Bioresource Technology 101 (2010) 2700–2705

Contents lists available at ScienceDirect

Bioresource Technology

journal homepage: www.elsevier.com/locate/biortech

Promoting sludge quantity and activity results in high loading rates in Anammox UBF

Jianwei Chen [a], Ping Zheng [a,*], Yi Yu [a], Chongjian Tang [a], Qaisar Mahmood [b]

[a] Department of Environmental Engineering, Zhejiang University, Hangzhou 310029, China
[b] Department of Environmental Sciences, COMSATS University, Abbottabad Campus, Pakistan

ARTICLE INFO

Article history:
Received 11 September 2009
Received in revised form 15 November 2009
Accepted 21 November 2009
Available online 24 December 2009

Keywords:
Anammox
Biomass growth
Specific activity
Nitrogen loading rate

ABSTRACT

The anaerobic ammonium oxidation (Anammox) was successfully started up in an upflow biofilm (UBF) reactor and operated for 435 days at 35 °C. The process development could be divided into four phases, i.e. endogenous denitrification without Anammox reaction (P1), functional biomass enrichment under relatively low nitrogen loading rate (NLR) (P2), specific Anammox activity (SAA) enhancement with high NLR (P3) and the final stable stage (P4). The maximal NLR was as high as 34.5 kg N/m^{3}d. Nitrogen mass balance showed that 88.84% of input nitrogen converted to dinitrogen gas, 1.74% was used for cellular synthesis and the rest was converted to nitrate. The final produced biomass was fast-growing with SAA of 1.8 g N/gVSS d and doubling time of 4.3–7.4 d. Granulation and biofilm formation contributed to the biomass enrichment, while appropriate recirculation as well as relatively high temperature (35 °C) helped to promote SAA. Effective retention time and promoted sludge activity were considered as the key factors for bacterial growth and efficient Anammox process.

1. Introduction

The Anammox (anaerobic ammonium oxidation) is a novel and promising alternative to the conventional nitrification–denitrification process treating wastewaters of low C/N ratio (Strous et al., 1998; van Dongen et al., 2001). It is carried out by bacterial species belonging to the order Planctomycetales that transform ammonium to nitrogen gas using nitrite as electron acceptor (Strous et al., 1999). Thus no external oxygen or carbon sources are required, which lower the operation cost. Besides, high nitrogen loading rates, e.g., 26 kg N/m^{3}d for lab-scale (Tsushima et al., 2007) and 9.5 kg N/m^{3}d for full-scale Anammox reactor (van der Star et al., 2007) are also very attractive.

Generally, high loading rates are dependent on two parameters, i.e. the quantity and activity of functional biomass in the reactor. The Anammox aggregates were estimated to have an extremely slow growth rate with doubling time of 11 days (Strous et al., 1998), leading to a longer start up period for the process. Many researchers have focused on the sludge retention strategies (Strous et al., 1998; Trigo et al., 2006; Fernandez et al., 2008), of which biofilm development and granulation are the most acceptable ones. The amendments in the liquid-induced shear force by shortening HRT may be an appropriate strategy to overcome slugging behavior of the Anammox reactor (Tang et al., 2009). The use of magnetic field (Liu et al., 2008) and sludge washout (Kieling et al., 2007) were also suggested to enhance the process performance. The addition of biocatalyst was another efficient way to accelerate Anammox start-up (Pynaert et al., 2004). Optimal conditions were commonly applied to enhance Anammox performance, including temperature of 35–40 °C (Dosta et al., 2008), pH of 8.0 (Strous et al., 1999), and an appropriate recirculation (Tsushima et al., 2007). Comparatively, very few reports exist that considered the improvement of Anammox biomass activity.

The main objectives of this work were as follows:

(1) The optimization of operational parameters based on the performance to increase sludge quantity and to promote sludge activity.
(2) The exploration of some macroscopic characteristics of Anammox biomass, which may be considered as indicators of the performance.
(3) To accomplish enrichment suitable as inoculum for the application of large-scale Anammox reactor.

2. Methods

2.1. Experimental set-up

A reactor of 8 L capacity was filled with hollow bamboo balls with diameters of 3–4 cm and used as Anammox upflow biofilter

* Corresponding author. Tel./fax: +86 57186971709.
E-mail address: pzheng@zju.edu.cn (P. Zheng).

0960-8524/$ - see front matter © 2009 Published by Elsevier Ltd.
doi:10.1016/j.biortech.2009.11.085

(UBF). The effluents were partially pumped back to the inlet as recirculation. The reflux ratio was originally set to 1:1, and changed when inlet substrates concentrations increased. Temperature was controlled at 35 ± 1 °C with a thermostatic jacket. And the influent pH was adjusted at neutral using 1 M H_2SO_4 solution. Inoculum was obtained from a full-scale wastewater treatment plant treating monosodium glutamate wastewater. The initial SS and VSS were 33.3 g L^{-1} and 14.6 g L^{-1}, respectively.

2.2. Feed stock

The reactor was fed with synthetic wastewater containing mg L^{-1} of KH_2PO_4 (0.01), $CaCl_2{\cdot}2H_2O$ (0.0056) (early periods) or (0.15) (later periods), $MgSO_4{\cdot}7H_2O$ (0.3) and $KHCO_3$ (1.25). Trace nutrient solutions 1 and 2 were also added by 1.25 mL L^{-1} each, which were prepared according to Strous et al. (1998). NH_4^+–N and NO_2^-–N were supplied in the form of $(NH_4)_2SO_4$ and $NaNO_2$, respectively.

2.3. Reactor operation

Initially, the Anammox reactor was fed with 30–50 mg L^{-1} NH_4^+–N and 50–70 mg L^{-1} NO_2^-–N at HRT of 9.6 h. This HRT was retained for 236 days, during which substrates were increased stepwise maintaining NO_2^-–N/NH_4^+–N ratio of 1.2–1.4. Thereafter, both HRT and substrate concentrations were adjusted to raise the NLR (Figs. 1 and 2). The NLR was not lifted till the effluent nitrite was lower than 50 mg L^{-1} for at least three HRTs.

Recirculation was employed to decrease the influent substrates concentrations, as the highest level in the feed stock reached 976 mg L^{-1} for NH_4^+–N and 1280.0 mg L^{-1} for NO_2^-–N, respectively. The reflux ratio varied for different substrate concentrations. When the influent ammonium and nitrite were below 190 mg L^{-1} and 250 mg L^{-1}, respectively, a reflux ratio of 1:1 was set, while 2:1 ratio was used at higher concentrations up to 300 mg L^{-1} (ammonium) and 400 mg L^{-1} (nitrite), respectively. A conservative ratio of 4:1 was available when higher strength (nitrite above 400 mg L^{-1}) influent was introduced.

2.4. Batch tests

The batch test was carried out according to methods reported by Dapena-Mora et al. (2007) and Dosta et al. (2008). Sludge taken from the reactor in period three was rinsed with phosphate buffer

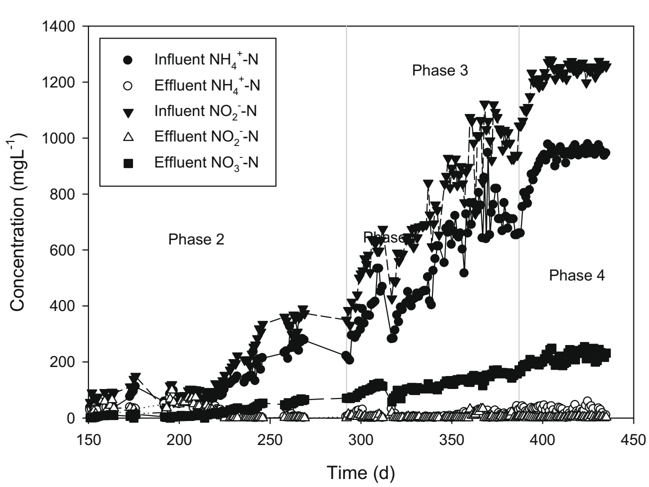

Fig. 1. Influent and effluent ammonium, nitrite and nitrate of Anammox reactor during the operation periods.

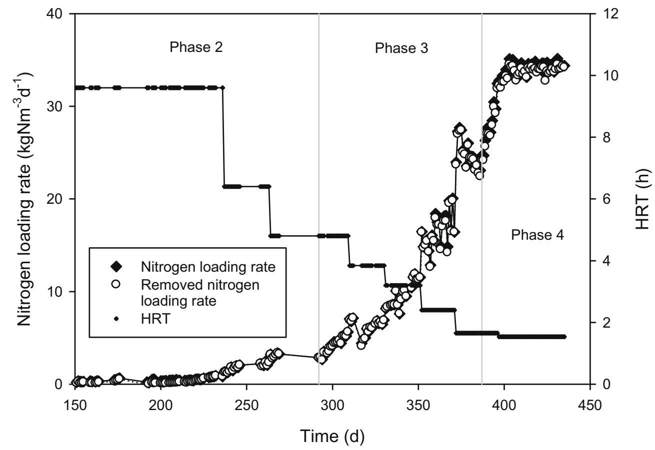

Fig. 2. The effect of HRT on nitrogen loading rate (NLR) with the passage of time.

(0.14 g L^{-1} KH_2PO_4 and 0.75 g L^{-1} K_2HPO_4) for five times. VSS in the test flask were controlled at 0.2–0.5 g L^{-1}. Temperature was maintained at 35 ± 0.1 °C and pH was adjusted to 7.0–8.0. Gas mixture comprising 95% Ar + 5% CO_2 was flushed for 15 min in order to create anaerobic conditions and to provide buffering capacity. Magnetic stirring at 150 rpm was carried out for mixing. The initial and final substrate concentrations were analyzed for mass balance calculation. Samples were taken using syringes and then filtered prior to measurement.

2.5. Analytical methods

The influent and effluent samples were collected on daily basis and were analyzed immediately or stored in a refrigerator at 4 °C until the analyses were carried out. NO_3^-–N, NO_2^-–N, NH_4^+–N, pH, SS and VSS were measured according to the standard methods (APHA, 1998). Ammonium was measured by using titrimetric method, nitrite was analyzed by using colorimetric method, and nitrate was analyzed by using ultraviolet spectrophotometric method. The suspended solids were measured by filtering the sample through a pre-dried filter paper. The filter was dried at 105 °C and then weighed after cooling. The volatile suspended solids was measured by burning the previous filter at 550 °C. Temperature was determined with a mercurial thermometer.

3. Results

3.1. UBF performance

According to the UBF performance, the operation could be divided into four different phases (Figs. 1 and 2).

During phase 1 (0–150 d), no significant Anammox was detected both in the continuous feeding and batch test. The effluent ammonium concentration was higher than that in the influent, and the input nitrite was almost fully consumed. No nitrate was generated, while a small volume of gas was collected. Hence, endogenous denitrification was regarded as the dominant reaction, since no organics were fed. This kind of phenomenon was also observed by other Anammox researchers during the start-up (Strous et al., 1997; Dapena-Mora et al., 2004b; Chamchoi and Nitisoravut, 2007; Wang et al., 2009).

Phase 2 started by day 151, when a small amount of ammonium was consumed. It lasted for 142 days. The Anammox evidently occurred as ammonium and nitrite were consumed (ammonium and nitrite removal efficiencies varied between 8.0–85% and 22.6–100%, respectively) and nitrate was produced in the range of 0.9–