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Performance of high-loaded ANAMMOX UASB reactors containing granular sludge

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ABSTRACT

The performance of high-loaded anaerobic ammonium oxidizing (ANAMMOX) upflow anaerobic sludge bed (UASB) reactors was investigated. Two ANAMMOX reactors (R1 with and R2 without effluent recycling, respectively) were fed with relatively low nitrite concentration of 240 mg-N L^{-1} with subsequent progressive increase in the nitrogen loading rate (NLR) by shortening the hydraulic retention time (HRT) till the end of the experiment. A super high-rate performance with nitrogen removal rate (NRR) of 74.3–76.7 kg-N m^{-3} day^{-1} was accomplished in the lab-scale ANAMMOX UASB reactors, which was 3 times of the highest reported value. The biomass concentrations in the reactors were as high as 42.0–57.7 g-VSS L^{-1} with the specific ANAMMOX activity (SAA) approaching to 5.6 kg-N kg-VSS^{-1} day^{-1}. The high SAA and high biomass concentration were regarded as the key factors for the super high-rate performance. ANAMMOX granules were observed in the reactors with settling velocities of 73–88 m h^{-1}. The ANAMMOX granules were found to contain a plenty of extracellular polymers (ECPs) such as 71.8–112.1 mg g-VSS^{-1} of polysaccharides (PS) and 164.4–298.2 mg g-VSS^{-1} of proteins (PN). High content of hemachrome (6.8–10.3 μmol g-VSS^{-1}) was detected in the ANAMMOX granules, which is supposed to be attributed to their unique carmine color.

1. Introduction

Anaerobic ammonium oxidation (ANAMMOX) is a promising biotechnology for the treatment of ammonium-rich wastewater (van der Star et al., 2007; Joss et al., 2009). Under anoxic conditions, the ANAMMOX bacteria accomplish autotrophic ammonium oxidation to dinitrogen gas employing nitrite as an electron acceptor (Strous et al., 1998). It offers several advantages over conventional nitrification-denitrification systems including higher nitrogen removal rate, lower operational cost and less space requirement (Jetten et al., 2005; van der Star et al., 2007; Joss et al., 2009). Combined with single reactor high activity ammonium removal over nitrite (SHARON) process in which half of ammonium is converted to nitrite, the first full-scale ANAMMOX process (70 m^3) was applied to treat sludge dewatering effluents in Rotterdam, The Netherlands in 2002 (van Dongen et al., 2001; van der Star et al., 2007). It stably operated achieving nitrogen removal rate (NRR) up to 9.5 kg-N m^{-3} day^{-1} (van der Star et al., 2007).

High-rate is one of the prime objectives for ANAMMOX process. The NRR of conventional nitrogen removal biotechnologies was less than 0.5 kg-N m^{-3} day^{-1} (Jin et al., 2008); while for ANAMMOX process, it was higher than 5 kg-N m^{-3} day^{-1} as obtained by a number of researchers using different reactors such as upflow biofilter, upflow anaerobic sludge blanket (UASB) reactor and gas-lift reactor (Sliekers

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Nomenclature

ANAMMOX	Anaerobic ammonium oxidation
ECP(s)	Extracellular polymer(s)
HLR	Hydraulic loading rate
HRT	Hydraulic retention time
NLR	Nitrogen loading rate
NRR	Nitrogen removal rate
PN	Protein
PS	Polysaccharide
SAA	Specific ANAMMOX activity
SEM	Scanning electron microscopy
SHARON	Single reactor high activity ammonium removal over nitrite
SVI	Sludge volume index
TEM	Transmission electron microscopy
TSS	Total suspended solids
UASB	Upflow anaerobic sludge bed
VSS	Volatile suspended solids

et al., 2003; Imajo et al., 2004; Isaka et al., 2007; van der Star et al., 2007; Tang et al., 2009a). To date, the highest NRR reported was 26.0 kg-N m^{-3} day^{-1} at hydraulic retention time (HRT) of 0.24 h (Tsushima et al., 2007). Previous works on anaerobic processes including anaerobic digestion (Thiele et al., 1990) and denitrifying process (Franco et al., 2006) attributed high volumetric removal rates to three main aspects. Firstly, the reactors should have high-quality sludge retention for sufficient biomass accumulation. Secondly, the microbial communities should aggregate as granular sludge or biofilms for optimum metabolic activity. Finally, the substrate requirements of ANAMMOX bacteria should be satisfied simultaneously avoiding substrate inhibition, especially nitrite inhibition (Strous et al., 1999; Isaka et al., 2007; Tsushima et al., 2007).

The granular sludge characterized by good settling property and high activity plays a pivotal role in the performance of high-rate bioreactors (Thiele et al., 1990; Franco et al., 2006; Zhang et al., 2008). The characteristics of granular sludge such as heterotrophic aerobic granules (Beun et al., 1999; Beun et al., 2002; Zheng and Yu, 2007; Adav et al., 2008), anaerobic granules (Hulshoff Pol et al., 2004; Show et al., 2004; Wu et al., 2009), hydrogen-producing granules (Mu and Yu, 2006; Zhang et al., 2008), denitrifying granules (Franco et al., 2006) and autotrophic nitrifying granules (Tsuneda et al., 2003; Liu et al., 2008; Belmonte et al., 2009) have been extensively studied. In case of ANAMMOX granules, the settling property, diameter distribution and substrate diffusion have been reported (Arrojo et al., 2006; Ni et al., 2009). The characteristics of carmine color of ANAMMOX granules and their associated extracellular polymers (ECPs) have also drawn considerable attention for the process optimization. The hydroxylamine oxidoreductase and hydrazine oxidoreductase are two important enzymes of the ANAMMOX pathway. Both of these enzymes are rich in heme *c* (Klotz et al., 2008; Schmid et al., 2008), which endows the granular sludge with the carmine color. The extracellular polymers are assumed to be a key factor in the formation of granular sludge, which can be secreted by ANAMMOX bacteria (Cirpus et al., 2006).

In the present study, two ANAMMOX UASB reactors were operated to investigate the performance of high-loaded reactors possessing carmine granular sludge.

Table 1 – Operational parameters of the ANAMMOX sludge and the two UASB reactors before the start of the experiment.

Characteristic	R1	R2
A: Characteristics of the sludge		
Diameter (mm)	1.9	2.1
TSS/VSS (%)	82	85
SAA (kg-N kg-VSS^{-1} day^{-1})	0.3	0.2
B: Operational characteristics of the two UASB reactors before the start of the experiment		
Influent ammonium concentration (mg-N L^{-1})	300	200
Influent nitrite concentration (mg-N L^{-1})	360	240
Effluent recycling ratio	0.5	–
HRT (h)	6.90	11.7
Sludge concentration (g-VSS L^{-1})	18.7	26.8
NRR (kg-N m^{-3} day^{-1})	6.0	2.9

2. Material and methods

2.1. Synthetic wastewater

Ammonium and nitrite were supplemented to mineral medium as required in the form of $(NH_4)_2SO_4$ and $NaNO_2$, respectively. The composition of the mineral medium was (g L^{-1} except for trace element solution) (Trigo et al., 2006): KH_2PO_4 0.01, $CaCl_2 \cdot 2H_2O$ 0.00565, $MgSO_4 \cdot 7H_2O$ 0.3, $KHCO_3$ 1.25, $FeSO_4$ 0.00625, EDTA 0.00625 and 1.25 mL L^{-1} of trace elements solution. The trace element solution contained (g L^{-1}): EDTA 15, H_3BO_4 0.014, $MnCl_2 \cdot 4H_2O$ 0.99, $CuSO_4 \cdot 5H_2O$ 0.25, $ZnSO_4 \cdot 7H_2O$ 0.43, $NiCl_2 \cdot 6H_2O$ 0.19, $NaSeO_4 \cdot 10H_2O$ 0.21, $NaMoO_4 \cdot 2H_2O$ 0.22 and $NaWO_4 \cdot 2H_2O$ 0.050 (adapted from van de Graaf et al. (1996)).

2.2. ANAMMOX bioreactors

The experimental work was carried out in two glass-made UASB reactors of 1.1 L capacity having internal diameter of 50 mm. Both reactors were completely covered with black cloth to avoid the growth of phototrophic organisms and the related oxygen production (van der Star et al., 2008). The reactors were fed with synthetic wastewater which was flushed with 95%Ar-5%CO_2 continuously to maintain anoxic conditions. The temperature was set at 35 ± 1 °C according to Tsushima et al. (2007) and the influent pH was controlled in the range of 6.8–7.0 (Tang et al., 2009b). The produced gas was