



Nitrous oxide production during nitrogen removal from domestic wastewater in lab-scale sequencing batch reactor

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Received 03 August 2007; revised 15 October 2007; accepted 29 November 2007

Abstract

The production of N₂O during nitrogen removal from real domestic wastewater was investigated in a lab-scale aerobic-anoxic sequencing batch reactor with a working volume of 14 L. The results showed that the total N₂O-N production reached higher than 1.87 mg/L, and up to 4% of removed nitrogen was converted into N₂O. In addition, N₂O led to a much higher greenhouse effect than CO₂ during aerobic reaction phase, this proved that N₂O production could not be neglected. The N₂O-N production during nitrification was 1.85 mg/L, whereas, during denitrification, no N₂O was produced, nitrification was the main source of N₂O production during nitrogen removal. Furthermore, during denitrification, the dissolved N₂O at the end of aeration was found to be further reduced to N₂. Denitrification thus had the potential of controlling N₂O production.

Key words: N₂O; domestic wastewater; nitrification; denitrification

Introduction

Biological nitrogen removal is an effective way to prevent eutrophication. Conventionally, N₂ was considered to be the end product of biological nitrogen removal. However, N₂O, one of the important greenhouse gases that may result in global warming and climate change (IPCC, 2001), was also found during biological nitrogen removal.

The mechanism of nitrous oxide emission commonly includes two processes: (1) nitrification, using nitrite as an alternate electron acceptor, thereby reducing it to N₂O; (2) denitrification, nitrate or nitrite reduction (Liu *et al.*, 2006). Wastewater treatment systems potentially contribute to N₂O production because nitrification and denitrification, being N₂O-producing processes (Beline *et al.*, 2001), are artificially enhanced in nitrogen removal.

At present, there are also disagreements on which process (nitrification and/or denitrification) mainly contributes to N₂O production (Hanaki *et al.*, 1992; Schulthess *et al.*, 1994; Schulthess *et al.*, 1995; Zheng *et al.*, 1994; Noda *et al.*, 2003). Under low dissolved oxygen concentration (DO ≤ 0.5 mg/L) or short sludge retention time (SRT), nitrification was the main source of N₂O (Park *et al.*, 2000; Okayasue *et al.*, 1997). But, under low chemical oxygen demand (COD)/total nitrogen (TN) ratio or low DO concentration, denitrification was the main source (Itokawa

et al., 2001; Schulthess *et al.*, 1994). In addition, N₂O production differed widely ranging from as high as 40% of removed nitrogen (Okayasue *et al.*, 1997; Kim *et al.*, 2000) to negligible removed nitrogen (Chiu and Chung, 2003). In simultaneous nitrification, denitrification, and phosphorus removal process, N₂O rather than N₂ was the major end-product of denitrification (Zeng *et al.*, 2003; Lemaire *et al.*, 2006). However, synthetic wastewater was used in this research. The N₂O accumulation might have been caused by the low microbial diversity facilitated by using synthetic wastewater. From the practical point of view, it is significant to investigate N₂O production from real wastewater treatment system. To date, limited research has been done on N₂O production during nitrogen removal from real domestic wastewater.

This study was carried out using a lab-scale sequencing batch reactor (SBR) with a working volume of 14 L. The principal aim of this study was to investigate N₂O production during biological nitrogen removal from real domestic wastewater.

1 Materials and methods

1.1 Wastewater composition

Table 1 shows the characteristics of domestic wastewater. The fresh domestic wastewater was taken from the campus of Beijing University of Technology, China. The wastewater was fed to SBR at the same time every day.

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Table 1 Characteristics of domestic wastewater

Parameter	Range	Mean±SE
COD (mg/L)	123.3–320.5	223.0±7.95
NH ₄ ⁺ -N (mg/L)	40.33–90.22	59.97±1.29
NO ₂ ⁻ -N (mg/L)	0–0.82	0.14±0.05
NO ₃ ⁻ -N (mg/L)	0–1.39	0.46±0.06
TN (mg/L)	55.60–100.62	69.32±1.84
COD/TN	2.17–3.80	3.20±0.14
pH	7.23–8.00	7.44±0.19

1.2 Reactor operation and setup

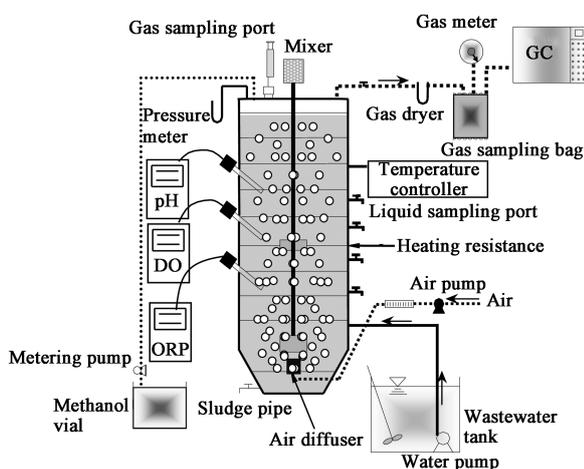
The working volume of lab-scale SBR was 14 L. The seeding sludge taken from a municipal wastewater treatment plant (WWTP) in Beijing, China, was acclimated to the wastewater prior to the start of the experiments for 1.5 months. The aerobic-anoxic SBR was used in the experiment. Each cycle of SBR consisted of six steps: 3 min feeding, 6 h aerobic reaction, 3 h anoxic reaction, 1.5 h settling (adding ethanol), 6 min decanting, and 1 min idling. Certain amount of excess sludge was disposed at the end of anoxic reaction. Ten liters of clarified supernatant was decanted. Influent pH was not controlled during the entire experimental period. The temperature was controlled at 28±1°C by a temperature controller. Airflow rate was kept constant at 100 ml/min and DO concentration during the aeration was approximately 1.5 to 2 mg/L. The mixed liquor suspended solid (MLSS) was maintained at approximately 2,000 to 2,500 mg/L. The effective SRT was approximately 15 d.

Figure 1 shows the schematic diagram of the experimental system. Sequencing batch reactor was gastight. The off gases were collected into gas sampling bags at intervals between 0.5 and 1 h, and at the same time liquid phase samples were taken to measure the dissolved N₂O. N₂O concentration in the gas samples were analyzed by gas chromatography (GC). The volume of the collected gases was measured with wet gas meter.

1.3 Analytical methods

1.3.1 Water quality analysis

COD, NH₄⁺-N, NO₃⁻-N, NO₂⁻-N, and MLSS were measured according to the standard methods (APHA,

**Fig. 1** Schematic diagram of the experimental system.

1998). Dissolved oxygen and pH were measured online using oxygen and pH meters (WTW 340i, WTW Company, Germany), respectively. Total organic carbon and TN were analyzed by Multi N/C 3000 (Analytik Jena AG, Germany).

1.3.2 N₂O analysis

All the gas samples were analyzed using a GC (Agilent 6890N, USA) with electron capture detector (ECD) and HP-Plot/molecular (30 m × 0.53 mm × 25 μm) column. The temperatures of ECD and the column were 300 and 180°C, respectively. Nitrogen was supplied as the carrier gas at a flow rate of 15 ml/min. All gas samples were analyzed in triplicate. The headspace method was used to analyze N₂O dissolved in the mixture. In this method, a 20-ml syringe was filled with 5 ml of mixture and 5 ml of nitrogen gas. In addition, 0.5 ml of 1,000 mg/L HgCl₂ was added to reduce microbial activity. After shaking the syringe it was left to stand for 1 h, the gas phase in the syringe was collected as the sample to analyze N₂O.

The total produced N₂O-N consists of two parts: the dissolved and emitted N₂O-N. The concentrations were determined according to literature (Noda *et al.*, 2003). The N₂O-N conversion rate (Y_{N_2O-N}) was calculated by Eq. (1).

$$Y_{N_2O-N} = \frac{C_{\text{produced}}}{C_{\text{TN}}} \quad (1)$$

where, C_{produced} (mg/L) and C_{TN} (mg/L) are the total produced N₂O-N concentration and the removed TN concentration, respectively.

2 Results and discussion

The activated sludge taken from municipal WWTP was cultivated using domestic wastewater. After cultivation for 45 d, ammonia in the influent was completely oxidized to nitrate during the aeration. Total nitrogen in the effluent was lower than 3 mg/L and kept stable. Thereafter, cyclic studies were performed every two weeks.

2.1 N₂O production in a typical cycle of aerobic-anoxic SBR

Figures 2 and 3 show the dynamics of NH₄⁺-N, NO₃⁻-N, NO₂⁻-N, pH, COD, and N₂O production in a typical cycle of SBR on day 65. At the beginning of aeration (60 min), COD was rapidly consumed and oxidized to CO₂, which was accompanied by slight decrease of NH₄⁺-N (Fig.2). The consumed NH₄⁺-N was mainly used as nitrogen source for synthesizing new cell tissue of heterotrophic bacteria. No N₂O emission or accumulation was observed (Fig.3).

Thereafter, nitrification began; NH₄⁺-N was oxidized to NO₂⁻-N and NO₃⁻-N. At the end of ammonia oxidation, NO₂⁻-N reached the maximum. Keeping on the aeration, NO₂⁻-N was further oxidized to NO₃⁻-N (Fig.2). During nitrification, the total produced N₂O-N and the emitted N₂O-N both increased to 2.3 mg/L, whereas the dissolved N₂O-N increased then decreased due to air stripping. After NH₄⁺-N was completely oxidized (i.e., during the

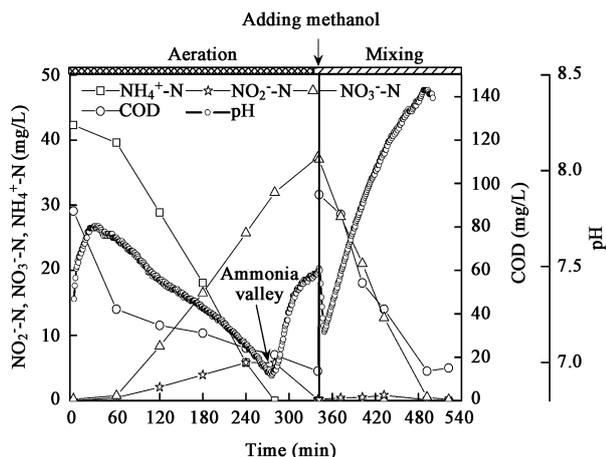


Fig. 2 Dynamics of $\text{NH}_4^+\text{-N}$, $\text{NO}_3^-\text{-N}$, $\text{NO}_2^-\text{-N}$, and COD in a typical cycle of SBR on day 65.

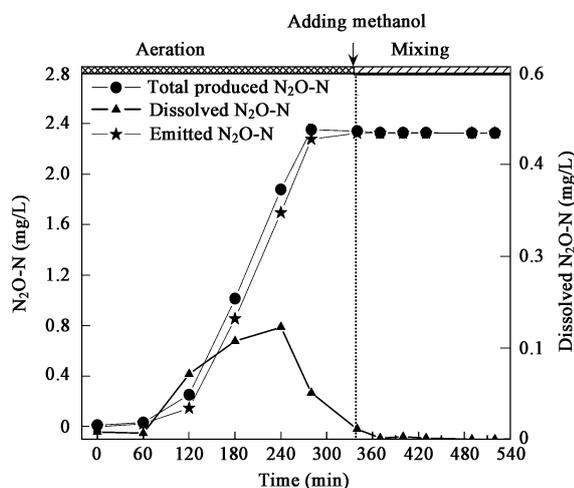


Fig. 3 N_2O production in a typical cycle of SBR on day 65.

accumulated nitrite oxidization), the total produced $\text{N}_2\text{O-N}$ and emitted $\text{N}_2\text{O-N}$ slightly increased.

After $\text{NH}_4^+\text{-N}$ was completely converted to $\text{NO}_3^-\text{-N}$, aeration was stopped and methanol was added as carbon source for denitrification. During denitrification, the total produced and the emitted $\text{N}_2\text{O-N}$ were kept constant, whereas the residual dissolved N_2O decreased to zero at the end of aeration (Fig.3), which was accompanied by the decrease of COD and nitrate (Fig.2).

The $\text{N}_2\text{O-N}$ production and conversion rate in the aerobic-anoxic SBR is summarized in Table 2. The total produced $\text{N}_2\text{O-N}$ reached 1.87 mg/L, and 4% of removed nitrogen was converted into N_2O . Treating 1 L domestic wastewater can produce as high as 4.11 mg N_2O . Because SBR is simple and compact, it has been widely applied for

Table 2 N_2O production and conversion rate during long-term SBR operation

Type of N	Range	Mean±SE
Total produced $\text{N}_2\text{O-N}$ (mg/L)	1.18–2.65	1.87±0.20
Emitted $\text{N}_2\text{O-N}$ (mg/L)	1.18–2.65	1.87±0.20
Removed TN (mg/L)	50.96–60.10	53.16±1.8
$Y_{\text{N}_2\text{O-N}}$ (%)	2–5	4±0.38

small towns wastewater treatment. According to the obtained results, assuming that TN was removed completely in a SBR plant with treatment capacity of 1×10^5 t/a, N_2O as high as 4.11 kg/a was assessed to be produced. Thus, N_2O production during nitrogen removal from domestic wastewater should not be ignored.

2.2 N_2O production during the aerobic reaction phase

The degradation of COD and nitrification were almost subsequently accomplished in the aerobic reaction phase of SBR, and two kinds of greenhouse gases, CO_2 and N_2O , were produced correspondingly. Table 3 summarizes CO_2 and N_2O production during the aerobic reaction phase for long-term operation (80 d). Assuming that the removed COD during aeration was completely oxidized to CO_2 , the calculated CO_2 production would be 1,590 mg in a cycle of SBR. N_2O production during aeration was observed to be approximately 41.14 mg. Intergovernmental Panel on Climate Change reported that the greenhouse effect of N_2O for 100 year span was 190 to 270 times higher than CO_2 on weight basis (IPCC, 2001). It thus can be concluded that N_2O lead to much higher greenhouse effect than CO_2 during the aerobic reaction phase, which further proved that N_2O production can not be neglected.

As shown in Tables 2 and 3, the total produced $\text{N}_2\text{O-N}$ during aerobic phase and during the entire aerobic-anoxic reaction phase were almost the same. From this result, it is clear that nitrification is the main source of N_2O during nitrogen removal from domestic wastewater. In addition, 44% of removed TN during nitrification was converted to N_2O (Table 3), indicating that N_2O was the main gaseous product during nitrification.

During the aerobic phase, the aeration rate influenced the dissolution and volatilization of the produced N_2O . High aeration rate accelerated the volatilization of the produced N_2O , thus, resulting in low dissolved $\text{N}_2\text{O-N}$ concentration and high off-gas $\text{N}_2\text{O-N}$ concentration. In this study, as aeration rate was maintained at 100 L/min, the volatilization of N_2O caused by aeration was constant during the entire aeration phase. $\text{N}_2\text{O-N}$ emission rate (R_{emission}) and $\text{N}_2\text{O-N}$ production rate ($R_{\text{production}}$) were calculated by Eqs.(2) and (3), respectively.

$$R_{\text{emission}} = \frac{\Delta C_{\text{off } \text{N}_2\text{O-N}}}{\Delta t} \quad (2)$$

$$R_{\text{production}} = \frac{\Delta C_{\text{produced}}}{\Delta t} \quad (3)$$

where, $C_{\text{off } \text{N}_2\text{O-N}}$ is the emitted $\text{N}_2\text{O-N}$ concentration.

Table 3 N_2O production during aerobic phase for long-term operation

	Range	Mean ± SE
Degradation of COD		
Removed COD (mg/L)	57.19–119.34	82.82±10.00
CO_2 production (mg)	1,101–2,300	1,594±192
Nitrification		
Removed TN (mg/L)	2.80–4.77	4.97±0.70
Total produced $\text{N}_2\text{O-N}$ (mg/L)	1.18–2.65	1.85±0.20
N_2O production (mg)	25.86–58.22	41.14±4.09
$\text{N}_2\text{O-N}$ conversion from removed TN (%)	20–64	44±10

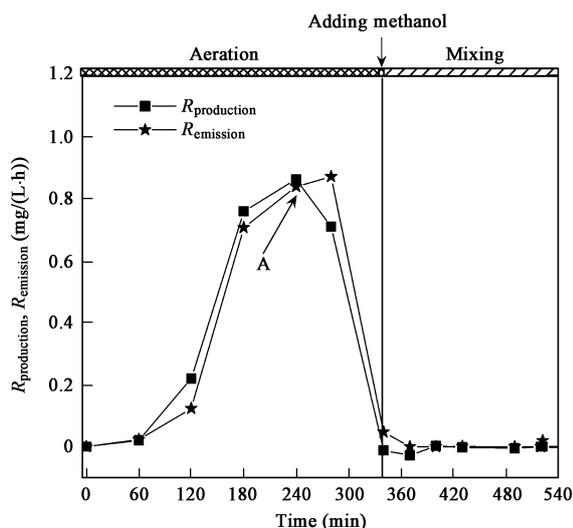


Fig. 4 N_2O emission and production rate in a typical cycle of SBR on day 65.

Figure 4 shows N_2O -N production and emission rates in a typical cycle of SBR on day 65. It could be clearly seen that at the first 240 min, N_2O -N production rate was a little higher than emission rate. When the maximal N_2O -N production rate reached 0.9 mg/(L·h) (point A in Fig.4), NH_4^+ -N, NO_2^- -N, and NO_3^- -N were 5.8, 5.84, and 25.7 mg/L, respectively (Fig.2). When NH_4^+ -N was lower than 5.8 mg/L (point A in Fig.4), N_2O -N production rate decreased sharply, indicating that NH_4^+ -N concentration had effect on N_2O production. This result also indicated that N_2O was mainly produced during the initial stage of aeration. Because the dissolved N_2O was continuously volatile from the mixture, at 240 min, N_2O emission rate increased to maximum, whereas N_2O production rate began to decrease. After ammonia was completely oxidized, if aeration was not stopped, the dissolved N_2O was emitted from the mixture. Thus, if aeration was stopped just at time of ammonia complete oxidation, over 3% of the total produced N_2O -N would be prevented to emit from the mixture.

As shown in Fig.2, because ammonia oxidization produces acid, at the beginning of nitrification, pH decreased gradually. When ammonia was oxidized completely, pH began to increase and “ammonia valley” appeared on the pH profile. “Ammonia valley” represented the end of com-

plete oxidation of ammonia. There is a slight possibility that using “ammonia valley” to control the aeration time would reduce N_2O emission from the mixture.

2.3 N_2O production during the anoxic reaction phase

Figure 5 shows N_2O -N production and conversion rate during denitrification. In denitrification, N_2O was rarely detected. Many researchers had reported that N_2O was mainly produced during denitrification (Itokawa *et al.*, 2001; Schulthess *et al.*, 1994). Many factors, such as the denitrifying microbial communities, wastewater qualities (Zeng *et al.*, 2003; Lemaire *et al.*, 2006), and the shortage of carbon source, influenced N_2O production (Kishida *et al.*, 2004). Because the wastewater treated in this study was real domestic wastewater, the microbial denitrifying communities in the activated sludge were diverse. Moreover, the dosage of carbon source was sufficient for denitrification. N_2O , being the intermediate product of denitrification, could be used as electron donor, consequently, denitrifiers could further reduce the dissolved N_2O to N_2 . Therefore, N_2O production and conversion rate during denitrification were zero or negative during the entire operation of SBR (80 d). During denitrification, not only no N_2O was produced, but it also favored the reduction of the dissolved N_2O . It should also be pointed out that N_2O would be produced if the dosage of carbon source was insufficient. This should be further investigated.

3 Conclusions

The lab-scale SBR fed with real domestic wastewater was operated to investigate N_2O production during nitrogen removal. Several conclusions could be made: (1) N_2O was produced during nitrogen removal from domestic wastewater. N_2O lead to much higher greenhouse effect than CO_2 did during aerobic reaction phase, which proved that N_2O production can not be neglected. (2) N_2O was mainly produced during the initial stage of aeration. (3) During denitrification, the dissolved N_2O at the end of nitrification was further converted to N_2 .

Acknowledgements

This work was supported by the National Natural Science Foundation of China (No. 50478040).

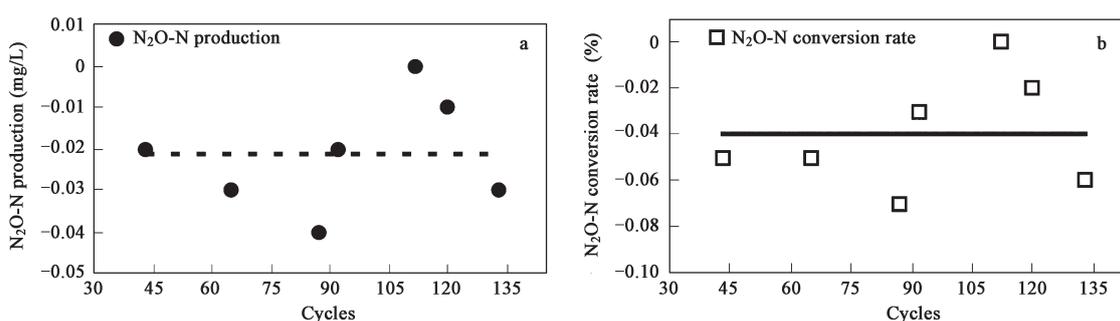


Fig. 5 N_2O -N production (a) and conversion (b) rate during denitrification.

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