consistently using the Perdew-Wang 91 (PW91) functional. Depending on the unit cell size and shape, the Brillouin zone integration was performed with a variable number of *k*-points generated by Monkhorst-Pack algorithm. The linear synchronous transit (LST)/quadratic synchronous transit (QST) methods were used to search transition state structure and calculate energy barrier.

Thermodynamic properties of the optimized structures were investigated with DMol³ module (Delley, 1996; 2000). The electron density functional was treated by the GGA with the exchange-correlation function of Perdew, Burke, and Ernzerhof (PBE). Double precision numerical basis sets combined with d-functions (DND) were used to describe the valence electrons, while all-electron core treatment was utilized

to describe the core electrons.

3. Results and discussion

3.1. Catalytic denitrification performance

The XRD spectrum of the photoelectrode in our work (Fig. 1a) was similar to that of the commercial TiO₂ nanoparticles P25 (Yuan et al., 2013). The TiO₂-coated electrode displayed cracked-mud structure (Fig. 1a). Clearly, the TiO₂ coating increased the electrode surface area without changing the electrode structure.

The current response vs time curves of the denitrification system with or without UV irradiation are shown in Fig. 1b. A higher current, with an average increase of $1.6 \pm 0.3 \text{ mA/m}^3$, was achieved under UV irradiation compared to the non-irradiated control. The change of electric current in response to irradiation occurred rapidly and repeatedly in multiple cycles, suggesting a stable and sensitive light response of the system.

Fig. 2a illustrates the changes of nitrate, nitrite, and ammonium concentrations in the bioelectro-photocatalytic denitrification system. The nitrate concentration decreased by about 60% within 24 h and the decreasing rate slowed down, confirming a high denitrification efficiency of this system. By contrast, neither nitrite nor ammonium concentration exhibited a significant increase, indicating a high selectivity of this system towards nitrate reduction to N₂ without other harmful by-products.

In addition, a comparison between the denitrification efficiencies in the

electrochemical, photocatalytic and bioelectro-photocatalytic systems (Fig. 2b) reveals much better performance of the bioelectro-photocatalytic system. This system had a significantly higher denitrification rate constant ($0.0308 \pm 0.0053 \text{ h}^{-1}$) than the individual photocatalytic ($0.0011 \pm 0.0015 \text{ h}^{-1}$) and electrochemical ($0.0072 \pm 0.0004 \text{ h}^{-1}$) systems (Fig. 2b), indicating that the bio-electrons could be effectively utilized to scavenge the photogenerated holes and improve the photocatalytic denitrification efficiency. Intermediates concentrations in the pure photocatalytic system are shown in SI (Fig. S3). The accumulation of nitrite and ammonia was also observed.

3.2 Catalytic reduction mechanisms

The photocatalytic denitrification reaction can be described as below: .

No nitrite was generated in the bioelectro-photocatalytic denitrification system, indicating a different reaction pathway compared with the conventional denitrification reactions (Supporting Information (SI), Fig. S1). The energy-minimized geometry structures of the reaction intermediates in the bioelectro-photocatalytic system are illustrated in Fig. 3. A combination of linear synchronous transit (LST) and quadratic synchronous transit (QST) methods were used to find the transition state (TS) and energy barrier by setting the reaction pathway of the reactant and the product structures. The thermodynamic and kinetic characteristics of elementary steps are listed in Table 1, whereas the energy barrier (E_a) for each step with the transition states is given in Fig. 4.

Anatase TiO_2 (101) is the most stable and frequently exposed surface of anatase (Gong et al., 2006; Gong et al., 2008). The initial step of this photocatalytic process is the adsorption of nitrate intermediate ($(\text{NO}_3^-)_{\text{ad}}$) onto the Ti atom of TiO_2 (101) surface, because 4 orbitals in 3d of Ti (the electronic arrangement is $[\text{Ar}]3d^24s^2$) are empty and could accept the lone pair electrons of O atom in NO_3^- to form Ti-O bond (Fig. 3b). The O_{ad} atom of $(\text{NO}_3^-)_{\text{ad}}$ connected on the surface is located on the site of lattice O in the above layer of TiO_2 (101) surface (SI, Fig. S2). The negative Gibbs free energy change ($\Delta_r G_m^\circ = -6.908 \text{ kcal/mol}$) indicates that the adsorption of NO_3^- is thermodynamically favored for the photocatalytic pathway. Then, the protons from 4 hydronium cations are added on the other O atoms of $(\text{NO}_3^-)_{\text{ad}}$ to form OH groups (Fig. 4c), which corresponds to the process from State 2 to State 3 (SI, Fig. S1) with a low energy barrier ($E_a = 1.658 \text{ eV}$).

The hydronium cation is a positively charged polyatomic ion with the chemical formula of H_3O^+ , a type of oxonium ion. This cation is often used to represent the nature of the proton in aqueous solution, which is highly solvated (bound to a solvent). Following the Step II, the intramolecular dehydration reaction between two the vicinal hydroxyl groups in State 3 occurs, resulting in State 4 (Fig. 3d). After that, the protons of 4 H_3O^+ cations bind with the O_1 and O_2 atoms of the adsorbed intermediates to form the structure with double bond between two N atoms ($\text{N}=\text{N}$) and 6 H_2O molecules (Fig. 3e). This process (from State 4 to State 5) has a more negative $\Delta_r G_m^\circ$ value, indicating that the protons attached on the O_1 and O_2 atoms with a high reactivity is energetically favorable and the bond cleavage of $\text{N}-\text{O}_1$ and $\text{N}-\text{O}_2$ occurs

spontaneously. Finally, molecular N_2 leaves the lattice oxygens (O_{ad}) on the (101) surface of TiO_2 (Fig. 3f), and the proton-coupled photogenerated electrons interact with the reactive O_{ad} to generate other two H_2O molecules to complete the photocatalytic denitrification reactions.

In the photocatalytic process, the negative $\Delta_r G_m^\ominus$ value of the overall reaction (Table 1) indicates that nitrate reduction on the TiO_2 (101) surface is thermodynamically favored at 1 atm and 298.15 K. The influence of temperature on $\Delta_r G_m$ of the overall photocatalytic denitrification pathway is also investigated. The $\Delta_r G_m$ of the overall reaction becomes more negative at higher temperatures (SI, Fig. S5), suggesting an elevated photocatalytic denitrification rate with the increasing temperature.

In the bioelectro-photocatalytic denitrification system established in this work, bio-electrons from the bioanode are efficiently utilized to restrain the recombination of the electrons and holes at the photocathode, resulting in a complete and selective denitrification. Expectedly, electrons generated from other origins, such as other fuel cell, photovoltaic cells or wind turbines, can also be used to drive this photocatalytic process. Thus, it provides a common way to increase the efficiency and selectivity of photocatalytic denitrification by coupling electron generation device with photocatalytic denitrification process.

The E_a of each step and the transition state structures of Steps IV and VI in our system are shown in Fig. 4. The total energy of the system changes with the evolution of intermediate structures in the multi-catalytic process. Step I is the adsorption

process for NO_3^- located on the surface. Moreover, the high E_a value of the rate-controlling step V (3.115 eV) indicates that the N-O bond cleavage in the structure shown in Fig. 3e should overcome a relatively high barrier to form the third valence bond between two N atoms. Elementary reactions with a negative E_a are typically barrierless reactions, in which the occurrence of reaction relies on the capture of molecules in a potential well. Thus, the negative E_a values of Steps IV and VI (Table 1) imply that the proton-coupled electron transfer reaction occurs without barrier, rapidly accomplishing the reduction reaction. Compared with the conventional denitrification mechanism shown in SI (Table S1 and Fig. S6), such a bioelectro-photocatalytic reaction pathway has a lower E_a (Fig. 4), suggesting that the complete photocatalytic reduction of nitrate to N_2 without accumulation of toxic intermediates is energetically feasible.

The complete photocatalytic reduction of nitrate to N_2 in our system should be attributed to the devoid of competitive nitrate absorption on the TiO_2 surface. Since the desirable product nitrogen contains two nitrogen atoms and nitrate has one nitrogen atom only, a pair of nitrate species has to meet on the active sites of the photocatalyst surface to form nitrogen. Compared with the dose of hole scavengers such as the most utilized formic acid (Zhang et al., 2005), whose adsorption energy (ΔE_{ad}) value on the TiO_2 (101) surface is almost five times larger than that of NO_3^- (Fig. 5), in our system less compounds will compete with nitrate for the absorption sites on the photocatalyst surface (Kormann et al., 1991). Thus, the scavenging of holes by the bio-electrons enables more homogeneous absorption and activation of

nitrate atoms pairs simultaneously for the final formation of N_2 from nitrate, resulting in the selective and efficient photocatalytic denitrification.

4. Conclusions

The bioelectro-photocatalytic system exhibits a high selectivity for photocatalytic reduction of nitrate to N_2 . This work demonstrates that bio-electrons from microbial metabolism of organic substrates could be effectively utilized to scavenge photogenerated holes and enhance photocatalytic denitrification. Compared with the conventional photocatalytic denitrification, this bioelectro-photocatalytic system has several advantages: (i) greater denitrification rate; (ii) higher selectivity and no accumulation of harmful by-products like nitrite or ammonium, (iii) no extra supply of hole scavenger, and (iv) cost effectiveness, because only a bioanode and a low-cost, facially-fabricated photocathode are required. In the future, improved design of a bioelectro-photocatalytic system and modification of photocatalyst are expected to further enhance the system performance. In addition, the bioelectricity-driven photoreduction approach demonstrated here might be also applicable to the effective reduction of other oxidative contaminants in water such as oxyanions, nitrosamines or even soluble arsenic, thus providing an efficient and environmentally-benign way for removing a variety of pollutants from water.

Acknowledgements

We thank the National Science Foundation of China (51208488, 21261160489 and 51538011) and the Collaborative Innovation Center of Suzhou Nano Science and Technology of the Ministry of Education of China for the support of this work.

Appendix A. Supplementary material

Supplementary data related to this article can be found online.

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Table 1 - Calculated thermodynamic and kinetic properties of the NO_3^- reduction in the bioelectro-photocatalytic denitrification system by LST/QST. The NO_3^- reduction on the anatase TiO_2 (101) surface at 298.15 K and 1 atm, and the energy barrier of every step corresponds to the bioelectron-assisted way shown in Fig. 4.

Step	$\Delta_r G_m^\ominus$	$\Delta_r H_m^\ominus$	$\Delta_r S_m^\ominus$	E_a
	(kcal/mol)	(kcal/mol)	(cal/mol K)	(eV)
$1 + 2\text{NO}_3^- \rightarrow 2$	-6.908	-11.145	-14.211	—
$2 + 4\text{H}^+ + 2\text{e}_{\text{cb}}^- \rightarrow 3$	-186.016	-178.117	26.494	1.658
$3 \rightarrow 4 + 2\text{H}_2\text{O}$	-68.227	-72.432	-14.105	2.265
$4 + 4\text{H}^+ + 4\text{e}_{\text{cb}}^- \rightarrow 5 + 2\text{H}_2\text{O}$	-269.880	-254.843	50.432	-2.559
$5 \rightarrow 6 + \text{N}_2$	20.949	19.797	-3.862	3.115
$6 + 4\text{H}^+ + 4\text{e}_{\text{cb}}^- \rightarrow 1 + 2\text{H}_2\text{O}$	-397.768	-391.597	20.697	-0.199
Total	-907.850	-888.337	65.445	—

Figure captions

Fig. 1. Characteristics of the photoelectrode. XRD spectrum (a) and SEM images (b) of the TiO_2 photoelectrode. A represents anatase and R represents rutile; and (c) Current vs time of the denitrification reactor under UV irradiation. Equilibrium current without UV irradiation was about 0.05 mA and peak current reached 0.4 mA with the UV light on.

Fig. 2. Performance of the bioelectro-photocatalytic denitrification system. (a) the change of nitrate, nitrite, and ammonium concentrations in denitrification process; (b) the relative concentration profiles of nitrate during denitrification process in the bioelectro-photocatalytic system (●: with bioanode to supply the bio-electrons but without irradiation; ○: with UV irradiation but without connecting to the bioanode; □: with both UV irradiation and bio-electrons supply from the bioanode).

Fig. 3. Optimized structures of every step in the photocatalytic denitrification process on the TiO_2 (101) surface. (a) Optimized structures of clean TiO_2 (101) surface; (b) NO_3^- adsorbed on the Ti atom with the O_{ad} atom on the site of lattice oxygen position; (c-e) Intermediates of reduction process with the partial top view in the insets; and (f) Nitrogen (N_2) generation.

Fig. 4. Comparison of energy barrier (E_a) for photocatalytic denitrification in the bioelectron-assisted way and the conventional pathway. The E_a of each step for NO_3^- reduction on the TiO_2 (101) surface with the transition state (TS) structures of Steps IV and VI in the bioelectro-photocatalytic system. Step I in the bioelectro-photocatalytic system is the adsorption of NO_3^- on the TiO_2 (101) surface.

Fig. 5. NO_3^- and formate (HCOO^-) adsorbed on Ti atom of the anatase TiO_2 (101) surface. (a) The calculated adsorption energy ($\Delta E_{\text{ad}} = E_{\text{TiO}_2} + E_{\text{NO}_3} - E_{\text{NO}_3\text{-TiO}_2}$) of NO_3^- on the TiO_2 (101) surface is 0.21 eV; and (b) the ΔE_{ad} of HCOO^- is almost five times larger than NO_3^-

Fig. 1

Fig. 2

Fig. 3

Fig. 4

Fig. 5

Graphical Abstract