

# Treatment of ammonium in slaughterhouse wastewater by UASB technology combined with EGSB using anammox and PVA gel

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## **Abstract:**

Slaughterhouse wastewater (SWW) possesses very high organic and nutrient concentrations and its residues are moderately solubilized, which leads to pollution affecting the environment and human health. The objective of this study was to investigate the effective removal of ammonium in slaughter wastewater by up flow anaerobic sludge blanket (UASB) technology combined with an expanded granular sludge bed (EGSB) using anammox and PVA gel as the biomass carrier. Ammonium loading rates (NLRs) increased from 0.25 kg N-NH<sub>4</sub><sup>+</sup>/m<sup>3</sup>.d to 0.75 kg N-NH<sub>4</sub><sup>+</sup>/m<sup>3</sup>.d with hydraulic retention times (HRTs) of 12, 6, and 4 h. The system was operated in 2 phases. In phase 1, the removal of ammonium by employing the combination of UASB technology and EGSB using anammox was examined. The removal efficiencies of nitrite were 52% (NLRs=0.25 kg N-NH<sub>4</sub><sup>+</sup>/m<sup>3</sup>.d), 69% (NLRs=0.5 kg N-NH<sub>4</sub><sup>+</sup>/m<sup>3</sup>.d) and 64% (NLRs=0.75 kg N-NH<sub>4</sub><sup>+</sup>/m<sup>3</sup>.d). On the other hand, the removal efficiencies of ammonium were about 37% (NLRs=0.25 kg N-NH<sub>4</sub><sup>+</sup>/m<sup>3</sup>.d), 64% (NLRs=0.5 kg N-NH<sub>4</sub><sup>+</sup>/m<sup>3</sup>.d) and 55% (NLRs=0.75 kg N-NH<sub>4</sub><sup>+</sup>/m<sup>3</sup>.d). In phase 2, a PVA gel was supplied to the EGSB as the biomass carrier for growing the anammox sludge. The result showed that the removal efficiencies of nitrite were about 55% (NLRs=0.25 kg N-NH<sub>4</sub><sup>+</sup>/m<sup>3</sup>.d), 77% (NLRs=0.5 kg N-NH<sub>4</sub><sup>+</sup>/m<sup>3</sup>.d), and 73% (NLRs=0.75 kg N-NH<sub>4</sub><sup>+</sup>/m<sup>3</sup>.d). In addition, the removal efficiencies of ammonium were about 56% (NLRs=0.25 kg N-NH<sub>4</sub><sup>+</sup>/m<sup>3</sup>.d), 68% (NLRs=0.5 kg N-NH<sub>4</sub><sup>+</sup>/m<sup>3</sup>.d), and 60% (NLRs=0.75 kg N-NH<sub>4</sub><sup>+</sup>/m<sup>3</sup>.d).

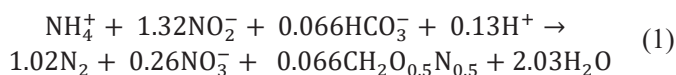
**Keywords:** ammonium removal, anammox, EGSB, PVA gel.

**Classification number:** 5.1

## **Introduction**

The main pollutant sources of wastewater from the slaughtering process are paunch, faeces, fat and lard, grease, undigested food, blood, suspended material, urine, loose meat, soluble proteins, excrement, manure, grit, and colloidal particles. SWW contains large amounts of biochemical oxygen demand (BOD), chemical oxygen demand (COD), total organic carbon (TOC), total nitrogen (TN), total phosphorus (TP), and total suspended solids (TSS). The treatment of SWW has been achieved by traditional methods such as aerobic and anaerobic biological systems.

Anammox (anaerobic ammonium oxidation) is a globally important microbial process of the nitrogen cycle that takes place in many natural processes. Anammox is a reaction that ammonium oxidation to dinitrogen gas using nitrite as the electron acceptor under anoxic conditions [1]. Since its discovery two decades ago, anammox-related research and its applications have experienced strong growth. Researchers have considered the anammox process as a method of treating the high-nutrient concentrations of wastewater. Based on mass balance from culture experiments using a sequencing batch reactor (SBR) to take account of the biomass growth, the anammox reaction has the following scaling coefficients [2, 3].



In comparison with traditional technologies, anammox has many advantages such as high nitrogen removal, low operational costs, and small space requirement [4]. Anammox has been successfully applied to treatment of wastewater on the laboratory scale, pilot scale, and full scale. Many types of wastewater have been surveyed with positive results. For example, the anammox process has been applied to the treatment of landfill leachate. This research

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showed that ammonium removal efficiency reached 88.1% and TN removal efficiency reached 80% [2]. However, in this study, the anammox process is applied in combination with PVA gel for the treatment of SWW. The purpose of the study is to assess slaughter wastewater treated by using UASB combined with EGSB technologies as well as to evaluate the factors that affect the treatment efficiency of these processes.

**Material and methods**

*Feed SWW*

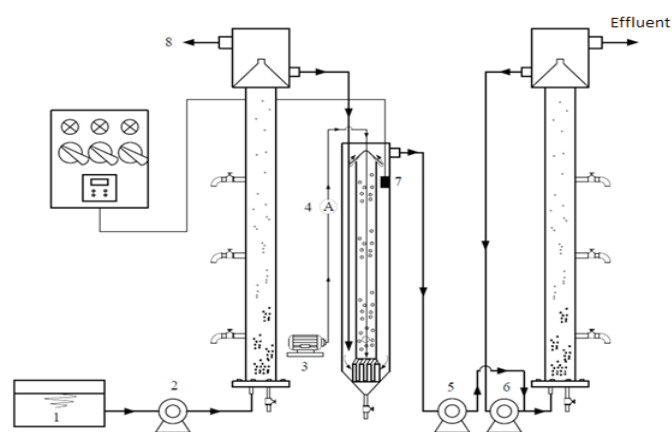
SWW was taken from the VISSAN Company’s wastewater treatment plant. The characteristic of the SWW is shown in Table 1.

**Table 1. Characteristics of SWW.**

Serial	Parameter	Unit	Value
1	pH		6.6-7.9
2	COD	mg/l	1,000-1,400
3	NH <sub>4</sub> <sup>+</sup> -N	mg/l	90-140
4	TKN	mg/l	130-170
5	NO <sub>2</sub> <sup>-</sup> -N	mg/l	0-1.58
6	NO <sub>3</sub> <sup>-</sup> -N	mg/l	0-2.50
7	Alkalinity	mg CaCO <sub>3</sub> /l	600-1,200
8	TP	mg/l	15-35
9	Temperature	°C	28-31

*Set-up of experiment and operational conditions*

The lab-scale system has three reaction tanks including the UASB, partial nitrification (PN), and EGSB is shown in Fig. 1.



**Fig. 1. Schematic diagram of the lab-scale system.** (1) Influent tank, (2) Influent pump, (3) Air pump, (4) Air valve, (5) Pump, (6) Circulating pump, (7) pH probe, (8) Biogas collection.

The wastewater pumped to the UASB was stored in a tank with volume of 90 l. The UASB is an acrylic tube with a working volume of 10 l with a height of 1.2 and 0.09 m internal diameter. On the column, there are 3 inspection valves. Each of these are 30 cm apart to collect wastewater and sludge samples. The PN also an acrylic tube. The working volume is 12.4 l with 0.78 m height and 0.14 m diameter. The central pipe is made of PVC and is composed of a 40 cm long section of pipe connected to a cone with a chisel around it. Air flow was supplied from the bottom of the tank through an air pump and adjusted through a valve. After passing the UASB-PN, wastewater will be stored in tanks with volume of 90 l and pumped into the EGSB tank. The EGSB tank is an acrylic tube with a working volume of 10 l, 1.2 m high and 0.09 m internal diameter. Water circulation in the tank is done through a circulating pump. The treatment efficiency of the system is analysed and evaluated.

*Enrichment of sludge and PVA gel*

*Enrichment of sludge:* anaerobic sludge is taken from the anaerobic tank and ammonia-oxidizing bacteria (AOB) sludge is taken from the aeration tank of the VISSAN wastewater treatment system. The anammox sludge is taken from the Institute of Tropical Biology, Ho Chi Minh city.

*PVA gel:* the PVA gel was provided by KURARAY AQUA CO., LTD. The PVA (Polyvinyl alcohol) gel is comprised of 4 mm spherical beads having a specific gravity of 1.025. One PVA-gel bead can hold up to 1 billion microorganisms depending on operating conditions [5].

*Operational conditions* (Table 2)

**Table 2. Operational conditions.**

Input flow (l/h)	HRT (h)	Ammonium loading rate (kg NH <sub>4</sub> <sup>+</sup> -N/m <sup>3</sup> .d)	DO PN (mg/l)	Operating time (d)
0.5	12	0.25	0.8-1.0	1-20
1	6	0.5	0.8-1.0	21-40
1.5	4	0.75	1-1.2	41-60

Wastewater was brought from the wastewater tank to the UASB through a pumping system. The reactor was operated in dark conditions by using a black plastic sheet fully covering the body to prevent the growth of algae. The mixed liquor suspended solids (MLSS) concentration of the reactor was maintained within 15,000 mg/l. The purpose of the UASB is to treat large quantities of organic matter in wastewater by converting organic nitrogen into ammonia to facilitate subsequent processing.

Water self-flowed from the UASB to the PN tank. The MLSS in the PN was kept in the range of 4,000-5,000 mg/l, the DO was adjusted from 0.8 to 1.2 mg O<sub>2</sub>/l, and the pH

was adjusted automatically through a pH controller and chemical pump.  $\text{NaHCO}_3$  salt was added to the PN tank to adjust the pH in the range of 7.5-8.5. The goal of the PN tank is to convert a part of  $\text{NH}_4^+$  into  $\text{NO}_2^-$  to a  $\text{NH}_4^+/\text{NO}_2^-$  ratio of 1/1.32 and to prevent the formation of  $\text{NO}_3^-$ , creating the most favourable conditions for the anammox process in the EGSB tank to take place.

The EGSB tank contains the activity of anammox microorganisms in anaerobic conditions. In addition, there is a water circulation pump that create a disturbance in the tank to increase the contact between the wastewater and microorganisms. The biological processes that take place in the tank will reduce the nitrogen content in the wastewater. The model is split into two stages. During stage one, the UASB/EGSB-anammox alone treated the SWW. In stage 2, the PVA gel was introduced into the model as a biomass carrier.

## Results and discussion

### The UASB/EGSB-anammox

*Partial nitrification (PN):* Figs. 2 and 3 show the loading rate of ammonium to be  $0.25 \text{ kg NH}_4^+\text{-N/m}^3\cdot\text{d}$  corresponding to an ammonium concentration of  $120 \pm 7.5 \text{ mg/l}$ . After the SWW passed through the UASB tank, the ammonium content increased to  $134 \pm 7.5 \text{ mg/l}$ . Nitrification process took place in the PN tank and the ammonium conversion efficiency was about 57%. The  $\text{NO}_2^-\text{-N}/\text{NH}_4^+\text{-N}$  ratio was about  $1.27 \pm 0.3$  and the highest ratio was 1.53 on the 20<sup>th</sup> day with an ammonium conversion efficiency of 63%. The DO in the PN tank at this stage was only about 0.8-1.0 mg/l, and the pH was in the range of 7.4-8.2 after long retention times to create conditions for AOB growth. The  $\text{NO}_3^-\text{-N}$  concentration of the effluent from the PN tank was very low ( $5 \pm 1.2 \text{ mg/l}$ ). This proved that the process in the PN tank was indeed the nitrification process, and the nitrification process was almost non-existent.

After the loading rate of ammonium was increased to  $0.5 \text{ kg NH}_4^+\text{-N/m}^3\cdot\text{d}$ , the input wastewater had a relatively stable ammonium content ( $123 \pm 8.8 \text{ mg/l}$ ). The ammonium concentration after passing through UASB tank increased to  $130 \pm 8 \text{ mg/l}$ . During the first few days during the loading process, the ratio of  $\text{NO}_2^-\text{-N}/\text{NH}_4^+\text{-N}$  was about 1.06 and the conversion rate was only about 51%. Because this value was quite low, the DO, pH and alkalinity parameters in the operation were adjusted to quickly improve the ratio. In the proceeding days, the ratio of  $\text{NO}_2^-\text{-N}/\text{NH}_4^+\text{-N}$  increased gradually day by day until the ratio reached its highest value on the 27<sup>th</sup> day, with an of  $\text{NO}_2^-\text{-N}/\text{NH}_4^+\text{-N}$  of 1.4 and conversion efficiency of nearly 57%. On the 30<sup>th</sup> day, the ratio of  $\text{NO}_2^-\text{-N}/\text{NH}_4^+\text{-N}$  was 1.31, which is similar to the

theoretical ratio, and the ammonium conversion efficiency reached 60%. In general, an average  $\text{NO}_2^-\text{-N}/\text{NH}_4^+\text{-N}$  ratio in the range of  $1.22 \pm 0.2$  is suitable for the anammox process in the EGSB tank.

After the first 10 days the loading rate of ammonium was up to  $0.75 \text{ kg NH}_4^+\text{-N/m}^3\cdot\text{d}$ , corresponding to HRT of 4 h, and the results showed that the conversion efficiency of ammonium decreased to 44%, the ratio of  $\text{NO}_2^-\text{-N}/\text{NH}_4^+\text{-N}$  fluctuated in the range of 0.9-0.93, and the lowest ratio, 0.79, was found on the 44<sup>th</sup> day. This proves that changing the load has a great impact on the processes. Increased load makes AOB sludge not able to adapt to the new living environment and other biological processes also become unstable. The process gradually stabilized in the following days. Then 10 days later, the ratio of  $\text{NO}_2^-\text{-N}/\text{NH}_4^+\text{-N}$  was  $1.1 \pm 0.04$  and relatively stable. On day 59, the ammonium conversion efficiency reached 59%.

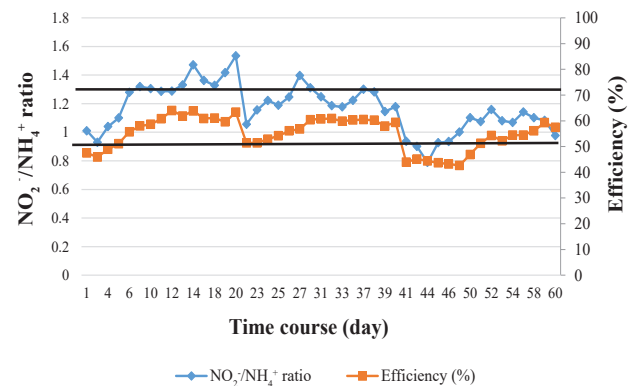


Fig. 2.  $\text{NO}_2^-\text{-N}/\text{NH}_4^+\text{-N}$  ratio in the survey process.

*Nitrogen removal efficiency:* the concentration of input and output nitrogen compounds of the EGSB tank is shown in Fig. 3. Over the first 20 days, the model was operated at a low loading rate of  $0.25 \text{ kg NH}_4^+\text{-N/m}^3\cdot\text{d}$  in order to allow the anammox bacteria to gradually adapt to SWW. The removal efficiency of  $\text{NO}_2^-\text{-N}$  increased with operation time, from the first day the removal efficiency was 22% and on the 20<sup>th</sup> day the removal efficiency was 52% with  $41.78 \text{ mg NO}_2^-\text{-N/l}$  removed. The average  $\text{NH}_4^+\text{-N}$  removal efficiency was 37% after 20 days of operation with  $18 \text{ mg NH}_4^+\text{-N/l}$  removed. At the same time, the amount of nitrate produced was  $1.8 \text{ mg NO}_3^-\text{-N}$ . This shows that the anammox bacteria began to adapt to the wastewater.

When the loading rate of ammonium was increased  $0.5 \text{ kg NH}_4^+\text{-N/m}^3\cdot\text{d}$  on the 21<sup>st</sup> day, the  $\text{NH}_4^+$  removal efficiency was 25% and the  $\text{NO}_2^-\text{-N}$  removal efficiency was 27%. This indicated that the anammox bacteria cannot adapt to new loads yet. After the loading rate increased, the processing efficiency increased markedly in the following days shown by an adjustment of the  $\text{NO}_2^-\text{-N}/\text{NH}_4^+\text{-N}$  ratio

in the range of 1.0-1.4 which created favourable conditions for the anammox bacteria. After 20 days of operation at an ammonium loading rate of 0.5 kg NH<sub>4</sub><sup>+</sup>-N/m<sup>3</sup>.d, the NH<sub>4</sub><sup>+</sup> removal efficiency was 64% and the NO<sub>2</sub><sup>-</sup>-N removal efficiency was 70%. The amount of NO<sub>3</sub><sup>-</sup>-N generated was about 8.8% compared to the amount of NH<sub>4</sub><sup>+</sup>-N consumed, which proves that the nitrate reduction process coexisted with anammox process.

When the loading rate of ammonium was increased 0.75 kg NH<sub>4</sub><sup>+</sup>-N/m<sup>3</sup>.d, the treatment efficiency had a sharp decline over the first few days. The NH<sub>4</sub><sup>+</sup> removal efficiency was 39% and the NO<sub>2</sub><sup>-</sup>-N removal efficiency was 18%. The main cause of this situation is that the annamox bacteria could not adapt to the sudden change in load. In the following days, the operating conditions reached a steady state whereby the removal efficiency increased gradually. On the last day, the performance reached 57 and 69%, with 26 mg NH<sub>4</sub><sup>+</sup>-N/l and 30.5 mg NO<sub>2</sub><sup>-</sup>-N/l removed. The average removal performance at this load was 55% (NH<sub>4</sub><sup>+</sup>-N) and 64% (NO<sub>2</sub><sup>-</sup>-N). The amount of NO<sub>3</sub><sup>-</sup>-N generated was about 5% compared to the amount of NH<sub>4</sub><sup>+</sup>-N consumed.

In general, the input NH<sub>4</sub><sup>+</sup>-N concentration was 134±5, 130±8, and 110±10 mg/l for ammonium loading rate of 0.25, 0.5, and 0.75 kg NH<sub>4</sub><sup>+</sup>-N/m<sup>3</sup>.d, respectively, and the ammonium treatment efficiency of the model reached 37, 64, and 55% respectively.

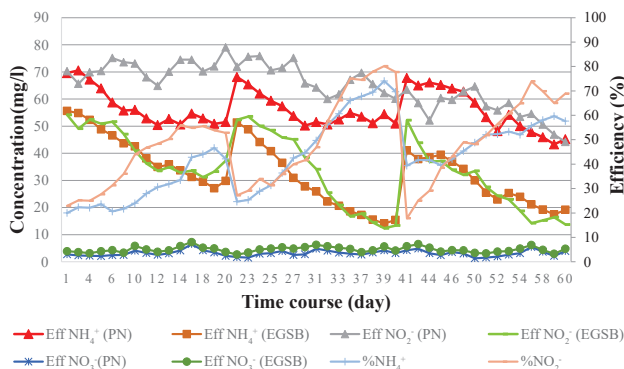


Fig. 3. Concentration of input and output nitrogen compounds of the EGSB tank.

**The UASB/EGSB-anammox combined with PVA gel**

PN: from Figs. 4 and 5 show that the 0.25 kg NH<sub>4</sub><sup>+</sup>-N/m<sup>3</sup>.d loading rate of ammonium, the input ammonium concentration was 126±10 mg/l. The ammonium conversion efficiency was about 58%, and the NO<sub>2</sub><sup>-</sup>-N/NH<sub>4</sub><sup>+</sup>-N ratio was 1.16±0.29. On the 7<sup>th</sup> day, the NO<sub>2</sub><sup>-</sup>/NH<sub>4</sub><sup>+</sup> ratio decreased to 0.75 because the system had problems during operation making the conversion rate from ammonium to nitrite lower

than required. After fixing the problem, the NO<sub>2</sub><sup>-</sup>/NH<sub>4</sub><sup>+</sup> ratio increased gradually, and on the 14<sup>th</sup> day the NO<sub>2</sub><sup>-</sup>/NH<sub>4</sub><sup>+</sup> ratio was 1.32 with ammonium conversion efficiency of 64%.

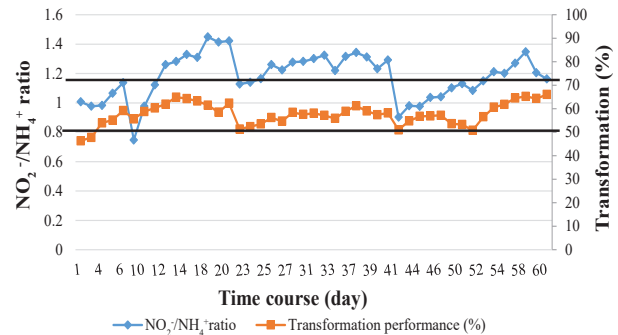


Fig. 4. NO<sub>2</sub><sup>-</sup>/NH<sub>4</sub><sup>+</sup> ratio in the survey process.

After increasing the loading rate of ammonium up to 0.5 kg NH<sub>4</sub><sup>+</sup>-N/m<sup>3</sup>.d, the ammonium conversion efficiency decreased to 51%. The input ammonium was 133±6 mg/l and the average NO<sub>2</sub><sup>-</sup>/NH<sub>4</sub><sup>+</sup> ratio was about 1.25±0.12. On the 32<sup>nd</sup> day, the ratio was 1.32. This ratio is the ideal theoretical ratio. While the ratio in this period was relatively unstable most of the ratios were in the range of 1.0-1.4 which meant they were still suitable for the next process.

On days 41 to 44, the loading rate of ammonium was increased to 0.75 kg NH<sub>4</sub><sup>+</sup>-N/m<sup>3</sup>.d corresponding to an HRT of 4 h. The results showed that the conversion efficiency of ammonium decreased to 51%. The ratio of NO<sub>2</sub><sup>-</sup>/NH<sub>4</sub><sup>+</sup> decreased to 0.9±0.07 because the sludge did not adapt to the change in loading rate. In the following days, the ratio of NO<sub>2</sub><sup>-</sup>/NH<sub>4</sub><sup>+</sup> increased gradually. On the 58<sup>th</sup> day, the highest ratio reached 1.34 with an ammonium conversion efficiency of 65%. The average ammonium conversion efficiency was 58%.

*Nitrogen removal efficiency:* the concentration of input and output nitrogen compounds from the EGSB tank is shown in Fig. 5. Over the first 20 days, the model was operated with a low loading rate of 0.25 kg NH<sub>4</sub><sup>+</sup>-N/m<sup>3</sup>.d, and the removal efficiency of NO<sub>2</sub><sup>-</sup> and NH<sub>4</sub><sup>+</sup> increased with operation time. On the first day, the removal efficiency of NO<sub>2</sub><sup>-</sup> was about 35% and the removal efficiency of NH<sub>4</sub><sup>+</sup> was about 42%. On the 6<sup>th</sup> day, the removal efficiency of NO<sub>2</sub><sup>-</sup> increased to 51% and the removal efficiency of NH<sub>4</sub><sup>+</sup> increased to 56%. On the 7<sup>th</sup> day, the removal efficiency of NO<sub>2</sub><sup>-</sup> unexpectedly dropped to 32.6% and the removal efficiency of NH<sub>4</sub><sup>+</sup> was about 35% because the NO<sub>2</sub><sup>-</sup>/NH<sub>4</sub><sup>+</sup> ratio was 0.75. After fixing a problem in the PN tank, the NH<sub>4</sub><sup>+</sup> and NO<sub>2</sub><sup>-</sup> treatment efficiency increased gradually and became relatively stable. The average processing efficiency was about 55% for NH<sub>4</sub><sup>+</sup> and 55% for NO<sub>2</sub><sup>-</sup>. At the same time, the production of NO<sub>3</sub><sup>-</sup> was about 6.4% of the influent NH<sub>4</sub><sup>+</sup>.



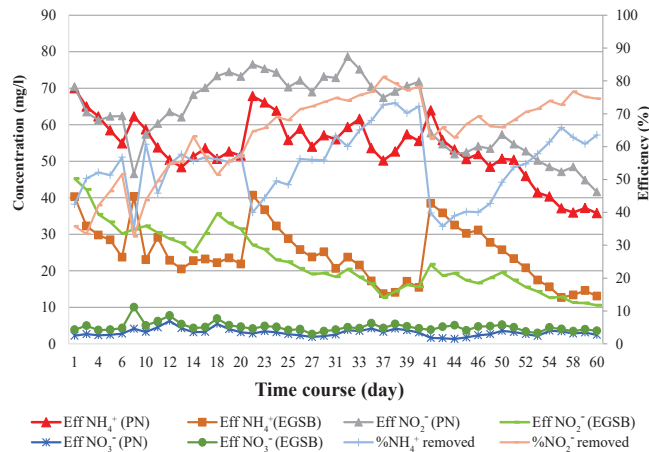


Fig. 5. Concentration of input and output nitrogen compounds of EGSB tank.

When increasing the loading rate of ammonium to 0.5 kg  $\text{NH}_4^+\text{-N}/\text{m}^3\cdot\text{d}$ , the removal efficiency of  $\text{NO}_2^-$  and  $\text{NH}_4^+$  was relatively stable. After 20 days of operation, the highest removal efficiency value was reached on day 17, with 73% for  $\text{NH}_4^+\text{-N}$  removal and 81% for  $\text{NO}_2^-\text{-N}$  removal. The average  $\text{NH}_4^+\text{-N}$  removal efficiency was about 68% and the average  $\text{NO}_2^-\text{-N}$  removal efficiency was about 77%. Over the last 5 days of this period, the output of ammonium nitrogen was approximately  $14 \pm 0.56$  mg/l. The amount of  $\text{NO}_3^-\text{-N}$  produced was about 5% of the influent  $\text{NH}_4^+$ .

The loading rate of ammonium was increased to 0.75 kg  $\text{NH}_4^+\text{-N}/\text{m}^3\cdot\text{d}$ . On the first day, the removal efficiency of  $\text{NO}_2^-$  was 62% and the removal efficiency of  $\text{NH}_4^+$  was 40%. Then, over the following days, the removal performance increased slowly. On the 54th day, the effect reached a steady state, and the  $\text{NH}_4^+\text{-N}$  removal efficiency was about 61% and the  $\text{NO}_2^-\text{-N}$  removal efficiency was about 74%. On the last day, the performance reached 63%  $\text{NH}_4^+\text{-N}$  removal and 75%  $\text{NO}_2^-\text{-N}$  removal, with 24.0 mg  $\text{NH}_4^+\text{-N}/\text{l}$  and 46

mg  $\text{NO}_2^-\text{-N}/\text{l}$  removed.

## Conclusion

The UASB/EGSB-anammox system was applied to treat SWW. HRTs were surveyed from 12, 6, and 4 h, and the ammonium removal efficiencies were 37, 64, and 55%, respectively. The nitrite removal efficiencies were 52, 69, and 64%, respectively. The PVA gel added during the second phase of the model showed an increase in pollution handling and model stability when operating at high loading rates. The ammonium removal efficiencies were 56, 68, and 60% for HRTs of 12, 6, and 4 h, respectively, and nitrite removal efficiencies were 55, 77, and 73%, respectively. This research model can be adapted to higher loads in order to assess its ability to handle critical conditions.

The authors declare that there is no conflict of interest regarding the publication of this article.

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