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# SUSTAINABLE WATER AND SANITATION SERVICES FOR ALL IN A FAST CHANGING WORLD

# Anammox treatment performances using polyethylene sponge as a biomass carrier

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## **REFEREED PAPER 1979**

Nitrogen removal using a polyethylene (PE) sponge biomass carrier was evaluated in a fixed-bed reactor for nitrogen removal by the anammox process. The fixed-bed reactor was operated continuously for 240 days. Average T-N removal efficiencies of each period increased from 38 % to 67 %, 72 %, 74 % to 75 % with stepwise increases in volumetric T-N loading rates. A T-N removal rate of 2.8 kg N/m<sup>3</sup>/day was obtained after 240 days of operation. After 3 months, anammox biomass fully covered the surface of the PE sponge carrier and the color of the material changed from white to red. Following 5 months of operation, biomass proliferated on the surface of the material and a dark-red color was observed. These results shown that anammox process using PE sponge materials as biomass carriers in the fixed-bed reactor will be suitable for NH<sub>4</sub>-N removal from wastewater containing high NH<sub>4</sub>-N. However, it is necessary to investigate whether PE sponge material can operate under high organic carbon concentrations in anammox process, because these wastewaters always contain high concentration of organic matter.

## Introduction

In 1995, the anammox process was discovered during experiments on a denitrifying wastewater treatment pilot plant where at Gist-Brocades (Delft, Netherlands) (Mulder et al., 1995). It was demonstrated that anammox is a new process by which ammonium and nitrite are transformed to dinitrogen gas as the main product under autotrophic anaerobic conditions. In the anammox pathway, hydroxylamine (NH<sub>2</sub>OH) and hydrazine (N<sub>2</sub>H<sub>4</sub>) have been verified to be important intermediates (Jetten et al., 2002). The anammox reaction is exergenic ( $\Delta G^0 = -358 \text{ kJ/mol NH}_4^+$ ) and provides the energy for the fixation of CO<sub>2</sub> (Van de Graaf et al., 1995). The stoichiometry of the anammox reaction was determined to be (Strous et al., 1998):

 $NH_4^+ + 1.32 NO_2^- + 0.066 HCO_3^- + 0.13H^+ \rightarrow$ 

 $1.02 \text{ N}_2 + 0.26 \text{ NO}_3^- + 0.066 \text{ CH}_2\text{O}_{0.5}\text{N}_{0.15} + 2.03 \text{ H}_2\text{O} (1)$ 

Anammox treatment performances for nitrogen removal using polyvinyl alcohol (PVA) gel beads and malt ceramics (MC) as biomass carriers achieved good results. PVA gel beads were used in a fluidized-bed reactor (Tran et al., 2006) where a maximum T-N removal rate of 3.0 kg N/m<sup>3</sup>/day was obtained. In another study, two fixed-bed reactors using MC material with 3 to 5 mm and 10 to 15 mm diameter pieces as biomass carriers for anammox treatment were also carried out. These two fixed-bed reactors both achieved high T-N removal rates of 3.1 kg N/m<sup>3</sup>/day (Tran et al., 2006). PE sponge is a new choice for nitrogen removal using anammox process

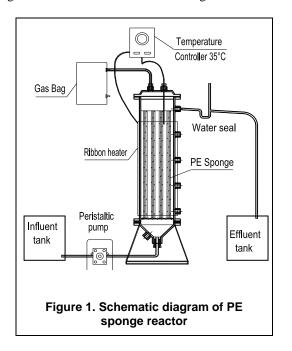
In this research, polyethylene (PE) sponge was used as biomass carrier for anammox sludge in a fixed-bed reactor to treat wastewater containing ammonium. The objective of this study is to investigate nitrogen removal capability of anammox process by using PE sponge as a biomass carrier.

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# Materials and methods

# Reactor set-up in laboratory-scale

The reactor used in this study was made from acrylic material with a total volume of 2.9 L. The reaction zone was 2.6 L and had an inner diameter of 9.0 cm and height (to effluent port) of 41 cm. A schematic diagram of the PE sponge reactor is shown in Figure 1. The reaction zone contains 6 vertically strips of sponge material for a total one-sided sheet area of 960 cm<sup>2</sup>. These 6 strips were folded as shown in Photograph 1 and fixed to a frame. The reactor had an airtight glass dome including vents for gas collection, gas emission and thermostatic sensing.





Photograph 1. PE sponge material frame

# Seed sludge

Anammox sludge taken from an existing nonwoven anammox reactor was used as seed sludge for the continuous-flow fixed-bed reactor. Before start-up, the seed sludge was circulated in the reactor for attachment on the surface of the PE sponge material.

## Composition of synthetic wastewater

Synthetic wastewater was prepared by adding ammonium and nitrite in the forms of  $(NH_4)_2SO_4$  and  $NaNO_2$ , respectively, to a mineral medium as described in Table 1. Tap water of groundwater origins was used for the preparation of this synthetic wastewater.

Table 1. Composition of synthetic wastewater	
Composition	Concentration
(NH <sub>4</sub> ) <sub>2</sub> SO <sub>4</sub> (mg N/L)	Variable (30-200)
NaNO <sub>2</sub> (mg N/L)	Variable (30-200)
KHCO <sub>3</sub> (mg/L)	125
KH <sub>2</sub> PO <sub>4</sub> (mg/L)	54
FeSO <sub>4</sub> .7H <sub>2</sub> O (mg/L)	9
EDTA (mg/L)	5

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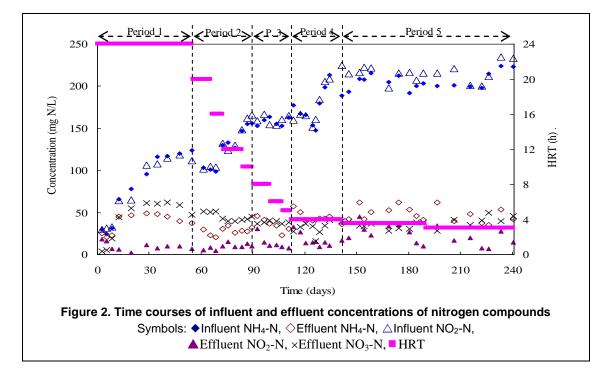
# **Results and discussion**

#### Removal efficiencies of nitrogen compounds

Influent NH<sub>4</sub>-N and NO<sub>2</sub>-N levels were increased from 30 to 200 mg N/L over a period of 240 days (Figure 2). During period 1 (the first 54 days), when the HRT was 24 h, effluent NH<sub>4</sub>-N levels were relatively high, ranging from 20 to 50 mg N/L, while effluent NO<sub>2</sub>-N levels were always very low with less than 10 mg N/L. NH<sub>4</sub>-N and NO<sub>2</sub>-N removal efficiencies were 44% and 81%, respectively. Furthermore, effluent NO<sub>3</sub>-N levels were also relatively high, from 20 to 60 mg N/L, with ratio of NO<sub>3</sub>-N production to influent T-N ranging from 25 to 38%, which was higher than the expected theoretical ratio of 10%. These results suggest the occurance of nitrification, with ammonium and nitrite being oxidized to nitrate under aerobic conditions resulting in low effluent nitrite and high effluent nitrate. These conditions are thought to be due to oxygen persisting inside the pores of PE sponge material because of the large pores of about 1060 µm in diameter.

During period 2 (days 55-89), HRT was decreased stepwise from 24 h to 20 h to 16 h to 12 h to 10 h and effluent  $NH_4$ -N levels progressively decreased to 20-30 mg N/L and  $NO_2$ -N levels to 5-15 mg N/L.  $NH_4$ -N and  $NO_2$ -N removal efficiencies were about 78% and 93%. In addition effluent  $NO_3$ -N levels were relatively high from 45 to 50 mg N/L and the ratio of  $NO_3$ -N production to influent T-N was from 15 to 25%, which it was lower than that of period 1.

During period 3 (days 90-111), the influent flow rate was increased from 6.2 L/d to 7.8 L/d to 10.4 L/d to 12.5 L/d for HRTs of 10 h, 8 h, 6 h and 5 h, respectively, and influent  $NH_4$ -N and  $NO_2$ -N levels were kept at 150mg N/L. At day 92, the effluent  $NH_4$ -N and  $NO_2$ -N levels were at 46 mg N/L and 30 mg N/L, respectively, which were higher than the values of previous runs. This is believed to be due to trouble with the temperature control system. After some days, effluent  $NH_4$ -N and  $NO_2$ -N concentrations decreased to less than 40 mg N/L and 20 mg N/L, respectively.  $NH_4$ -N and  $NO_2$ -N removal efficiencies of these 21 days of 78% and 92% were obtained, respectively. The ratio of  $NO_3$ -N production to influent T-N was 12 - 13% lower than that of period 2, which was closer to the theoretical ratio of 10%.

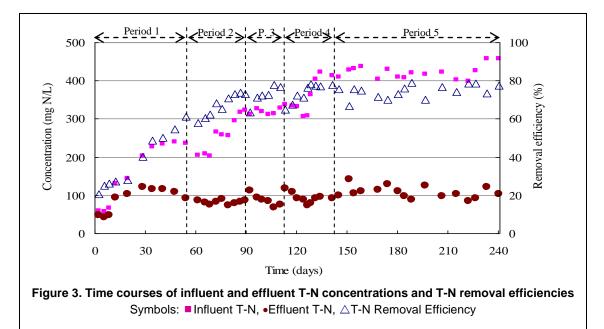


During period 4 (days 112-141), the influent flow rate was increased up from 12.5 L/d to 15.6 L/d for HRTs of 5 h to 4 h, respectively, and the influent  $NH_4$ -N and  $NO_2$ -N concentrations were increased stepwise from 150 mg N/L to 200 mg N/L.  $NH_4$ -N removal efficiency was 75% and  $NO_2$ -N removal efficiency was as high as 90%. The ratio of  $NO_3$ -N production to influent T-N was 8 – 12%, which was equal to the theoretical ratio. Therefore, is appears the anammox process had become dominant in comparison with nitrification.

During period 5 (days 142-240), the influent flow rate was increased up from 17.8 L/d to 20.8 L/d for HRTs of 3.5 h and 3 h, respectively, and the influent NH<sub>4</sub>-N and NO<sub>2</sub>-N concentrations were kept at 206 mg N/L and 215 mg N/L, respectively. NH<sub>4</sub>-N removal efficiency was 77% and NO<sub>2</sub>-N removal efficiency was as high as 90%, demonstrating stable and efficient anammox treatment during period 5.

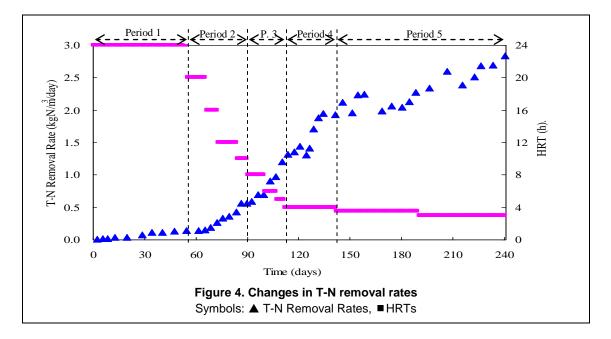
#### **T-N removal efficiency**

Influent and effluent T-N and T-N removal efficiencies are shown in Figure 3. Influent T-N was increased from 60 to 423 mg/L. During period 1 (the first 54 days), effluent T-N levels were very high reaching a maximum of 120 mg/L and average T-N removal efficiency was only 38% with minimum of 20% and maximum of 60%. During period 2 (days 55-89), effluent T-N levels were lower at 80-100 mg /L. T-N removal efficiency was changed from 58% to 74% and average value was higher at 67%. During period 3 (days 90-111), influent T-N was maintained at a level of 300 mg/L and effluent T-N decreased from 113 mg/L to 70 mg/L, while average value was increased to 72%. During period 4 (days 112-141), influent T-N was increased from 300 mg/L to 400 mg/L and effluent T-N levels fluctuated from 117 mg/L to 73 mg/L. Average T-N removal efficiency was 74%, which was higher than period 3. During period 5 (days 142-240), the average influent T-N was 423 mg N/L and effluent T-N was about 108 mg N/L and average T-N removal efficiency was more stable at 75%. During an operational period of 240 days, average T-N removal efficiencies improved stepwise from 38% to 75% and the highest value was 80%. These results shown that, after each period which increasing the flowrate or T-N concentration, the reactor had to adapt with new operational parameters. Therefore, the T-N removal efficiency was decreased slightly after increasing the flowrate or T-N concentration.



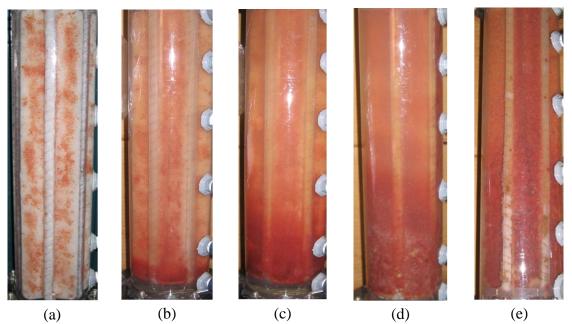
#### **T-N removal rates**

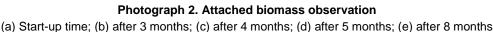
The reactor has been operated continuously for 8 months and T-N removal rate increased very slowly from 0.05 to 0.1 kg N/m<sup>3</sup>/day during period 1 (the first 54 days) as shown in **Figure 4**. This was due to the co-existing nitrification and the anammox processes in the reactor. However, T-N removal rates increased quickly from 0.1 to 0.6 kg N/m<sup>3</sup>/day during period 2 (days 55-89). Especially, T-N removal rate increased two time from 0.6 to 1.2 kg N/m<sup>3</sup>/day during period 3 (days 90-111). Consequently, the anammox bacteria were more active during this period than the previous period. In addition, T-N removal rates increased stepwise from 1.2 to 2 kg N/m<sup>3</sup>/day for a short time during period 4 (days 112~141). Subsequently, a T-N removal rate of 2.8 kg N/m<sup>3</sup>/day was obtained during period 5 (days 142-240), showing that the anammox bacteria were very active at this period.



## Attached biomass observation

Photograph 2 shows the variations in biomass color of the PE sponge reactor at start-up and after 3 months, 4 months, 5 months and 8 months of operation. After 3 months, biomass was attached strongly on the surface of the PE sponge material and its white color had changed to a reddish color. After 5 months, anammox biomass had proliferated on the surface of the PE sponge material and the color of biomass carrier changed to dark-red color. This observation shows that anammox bacteria grew quickly on the large porous PE sponge biocarrier. Influent wastewater was supplied from bottom of the reactor so that substrate concentration at bottom of the reactor is highest. Consequently, biomass grew better at the bottom of the reactor than at the upper part of the reactor.





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# Conclusions

In the continuous fixed-bed reactor using the PE sponge sheet as a biomass carrier, NH<sub>4</sub>-N and NO<sub>2</sub>-N removal efficiencies improved over the period of testing from 44% to 77% and 81% to 90%, respectively. During the 240 days of operation, T-N removal efficiencies increased stepwise from 38% to 75% as influent T-N was also increased stepwise from 60 to 423 mg N/L. The reactor was operated continuously for 8 months and the T-N removal rate reached 2.8 kg N/m<sup>3</sup>/day after 240 days of operation. This showed the effectiveness of the PE sponge sheet biomass carrier as a support material for the extremely slowly growing anammox bacteria and suitable for wastewater treatment with high ammonium concentration. After 8 months of operation, anammox biomass was attached strongly on the surface of the PE sponge material and dark-red color biomass was observed. This observation confirmed that anammox bacteria grew quickly within the large, 1060  $\mu$ m pores of the PE sponge biomass carrier, thus demonstrating its suitability as a biomass carrier for anammox bacteria. These results shown that anammox process using PE sponge materials as biomass carriers in the fixed-bed reactor will be suitable for NH<sub>4</sub>-N removal from wastewater containing high NH<sub>4</sub>-N. However, it is necessary to investigate whether PE sponge material can operate under high organic carbon concentrations in anammox process, because these wastewaters always contain high concentration of organic matter.

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