Desalination and Water Treatment
www.deswater.com

doi: 10.1080/19443994.2015.1101024

57 (2016) 19411–19417
September

Taylor & Francis
Taylor & Francis Group

Nitrate removal by a combined bioelectrochemical and sulfur autotrophic denitrification (CBSAD) system at low temperatures

Dan Chen, Tao Dai, Hongyu Wang, Kai Yang*

School of Civil Engineering, Wuhan University, Wuhan 430072, China, emails: danchen1203@126.com (D. Chen), daitao2013whu@163.com (T. Dai), hongyuwangcc@126.com (H. Wang), kaiyangcc@126.com (K. Yang)

Received 6 April 2015; Accepted 23 September 2015

ABSTRACT

A newly combined bioelectrochemical and sulfur autotrophic denitrification system was developed and steadily operated to treat nitrate polluted water at 10˚C for 270 d. The parameters hydraulic retention time (HRT) and current were continuously adjusted to achieve better nitrate removal efficiency. Results demonstrated that the sulfur autotrophic denitrification part (S-part) efficiency was effectively improved by adjusting the parameter HRT, while current was the key control parameter for the electrochemical part (E-part) at low temperatures in this CBSAD reactor. This CBSAD system had the ability to maintain pH balance, and the effluent SO_4^{2-} concentration could be effectively controlled at cold temperatures in this combined autotrophic system.

Keywords: Nitrate; CBSAD; Cold temperature

1. Introduction

Nitrate removal approaches have received considerable attention in recent years because nitrate is harmful to human health and environment when it accumulates in water [1]. There are many physicochemical and biological methods for nitrate removal from water, but the most economical and effective method is biological since conventional physicochemical methods are not selective and always bring about secondary contaminations [2]. Biological methods contain heterotrophic denitrification and autotrophic denitrification. Heterotrophic denitrification takes organic matters as nutrition so that micro-organisms proliferate rapidly; however, this process always needs additional organic carbon source and causes secondary pollution. Compared to heterotrophic denitrification, autotrophic process uses inorganic carbon compounds such as CO_2 and HCO_3^- as denitrifying carbon source and inorganic matters such as H_2, reduced sulfur, and Fe^{2+} as electron donor for autotrophic micro-organisms to convert nitrate to nitrogen gas. This autotrophic technology arouses many researchers' interest because this process is cost-effective, non-polluting, and causes low biomass [3]. Among autotrophic methods, sulfur autotrophic denitrification and hydrogen autotrophic denitrification attract the most attention.

Sulfur autotrophic process consumes alkalinity resulting in decreased pH, so that lime is often needed to neutralize acid in this denitrification system. Moreover, this process will produce excessive SO_4^{2-} under high nitrate loading condition [4]. Although hydrogen is a clean and excellent electron donor for autotrophic denitrification, the easy explosibility and low solubility limit its wide application. Currently, bioelectrochemical

---

*Corresponding author.

process has been extensively researched because many studies demonstrate that the hydrogen which produces on the cathode surface can be effectively utilized for autotrophic denitrification and the utilization ratio can reach 100% [5].

Based on the characteristics of sulfur autotrophic denitrification and bioelectrochemical autotrophic denitrification, the combined bioelectrochemical and sulfur autotrophic denitrification process is an excellent approach for nitrate removal because H^+ caused by sulfur autotrophic part will be used as a precursor for hydrogen production through the electrochemical part, leading to a balanced pH value; and the effluent SO_4^{2-} will also be controlled since the electrochemical part shares the nitrate loading [4]. Wang and Qu [3] studied a combined bioelectrochemical and sulfur autotrophic denitrification system to treat nitrate polluted drinking water, and achieved 90–100% efficiency for 30 mg/L nitrate at a hydraulic retention time (HRT) range of 1.9–5.0 h and current range of 3–16 mA. Wan et al. [1] developed a combined sulfur autotrophic and proton-exchange membrane electrodialytic denitrification system for nitrate removal from groundwater, and they reported that HRT was the controlling factor for sulfur autotrophic part, while current was the controlling factor for proton-exchange membrane electrodialytic denitrification part. In addition, the system achieved 95.8% nitrate removal efficiency at an HRT of 2 h and a current of 350 mA with non-excessive SO_4^{2-} and neutral pH value. Tong et al. [6] used a heterotrophic/biofilm-electrode autotrophic denitrification reactor to deal with nitrate groundwater, and investigated that the optimal C/N and current density were 1.13 and 239.6 mA/m^2 for maximum performance.

Although this combined process achieves better efficiency under appropriate environment, biological approaches often face great challenges in the winter due to the fact that lower temperature can inhibit the activities of autotrophic micro-organisms and previous researches have proved that denitrification process was strongly inhibited at 10℃ [7]. The operating parameters such as HRT and current are often adjusted and optimized in order to improve the denitrification efficiency at cold temperatures [8]. In the present study, a combined bioelectrochemical and sulfur autotrophic denitrification system was developed and operated at 10℃ (typical water temperature during winter of Wuhan, China). The HRT and current were continuously adjusted to achieve better performance.

2. Materials and methods

2.1. Reactor operation

A schematic diagram of the combined bioelectrochemical and sulfur autotrophic denitrification (CBSAD) reactor used in this work is shown in Fig. 1. The total diameter and height of the CBSAD reactor were 100 and 300 mm. The E-part was on the upside, and the S-part was on the underside. E-part consisted of an anode (carbon rod, diameter 6 mm, height 150 mm), a cathode (graphite felt, specific surface area 2,631 m^2, which closed to the interior walls of the reactor), and a DC-regulated power supply (RPS-3005D, 0–30 V, 0–5 A) for electricity supply. The S-part was filled with sulfur granules, whose diameter was in the range of 5.0–8.0 mm.

At acclimation stage, the continuous-flow reactor was covered with anaerobic sludge from Erlangmiao Municipal Wastewater Treatment Plant, Wuhan, China

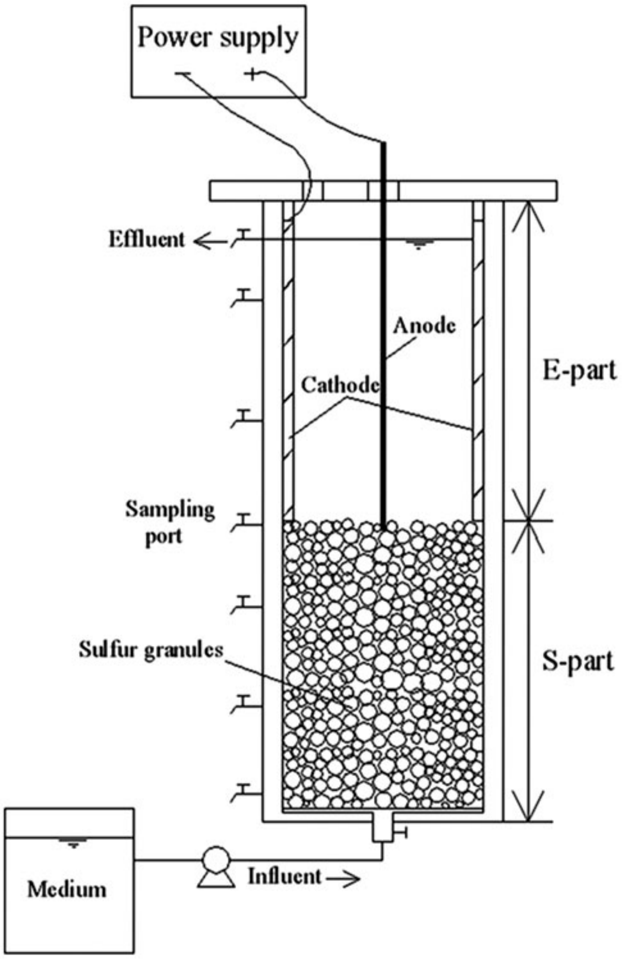

Fig. 1. Schematic representation of the CBSAD system.

and operated for 35 d at a current of 10 mA, an HRT of 21 h, nitrate concentration of 30 mg/L with synthetic wastewater containing NO_3^--N 30 mg/L, HCO_3^- 80 mg/L, $MgSO_4$ 10 mg/L, $MgCl_2$ 10 mg/L, $ZnCl_2$ 0.52 mg/L, $CoCl_2$ 1.90 mg/L, $MnSO_4$ 1.00 mg/L, $MgCl_2$ 0.25 mg/L, $NiCl_2$ 0.24 mg/L, $CuCl_2$ 0.29 mg/L, $FeSO_4$ 0.25 mg/L, $CaCl_2$ 0.50 mg/L, and Na_2MoO_4 0.36 mg/L. At last, the cathode surfaces and sulfur granules surfaces formed a dense biofilm and the CBSAD denitrification efficiency reached 90%.

Then the CBSAD reactor was continuously and steadily operated for 270 d at 10℃. The experimental design is shown in Table 1.

2.2. Analysis methods

Ammonia nitrogen (NH_4^+-N), nitrate (NO_3^--N), nitrite (NO_2^--N), and sulfate (SO_4^{2-}-S) were determined according to Standard Methods for the Examination of Water and Wastewater [9]. Nitrogen gas (N_2) and nitrous oxide (N_2O) were measured by an Agilent HP4890D gas chromatography. The pH was measured by a pH meter (PHS-3C, Kexiao Instrument, China). The water temperature was measured by a thermometer (TM827, Zhugongda Instrument, China).

3. Results and discussion

3.1. Effects of current on combined denitrification system at low temperatures

Fig. 2 shows the nitrate removal efficiency of this CBSAD reactor for different current values at 10℃ in Stage 1. The S-part nitrate removal efficiency presented no obvious change when the current increased from 20 to 400 mA, suggesting that current did not directly affect the S-part, which was in accordance with the study of Wang and Qu [3]. However, the

Table 1
Experimental design

Stage		Current (mA)	HRT (h)	Initial nitrate concentration (mg/L)
Stage 1 (day 1–60)	Day 1–10	20	9	30
	Day 11–20	50	9	30
	Day 21–30	100	9	30
	Day 31–40	200	9	30
	Day 41–50	300	9	30
	Day 51–60	400	9	30
Stage 2 (day 61–100)	Day 61–70	100	9	30
	Day 71–80	100	16	30
	Day 81–90	100	21	30
	Day 91–100	100	32	30
Stage 3 (day 101–140)	Day 101–110	100	9	60
	Day 111–120	100	9	60
	Day 121–130	100	9	60
	Day 131–140	100	9	60
Stage 4 (day 141–170)	Day 141–150	50	16	60
	Day 151–160	80	16	60
	Day 161–170	100	16	60
Stage 5 (day 171–200)	Day 171–180	150	16	100
	Day 181–190	200	16	100
	Day 191–200	300	16	100
Stage 6 (day 201–220)	Day 201–210	150	21	100
	Day 211–220	200	21	100
Stage 7 (day 221–250)	Day 221–230	150	21	150
	Day 231–240	200	21	150
	Day 241–250	300	21	150
Stage 8 (day 251–270)	Day 251–260	200	32	150
	Day 261–270	300	32	150

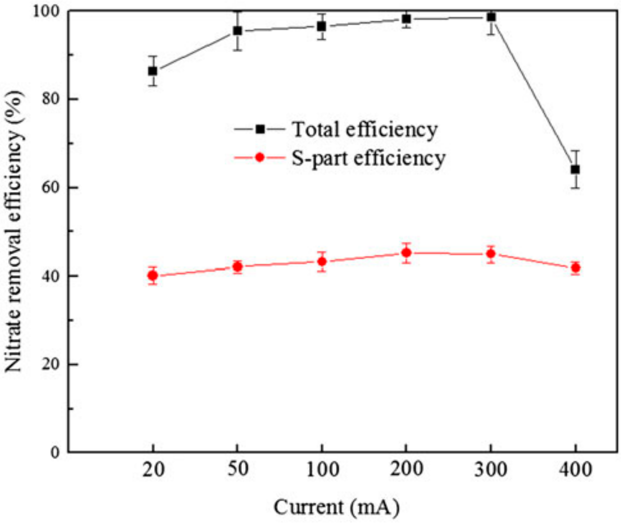

Fig. 2. Effects of current on CBSAD system at low temperatures.

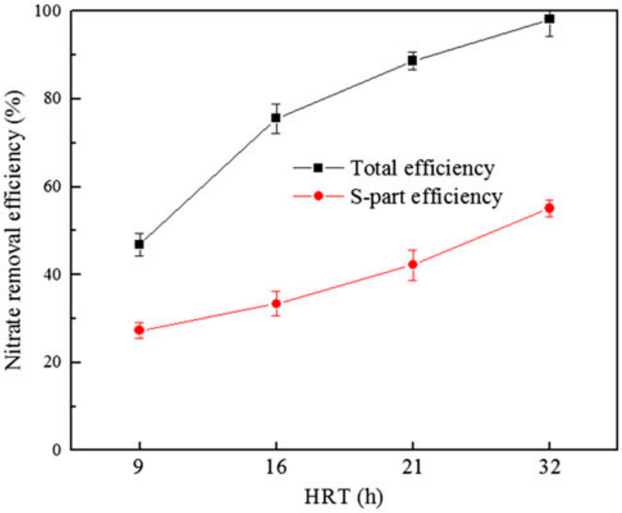

Fig. 3. Effects of HRT on CBSAD system at low temperatures.

total nitrate removal efficiency increased from 86.4 to 98.6% as the current increased from 20 to 300 mA. If the current was further increased to 400 mA, the total efficiency dramatically dropped to 64.1%. This result was attributed to the fact that the bioelectrochemical denitrification part was totally related to the current [10]. The E-part nitrate removal efficiency increased as the current increased from 20 to 300 mA due to the reason that more hydrogen would appear onto the cathode surface accelerating the activities of autohydrogenotrophic bacteria and autotrophic denitrification at higher current. However, excessive hydrogen would cause inhibition effect [11] on the bioelectrochemical denitrification part so that the efficiency significantly decreased. In this study, hydrogen inhibition effect would appear at 400 mA current. Moreover, NH_4^+-N concentrations were always close to zero, and the NO_2^--N concentration varied from 0.1 to 2.5 mg/L. Additionally, N_2O and NO concentrations remained at a low level below 2.7 mg/L, while maximum N_2 concentration reached 26.32 mg/L (initial nitrate concentration of 30 mg/L) Therefore, this CBSAD reactor could achieve satisfactory denitrification performance for appropriate current at cold temperatures.

3.2. Effects of HRT on combined denitrification system at low temperatures

Fig. 3 shows the total and S-part nitrate removal efficiencies under increasing HRT at 10℃ in Stage 2. For the S-part, nitrate removal efficiency increased from 27.2 to 55.1% as the HRT increased from 9 to 32 h, as a result sulfur autotrophic micro-organisms had enough time to convert nitrate at long HRTs. The appropriate HRT for this CBSAD reactor was longer than that of Wan et al. [1,4] (2 h, 2.1–4.2 h) and Wang and Qu [3] (1.9–5.0 h), which was attributed to the fact that this CBSAD reactor was operated at cold temperature, so that the autotrophic micro-organisms presented relatively low activity and would take longer HRTs to achieve better performance. For the total denitrification system, nitrate removal efficiency also increased form 46.9 to 98.1% as the HRT increased from 9 to 32 h, indicating that the perfor-

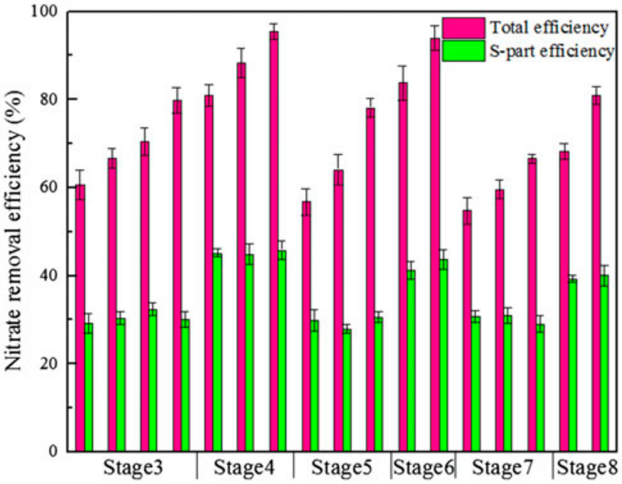

Fig. 4. Optimal current and HRT for maximum nitrate removal efficiency.

mance of the bioelectrochemical denitrification part was also improved at higher HRTs and at 10℃. During this stage, NH_4^+-N and NO_2^--N concentrations always remained at a low level below 2.0 mg/L.

3.3. Optimal current and HRT for maximum nitrate removal efficiency

As shown in Fig. 4, for Stage 3, the total nitrate removal efficiency increased from 60.5 to 79.9% as the current increased from 50 to 200 mA, while the S-part efficiency did not increase obviously along with the growth of current. From Stage 3 to Stage 4, when the HRT increased from 9 to 16 h, the total nitrate removal efficiency significantly increased to the maximum value 95.5% as the current increased to 100 mA, and the S-part nitrate removal efficiencies showed obvious increase. Therefore, the optimal current and HRT for 60 mg/L nitrate at 10℃ were 100 mA and 16 h.

In Stage 5 and Stage 6, the nitrate loading was increased to 100 mg/L. For Stage 5, the S-part efficiency varied from 27.9 to 30.6% when current increased from 150 to 300 mA, while the total efficiency increased from 56.7 to 78.1% under an HRT of 16 h. For Stage 6, the longer HRT of 21 h caused tremendous increases of the S-part efficiencies and total efficiencies. A maximum total nitrate removal efficiency of 93.9% was achieved at an HRT of 21 h and a current of 200 mA at 10℃.

For 150 mg/L nitrate in Stage 7 and Stage 8, the total nitrate removal efficiency increased from 54.8 to 66.6% as the current increased from 150 to 300 mA at an HRT of 21 h in Stage 7, and this efficiency further increased to 80.9% at a current of 300 mA and an HRT of 32 h in Stage 8. The S-part efficiency remained the same with increasing current, but increased as the HRT increased from 21 to 32 h from Stage 7 to Stage 8. These results demonstrated that the S-part efficiency was effectively improved by adjusting the parameter HRT, while current was the key control parameter for the E-part at low temperatures in this CBSAD reactor.

3.4. pH value in the influent and effluent

As reported, a suitable pH environment was important for effective nitrate removal, while an uncontrolled pH environment always resulted in incomplete and inefficient denitrification [12]. Many researchers reported that the optimal pH value for effective autotrophic denitrification process was 6.5–8.0 [13–15]. If pH is higher or lower than the suitable value, the autotrophic denitrification efficiency would obviously decrease because the activities of the autotrophic micro-organisms would be inhibited under improper pH environment. It can be seen from Fig. 5 that pH values in the influent and effluent always fluctuated in the range of 6.4–7.6 in this CBSAD reactor at 10℃ from Stages 1 to 8, which demonstrated that this CBSAD system had the ability to maintain pH balance at cold temperatures. In addition, alkalinity concentrations in the influent and effluent remained at 437–478 mg/L$CaCO_3$ and 425–465 mg/L$CaCO_3$, respectively. In the S-part, nitrate is reduced to nitrogen gas, then H^+ and SO_4^{2-} are generated (Eq. (1)).

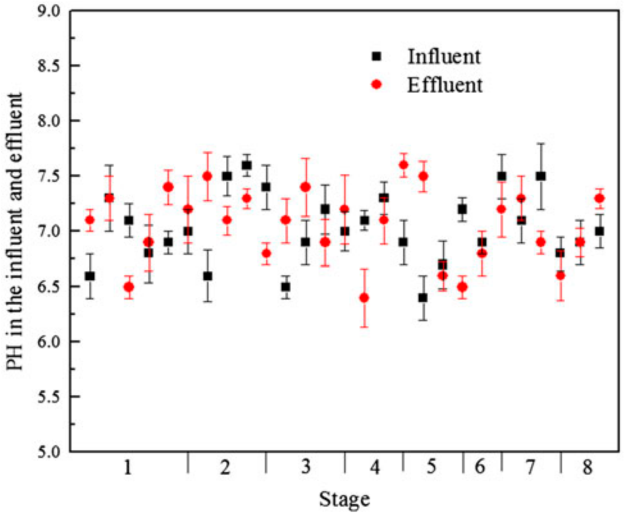

Fig. 5. pH value in the influent and effluent in this CBSAD reactor.

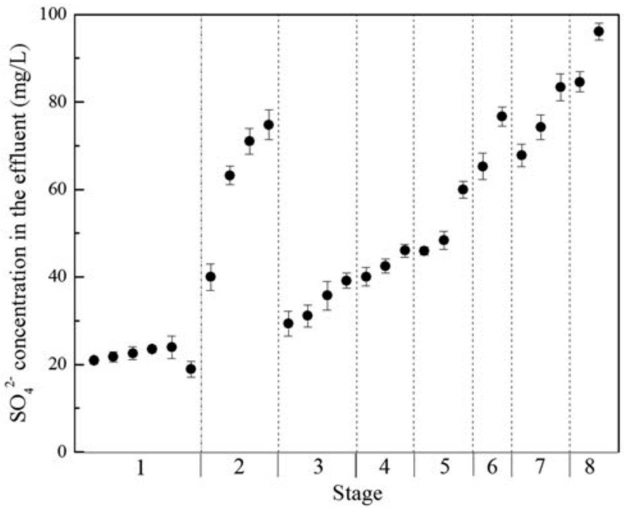

Fig. 6. Effluent SO_4^{2-} concentrations in this CBSAD reactor.

Table 2
Nitrate removal rate in this system compared with literatures

Current (mA)	HRT (h)	Initial nitrate (mg/L)	Temperature (°C)	Efficiency (%)	Refs.
350	2.0	32.0	20	95.8	[1]
3–16	1.9–5.0	30.0	25	90–100	[3]
30-1,000	2.1–4.2	20.9–22.0	21	95–100	[4]
300	9.0	30.0	10	98.6	This study

$$1.06\,NO_3^- + 1.11\,S + 0.3\,CO_2 + 0.785\,H_2O \rightarrow 0.06\,C_5H_7O_2N + 0.5\,N_2 + 1.11\,SO_4^{2-} + 1.16\,H^+ \quad (1)$$

While in the E-part, nitrate is reduced to nitrogen gas, and H^+ is consumed (Eq. (2)).

$$1.06\,NO_3^- + 0.3\,CO_2 + 3.34\,H_2 + 1.06\,H^+ \rightarrow 0.06\,C_5H_7O_2N + 0.5\,N_2 + 3.66\,H_2O \quad (2)$$

Results show that no excessive H^+ appeared in this reactor, which also indicated that the H^+ generated by the S-part was effectively utilized by the E-part for electrochemical reaction [4] in this combined autotrophic system.

3.5. SO_4^{2-} concentration in the effluent

Fig. 6 shows the effluent SO_4^{2-} concentrations in this CBSAD reactor from Stage 1 to Stage 8 at 10°C. The effluent SO_4^{2-} was generated by the S-part through sulfur autotrophic denitrification reaction, and its concentration increased along with the increase of nitrate loading, which was in accordance with the research of Wan et al. [4]. The maximum SO_4^{2-} concentration in this CBSAD reactor from Stage 1 to Stage 8 was 96.1 mg/L, which was lower than 250 mg/L (China EPA, Drinking Water Standard). In addition, Eq. (1) shows that 1.62 g of sulfate produced from 1 mg of nitrate removal. In this study, the experimentally produced sulfate was lower than the theoretical values. This result demonstrated that the effluent SO_4^{2-} concentration could be effectively controlled at cold temperatures in this combined autotrophic system.

For real-scale applications, operating parameters such as HRT and current should be amended because practical wastewater has different properties (pH, temperature, pollutant concentration). Moreover, the operational cost of this system was mainly depended on electric energy consumption and carbon source consumption. Further research was going to focus on energy-efficient on the premise of high removal efficiency. Table 2 shows the nitrate removal rate in this system compared with literatures, which demonstrates that this system achieved excellent efficiency as other literature at same initial nitrate concentration. The optimal HRT was longer than that of other researches because this system was operated at cold temperature.

4. Conclusions

The CBSAD system was proved to exhibit satisfactory denitrification capacity for treatment of nitrate polluted water at cold temperatures. The combined reactor presented excellent denitrification performance by adjusting current and HRT to appropriate values, and it had the ability to control pH at neutral and SO_4^{2-} lower than the standard value at 10°C. The maximum total nitrate removal efficiency reached 98.6% for a current of 300 mA and an HRT of 9 h at initial nitrate concentration of 30 mg/L. When initial nitrate concentration increased to 60 mg/L, the total nitrate removal efficiency significantly increased to 95.5% at a current of 100 mA and an HRT of 16 h. The maximum total nitrate removal efficiency of 93.9% was achieved for an HRT of 21 h and a current of 200 mA at initial nitrate concentration of 100 mg/L. Moreover, 80.9% efficiency was observed for initial nitrate concentration of 150 mg/L at a current of 300 mA and an HRT of 32 h.

Acknowledgments

This work was financially supported by the National Natural Science Foundation of China (NSFC) (No. 51378400) and the National Science and Technology Pillar Program (2014BAL04B04).

References

[1] D. Wan, H. Liu, R. Liu, J. Qu, Study of a combined sulfur autotrophic with proton-exchange membrane electrodialytic denitrification technology: Sulfate control and pH balance, Bioresour. Technol. 102 (2011) 10803–10809.

[2] Y. Sun, M. Nemati, Evaluation of sulfur-based autotrophic denitrification and denitritation for biological removal of nitrate and nitrite from contaminated waters, Bioresour. Technol. 114 (2012) 207–216.

[3] H. Wang, J. Qu, Combined bioelectrochemical and sulfur autotrophic denitrification for drinking water treatment, Water Res. 37 (2003) 3767–3775.
[4] D. Wan, H. Liu, J. Qu, P. Lei, S. Xiao, Y. Hou, Using the combined bioelectrochemical and sulfur autotrophic denitrification system for groundwater denitrification, Bioresour. Technol. 100 (2009) 142–148.
[5] B. Fan, J. Qu, S. Liu, P. Lei, G. Meng, Nitrate removal from drinking water by three-dimension electrode electrochemical-biofilm reactors, J. Acta Sci. Circumstantiae. China 21 (2001) 39–43.
[6] S. Tong, N. Chen, H. Wang, H. Liu, C. Tao, C. Feng, B. Zhang, C. Hao, J. Pu, J. Zhao, Optimization of C/N and current density in a heterotrophic/biofilm-electrode autotrophic denitrification reactor (HAD-BER), Bioresour. Technol. 171 (2014) 389–395.
[7] N. Sundaresan, L. Philip, Performance evaluation of various aerobic biological systems for the treatment of domestic wastewater at low temperatures, Water Sci. Technol. 58 (2008) 819.
[8] S. Yao, J. Ni, T. Ma, C. Li, Heterotrophic nitrification and aerobic denitrification at low temperature by a newly isolated bacterium, *Acinetobacter* sp. HA2, Bioresour. Technol. 139 (2013) 80–86.
[9] APHA, Standard Methods for the Examination of Water and Wastewater, American Public Health Association, Washington, DC, 2005.
[10] S. Islam, M.T. Suidan, Electrolytic denitrification: Long term performance and effect of current intensity, Water Res. 32 (1998) 528–536.
[11] S. Ghafari, M. Hasan, M.K. Aroua, Nitrate remediation in a novel upflow bio-electrochemical reactor (UBER) using palm shell activated carbon as cathode material, Electrochim. Acta 54 (2009) 4164–4171.
[12] C. Glass, J. Silverstein, Denitrification kinetics of high nitrate concentration water: pH effect on inhibition and nitrite accumulation, Water Res. 32 (1998) 831–839.
[13] S. Ghafari, M. Hasan, M.K. Aroua, A kinetic study of autohydrogenotrophic denitrification at the optimum pH and sodium bicarbonate dose, Bioresour. Technol. 101 (2010) 2236–2242.
[14] M. Zhou, W. Fu, H. Gu, L. Lei, Nitrate removal from groundwater by a novel three-dimensional electrode biofilm reactor, Electrochim. Acta 52 (2007) 6052–6059.
[15] C.M. Ho, S.K. Tseng, Y.J. Chang, Autotrophic denitrification via a novel membrane-attached biofilm reactor, Lett. Appl. Microbiol. 33 (2001) 201–205.