B16.9.1 Development of Appropriate Wastewater and Sludge Treatment Technology for Controlling CH_4 and N_2O emission applicable to China

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Abstract

Alcaligenes faecalis, a typical heterotrophic nitrifying and denitrifying bacterium, was added into activated sludge and was made to be a dominant genus of the activated sludge ecosystem. Influent COD to total nitrogen (TN) ratios and quantities of the inoculated *A. faecalis* were changed to test the responses of the systems. Behaviors of the systems to N_2O generation and wastewater treatment were evaluated. Experimental results showed that: N_2O production of the systems containing *A. faecalis* was generally about 50% of those of the control reactors containing only activated sludge. In addition, the former systems had a higher total nitrogen removal capability, their average total nitrogen removal efficiencies were generally over 30% higher than those of the control activated sludge systems. Moreover, COD removal efficiencies of the systems containing *A. faecalis* were higher than those of the control systems. On the other hand, more alien *A. faecalis* addition and higher influent COD to TN ratio led to lower N_2O emission and better effluent water qualities. Therefore, increasing the quantity of *A. faecalis* in activated sludge should be encouraged for controlling N_2O emission and upgrading wastewater treatment capability.

Key Words: *Alcaligenes faecalis*, global warming, N_2O emission, nitrogen removal

1. Introduction

China is the largest developing country in the world and its pollution situation is very serious. Because of its economic and backward technological status, international cooperation is wished to lighten its pollution burden and will be beneficial to global environment and the whole human being.

China's lower economic status and backward technological level ask the Advanced technologies with high wastewater treatment efficiency and low cost, in which microbial function will be modified and strengthened and new processes and reactors, like membrane reactor, genetic engineering microorganism, etc., will be developed.

Last two-year's experiments showed that *Alcaligenes faecalis* could play important roles in reducing N_2O production in mixed microbial cultures under certain conditions (H7) and in activated sludge system under aerobic conditions (H8). Since mass cultivation of *A. faecalis*, a typical heterotrophic nitrifying denitrifying bacterium, is far easier than that of autotrophic nitrifiers like *N. europea*, and since anoxic processes are world-widely applied for nitrogen removal, this year's experiments were made to extend the previous researches that *A. faecalis* can play some important roles in both controlling N_2O emission and increasing nitrogen removal under anoxic conditions. In this year's studies, *A. faecalis* was introduced in activated sludge system and experiments were made under anoxic conditions.

2. Research Objective

One of this-year`s research objective was to test the functions of A. *faecalis* in reducing N_2O emission and upgrading nitrogen removal capability in anoxic activated sludge wastewater treatment process with the further aim of developing some high-effect wastewater treatment processes with low cost applicable for China.

3. Research Method

A. faecalis (IFO14479) was mass cultivated and harvested with the methods as described in reference 1). Different amount of the condensed biomass was then added into different reactors (total volume, 2.7 l) to form its different initial concentrations in the reactors.

Two experiments were made in dark with the ambient temperature kept at 20°C. They were, the experiments testing the responses of the systems to the variation of influent COD to TN ratio (CN Experiments, hereafter) and the experiments determining the responses of the systems to the variation of the inoculated amount of *A. faecalis* (Amount Experiments, hereafter). In both the two experiments, artificial wastewater was supplied as substrate with its composition shown in Table 1. Operational conditions are illustrated in Table 2.

Table 1 Composition of artificial wastewater

NaCl	65.6
$MgSO_4 \cdot 7H_2O$	81.6
KH_2PO_4	185.6
KCl	134.4
$NaNO_3$	1291.2
Dextrin	304.4
Bacterpeptone	654.4
Yeast Extract	654.4
Meat Extract	745.6
$NaHCO_3$	957.2
Distilled water	10 l

Units are " mg " except that for distilled water

Samples was taken and analyzed after 30 days, when operation of the reactors was thought to reach to their stable state. Influent and effluent concentrations of NH_4-N, NO_2-N, NO_3-N, and total nitrogen (TN) were measured by automated colorimetric methods on a TRAACS-800 instrument (Roy and Knowles, 1994). N_2O concentration was determined by gas chromatography as described by Wu *et al.* (1996). Influent and effluent COD was detected as COD_{Mn} (COD, hereafter) (Japan Sewage Work Association).

Table 2 Operation parameters

Experiment	Amount Experiments				CN Experiments					
Reactor No	1	2	3	4	5	6	7	8	9	10
C:N ratio	3.8:1				4.26:1		3.8:1		2.35:1	
Amount of *A. faecalis* (10^{10} CFU l^{-3})	0	0.1	1	10	0	1	0	1	0	1
SRT (for sludge) (day)	14									
HRT (hr)	8									
Temperature (℃)	20									

4. Results

4.1 N_2O *concentrations in gas phase*

Variation of N_2O concentration in the gas phase of all the experiments is shown in Figure 1. Their arithmetic average values of the first 30 days are showed in Figure 2.

In Amount Experiments, N_2O concentration in gas phase of the AF reactors were much lower than that of AS reactor in the first stage (about 30 days after the sampling started and about 2 months after the experiments start-up), which suggested that, like the case under aerobic conditions (Wu, *et al.*, 1997), addition of *A. faecalis* was beneficial for reducing N_2O emission. The results can be also easily observed in Figure 3 (A). Assuming that the average N_2O concentration of the control was 1, average N_2O concentrations of the AF reactors with the inoculated *A. faecalis* of 10^9, 10^{10}, 10^{11} CFU l^{-1} were 0.53, 0.44 and 0.37 respectively. That is to say, addition of *A. faecalis* can reduce N_2O emission by 47~63%, and the more *A.*

faecalis was in the activated sludge system, the less N_2O generated.

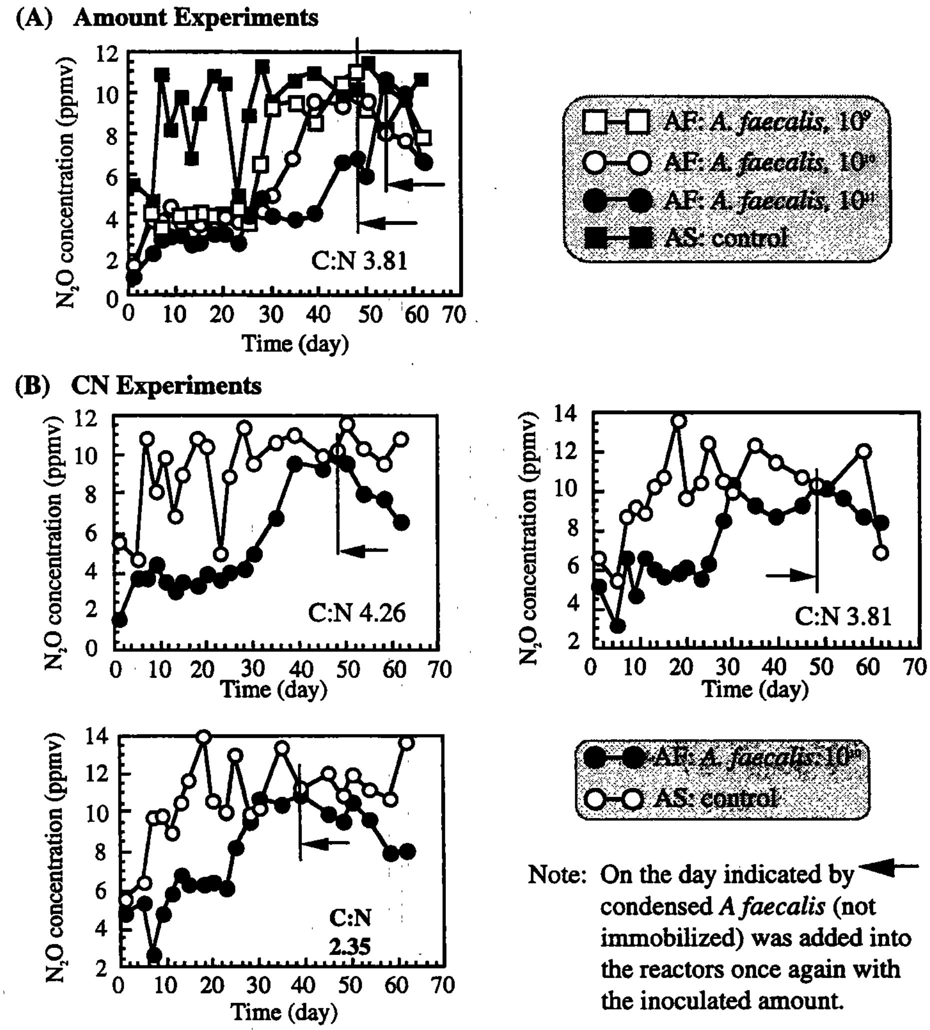

Figure 1 Variation of N_2O concentrations in gas phase versus time

In CN Experiments, the higher the C:N ratio was, the less N_2O emitted from both the AF and AS reactors. On the other hand, N_2O concentrations of the three AF reactors with different C:N ratios were lower than those of the three corresponding controls. Assuming the average N_2O concentrations of the control AS reactors were 1 in the first 30 days, corresponding average N_2O concentrations of the AF reactors with C:N ratio of 4.26:1, 3.81:1 and 2.35:1 were 0.62, 0.64 and 0.43, respectively. Higher C:N ratio was hence useful to reduce N_2O yield, and to control N_2O emission. Therefore, to keep enough available carbon in the reactor will be important to control N_2O emission for the *A. faecalis*-addition activated sludge process as well as for the conventional activated sludge process under anoxic conditions.

4.2 *Total nitrogen removal*

Average TN removal efficiencies of different reactors in the first-30-day are shown in Figure 3.

Similar to the case of N_2O emission, AF reactors had higher TN removal efficiencies than the AS reactors. Effluent TN concentrations of the AF reactors were generally lower than the AS reactors in both the two experiments (data not shown).

In Amount Experiments, assuming the average TN removal efficiency of the control reactor was 1, average TN removal efficiencies of the AF reactors with initial concentrations of *A. faecalis* of 10^9, 10^{10}, 10^{11} CFU·l^{-1} were 1.35, 1.47 and 1.68 respectively. Namely, addition of *A. faecalis* could increased TN removal efficiency by 35~68% of that of the control AS reactor.

In addition, the average TN removal efficiency of the reactor with the most *A. faecalis* inoculated, 10^{11} CFU·l^{-1}, was about 1.24 times of that of the reactor containing the least alien *A. faecalis*, which suggested that the more the *A. faecalis* was inoculated, the higher the TN removal efficiencies achieved in the reactors.

In CN Experiments, as the C:N ratio increased, the TN removal efficiency increased. Moreover, the difference of the average TN removal efficiencies between AF reactors and the control AS reactors changed as the C:N ratio changed. The C:N ratio of 3.81:1 led to the highest difference, assuming the TN removal efficiency of the control reactor was 1, that of the AF reactor was about 1.47. In contrast, C:N ratio of 2.35:1 led to the least difference. In this case, TN removal efficiency of the AF reactor was only 1.09 when assuming that of the control was 1. Therefore, at the aspect of making the advantages of inoculated *A. faecalis* to economically remove nitrogen, C:N ratio should be around 3.81.

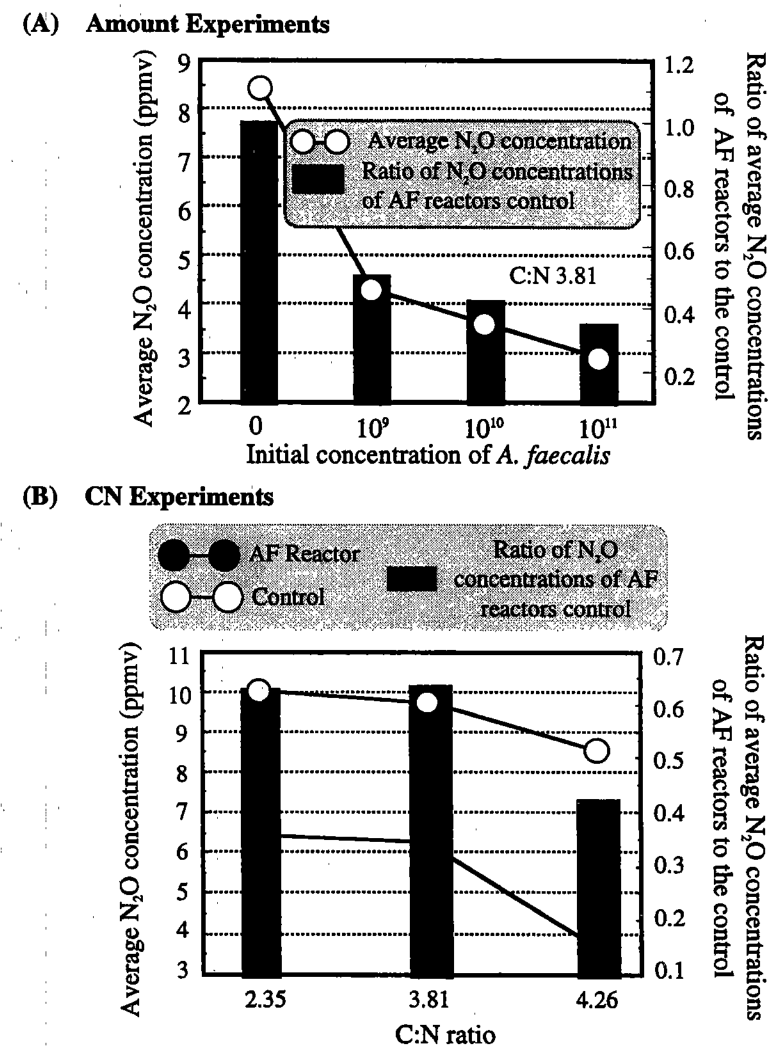

Figure 2 Average N_2O concentration of the first 30 days

4.3 *COD removal*

As one of the most important objectives, to remove organic from wastewater should be considered in any cases involved in wastewater treatment. The 30-day average COD removal efficiencies of different reactors under different conditions are compared in Figure 4.

In Amount Experiments, average COD removal efficiency of the control was 61.5%, while those of the AF reactors were from 68.4~78.3. Namely, addition of *A. faecalis* could lead to the increase of COD removal efficiency by about 7~17%. Again, the more the *A. faecalis* inoculated, the higher the COD removal efficiency achieved.

In CN Experiments, average COD removal efficiencies were from 70.0% to 91.0% in the AF reactors, higher than those of the control reactors, 60.5%-86.1%. The higher the C:N ratio was, the higher the average COD removal efficiencies were in both AF and AS reactors. However, as the C:N ratio increased, the difference between the AF reactors and the control AS reactors decreased from about 1.19 to 1.05, suggesting that lower C:N ratio could lead to higher advantages of *A. faecalis* addition.

4.4 *Endurance of the advantages of the added A. faecalis*

N_2O concentrations increased (as shown in Figure 1) and TN removal efficiencies decreased (data not shown) after about 30 days (namely, about 2 months after the *A. faecalis* added into the activated sludge systems). In the case of N_2O emission in Amount Experiments, N_2O concentrations of the AF reactor with the initial concentration of *A. faecalis* of 10^9 CFU·l^{-1}

increased sharply at the 26th day (after the sampling began). It was at the 28th day that N_2O concentration began to increase quickly in AF reactor with the initial amount of *A. faecalis* of 10^{10} CFU·l^{-1}. Then, at the 48th day (76th day after the experiments began), N_2O concentrations of the above two reactors were almost the same as that of the control AS reactor, which suggested the advantages of the alien *A. faecalis* in reducing N_2O emission had almost lost. On the 48th day, *A. faecalis* was added again into the two reactors with the same amounts as it was added at the beginning of the experiments. From the 50th day, N_2O concentrations started to decrease and kept decreasing till the end of the experiments. In the AF reactors with the initial concentration of *A. faecalis* 10^{11} CFU · l^{-1}, the increase of N_2O concentration happened later than the above two cases. It was at the 40th day for the increase of N_2O concentration to take place, and N_2O concentration of the AF reactor increased to the same level of that of the control at the 54th day. After the *A. faecalis* with the same amount as that to form the initial concentration of 10^{11} CFU·l^{-1} added into the AF reactor on the 54th day, N_2O concentration of the reactor decreased quickly.

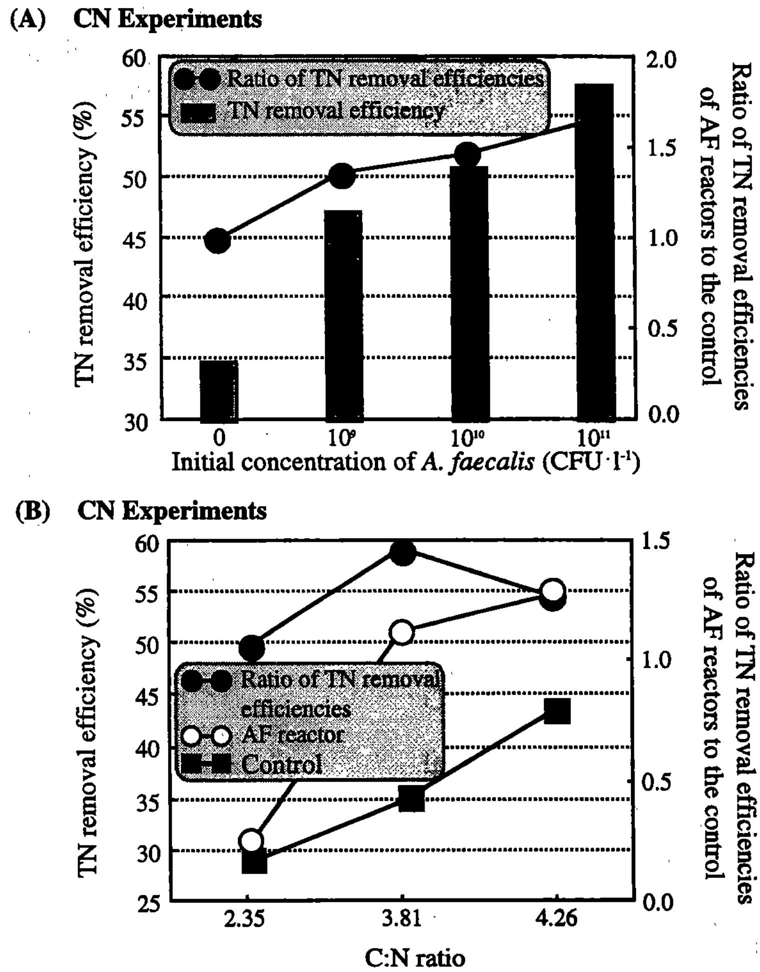

Figure 3 Average TN removal efficiency

In CN Experiments, it was at the 48th, 48th and 40th day that the N_2O concentrations of the AF reactors (with C:N ratio of 4.26:1, 3.81:1 and 2.35:1 respectively) became almost the same of those of the control AS reactors, which suggested that the advantages of the added *A. faecalis* could lasted for about 76, 76 and 68 days in the CN Experiments. On these days, *A. faecalis* was added into the three AF reactors with the same amounts as those were at the beginning of the experiments, and N_2O concentration decreased then.

The similar phenomena occurred in the performance of the AF reactors to TN removal. While it appeared that COD removal could keep relatively stable.

5. Discussion

Since *A. faecalis* was suspended in the AF reactors, the lose of its function of reducing N_2O emission and improving TN removal may be due to the following two reasons: 1) its amount may become less and reached a dynamic equilibrium in the activated sludge ecosystem because of dilution, the washing out and some other mechanisms; 2) influenced by the activated sludge ecosystem, its activities and dominance may lose. Consequently, its advantages lost gradually. Because of the limitation of detecting *A. faecalis* in a activated sludge system, *A. faecalis* was not enumerated in the experiments. However, the experimental phenomena could suggest that the suspended-growth manner of alien *A. faecalis* could keep it dominant genus for about 3 months. Practical detecting methods should be developed, and measures or strategies, like the application of the immobilization technique, should be applied as well.

The following results could be concluded then:

High amount of *A. faecalis* in activated sludge was beneficial for reducing N_2O emission and for increasing total nitrogen and COD removal under anoxic conditions. N_2O emission from the reactors containing both *A. faecalis* was about half of those from the control activated sludge systems. Moreover, average total nitrogen removal efficiencies of the former cases were about more that 35% higher than those of the latter activated sludge systems. In addition, introduction of *A. faecalis* into activated sludge led to higher COD removal efficiency.

Under the experimental conditions, higher amount of *A. faecalis* inoculated in the reactors and higher influent COD:TN ratio led to less N_2O emission, higher TN and COD removal efficiencies.

Since the introduced *A. faecalis* was kept in suspended-growth, its advantages of reducing N_2O emission and upgrading wastewater treatment could last for about 2~3 months. Strategies and measures should made or taken for keeping competitive ability of *A. faecalis* in the activated sludge systems.

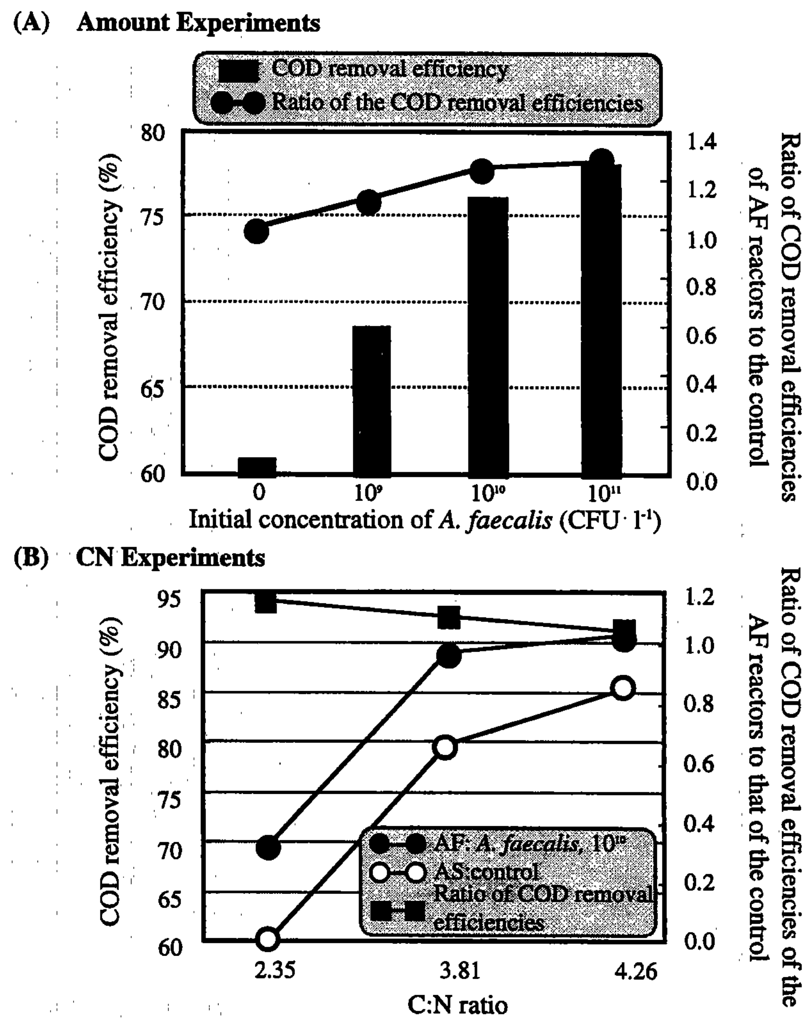

Figure 4 Average COD removal efficiency

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