

# The evaluation of enhanced nitrification by immobilized biofilm on a clinoptilolite carrier

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## Abstract

This study was conducted to evaluate the effect of clinoptilolite on nitrification in activated sludge (AS), and was focused on a relationship between ammonium exchange capacity of this mineral and improvement of nitrification. In batch experiments, the adsorption property of biofilm-attached clinoptilolite did not show substantial difference from that of natural clinoptilolite, indicating that bioregeneration became completely achieved without any regenerant in the AS. The AS with added clinoptilolite (ZR) was compared to the control AS (CR) when the ratio of chemical oxygen demand (COD) to total kjeldahl nitrogen (TKN) of influent, i.e. C/N ratio, was varied from 3.25 to 7.5 at a hydraulic retention time (HRT) of 3 h. Enhanced nitrification was comparatively observed for the ZR as C/N ratio gradually increased. The results indicated that the clinoptilolite provided a relatively low C/N ratio for nitrifiers, due to ammonium adsorption of this mineral, and consequently nitrification was accelerated. © 2002 Published by Elsevier Science Ltd.

**Keywords:** Nitrification; Clinoptilolite; Ammonium exchange capacity; C/N ratio; Bio-carrier

## 1. Introduction

Zeolite is a kind of natural or synthetic mineral ion exchanger, which is well known for its ability to remove ammonium ion from wastewater, due to preferential selectivity for this ion. Especially, clinoptilolite was reported to show high selectivity of ammonium ion among various zeolites (Ćurković et al., 1997; Roić et al., 2000). For ammonium removal from water and wastewater, zeolite has been usually employed in a downflow, packed-bed, column with chemical regenerants used, resulting in high cost (Klieve and Semmens, 1980; Semmens and Martin, 1988; Nishimura et al., 1996). To overcome this problem, bioregeneration was originally proposed by Semmens and Goodrich (1977). They reported that ammonium ion absorbed by zeolite was regenerated through biological nitrification, called 'bioregeneration', greatly reducing the cost of chemical regenerants. In addition, it was proposed that zeolite with ion exchangeability could act as a bio-carrier for

nitrifying bacteria (Semmens et al., 1977). Lahav and Green (1998) also emphasized the importance of bioregeneration in development of high concentrations of nitrifiers attached onto the surface for the minimal competition with heterotrophs. Based on these results, powdered zeolite has been employed to improve nitrification and sludge settling property in the AS. Olah et al. (1989) noted that the application of zeolite and trivalent cationic salts (e.g., Al<sup>3+</sup>, Fe<sup>3+</sup>) increased biological activity by 25% and led to much higher removal efficiency of suspended solid (SS) over conventional AS, together with enhancement of nitrification. Son et al. (2000) recently reported that zeolite addition into anoxic and oxic processes also efficiently removed ammonium ion, accordingly achieving reliable nitrogen removal. They also proposed that the ammonium exchange property of zeolite offered a comparatively high concentration of ammonium to microbes, and subsequently nitrification became improved. However, these experiments were extremely insufficient for demonstrating the relationship between ammonium exchange capacity of zeolite and improved nitrification in the AS.

This study was carried out to evaluate the feasibility of bioregeneration without any regenerant in the

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clinoptilolite-added AS and to determine the relationship between ammonium exchange capacity of this mineral and nitrification.

## 2. Methods

### 2.1. Materials

The zeolite used in the experiment was a natural clinoptilolite, one of the Heulandite groups, distributed by Pohang, Korea. The specification of the clinoptilolite is shown in Table 1.

The synthetic wastewater for continuous reactors consisted of peptone (45 mg/l), L-glutamate (75 mg/l),  $\text{CH}_3\text{COONH}_4$  (55 mg/l),  $\text{NH}_4\text{Cl}$  (48.5 mg/l) and  $(\text{NH}_4)_2\text{SO}_4$  (16.2 mg/l) at normal condition of 130 mg COD/l and 40 mg TKN/l. It also included minerals, Na, Mg, Fe, Ca and  $\text{PO}_4^{3-}$ , ranging from 1 to 2.5 mg/l. To inject peak loads of organics and nitrogen, glucose and  $\text{NH}_4\text{Cl}$  were added into synthetic wastewater for several days corresponding to the given shock load.

### 2.2. Batch test

Ammonium exchange characteristics of the clinoptilolite were determined by jar test. The experiment was carried out with clinoptilolite dosage varied at 25 °C. The synthetic wastewater was made by diluting ammonium chloride in distilled water (40 mg  $\text{NH}_4^+\text{-N/l}$ ). In addition, a variation of ammonium exchange property of this mineral was observed when the concentrations of potassium and sodium were varied from 100 to 800 mg/l. Clinoptilolite was added ranging from 5 g to 40 g to 2 l of wastewater. The suspension was immediately mixed at 180 rpm for 2 h. To investigate an influence of biofilm formation on ammonium exchange capacity of the

Table 1

The physico-chemical characteristics of the clinoptilolite employed in the experiment

Physical characteristics		Chemical composition	
Specific surface area <sup>a</sup> ( $\text{m}^2/\text{g}$ )	320	Component	% (w/w)
Average pore radius <sup>a</sup> (Å)	38	$\text{SiO}_2$	72.17
Mean diameter <sup>b</sup> ( $\mu\text{m}$ )	19.75	$\text{Al}_2\text{O}_3$	11.88
Density <sup>c</sup> ( $\text{g}/\text{cm}^3$ )	2.42	$\text{Fe}_2\text{O}_3$	1.98
Ammonia exchange capacity <sup>d</sup> (mg $\text{NH}_4^+\text{-N/g}$ )	2.02	CaO	1.30
		MgO	0.86
		$\text{K}_2\text{O}$	2.86
		$\text{Na}_2\text{O}$	2.74
		Ignition loss	5.73

<sup>a</sup> Surface area and pore radius were determined by B.E.T. test and SEM.

<sup>b</sup> Mean diameter was measured by coulter multisizer.

<sup>c</sup> Density of carrier was analyzed by pycnometer.

<sup>d</sup> Ammonia exchange capacity was determined by batchwise jar test.

clinoptilolite, the parallel ammonium adsorption tests were conducted with mixed liquor containing biofilm-formed clinoptilolite withdrawn from the ZR or control one. Before the adsorption test, the mixed liquor from ZR was put into a 1 l beaker into which diffused air was supplied to maintain 3 to 4 mg/l of dissolved oxygen (DO), for 1 h. The amount of clinoptilolite from the mixed liquor was measured as mixed liquor fixed suspended solid (MLFSS) by the standard method (APHA, 1998). After the adsorption procedure, the sample collected in the jar was immediately filtered through a 0.1  $\mu\text{m}$  membrane to determine the concentration of residual ammonium in the solution.

### 2.3. Operating of continuous reactor

Two AS reactors equipped with 5 l of oxic basin and 2 l of clarifier were concurrently operated as described in Fig. 1; one was tested as a control without dosing the

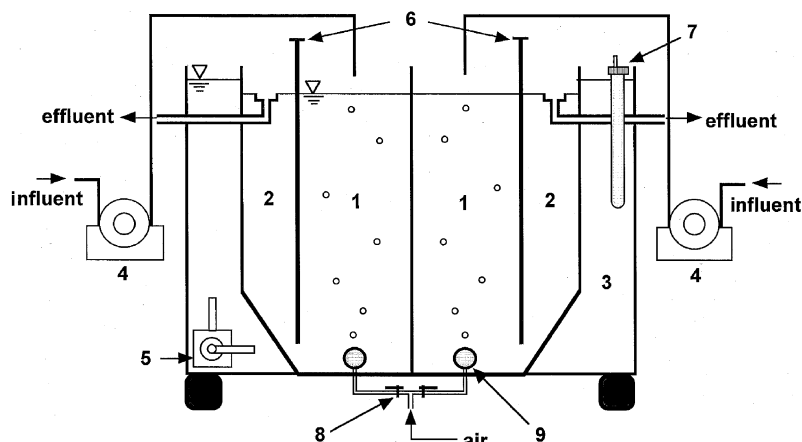


Fig. 1. Schematic diagram of activated sludge reactors, one was operated as the control and the other was to maintain the clinoptilolite of 4000 mg/l under the same condition. 1. oxic basin, 2. clarifier, 3. water jacket, 4. feeding pump, 5. circulation pump, 6. baffle, 7. temperature controller, 8. air valve, 9. air diffuser.

clinoptilolite and the other was operated with the mineral added.

The AS reactors were seeded with sludge of 1500 mg MLVSS/l from a domestic wastewater treatment plant, and were operated at 25 °C. DO was controlled at 2.5–3.5 mg/l, and bicarbonate buffer provided with the influent to adjust pH at 7.6–7.8. The clinoptilolite was uniformly added into the aeration basin to maintain 4000 mg/l, which was manually supplemented corresponding to the amount of MLVSS in the excess sludge, assuming that MLVSS was completely equivalent to the concentration of clinoptilolite in the basin. At this time, sludge was consistently withdrawn from the aeration basin to achieve a solids retention time (SRT) of 8 days, ignoring effluent SS. The experiment was performed at the normal concentrations of 130 mg COD/l and 40 mg TKN/l, and hydraulic retention time (HRT) of 3 h. For the specific operating periods at the 42 day and 112 day, organic load was abruptly elevated to 206 and 327 mg COD/l with other parameters being fixed. Ammonia load was also increased to 81.7 and 150 mg TKN/l, respectively, at the 88 day and 96 day. To evaluate the relationship between ammonium exchange capacity of clinoptilolite and nitrification, potassium was added stepwise from 100 to 800 mg K<sup>+</sup>/l under the normal condition.

#### 2.4. Analyses

TKN and NH<sub>4</sub><sup>+</sup>-N concentrations were determined by standard macro-kjeldahl and titrimetric methods (APHA, 1998). In addition NO<sub>2</sub><sup>-</sup>-N and NO<sub>3</sub><sup>-</sup>-N concentrations were measured by using ion chromatography (DX-500 System, Dionex). Mixed liquor suspended solid (MLSS), mixed liquor volatile suspended solids (MLVSS), MLFSS and chemical oxygen demand were measured by standard methods (APHA, 1998).

The activity of nitrifying microbes was evaluated from a nitrate formation rate (NFR) test, and heterotrophic activity was measured by oxygen uptake rate (OUR) at 25 °C. The NFR was determined by placing 400 ml of MLSS from each reactor into a 2 l beaker, and 50 mg/l of NH<sub>4</sub><sup>+</sup>-N was introduced from a stock solution of 20,000 mg/l of NH<sub>4</sub>Cl. Then, diffused air and bicarbonate were supplied into the beaker so that DO and pH could be maintained at 3–4 mg/l and 7.2–7.5, respectively. MLVSS, DO and pH were monitored during the aeration. A sample was taken every 30 min and the nitrate concentration was checked. For the OUR, 300 ml of MLSS was shifted to a biochemical oxygen demand (BOD) bottle and the COD of the contents was brought to 100 mg/l by a stock solution of 10,000 mg/l of glucose and then DO drop was continuously observed at 30 s intervals with a DO meter (YSI 50).

### 3. Results and discussion

#### 3.1. Ammonium exchange property and bioregeneration of the clinoptilolite

To investigate the influence of biofilm formation on ammonium exchange capacity of the clinoptilolite, the ammonium exchange properties were compared between biofilm-formed clinoptilolite from the ZR and the control. The ion exchange of the clinoptilolite between NH<sub>4</sub><sup>+</sup>-N and Na<sup>+</sup> or K<sup>+</sup> ions finally came to equilibrium. At this adsorption equilibrium, the distribution of NH<sub>4</sub><sup>+</sup>-N ion between exchanged phase and solution phase could be described by the Langmuir isotherm (Weber, 1972).

$$Q_e = Q_{\max} \times K \cdot C_N \cdot (1 + K C_N), \quad (1)$$

where  $Q_e$  is the amount of NH<sub>4</sub><sup>+</sup>-N exchange per unit weight of clinoptilolite (mg NH<sub>4</sub><sup>+</sup>-N/g clinoptilolite),  $Q_{\max}$  the maximal NH<sub>4</sub><sup>+</sup>-N exchange per unit weight of clinoptilolite (mg NH<sub>4</sub><sup>+</sup>-N/g clinoptilolite),  $K$  the Langmuir energy constant (l/mg NH<sub>4</sub><sup>+</sup>-N), and  $C_N$  is the NH<sub>4</sub><sup>+</sup>-N concentration in the solution (mg NH<sub>4</sub><sup>+</sup>-N/l).

$Q_{\max}$  and  $K$  can be determined by employing Eq. (1). 3.8 mg NH<sub>4</sub><sup>+</sup>-N/g clinoptilolite and 0.19 l/mg NH<sub>4</sub><sup>+</sup>-N were obtained for  $Q_{\max}$  and  $K$  of biofilm-formed clinoptilolite. For the control, those values were 3.5 mg NH<sub>4</sub><sup>+</sup>-N/g-clinoptilolite and 0.18 l/mg NH<sub>4</sub><sup>+</sup>-N, respectively, as shown in Fig. 2.

It was concluded that biofilm formation had little influence on the exchange property of this mineral. Furthermore, this result indicated that the ammonium exchange capacity of the clinoptilolite was completely recovered by nitrification, called ‘bioregeneration’ (Lahav and Green, 1998). It has been usually said that suspended microbes oxidize ammonium ion liberated in the solution and subsequently, exhausted clinoptilolite is

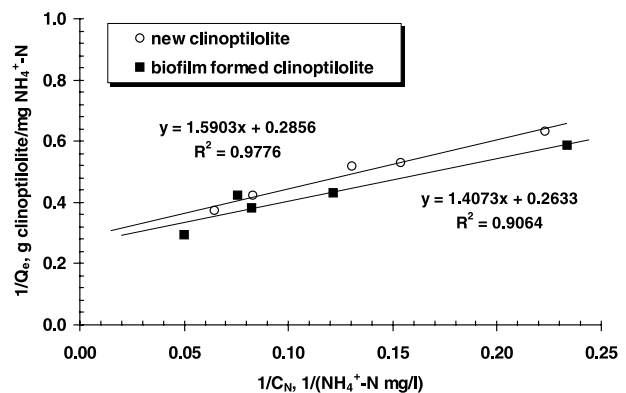
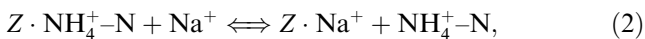


Fig. 2. The influence of biofilm formation on ammonium exchange capacity of the clinoptilolite, that ranging from 5 to 40 g was added to 2 l of synthetic wastewater containing 40 mg NH<sub>4</sub><sup>+</sup>-N/l. The suspension was mixed at 180 rpm for 2 h. Water temperature was maintained at 25 °C.

regenerated by the equilibrium reaction described at Eq. (2) (Semmens et al., 1977).



where  $Z \cdot \text{NH}_4^+-\text{N}$  is the exchanged ammonium ion to the clinoptilolite (mg/l), and  $\text{NH}_4^+-\text{N}$  the liberated ammonium ion in the solution (mg/l).

They proposed that the rate of nitrification is much slower at bioregeneration than it is when the ammonium is present in the solution since the ammonium ions within crystalline cages of the clinoptilolites are not available for nitrifying microorganisms, contradicting the enhancement of nitrification by clinoptilolite addition in the AS. Lahav and Green (1998) additionally investigated bioregeneration of ammonium saturated clinoptilolite after displacing ammonium ion with cationic regenerant. They consequently emphasized the necessity of the regenerant for recovery of exhausted clinoptilolite during the bioregeneration. However, in the present study, bioregeneration was completely achieved by only supplying air, without its necessity of liberating ammonium ion into the solution by regenerant. It indicates that ammonium ion exchanged inside the clinoptilolite is accessible to nitrifying bacteria, which was supported by other studies. Preston and Alleman (1993) found that nitrifiers co-immobilized with clinoptilolite improved mass transfer of ammonium ion inside the pellets and nitrification considerably accelerated. In addition, Yang (1997) reported that higher concentration of ammonium applied to a fluidized-bed system of co-immobilized nitrifiers and zeolite, improved ammonium exchange, which thus increased the availability of ammonium substrate supplied to the nitrifying bacteria.

### 3.2. The influence of clinoptilolite in a continuous reactor on nitrification

To evaluate the effect of clinoptilolite on nitrification, experiments for both ZR and CR were concurrently conducted at an HRT of 3 h and a SRT of 8 day. As illustrated in Fig. 3(a), periods of approximately 9 and 16 days were required for the ZR and the CR, respectively, to acclimate to normal condition (3.25 C/N ratio). After the acclimation, nitrification gradually increased in both reactors, and the ZR showed maximal nitrification of 97%, though the CR had only 88% nitrification. However, significant difference of nitrification efficiency was found in the relatively high C/N ratio given. For 245 mg COD/l of the first organic peak load (6.1 C/N ratio), the nitrification efficiency in the ZR was reduced to 74%, while it was still seriously deteriorated (52%) in the CR. At this time, the effluent ammonium concentration of the ZR and the CR also rose to 11.2 and 15.1 mg  $\text{NH}_4^+-\text{N}/\text{l}$ . The improved nitrification in the ZR was correspondingly detected with the

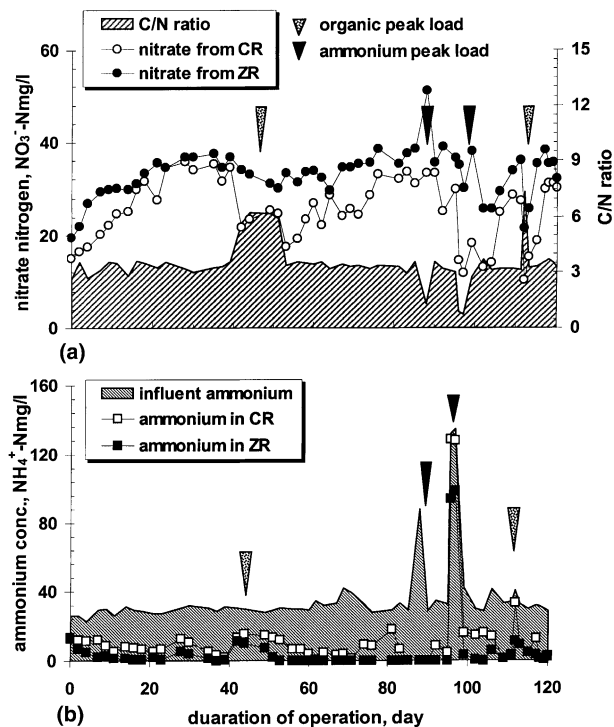


Fig. 3. Variation of nitrification corresponding to influent loading at both ASs: (a) influent C/N ratio versus effluent  $\text{NO}_3^-$ -N; (b) influent  $\text{NH}_4^+-\text{N}$  versus effluent  $\text{NH}_4^+-\text{N}$ .

300 mg COD/l of the second organic peak load (7.5 C/N ratio). The efficiencies of nitrification in the ZR and the CR were 52% and 25%, respectively.

In theory, a high C/N ratio of substrate results in competition for DO and space in the biofilm between nitrifying and heterotrophic bacteria. In this competition, heterotrophs overwhelm nitrifiers because of their higher yields and growth rates (Wiesmann, 1994). Okabe et al. (1996) reported that the population of nitrifying microbes was proportionally decreased as influent C/N ratio was elevated, and they found that the period of acclimation for nitrification was increased by a factor of three when the C/N ratio was increased twice. Thus, in the present study, it is not surprising that nitrification was highly deteriorated in both reactors with a high C/N ratio. Nevertheless, ZR achieved better efficiency of nitrification than CR as well as faster acclimation when the normal condition recovered.

The effect of clinoptilolite on ammonium shock loads was determined (Fig. 3). For 88 mg/l of ammonium peak load, the nitrification efficiencies of the ZR and the CR were 54% and 35%, respectively. The nitrification efficiency of the ZR was also increased by 2.5 times compared to that of the CR for 135 mg/l of  $\text{NH}_4^+-\text{N}$  (the second ammonium peak load). Moreover, the ZR also showed rapid acclimation when back to the normal condition, although it took about 12 days in the CR.

All of the results proposed a possible mechanism of nitrogen removal in the ZR, that clinoptilolite provided

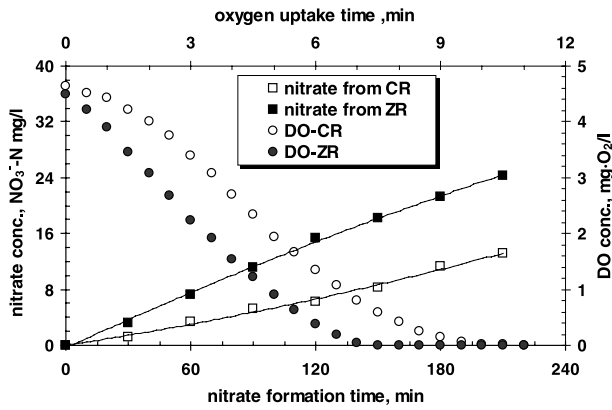


Fig. 4. Results of NFR and OUR test with both ASs.

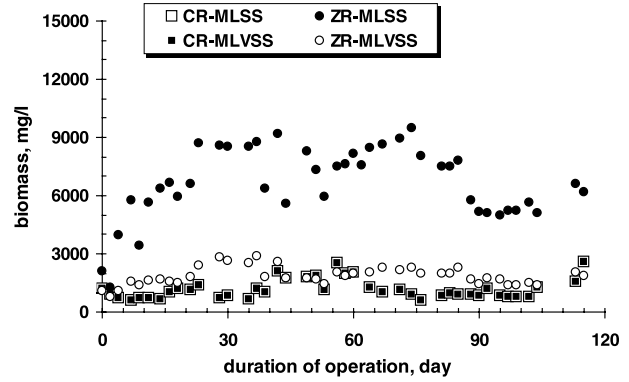


Fig. 5. The effect of powdered clinoptilolite on biomass in AS; MLVSS illustrated amount of biomass in the AS, the concentrations of clinoptilolite were calculated as MLFSS (i.e. difference between MLSS and MLVSS).

Table 2  
The enhancement of nitrification by clinoptilolite at different conditions

Specific influent conditions	CR			ZR		
	NE <sup>a</sup>	NFR <sup>b</sup>	OUR <sup>c</sup>	NE <sup>a</sup>	NFR <sup>b</sup>	OUR <sup>c</sup>
3.25 of C/N ratio	62 [14]	4.40 [0.8]	27 [2.2]	85 [11]	8.11 [0.1]	30 [2.0]
6.1 of C/N ratio	52 [8]	3.41 [0.3]	35 [1.5]	74 [6]	7.28 [0.5]	37 [1.6]
7.5 of C/N ratio	25 [1]	3.11 [0.3]	38 [1.2]	52 [2]	5.23 [1.8]	38 [2.2]
88 mg NH <sub>4</sub> <sup>+</sup> -N/l	35 [5]	4.25 [1.2]	22 [2.8]	54 [2]	8.49 [1.0]	30 [0.8]
135 mg NH <sub>4</sub> <sup>+</sup> -N/l	8 [2]	4.23 [0.8]	22 [1.5]	21 [1]	8.92 [0.3]	31 [1.2]

Data were averaged; [ ] standard deviation.

<sup>a</sup> Nitrification efficiency (%).

<sup>b</sup> Nitrification formation rate (mg NO<sub>3</sub><sup>-</sup>-N/l h).

<sup>c</sup> Oxygen uptake rate (mg O<sub>2</sub>/l h).

a relatively low C/N ratio for nitrifier in the ZR due to its sustained ammonium absorption capacity and the immobilization of microbes on the surface and interior, and as a role of bio-carrier. Thus, Olah et al. (1989) reported that zeolite addition into the AS improved nitrification by 60% over the control, due to the high concentration of nitrifying microorganisms attached to the mineral. Sim et al. (1973) also proposed that zeolite application into the AS significantly improved nitrification. It is inferred that a repetition of bioregeneration increases the concentration of nitrifiers, because ammonium exchange property of the clinoptilolite was independent of biofilm formation of this mineral.

The amount of nitrifiers in the microorganisms was precisely assessed by comparing OUR and NFR values between the ZR and the CR. As shown in Fig. 4, the OUR values were 27 and 30 mg O<sub>2</sub>/l min in the CR and the ZR, respectively, which indicated that clinoptilolite did not influence the heterotroph activity. However, the ZR presented a relatively higher NFR of 8.11 mg NO<sub>3</sub><sup>-</sup>-N/l h, which was a factor of two as compared to that of the CR. The results indicated that nitrifiers were significantly accumulated in the ZR.

Fig. 5 demonstrates that biomass was augmented by clinoptilolite dosage in continuous AS. For the ZR, biomass gradually increased and then reached steady-state ranging from 2380 to 2910 mg MLVSS/l. The maximal concentration of clinoptilolite was 6560 mg/l; although some of the minerals were constantly washed out from the settling basin. For the CR, MLSS was

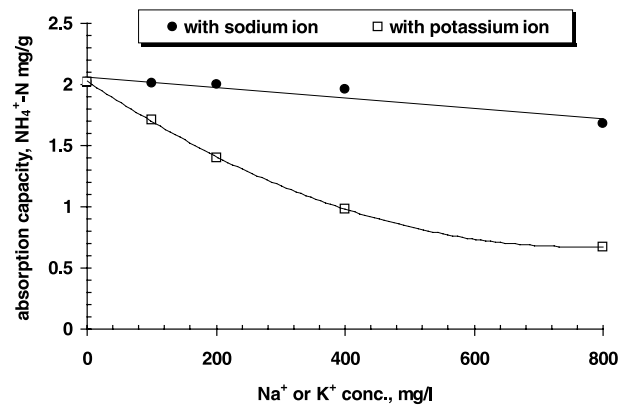


Fig. 6. The ammonium exchange characteristics of clinoptilolite at different sodium and potassium concentrations.

Table 3  
The influences of potassium concentration on nitrification

Potassium concentration (mg/l)	CR			ZR		
	NE <sup>a</sup>	NFR <sup>b</sup>	OUR <sup>c</sup>	NE <sup>a</sup>	NFR <sup>b</sup>	OUR <sup>c</sup>
0	80 [7]	3.83 [0.6]	32 [2.8]	95 [2]	5.26 [0.4]	35 [2.1]
100	85 [2]	3.83 [0.3]	27 [3.5]	86 [3]	4.86 [0.3]	30 [2.1]
200	73 [1]	3.54 [0.4]	35 [2.4]	74 [3]	4.01 [0.1]	37 [2.5]
400	83 [2]	3.71 [0.4]	38 [2.4]	81 [4]	4.27 [0.2]	38 [2.3]
800	54 [2]	3.22 [0.2]	33 [2.1]	59 [3]	3.51 [0.2]	34 [1.9]

Data were averaged; [ ] standard deviation.

<sup>a</sup> Nitrification efficiency (%).

<sup>b</sup> Nitrification formation rate (mg NO<sub>3</sub><sup>-</sup>-N/l h).

<sup>c</sup> Oxygen uptake rate (mg O<sub>2</sub>/l h).

maintained from 1140 to 2400 mg/l and MLVSS was ranged from 980 to 2050 mg/l. Therefore, it was concluded that the clinoptilolite resulted in high microbial concentration, which was caused by its role of bio-carrier. Biofilm development on the surface of the zeolite was also reported by Son et al. (2000). They examined nitrogen removal in oxic and anoxic processes with zeolite circulation and observed that a formation of biofilm on the zeolite surface was progressively augmented with time, by using a dye adsorption method.

Consequently, it can be concluded that clinoptilolite results in high concentration of nitrifiers by providing a relatively low C/N ratio and playing a role of bio-carrier, thus nitrification is competently improved as summarized at Table 2.

### 3.3. The influence of potassium ion on nitrification

Adsorption experiments were conducted to determine the degree of affinity of NH<sub>4</sub><sup>+</sup>-N on the clinoptilolite in the presence of Na<sup>+</sup> or K<sup>+</sup> ions. The equilibrium of exchange reactions of these ions can also be described by Eq. (2). Ammonium exchange capacities of clinoptilolite were observed with different concentrations of potassium and sodium ranging from 100 to 800 mg/l. As illustrated in Fig. 6, ammonium exchange capacity, initially 2.02 mg NH<sub>4</sub><sup>+</sup>-N/g, was decreased to 1.678 and 0.674 mg NH<sub>4</sub><sup>+</sup>-N/g, respectively, at 800 mg/l of sodium or potassium concentrations.

Especially, Fig. 6 shows that the ammonium exchange property of clinoptilolite was significantly decreased at over 200 mg/l of potassium. Semmens and Martin (1988) reported that clinoptilolite had a greater selectivity for potassium than for other ions such as ammonium, barium and calcium. Furthermore, Lahav and Green (1998) examined ammonium ion elimination with a biological exchange system using chabazite as ion exchanger; and it was noted that the worst NH<sub>4</sub><sup>+</sup>-N breakthrough curve was reached for a resin presaturated with potassium.

To confirm the relationship between ammonium exchange capacity of the clinoptilolite and nitrification in

the AS, continuous reactors were operated with potassium concentration elevated in normal condition (3.25 C/N ratio). At this condition, sodium ion concentration, resulting from adding sodium bicarbonate to adjust pH, was about 126 mg Na<sup>+</sup>/l. For the case of no potassium ion, the ZR showed 95% of nitrification in contrast to 80% obtained in the CR. However, the differences of nitrification between the two reactors were gradually decreased in line with the increase in potassium concentration, and consequently 59% and 54% of nitrification were obtained in the ZR and the CR, respectively, at 800 mg/l of potassium. Table 3 thoroughly describes the effect of potassium concentration on nitrification in both reactors. NFR values were 5.26 and 3.83 mg NO<sub>3</sub><sup>-</sup>-N/l h in the ZR and the CR without potassium addition, respectively. For the case of 800 mg/l of potassium concentration, NFR of the CR was decreased by 16% while that of the ZR was significantly decreased by 33% compared to that for normal condition.

## 4. Conclusions

It was concluded that clinoptilolite provided a relatively low C/N ratio for nitrifiers in the ZR, due to its ammonium absorption capacity. In addition, it was confirmed that biofilm formed on clinoptilolite was almost completely regenerated by nitrification, and biomass was augmented to 2910 mg MLVSS/l in the ZR. Therefore, it was decided that exchanged ammonium ion on the clinoptilolite was available for microbes, especially nitrifiers, and that this mineral worked as an efficient bio-carrier. The increase of potassium concentration decisively showed the relationship between nitrification and ammonium exchange property of the clinoptilolite. At 800 mg/l of potassium concentration, NFR of the ZR was significantly reduced by 33% though that of the CR was only decreased at 16%. These results showed that with added clinoptilolite, nitrification enhancement is caused by ammonium adsorption and consecutive bioregeneration, resulting in a high concentration of nitrifying bacteria.

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## References

- APHA, AWWA, WEF, 1998. Standard Methods for the Examination of Water and Wastewater, 20th ed.
- Ćurković, L., Cerjan-Stefanović, S., Filipan, T., 1997. Metal ion exchange by natural and modified zeolites. *Wat. Res.* 31, 1379–1382.
- Klieve, J.R., Semmens, M.J., 1980. An evaluation of pretreated natural zeolites for ammonium removal. *Wat. Res.* 14, 161–168.
- Lahav, O., Green, M., 1998. Ammonium removal using ion exchange and biological regeneration. *Wat. Res.* 32, 2019–2028.
- Nishimura, F., Somiya, I., Tsuno, H., Iwabu, H., 1996. Development of a combined BAC and BZ reactor for removal of nitrogen in wastewater from sludge drying process. *Wat. Sci. Tech.* 34, 145–151.
- Roić, M., Cerjan-stefanović, S., Kurajica, S., Vančina, V., Hodžić, E., 2000. Ammoniacal nitrogen removal from water by treatment with clays and zeolites. *Wat. Res.* 34, 3675–3681.
- Okabe, S., Oozawa, Y., Hirata, K., Watanabe, Y., 1996. Relationship between population dynamics of nitrifiers in biofilms and reactor performance at various C:N ratios. *Wat. Res.* 30, 1563–1572.
- Olah, J., Papp, J., Meszaros-kis, A., Mucsi, G.Y., Kallo, D., 1989. Simultaneous separation of suspended solids, ammonium and phosphate ions from wastewater by modified clinoptilolite. In: Karge, H.G., Weitkamp, J. (Eds.), *Zeolites as Catalysts, Sorbents and Detergent Builders*. Elsevier, Amsterdam, pp. 711–719.
- Preston, K.T., Alleman, J.E., 1993. Co-immobilization of nitrifying bacteria and clinoptilolite for enhanced control of nitrification. In: *Proceedings of the 48th of Industrial Waste Conference*, Purdue University, West Lafayette, Indiana, USA, pp. 407–412.
- Semmens, M.J., Martin, W.P., 1988. The influence of pretreatment on the capacity and selectivity of clinoptilolite for metal ions. *Wat. Res.* 22, 537–542.
- Semmens, M.J., Goodrich Jr., R.R., 1977. Biological regeneration of ammonium-saturated clinoptilolite-I. Initial observation. *Environ. Sci. Tech.* 11, 255–259.
- Semmens, M.J., Wang, J.T., Booth, A.C., 1977. Biological regeneration of ammonium-saturated clinoptilolite-II. Mechanism of regeneration and influence of salt concentration. *Environ. Sci. Tech.* 11, 260–265.
- Sim, R.C., Little, L.W., Brown, J.C., 1973. Application of clinoptilolite in biological nitrification systems. In: *46th Annual Water Pollution Control Federal Conference* Cleveland, Ohio.
- Son, D.H., Kim, D.W., Chung, Y.C., 2000. Biological nitrogen removal using a modified oxic/anoxic reactor with zeolite circulation. *Biotechnol. Lett.* 22, 35–38.
- Weber, W.J., 1972. In: *Physicochemical Processes for Water Quality Control*. Wiley/Interscience, New York, pp. 261–304 (Chapter 6).
- Wiesmann, U., 1994. Biological nitrogen removal from wastewater. In: Fletcher, A. (Ed.), *Advances in Biochemical Engineering Biotechnology*, vol. 51. Springer, Berlin, pp. 113–154.
- Yang, L., 1997. Investigation of nitrification by co-immobilized nitrifying bacteria and zeolite in a batchwise fluidized bed. *Wat. Sci. Tech.* 35, 169–175.