Wastewater Treatment in a Hybrid Biological Reactor (HBR) : Nitrification Characteristics[1]

JIAN-LONG WANG[‡,2] AND LI-BO WU[#]

[‡]Laboratory of Environmental Technology, INET, Tsinghua University, Beijing 100084, China;
[#]College of Environmental Science and Engineering, Nankai University, Tianjin 300071, China

Objective To investigate the nitrifying characteristics of both suspended- and attached-biomass in a hybrid bioreactor. **Methods** The hybrid biological reactor was developed by introducing porous ceramic particles into the reactor to provide the surface for biomass attachment. Microorganisms immobilized on the ceramics were observed using scanning electron microscopy (SEM). All chemical analyses were performed in accordance with standard methods. **Results** The suspended- and attached-biomass had approximately the same nitrification activity. The nitrifying kinetic was independent of the initial biomass concentration, and the attached-biomass had a stronger ability to resist the nitrification inhibitor. **Conclusion** The attached biomass is superior to suspended-biomass for nitrifying wastewater, especially that containing toxic organic compounds. The hybrid biological reactor consisting of suspended- and attached-biomass is advantageous in such cases.

Key words: Attached biomass; Hybrid biological reactor; Nitrification; Suspended biomass; Nitrification inhibitor

INTRODUCTION

Biological nutrient removal (BNR) is becoming increasingly common in both domestic and industrial wastewater treatment. While the removal of phosphorus can be achieved both chemically and biologically, nitrogen removal is almost exclusively done biologically. The removal of ammonium is an important problem in modern wastewater treatment systems. Ammonium is a common pollutant and can cause eutraphication of receiving watercourse. It is generally eliminated from wastewater by a combination of two processes, i.e. nitrification and denitrification. Ammonium is oxidized under aerobic conditions to nitrite and then nitrate (nitrification) which is subsequently reduced to nitrogen gas under anoxic conditions (denitrification).

The mechanism of nitrification can be described in the following two reactions.

$$2H^{+}+NH_3+2e^{-}+O_2 \rightarrow NH_2OH+H_2O \quad (1)$$

$$NH_2OH+H_2O \rightarrow HNO_2 + 4H^{+} + 4e^{-} \quad (2)$$

The oxidation of NH_3 to NO_2^- by ammonia-oxidizing bacteria (AOB) is a two-step process, proceeding via hydroxylamine. Ammonia monooxygenase (AMO) catalyzes the

[1]This work was supported by National Natural Science Foundation of China (Grant No. 59978020).

[2]Correspondence should be addressed to Dr. Jian-Long WANG, Tel: 86-10-62784843. Fax: 86-10-62771150. E-mail: wangjl@tsinghua.edu.cn

Biographical note of the first author: Jian-Long WANG, male, born in 1964, Ph. D., majoring in the field of water pollution control and environmental biotechnology.

0895-3988/2004
CN 11-2816

oxidation of NH_3 to NH_2OH, and hydroxylamine oxidoreductase (HAO) catalyzes the oxidation of NH_2OH to NO_2^-. Two of the electrons produced in the second reaction are used to compensate for the electron input of the first reaction, whereas the other two are passed *via* an electron transport chain to the terminal oxidase, thereby generating a proton motive force.

$$2H^+ + 0.5O_2 + 2e^- \rightarrow H_2O \quad (3)$$

The sum reaction is given as follows:

$$2NH_3 + 3O_2 \rightarrow 2HNO_2 + 2H_2O \quad (4)$$

Nitrite oxidation process:

$$NO_2^- + 0.5O_2 \rightarrow NO_3^- \quad (5)$$

In biological wastewater treatment systems, the biomass may be in the suspended growth form, as in the activated sludge process, or in the attached growth form, as in the trickling filters and rotating biological contactors. Some treatment systems such as fluidized bed systems contain both suspended and attached biomass and can be called hybrid growth systems[1,2].

The biomass concentration in the biological reactors can be increased by various immobilization techniques. The passive immobilization can be achieved by providing solid surfaces to facilitate the natural process of microbial attachment. This application of support materials to the activated sludge process combines the advantages of both the attached growth systems and suspended growth systems. By means of biomass carriers, it is possible to obtain about two fold increase in biomass concentration in the aeration tanks compared to that in the conventional activated sludge process[3]. Activated sludge technologies combined with immobilized biomass such as fixed film reduce the reactor size while maintaining the same performance. A wide range of biofilm support materials have been developed and applied in wastewater treatment systems. However, to date, little is known about the nitrification performance in this kind of hybrid system. Because research of fixed film on nutrient removal is new, there is much knowledge to be developed which could increase the widespread application of this technology to nutrient removal[4,5].

Immobilization of biocatalysts (enzymes and cells) has received increasing interest in recent years, which offers a promising potential for the improvement of the efficiency of bioprocess. Compared with the free cell, the immobilized cell has several advantages: (1) It can increase the biodegradation rate through a higher cell loading. (2) The bioprocess can be controlled more easily. (3) The continuous process can take place at a high dilution rate without washout. (4) The catalytic stability of biocatalysts as well as the tolerance against toxic compounds can be improved. Nitrification and denitrification by using immobilized microbial cells have been reported[6-9]. However, to date, there is no research on the comparison of nitrifying activity between free and immobilized microbial cells.

In this project, we developed a hybrid biological reactor containing both the suspended growth biomass and the attached growth biomass, which was to be accomplished by introducing a certain amount of porous materials. In the previous paper we investigated the effect of organic loading rates in a hybrid biological reactor[3]. The objective of this study was to compare the nitrifying characteristics of the suspended- and attached-biomass in a hybrid biological reactor.

MATERIALS AND METHODS

Reactor and Carrier

A concentric-tube airlift reactor was used. The reactor had a working volume of 3 liters.

The porous ceramic particles, with a diameter of 0.6-0.9 mm, were used as a carrier for microbial attachment. The carrier concentration in the reactor was 10 g/L.

Wastewater Composition

Synthetic wastewater was used containing NH_4Cl (600 mg/L), $NaHCO_3$ (500 mg/L), K_2CO_3 (1000 mg/L), K_2HPO_4 (152 mg/L) and KH_2PO_4 (60 mg/L). Inorganic carbonate was used as the primary carbon source, and no other carbon source was added to the medium. The temperature was maintained at 30℃ and the pH was controlled at about 7 by the addition of 1 mmol/L sodium hydroxide.

Acclimation and Operation

Seed sludge was obtained from a sewage treatment plant and acclimated to the synthetic wastewater of 600 mg/L NH_4Cl for about 2 months. Once acclimated, the reactor was operated for more than one month. The solid retention time (SRT) of the suspended biomass was maintained for about 10 days.

Analytical Methods

All chemical analyses were performed in accordance with standard methods[10].

Microorganisms immobilized on the ceramics were observed by scanning electron microscope. Specimen preparation was as follows. Firstly, it was fixed by 2.5% glutaraldehyde fixing solution and 1% osmic acid solution, and then washed three times with phosphate buffer. Secondly, the specimen was exposed to sequential ethanol dehydration from 30% to 100% in 20% increment with 20 min exposure at each concentration, and then it was replaced by acetate iso-amylester. After dehydration, it was dried at CO_2 critical point. At last, the specimen was sputtered with gold by ion-coater for 2 min at an applied current of 50 mA (Eiko, IB-3 Ion-coater) and then examined under scanning electron microscope (HITACHI S-570).

RESULTS

Nitrification Activity of Suspended- and Attached-Biomass

After two months' operation, the amount of biomass attached to the carriers was 29.6 mg VSS/g carrier, which was corresponding to a biomass concentration of 296 mg/L. The

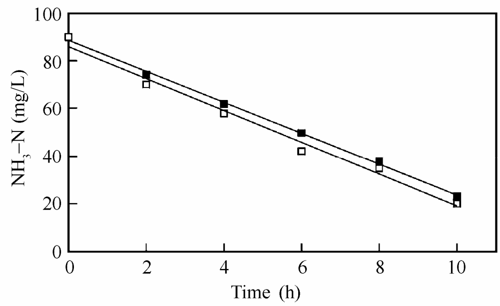

FIG. 1. Nitrification process by the suspended- and attached-biomass. (□) represents the attached biomass; (■) represents the suspended biomass.

concentration of suspended-biomass was 290 mg/L. In order to compare nitrification characteristics of the suspended-and attached-biomass, the experiments were performed in a batch reactor. Figs. 1 and 2 show the nitrifying activity and the kinetics rate constants of the suspended- and attached-biomass.

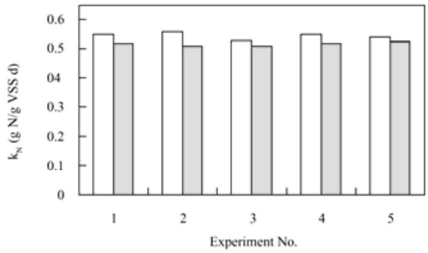

FIG. 2. Comparison of nitrification kinetics. (□) represents the suspendedbiomass; (■) represents the attached biomass.

Observation of the Attached- and Suspended- Biomass by SEM

Porous ceramic particles were excellent carrier with lots of pores inside. Our prior experiments showed that they were suitable for immobilization of microbial cells. The

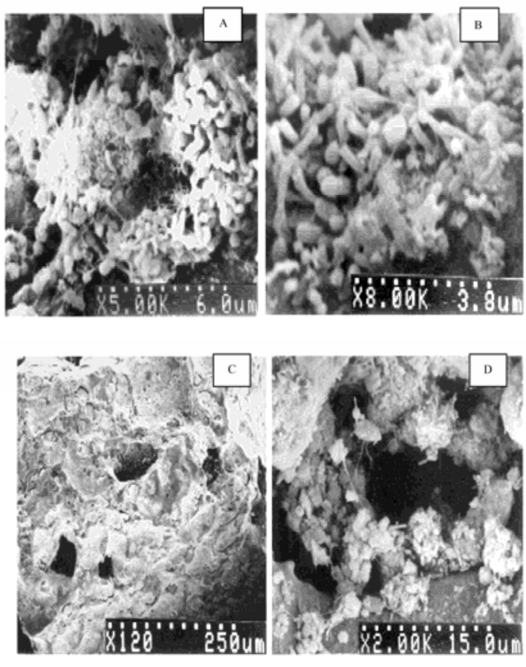

FIG. 3. SEM microscopy of biomass.

observation of suspended- and attached-biomass was carried out by scanning electronic microscopy (SEM). The SEM photographs are shown in Fig. 3.

Influence of X_0 on Nitrification Kinetics

The influence of initial biomass concentration X_0 (expressed in mg VSS/l) on nitrification kinetics was studied and compared. The result is presented in Fig. 4. It showed that the nitrification kinetics was independent of the initial biomass concentration in the range of 50-300 mg VSS/l for both suspended- and attached-biomass.

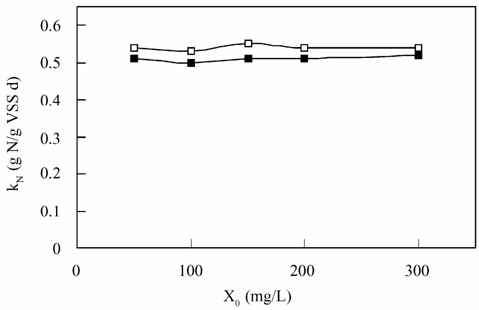

FIG. 4. Effect of X_0 on nitrification kinetics. (□) represents the suspended-biomass; (■) represents the attached-biomass.

Nitrification Inhibition

The nitrifying bacteria were the most sensitive microorganisms within the biological wastewater treatment process. The influence of toxic organic compounds, which commonly existed in industrial wastewater, such as phenol, on the nitrification inhibition for both suspended- and attached-biomass was investigated and the results are depicted in Fig. 5.

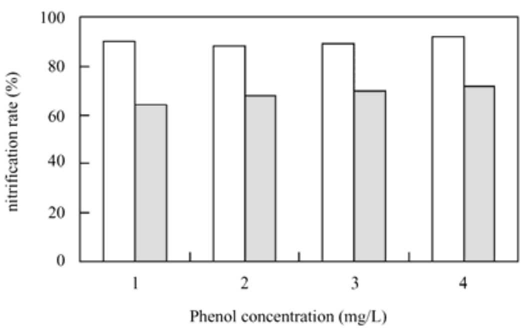

FIG. 5. Effect of phenol on nitrification inhibition. (□) represents the attached-biomass; (■) represents the suspended-biomass.

It revealed that the attached-biomass had a stronger ability to resist the nitrification inhibition caused by toxic organic compounds.

DISCUSSION

From Figs. 1 and 2, we can see that both the suspended- and attached-biomass had approximately the same nitrification activity, that is to say, the adsorption of biomass on the ceramic support had no effect on nitrification. The kinetic rate constants of nitrification for the suspended- and attached-biomass were 0.55 and 0.52 g N/g VSS d, respectively.

SEM could show the different patterns of the suspended- and attached-biomass. Fig. 3A indicates the attached-biomass, which contained more rod bacteria than suspended-biomass (Fig. 3B). Fig. 3C shows the surface of ceramic particles immersed in the reactor for more than one month, which indicated that the biofilm developed on the surface of ceramic was not too much. Fig. 3D demonstrates the development of microcolonies inside the ceramic particle pores. It can be seen that a large amount of microbial cells retained in the inner pore and attached to the inside surface of ceramic particles.

The results suggested that the attached-biomass was superior to the suspended-biomass for nitrification of wastewater, especially containing organic compounds. The hybrid biological reactor consisting of the suspended- and attached-biomass was advantageous in such cases[11,12].

CONCLUSIONS

A hybrid biological reactor was developed by introducing porous ceramic particles as immobilizing carrier for microbial attachment into the bioreactor. In this system, there exist two kinds of biomass, that is, suspended- and attached- biomass. The nitrifying characteristics of both the suspended- and attached- biomass can be determined. The results show that the kinetic rate constants of nitrification for the suspended- and attached-biomass are 0.55 and 0.52 g N/g VSS d, respectively, which are independent of the initial biomass concentrations. The attached biomass has a stronger ability to resist the nitrification inhibitor.

删除的内容:

分页符

REFERENCES

1. Wang, J. L., Shi, H. C., and Qian, Y. (1997). The advances in hybrid biological reactor applied in biological wastewater treatment. *Industrial Water Treatment* **17**(1), 6-10. (In Chinese)
2. Tyagi, R. D. (1990). Wastewater treatment by immobilized cells. Florida, CRC Press. pp 103-134.
3. Wang, J. L., Shi, H. C., and Qian, Y. (2000). Wastewater treatment in a hybrid biological reactor (HBR): effect of organic loading rates. *Process Biochem*. **36**(3), 297-303.
4. Shin, H. S. and Park, H. S. (1989). Enhanced nutrient removal of immobilized activated sludge system. *Biotechnol. Lett*. **11**(4), 293-298.
5. Shin, H. S. and Park, H. S. (1989). Enhanced nutrient removal in porous bimass carrier sequencing batch reactor (PBCSBR). *Wat. Sci. Technol*. **23**(6), 719-728.
6. Arvidsson, P., Nilsson, K., Hakanson, H., and Mattison, B. (1998). Monitoring nitric oxide from immobilised denitrifying bacteria, *Pseudomonas stutzeri*, by the use of chemiluminescence. *Appl. Microbiol. Biotechnol.* **49**(7), 677-681.
7. Garbayo, I., Leon, R., Vigara, J., and Vilchez, C. (2002). Inhibition of nitrate consumption by nitrite in entrapped *Chlamydomonas reinhardtti* cells. *Bioresource Technol*. **81**(2), 207-215.
8. Shan, H. and Obbard, J. P. (2001). Ammonia removal from prawn aquaculture water using immobilized nitrifying bacteria. *Appl. Microbiol. Biotechnol*. **57**(9), 791-798.
9. Tal, Y., Rijn, J., and Nussinovitch, A. (1999). Improvement of mechanical and biological properties of freeze-

dried denitrifying alginate beads by using starch as a filter and carbon source. *Appl. Microbiol. Biotechnol.* **51**(8), 773-779.
10.APAH (1989). Standard Methods for the Examination of Water and Wastewater. American Public health Association, Washington, DC.
11.Wang, J. L. (2002). Microbial Immobilization Techniques and Water Pollution Control. Science Press, Beijing. (In Chinese)
12.Wu, L. B., Wang, J. L., Qian, Y., and Zhan, X. M. (1999). Study on nitrification and denitrification of sludge in hybrid biological reactor. *Water and Wastewater Engineering* **25**(1), 15-17.

(*Received December* 1, 2003 *Accepted February* 26, 2004)