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APPLICATION OF ASBR IN ANAEROBIC AMMONIA OXIDATION

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Abstract: The effect of continuous anaerobic ammonia oxidation in an ASBR (Anaerobic sequencing batch reactor) was investigated. It was found that mature anaerobic ammonia oxidation activated sludge could quickly adapt to the reactor within one month and have a stable effluent. After 67 days of cultivation, the concentration of ammonia nitrogen had been in a position meeting the NH_4^+-N first-level A standard of Discharge standard of pollutants for municipal wastewater treatment plant (GB 18918-2002) continuously and stably. After 81 days, the anaerobic ammonium oxidation activated sludge SS increased from 2880 mg/l to 5580 mg/l.

Keywords: ASBR; anaerobic ammonia oxidation; continuous operation.

Foreword

Anaerobic ammonia oxidation process, does not need to add additional organic carbon source. Also does not need aeration compared with traditional sewage treatment process. Ammonia nitrogen and the nitrite nitrogen can be proportional to remove and generate nitrogen directly at the same time. However, anammox bacteria have strict requirements on their own living environment.

Shen [1] used anaerobic ammonia-oxidizing bacteria stably running for more than 300 days to study the effects of different concentrations of sodium acetate on anaerobic ammonia oxidation, and found that sodium acetate had a catalytic effect on anaerobic ammonia oxidation at low concentrations (0-200 mg/l), equivalent COD at 0-116 mg/l. Chamchoi [2] found that when the COD concentration was higher than 300mg/l, the strains were inactivated in UASB reactor. Zu[3] further demonstrated the effect of COD on anammox bacteria by activating anaerobic ammonia oxidation microbial fuel cells, and the low glucose concentration (100~200 mg/l), the electrical performance of **anaerobic ammonia oxidation**-microbial fuel cells (ANAMMOX-MFC) denitrification was enhanced. When the glucose concentration was higher than 300 mg/l, the electricity generation performance was degraded. Therefore, when

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the anaerobic ammonia oxidation reaction is performed, it is necessary to draw attention to the COD concentration in the reactor.

Anaerobic ammonia-oxidizing bacteria are anaerobic bacteria that are very sensitive to dissolved oxygen (DO). When the DO in water was 0.5% -2% of the air saturation, the activity of anaerobic ammonia-oxidizing bacteria was completely inhibited; the inhibitory concentration was more than 0.5% air saturation [4]. The test [5] showed that DO could inhibit anaerobic ammonia oxidation activity, but the activity of anammox bacteria rapidly recovered after removing high dissolved oxygen from the sewage. Studies [6] had shown that when the dissolved oxygen was 2-5 mg O_2/L, the activity of anammox bacteria was not affected.

The effect of pH on the activity of anaerobic ammonia-oxidizing bacteria in the process of nitrogen removal is mainly shown in two aspects: a. the influence of pH on the bacteria itself; b. the influence of pH on the substrates such as ammonia nitrogen and nitrite nitrogen, and then affects the physiological activity of anaerobic ammonia-oxidizing bacteria.

When the optimum pH of anammox bacteria was 6.7-8.3, and lower than 6.0 or higher than 9.5, the activity of anammox bacteria will be weakened [4]. Zheng Meng[7] studied the preservation of anaerobic ammonia-oxidizing bacteria and found that the anaerobic ammonia-oxidizing bacteria had the highest activity at the pH of 8 at 15 °C.

Yang [8] studied the effect of temperature on the activity of anammox sludge by measuring the anaerobic ammonia oxidation rate. The optimum temperature of anammox bacteria was 30-35°C, and the physiological activity of anaerobic ammonia-oxidizing bacteria decreased above 45°C or below 15°C.Li [9] stored anaerobic ammonia sludge particles at different temperatures and studied the sludge activity. It was noted that the anaerobic ammonia oxidation activity had little effect at room temperature and low temperature. Zeng [10] found that the abundance of anammox bacteria in summer subsurface wetlands was higher than that in autumn, but the diversity of anammox bacteria in autumn was higher than in summer. After running the SBBR reactor at 15°C for a long time, the temperature suddenly dropped to 11°C, and the activity of anaerobic ammonia-oxidizing bacteria was inhibited.[11]

In realistic applications, whether anaerobic ammonia oxidation process can be started quickly and run smoothly has a direct relationship with the reactor and operating method used.

It is generally believed that the doubling time of anammox bacteria is about 21 days. The commonly used reactors include fluidized bed reactor, fixed bed biofilm reactor, sequencing batch reactor (SBR), membrane bioreactor (MBR), upflow anaerobic sludge bed reactor (UASB) and so on. The ideal anammox reactor should satisfy: (1) maintaining a sufficient amount of microorganisms; (2) stable and reliable operation; (3) strictly anaerobic and light-proof; (4) no dead zone or no local high substrate concentration zone (5) good mass transfer performance [12].

With the deepening of research, many researchers had used different reactors to cultivate anammox bacteria, and also obtained a variety of conclusions. Wang[13] used MBR reactor to culture anaerobic ammonia-oxidizing bacteria from a start-up time of 60 days; Gong [14] studied ANAMMOX against NH_4^+-N and NO_2^--Nin a UASB reactor. Thesuitable concentration load was 220 mg/L, the hydraulic retention time 4 h, the optimum temperature 35 °C, and the optimum pH 8.0. Under these conditions, the removal rates of NH_4^+-N, NO_2^--N and TN were respectively up to 97%, 98.5% and 88%. Chen [15] used EGSB process to cultivate anammox bacteria, in which volumetric load and volumetric nitrogen removal rate were above other the record.

There are many factors affecting the activity of anammox bacteria mentioned above. Strous[16] proposed this use of sequencing batch reactors for anaerobic ammonia oxidation. This reactor cannot only reduce the loss of bacteria, but also enrich it. In this study, a self-designed SBR reactor was utilized to culture anaerobic ammoxidation bacteria in order that the reactor could run stably.

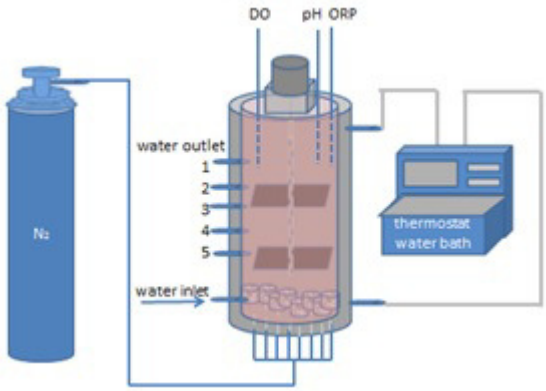

Fig. 1 Simple device for anaerobic SBR reactor

1.1 Experimental water

Artificial water was used for the experimental water according to (g/L) KH_2PO_4 0.025, $CaCl_2 \cdot 2H_2O$ 0.3, $MgSO_4 \cdot 7H_2O$ 0.2, $FeSO_4$ 0.006 25, $KHCO_3$ 1.25, EDTA 0.006 25 and trace element liquid 1.25 mL/L. The trace element liquid composition was (g/L): EDTA 15;$ZnSO_4 \cdot 7H_2O$ 0.43;$NaSeO_4 \cdot 10H_2O$ 0.21;H_3BO_4 0.014;$CoCl_2 \cdot 6H_2O$ 0.24;$MnCl_2 \cdot 4H_2O$ 0.99;$CuSO_4 \cdot 5H_2O$ 0.25;$NaMoO_4 \cdot 2H_2O$ 0.22;Ni $Cl_2 \cdot 2H_2O$ 0.19 and $NaWO_4 \cdot 2H_2O$ 0.050. NH_4Cl and $NaNO_2$ were provided as main inorganic nitrogen sources for anammox bacteria, and their concentrations were increased later.

1.2 Detection methods

NH_4^+-N was detected by Nessler's reagent spectrophotometric method; NO_2^--N was determined by N-(1-naphthy)-ethylene diamine spectrophotometry; NO_3^--N was determined by ultraviolet spectrophotometry; SS was determined by gravimetric method. The pH, DO, and ORP were monitored using an HQ40d Hach Portable Meter.

1.3 Reactor operation

The reactor used in this study was a hollow column anaerobic SBR with a height of 30cm and an internal diameter of 8cm. In the reactor, domesticated mature anaerobic ammonia oxidation activated sludge was added. Removal loads of ammonia nitrogen and nitrite nitrogen were 100% and 98.84% respectively, and the SS was 2880 mg/l. The dissolved oxygen in the reactor was controlled to be lower than that of 0.3mg/l by nitrogen blowing. Hydraulic retention time (HRT) was placed at 24h. The temperature was controlled at about 35 °C, and the outer wall of the reactor was wrapped with tin foil, reducing photosynthetic bacteria to compete for substrates. Every day, 1000ml of water was entered and 1000ml of water was discharged, and the water quality was tested, including ammonia nitrogen, nitrite nitrogen and nitrite nitrogen.

Results and Discussion

Anaerobic ammonia oxidation activated sludge was put into an anaerobic SBR, and the reactor had been continuously operated for 81d. By analyzing the influent and effluent concentrations of ammonia nitrogen, nitrous nitrogen, and nitrate nitrogen, as well as ΔNO_2^--N/ΔNH_4^+-N and ΔNO_3^--N/ΔNH_4^+-N, the entire process could be divided into

adaptation stage and activities raise the stage.

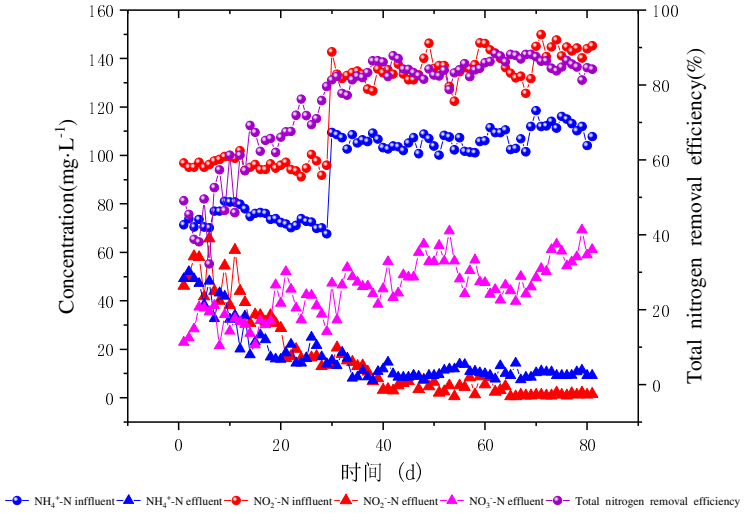

Figure 2 Total nitrogen removal efficiency during ASBR operation

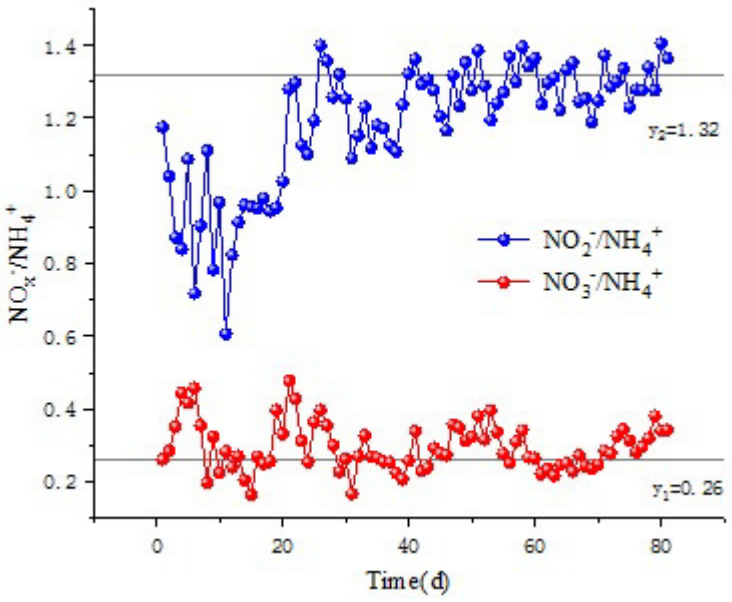

Fig.3 Change of ΔNO_2^--N/ΔNH_4^+-N and ΔNO_3^--N/ΔNH_4^+-N

2.1 Adaptation stage

The influent ammonia nitrogen and nitrite nitrogen concentrations were75 mg/l and 95 mg/, respectively. After 29 days of cultivation, the anaerobic ammonia oxidation activated sludge was in a position to adapt to the anaerobic SBR reactor. The total nitrogen removal efficiency was given in Figure 2 from 49.05% to 79.59% (day 29), and the effluent quality was stable.

$$NH_4^+ + 1.32\ NO_2^- + 0.066HCO_3^- + 0.13H^+ \rightarrow 1.02N_2 + 0.26\ NO_3^- + 0.066CH_2O_{0.5}N_{0.15} + 2.03H_2O$$

As showed in the above, anaerobic ammonia oxidation biochemical reaction equation, the reduction ofNO_2^--N/ the reduction of NH_4^+-N is 1.32, and the increase of NO_3^--N/ the reduction of NH_4^+-N is 0.26. It could also be seen in figure 3 that the theoretical values of the initial ΔNO_2^--N/ΔNH_4^+-N and ΔNO_3^--N/ΔNH_4^+-Nas quite different from those of the equation of anaerobic ammonia oxidation and were unstable. At the end of 29 days, their values was relatively stable, indicating that they had been adjusted to adapted to the operation of the reactor.

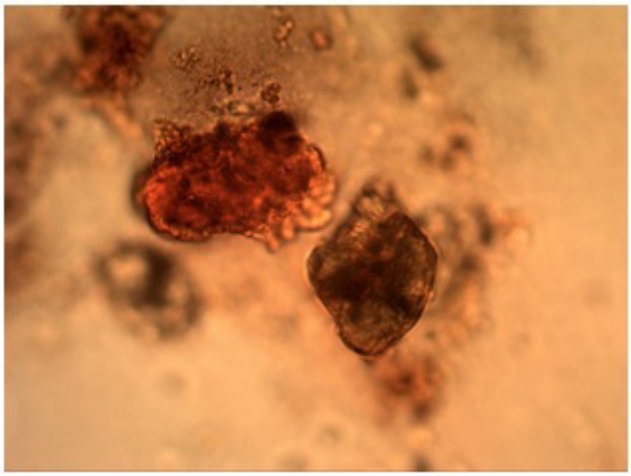

Figure 4: Anaerobic ammonia-oxidizing bacteria amplified 1000 times under light microscope

2.2 activities raise stage

For increasing the anaerobic ammonia oxidation activated sludge's activity that had adapted to ASBR, the influent concentrations of ammonia nitrogen and nitrous nitrogen were increased to 105 mg/l and 135 mg/l, respectively. At this stage, the reactor continued to operate stably, and the concentration of ammonia-nitrogen effluent had been reduced to 1.26 mg/l at the time of culturing to 67 days. This concentration of ammonia nitrogen had been far below 5mg/l- the Class A standard of Discharge standard of pollutants for municipal wastewater treatment plant (GB 18912-2002). The total nitrogen removal efficiency was stable at 85.57% (67-81d). This value was not higher, because the anaerobic ammonia oxidation reaction was the process of producing nitrate. As shown in the equation, while removing 1mol of ammonia nitrogen, 0.26mol of nitrate nitrogen producing, so the total nitrogen removal efficiency of the entire reactor was not very high.

ΔNO_2^--N/ΔNH_4^+-N and ΔNO_3^--N/ΔNH_4^+-N fluctuated around their respective theoretical

values at this stage, but the range of fluctuation was significantly smaller than before, and SS was 5580 mg/l at 81 d. It showed that anammox activated sludge could fully adapt the reactor and be enriched in the reactor.

In conclusion

Anaerobic ammonia oxidation activated sludge can adapt to the anaerobic SBR designed in this study within 30 days and can be enriched. The reactor can be operated stably to cultivate anaerobic ammonia oxidizing bacteria and increase its activity. The total nitrogen removal efficiency couldreach 85.57% when the reactor was operated after 67 days.

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