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Contributions of biofilm and suspended sludge to nitrogen transformation and nitrous oxide emission in hybrid sequencing batch system

Ingwei Wayne Lo[1,*], Kwang Victor Lo[1], Don S Mavinic[1], Dean Shiskowski[2], William Ramey[3]

1. Department of Civil Engineering, University of British Columbia, Vancouver, BC, V6T 1Z4, Canada. E-mail: waynelo.ubc@gmail.com
2. Associated Engineering Ltd., Burnaby, BC, V5G 4M5, Canada
3. Department of Microbiology & Immunology, University of British Columbia, Vancouver, BC, V6T 1Z4, Canada

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Abstract

Hybrid system combines the nature of suspended growth and attached growth has been widely applied to wastewater treatment. In this research, the contributions to N transformation and N_2O emission by biofilm and suspended sludge in the hybrid sequencing-batch reactor for a simultaneous nitrification, denitrification and phosphorus removal process were investigated. For the hybrid system, nitrification occurred mostly in the suspended sludge, while the biofilm played the major role in denitrification. The interaction of the biofilm and the suspended sludge in the same reactor resulted in a better overall nitrogen removal performance with simultaneous nitrification and denitrification. However, N_2O emission was the main end product of nitrogen removal for the hybrid system; while it was N_2 for the biofilm. The relative low N_2O emissions from the pure biofilm and the pure suspended sludge corresponded to the relatively high nitrate at the end of the aeration period compared with the hybrid system.

Key words: N_2O emission; biofilm; suspended sludge; hybrid system

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Introduction

Excess nitrogen and phosphorus in wastewater effluent have a serious environmental impact on the receiving water bodies. To lessen these effects, various biological nutrient removal (BNR) processes have been applied to wastewater treatment. The process of simultaneous nitrification and denitrification provides a relatively simple, compact configuration to achieve nitrogen removal. In addition, some microorganisms, namely denitrifying phosphorous accumulating organisms (DPAOs), are able to use nitrate and/or nitrite as an electron acceptor for phosphate uptake. This makes it possible to have simultaneous nitrification, denitrification and phosphorus removal from wastewaters (Lee et al., 2001; Turk and Mavinic, 1986).

Four major microbial populations are included in a simultaneous nitrification, denitrification, and phosphorus removal (SNDPR) system: ammonia oxidation bacteria (AOBs), nitrite oxidation bacteria (NOBs), phosphorous accumulating organisms (PAOs, including DPAOs) and other denitrifiers. Generally, NOBs need a longer solids retention time (SRT) than AOBs (van Dongen, 2001). On the other hand, to maintain good P-removal, sludge containing stored poly-P needs to be wasted from the reactor and new cells grown to take up phosphorus from the bulk solution. Therefore, the SRT for PAOs has to be maintained below a certain level (Chuang et al., 1997). The hybrid system integrating biofilm within a suspended-growth system, maximizes the SRT of sludge in the biofilm, but has a potential of operating the suspended sludge phase with a relatively short SRT. The hybrid system also encourages a rapid denitrification of nitrite, when it is produced by the ammonium oxidizers (Chung et al., 2006). In addition, a hybrid system is stable with the advantage of a short hydraulic retention time (HRT) and a small system volume, due to the accumulation of biofilm biomass along with suspended biomass.

In a hybrid system, the biofilm and the suspended sludge may function differently in terms of nitrogen and phosphorus removal. Falkentoft et al. (2001) mentioned that diffusion limitations hindered biological phosphorus removal in the biofilm. Kumar and Chaudhari (2003) also found that both the suspended sludge and the hybrid system achieved greater than 90% ammonia oxidation. However, the nitrogen removal efficiency was 48% in a pure suspended growth SBR and in excess of 62% in pure biofilm SBRs.

SNDPR has been reported in biofilm systems by several authors (Zeng et al., 2003; Castillo et al., 1999). Helness and Odegaard (2001) used a sequencing batch moving bed biofilm reactor for SNDPR. Rogalla et al. (2006) compared the phosphorus removal of biofilm systems and hybrid systems. That research suggested that very low

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[*] Corresponding author. E-mail: waynelo.ubc@gmail.com

nitrogen and phosphorus effluent could be demonstrated both on pilot and full scale. Lo (2008) also reported a better overall SNDPR performance of a hybrid system than a conventional suspended growth system in SBRs.

Nitrous oxide, a green house gas with a global warming potential 310 times that of the contribution of carbon dioxide (IPCC, 2006), is produced from both heterotrophic and autotrophic processes. Research has found that the emission of N_2O has a close relationship with the concentration of nitrite (NO_2^-) and dissolved oxygen (DO) in the bulk solution (Shiskowski, 2004; Lemaire et al., 2006). Lo et al. (2008) found these emissions from the hybrid system were less than those from the suspended growth system.

To date, research has been conducted on the treatment performance of hybrid systems. However, no sufficient study has examined the relative effects of the biofilm and the suspended sludge on N and P removal efficiencies and nitrous oxide emission in a hybrid SNDPR system. In this study, the effects of sludge makeup on SNDPR and N_2O emissions were therefore investigated. The nitrogen balances were calculated to understand the transformation of nitrogen during the treatment. In order to control the compositions of the nutrients, synthetic wastewater was prepared for the experiment. Further study of using different types of real wastewater is required for supporting the result from this investigation.

1 Materials and methods

1.1 SBR system

Hybrid SBRs, which had suspended-growth sludge and biofilm attached to plastic material was designed for use in the SNDPR process. The working volume of each reactor was 10.5 L. The decanting volume was 3.5 L. Bio-carriers (K3 type, Kaldnes) filled 30% of the working volume (450 carriers in total). Each SBR cycle (8 hr) included six stages: feeding (10 min), anaerobic (60 min including 10 min feeding), aerobic (4 hr), anoxic (2 hr), post aeration (30 min) and settling/decanting (30 min). The schedule of air supply was set by an electronic (programmable) timer. The reactors were operated at an ambient temperature of (22 ± 2)°C. The SRT was maintained at 10 days by wasting 10% of the suspended mixed liquor at the end of the post aeration period. Sludge seed was obtained from the wastewater treatment pilot plant at the University of British Columbia in Vancouver, British Columbia, Canada. This wastewater treatment pilot plant was operated in an enhanced biological phosphorus removal mode.

Synthetic wastewater was prepared as the influent for the treatment system. Each 1 L of the synthetic wastewater consisted of (g): sodium acetate 0.3, lactose 0.15, beef extract 0.15 (Difco), NH_4Cl 0.1 g, KH_2PO_4 0.025, KH_2PO_4 0.025, $MgSO_4 \cdot 7H_2O$ 0.05, $CaCl_2 \cdot H_2O$ 0.03, $NaHCO_3$ 0.03 and nutrient solution as previously described (Lo et al., 2008). The complete influent contained chemical oxygen demand (COD) of approximately 550 mg/L, total nitrogen (TN) of 35 mgN/L, and total phosphorous (TP) of 12 mg/L. The pH was approximately 7.

The N_2O off-gas was monitored by an infrared N_2O monitor (3010, Bacharach, UK). In order to reduce the interference of CO_2, the off-gas bubbled through 5 mol/L KOH contained in a 30 mL impinger flask to remove CO_2. The gas then passed through an ice-chilled impinger flask to condense moisture before it entered the mouth of the monitor (Shiskowski and Mavinic, 2005). DO, pH and oxidation-reduction potential (ORP) probes were installed in each reactor. The monitoring data were recorded in a data acquisition developed to operate on a Microsoft computer.

1.2 Chemical analyses

NH_3/NH_4^+, NO_2^-, NO_3^- and *ortho*-P were determined by the flow injection spectrophotometric analysis (Quikchem 8000, Lachat, USA). Total organic carbon (TOC) and TN were measured by a TOC/TN analyzer (IL TOC-TN, Lachat, USA). Mixed liquor suspended solids (MLSS), and total solids (TS) were determined according to Standard Methods for Examination of Water and Wastewater (APHA, 2005). Total Kjeldahl nitrogen (TKN) was digested according to Standard Methods for Examination of Water and Wastewater and measured by flow injection analysis of spectrophotometry (Quikchem 8000, Lachat, USA). Dissolved nitrous oxide concentrations were measured using a modified version of the procedure described by Shiskowski and Mavinic (2005). A mixed liquor of 20 mL was collected, and preserved by phenyl mercuric acetate in a 60 mL plastic syringe containing 20 mL of air (for the biofilm tests, a carrier with biofilm was put into a 60 mL plastic syringe with 20 mL effluent and 20 mL air). Immediately, the syringe was vigorously shaken by hand for 30 sec, to partition the nitrous oxide between the mixed liquor and the syringe headspace. After a waiting period of 5 min, a 100 μL headspace sample was collected in a gas-tight syringe for GC injection using the same method as for gaseous N_2O determination.

1.3 Determination of the size of floc and the thickness and mass of biofilm

The size of the sludge floc was analyzed by a particle analyzer (Hydro 2000S, Malvern, USA). The thickness of the biofilm was measured using a stereo zoom microscope (SMZ-168, Motic, USA) and computer photographic software-Motic Images plus 2. The biomass of the biofilm was obtained by measuring the difference of mass between new carriers dried at 105°C and bioflim grown carriers also dried at 105°C.

1.4 SND efficiency

Simultaneous nitrification denitrification (SND) in this study is defined as the loss of nitrogen during the aeration period. The SND efficiency is given in Eq. (1).

$$\text{SND} = \left(1 - \frac{\text{NH}_{4,\text{e}}^{+} + \text{NO}_{2,\text{e}}^{-} + \text{NO}_{3,\text{e}}^{-}}{\text{NH}_{4,\text{i}}^{+} - \text{NH}_{4,\text{e}}^{+}}\right) \times 100\% \qquad (1)$$

where, $\text{NH}_{4,\text{e}}^{+}$ (N mg/L) is nitrogen in ammonium and ammonia at the end of aeration; $\text{NO}_{2,\text{e}}^{-}$ (N mg/L) and

$NO_{3,e}^-$(N mg/L) are nitrogen in nitrite and nitrate at the end of aeration; $NH_{4,i}^+$ (N mg/L) is nitrogen in ammonium and ammonia at the beginning of aeration.

1.5 N_2O off-gas production

N_2O off-gas production in this study was presented as the accumulated nitrogen of N_2O, divided by the total nitrogen of the influent.

$$N_2O_{\text{off-gas production}} = \frac{N_2O\text{-N}}{TN} \times 100\% \quad (2)$$

where, N_2O-N (mg/L) is nitrogen of N_2O.

1.6 Experimental design

1.6.1 Track studies of the biofilm, the suspended sludge and hybrid system

When tested the relative effects of biofilm and suspended sludge, all biofilm carriers were removed from the mixed liquor in the reactor and transferred to a separate reactor containing effluent of treated synthetic wastewater. Thus, one reactor had suspended sludge only, and another reactor had biofilm only. The original reactor was the complete hybrid system. In order to calculating N_2O emission, three systems were carried out under the conditions with an aeration rate of 200 mL/min. Given that the biomass in the suspended sludge and the biofilm were less than that in the hybrid system, the wastewater strength of the influent for the biofilm and the suspended sludge was reduced to 65% of the influent for the hybrid system. The reaction rates were then considered as per unit mass.

1.6.2 Heterotrophic denitrification tests

Heterotrophic denitrification is defined here as denitrification which occurs when organisms use organic carbon as an energy source. Anaerobic heterotrophic denitrification by the biofilm, the suspended sludge and the hybrid systems were tested under conditions without DO and ammonia. In these tests, a final concentration of $NaNO_3$ 7 mg N/L was spiked into the reactors instead of feeding influent.

2 Results and discussion

The hybrid system was operated for 620 days. The effluent qualities were relatively stable in terms of TOC (approximately 10 mg/L), ammonia/ammonium (approximately 0 mg/L), nitrate and nitrite (approximately 3 mg/L), and *ortho*-P (approximately 1 mg/L; data not shown). The present study was conducted after day 550 of the operation. In these tests, the MLSS of the suspended sludge was around 1200 mg/L (a total 12.6 g in the reactor). The size of the suspended sludge was distributed in a range from 7 to 320 μm, with a mean value of 70 μm. For the biofilm, the thickness was approximately 2.2 mm. The biomass was (99 ± 17) mg per carrier and 44.6 g in total mass. The ratio of total biomass in suspended sludge reactor and the biofilm was 1 to 3.5. The total biomass in the hybrid system was 57.2 g.

2.1 Nitrogen removal

The aeration time lasted until the ammonium was fully or nearly oxidized. The results indicated that the ammonia oxidation rates (AOR) for the three tests were different. The required aeration time for the suspended sludge and hybrid system tests were 240 and 270 min, respectively, while it was 330 min for the biofilm system, due to a relatively low AOR. The respective total nitrogen removal of the biofilm, suspended and hybrid system were around 49.5%, 26.7% and 71.5% of total nitrogen in the influent. The results of the nitrogen removal test are summarized in Table 1. Time profiles for nitrogen compounds, DO, ORP and N_2O off-gas of the biofilm, the suspended sludge and the hybrid systems are shown in Figs. 1–3, respectively.

Table 1 Summary of the nitrogen removal of the biofilm, the suspended sludge, and the hybrid system

	Biofilm	Suspended sludge	Hybrid system
AOR (N mg/(hr·g biomass))	0.24	1.54	0.38
DNR (N mg/(hr·g biomass))	0.57	0.14	0.51
SND (%)	41.0	16.7	75.9
N_2O emission (% as TN)	0.5	4.2	21.2

Aeration rate: 200 mL/min.
AOR: ammonia oxidation rate; DNR: denitrification rate; SND: simultaneous nitrification denitrification.

2.1.1 Ammonia oxidation and nitrite oxidation

In the biofilm test, the ammonia oxidation was relatively slow at 0.24 mg N/(hr·g biomass). At this lower AOR, the slower oxygen uptake resulted in a relatively high DO level in the reactor (Figs. 1b, 2b, 3b). This higher DO in the biofilm reactor facilitated nitrite conversion to nitrate (Fig. 1a). Nitrite build-up did not happen in the biofilm system and nitrite concentrations throughout the test were lower than 0.2 mg/L.

The slow AOR in the biofilm test led to a low nitrite production rate. Once the rate of nitrite conversion was greater or equal to the nitrite production rate, nitrite concentrations in the solution would be very low or even zero. This consumption of nitrite might be caused by nitrite oxidation via heterotrophic denitrification and/or autotrophic denitrification. Nitrite is normally oxidized to nitrate if oxygen is present. The thickness of the biofilm can hinder the transfer of oxygen. As a result, the inner layer of the biofilm is in an anaerobic condition (Chang et al., 2003; Stewart, 2003). Some publication reported on a limited penetration depth of biofilm (e.g., 0.5 mm) under low DO conditions (e.g., 1 mg/L) in the bulk solution (Stewart, 2003). Li and Bishop (2004) used micro DO sensor to measure the DO gradient in a biofilm and found the penetration limitation of 1 mg/L DO to 0.5 mm biofilm thickness. However, the relatively high DO of 2 to 5 mg/L used in the present study could partially penetrate the biofilm (2.2 mm thick), and should result in a high level of nitrate. It has also been proved that nitrite could coexist with oxygen as an electron acceptor, and be denitrified at a relatively slow rate (Henze, 2000).

For the suspended sludge test, the specific ammonia

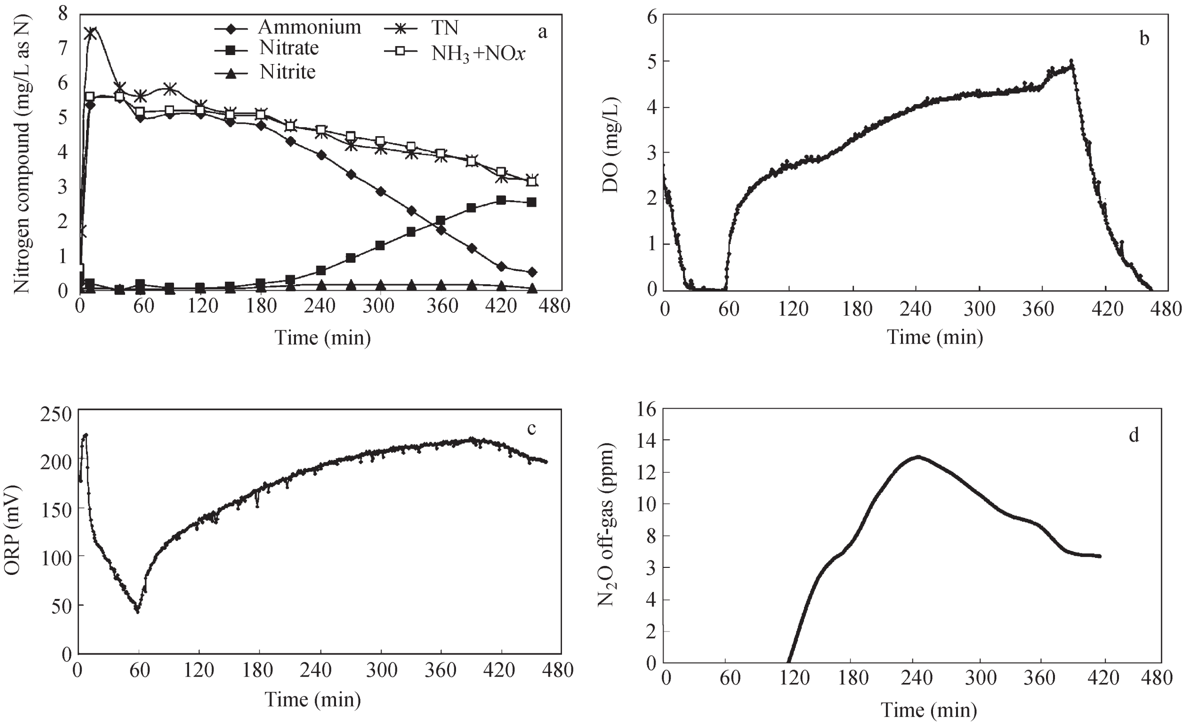

Fig. 1 Time profiles of the biofilm test. (a) nitrogen; (b) dissolved oxygen (DO); (c) oxidation reduction potential (ORP); (d) N_2O. The aeration was from time 60 min to 390 min.

oxidation rate averaged 1.54 mg N/(hr·g biomass), which was 6.4 times higher than the AOR of the biofilm system. Since the biomass of the biofilm was 3.5 times the biomass of the suspended sludge, the overall contribution of the suspended sludge to ammonia oxidation was 1.8 times (6.4/3.5) higher than that of the biofilm. However, the DO level in the biofilm test was higher than the test level with the suspended sludge. This different of DO resulted in a higher AOR. That is, if DO conditions in the two reactors were the same, the contribution of the suspended sludge to ammonia oxidation in the hybrid reactor would be greater than 1.8 times compared with the contribution by the biofilm. Thus, the suspended sludge played the major role in ammonia oxidation. Nitrite accumulation was also found in the suspended sludge (Fig. 2a). The highest nitrite concentration accumulated in the bulk solution was around one third of the total nitrogen in the feed. The accumulation of nitrite implies that the performance of NOBs is lower than that of AOBs.

After 240 min, the AOR slowed down when the ammonia concentration decreased. From Fig. 2b, it can be seen that there was a rapid rise in DO near the end of the aeration period. Thus, the rate of nitrite oxidation was enhanced and led to a breakthrough in the nitrite built-up. A corresponding rapid rise also occurred in the nitrate profile. The TN profile (Fig. 2a) of the suspended sludge was flat and decreased relatively slowly, which pointed to an inefficient nitrogen removal. Nitrate was the major end product of ammonia oxidation in this test.

In the hybrid system, the average AOR was 0.38 mg N/(hr·g biomass). The concentrations of nitrite and nitrate were relatively low during the test. From the TN profile, the hybrid system had a better overall nitrogen removal than did the biofilm and the suspended sludge, when they were tested individually.

Simm et al. (2005) applied the technology of fluorescence *in-situ* hybridization (FISH) to identify the spatial distribution of AOBs and NOBs in sludge floc and concluded that AOBs tend to grow in the inner part of the floc and NOBs in the outer part of the sludge floc. This may be the result of competition for substrates by different microbial populations. Schramm et al. (2000) suggested the oxygen half saturation coefficient for AOBs may be significantly lower than that for NOBs. Other research reported that NOBs were out-competed at low DO levels and this was beneficial for the accumulation of nitrite for the processes of autotrophic nitrogen removal over nitrite (Ciudad et al., 2007; Gali et al., 2007). The DO of the hybrid system was usually maintained at between 0 to 1.5 mg/L. The low DO and the denitrifying competition for utilization of nitrite limited the growth of NOBs. This may be the reason for the nitrite accumulation in the suspended sludge test. In this case, there was an insufficient evidence suggesting that the better NOB performances in the hybrid system and biofilm system were due to low levels of nitrite. Nitrite might be diminished by heterotrophic denitrification.

Theoretically, denitrification of the biofilm system should be more efficient than is the case with the suspended sludge and the hybrid systems because a larger portion of the biomass should be relatively depleted of air. However, as mentioned previously, a low degree of denitrification in the biofilm test was observed due to the higher DO in the bulk solution. A further test of heterotrophic denitrification under conditions of no DO therefore had to be investigated, in order to better understand the reactors performance

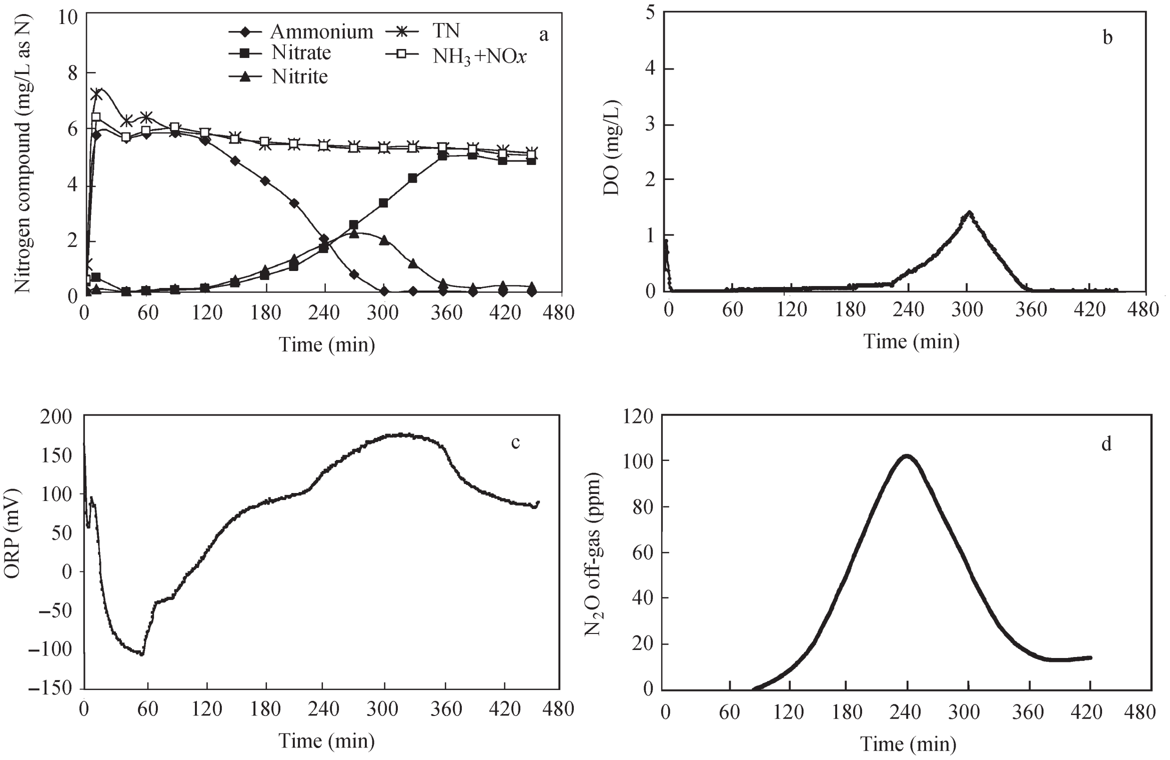

Fig. 2 Time profiles of the suspended sludge test (a) nitrogen; (b) DO; (c) ORP; (d) N_2O. The aeration was from time 60 min to 300 min.

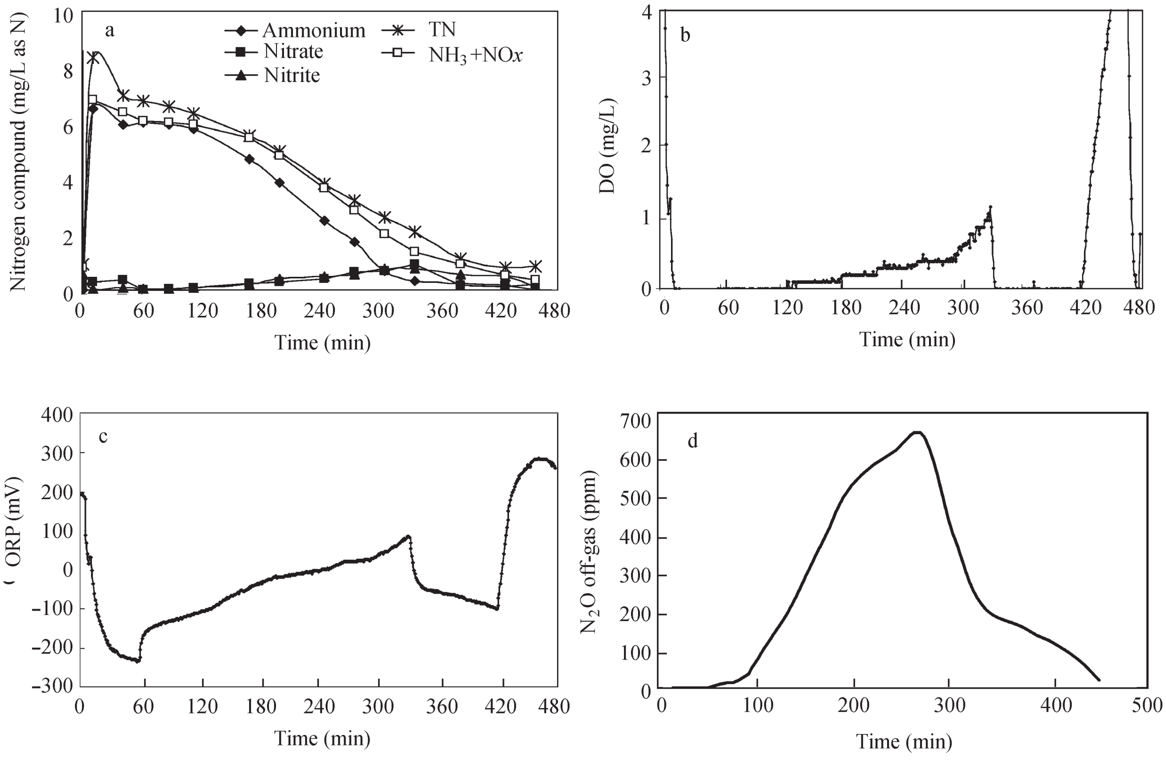

Fig. 3 Time profiles of the hybrid system (a) nitrogen; (b) DO; (c) ORP; (d) N_2O. The aeration was from time 60 min to 330 min.

(reported in the next section).

The ORP profiles at the end of aeration of the biofilm and the suspended sludge tests were relatively high (up to 220 and 180 mV; Fig. 1c and Fig. 2c, respectively) compared with the hybrid system (below 100 mV; Fig. 3c). A greater abundance of nitrate in the bulk solution of the biofilm and the suspended systems than in the hybrid system indicated that under a higher ORP range nitrite tended to be oxidized to nitrate than to be reduced to nitrous oxide or/and nitrogen gas.

2.1.2 Heterotrophic denitrification

Under zero DO, heterotrophic denitrification occurred when there was no ammonia in the solution. Specific heterotrophic denitrification rates of 0.57, 0.14 and 0.51 mg N/(hr·g biomass) were observed for the biofilm, the suspended sludge and the hybrid systems, respectively. The specific denitrification rate for the biofilm was 4.1 times more than the rate of the suspended sludge. Considering the total biomass of the biofilm was 3.5 times that of the suspended sludge, the overall contribution to denitrification from the biofilm in the hybrid reactor was 14.3 times greater than for the suspended sludge in the hybrid reactor. Thus, in the hybrid system, the contribution to denitrification was mainly from the biofilm.

2.2 Simultaneous nitrification denitrification

There were 41.0% and 16.7% SND efficiencies for the biofilm and the suspended sludge reactors at the end of aeration, while the hybrid system had an SND efficiency of about 75.9%. The unexpected low SND achieved in the biofilm test was potentially due to the inhibition of denitrification by high DO in the bulk solution because the efficiency of SND is dependent on a low DO (Pochana and Keller, 1999). The SND efficiency of the suspended sludge test, however, was also relatively low, even though the level of DO usually remained below 1 mg/L. Most of the nitrogen was converted to nitrate at the end of the aeration in the suspended sludge test. However, the hybrid system facilitated the SND process. It had a high oxygen uptake rate, due to the suspended sludge, resulting in a low DO, and consequent denitrification of the biofilm.

2.3 N_2O off-gas

Nitrogen removal might occur via the emission of dinitrogen gas (N_2) or gaseous nitrous oxide. During the aeration period, the biofilm reactor had the lowest N_2O emission of 0.5% of the influent TN (Fig. 1d). It was 4.2% for the suspended sludge reactor (Fig. 2d) test. The hybrid system had the highest N_2O off-gas, at about 21.2% (Fig. 3d).

The production of nitrous oxide is affected by TN, carbon source, DO, pH, temperature, salt (Wicht, 1996; Tsuneda et al., 2005), and by the population of microorganisms. It was ascertained that N_2O emissions were affected mostly by DO level and microbial populations in this work. The following reasons may explain the low N_2O emissions from the biofilm test. First, heterotrophic microbes were the majority of the microorganisms in the biofilm and the inner part of the biofilm provided anaerobic conditions suitable for reducing N_2O. Second, the AOR by AOBs was relatively low and resulted in a relatively slow N_2O production. This gave sufficient time for the N_2O gas to be reduced. Third, the relatively high DO in the biofilm test might have enhanced the rate of nitrite oxidation and diminished the opportunity for nitrite reduction to nitrous oxide gas.

For the suspended sludge test, there was no essential nitrous oxide emission. As discussed previously, most of the nitrogen was oxidized to nitrate, and little denitrification occurred. From the N_2O off-gas profiles shown in Fig. 2d, N_2O was still being emitted from the reactor at the end of the cycle, due to the carry-over denitrification. This suggests that although the N_2O off-gas was not abundant during the treatment, N_2O off-gas might still be emitted to the air when denitrification occurs in the receiving waters.

The hybrid system facilitated SND. Low concentrations of both nitrite and nitrate were observed in the hybrid system. This implied the shortcut of denitrification via nitrite occurred in the reactor. However, N_2O off-gas was around 20% of the TN in the influent. High N_2O emissions from a BNR system (Zeng et al., 2003; Hwang et al., 2006) and the amount of N_2O emissions exceeded 50% of TN (Itokawa et al., 1996) have been observed. In this hybrid system, it seems likely that the N_2O production rate was higher than the N_2O reduction rate because of the limite reduction potential which was corresponded with aeration in the reactor. Hence, the accumulated N_2O was stripped out by aeration.

2.4 Nitrogen balance

TN, NH_4^+, NO_2^-, NO_3^- and N_2O (both gas and in liquid) were measured for the nitrogen balance. Soluble organic N was determined by soluble TN minus NH_4^+, NO_2^- and NO_3^-. Nitrogen used for assimilation was estimated by the difference of TKNs percentage of the biomass and the growth of the biomass, which was also the waste sludge in this stable system. TKNs were determined to be (7.4 ± 0.2)% and (8.2 ± 0.3)% for the biofilm and the suspended sludge, respectively. The determination of N_2 was calculated, rather than measured. It was determined as the initial TN minus measured N species, minus soluble organic N (org-N), and minus assimilation of N. In the aeration phase in both the suspended sludge and the hybrid systems, nitrogen gas was not the main end product of denitrification. It was only in the biofilm tests that nitrogen gas was the main product of denitrification. The thickness of the biofilm provided the ability for reducing nitrogen compounds to nitrogen gas. The nitrogen balance of the

Table 2 Summary of nitrogen balance at the end of aeration

	Percentage of TN						
	NH_4^+	NO_3^-	NO_2^-	N_2O	N_2	Soluble org-N	New cell-N
Biofilm	18.1	27.7	2.5	0.5 (0)	29.1	5.5	16.8
Suspended sludge	0.6	51.9	23.6	4.2 (0.2)	1.9	0.7	17.4
Hybrid system	6.4	8.8	9.5	21.2 (1.5)	14.9	1.3	33.5

Data outside the parenthesis were N_2O off-gas; in parenthesis were N_2O in liquid; N_2 was determined by calculation; new cell-N was determined by the differences of TKN of the biomass and the waste sludge.
Data were collected at 330, 240 and 270 min for the biofilm, the suspended sludge and the hybrid system, respectively.

biofilm, the suspended sludge and the hybrid system at the end of the aeration are summarized in Table 2. The main end product of nitrogen removal for the biofilm system was N_2 (around 29% of TN); while was N_2O for the suspended sludge and the hybrid system, the emissions were around 4% and 21% of initial TN, respectively. In the suspended sludge system, nitrate and nitrite were 52% and 24% of TN, which would require advanced denitrification and might have the potential of N_2O release. The organic nitrogen mainly from beef extract (the initial concentration was approximately 3 mg/L) was quickly uptaken by cells or converted to ammonium. During the treatment, some org-N might release from the decayed cells to form soluble org-N. At the end of the cycle, the amounts of soluble org-N were approximately 6%, 1% and 1% of TN for the biofilm, the suspended sludge and the hybrid system, respectively. In the case of the hybrid system, around 34% of initial TN was converted to new cell-N implied that using the growth of the biomass could be an alternative for nitrogen removal.

3 Conclusions

In the hybrid system, the main contributions from the suspended sludge were ammonia oxidation and nitrite oxidation. In this study, the specific AOR of the suspended sludge was about 6.4 times higher than the case with the biofilm (1.8 times higher based on the amount of the mass). Denitrification was the major contribution of the biofilm. The specific denitrification rate of the biofilm was about 4.1 times the rate of the suspended sludge (the normalized rate was 14.3 times higher based on the amount of the mass). A good performance in simultaneous nitrification and denitrification was achieved when the biofilm and the suspended sludge grew together in the same reactor. Total nitrogen removals were 49.5%, 26.7% and 71.5% for the biofilm, the suspended sludge and the hybrid system, respectively. SND was 75.9% for hybrid system, and were 41.0% and 16.7% for biofilm and the suspended sludge system, respectively. However, there was significant N_2O emission form the hybrid system, approximately 21% of TN. N_2O emissions from the biofilm and the suspended sludge were lower than the hybrid system, due to relative large amount of ammonia converted to NO_3^- at the end of the treatment. The high levels of nitrate in the effluent might cause N_2O emission when denitrification happens in the water receiving bodies. Based on the nitrogen balance analysis, N_2 was the main end product of nitrogen removal for the biofilm. For the suspended sludge and the hybrid system, it was N_2O. There was approximately 1/3 of initial TN converted to new cells. High biomass yield processes could be a potential approach for nitrogen removal from wastewater.

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