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# Characteristics of nitrogen and phosphorus removal in SBR and SBBR with different ammonium loading rates

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**Abstract**−Laboratory scale experiments were conducted to study the deterioration of enhanced biological phosphorus removal (EBPR) due to influent ammonium concentration, and to compare the performance of two types of sequencing batch reactor (SBR) systems, a conventional SBR and sequencing batch biofilm reactor (SBBR). Both in SBR and SBBR, the total nitrogen removal efficiency decreased from 100% to 53% and from 87.5% to 54.4%, respectively, with the increase of influent ammonium concentration from 20 mg/*l* to 80 mg/*l*. When the influent ammonium concentration was as low as 20 mg/*l* (C : N : P=200 : 20 : 15), denitrifying glycogen-accumulating organisms (DGAOs) were successfully grown and activated by using glucose as a sole carbon source in a lab-scale anaerobic-oxic-anoxic ($A_2O$) SBR. In the SBR, due to the effect of incomplete denitrification and pH drop, the nitrogen and phosphorus removal efficiency decreased from 77% to 33.3% when the influent ammonium concentration increased from 20 mg/*l* to 80 mg/*l*. However, in the SBBR, simultaneous nitrification/denitrification (SND) occurred, and the nitrification rate in the aerobic phase did not change remarkably in spite of the increase in influent ammonium concentration. Phosphorus removal was not affected by the increase of influent ammonium concentration.

Key words: Ammonium Concentration, Deterioration, EBPR, SBR, SBBR

## INTRODUCTION

When nutrients such as nitrogen and phosphorus are not removed from wastewater, oxygen depletion, nuisance algal blooms and eutrophication in water are accelerated. Biological methods have been used successfully at municipal and industrial levels to remove these nutrients [1,2].

In enhanced biological phosphorus removal (EBPR), under anaerobic conditions, phosphate-accumulating organisms (PAOs) take up volatile fatty acids (VFAs) and store them internally in the form of polyhydroxyalkanoates (PHAs). The reducing power is derived from the glycolysis of the intracellular glycogen, while the energy is obtained partly from the glycogen utilization but mostly from the hydrolysis of the intracellular stored polyphosphate (polyP), which results in the release of ortho-phosphate. In the subsequent aerobic conditions, PAOs take up an amount of ortho-phosphate to recover the intracellular polyP level by oxidizing the stored PHAs [3].

Recently, glycogen accumulating organisms (GAOs) have also been found to be able to proliferate under alternating anaerobic and aerobic conditions [4]. GAOs take up VFA under anaerobic conditions and store them as intracellular PHAs, with the required reducing power and energy both being derived from the glycolysis of glycogen. Under the subsequent aerobic conditions, GAOs grow and replenish the glycogen pool by using the intracellular PHA as both the carbon and energy sources.

Generally, biological methods are continuous systems with suspended biomass which require the internal recirculation of sludge and/or wastewater, inside a train of reactors in which the necessary conditions are provided so that the different biological processes may take place. The conventional activated sludge process for nutrient removal is a space-oriented system. Flow moves from one tank to the next tank continuously and virtually all tanks in system are occupied by liquid. However, a sequencing batch reactor (SBR) is a time-oriented system. SBR has already shown many advantages including nitrogen and phosphorus removal with less bulking [5,6].

Application of SBR to biofilm reactors was suggested by Wilderer to overcome the difficulties about the growth and maintenance of suspended activated sludge flocs [7]. This combined system is called a sequencing batch biofilm reactor (SBBR). SBBR is considered to be the hybrid of fully developed SBR technology and biofilm system technology. SBBR can offer some benefits with little risk in field applications considering the results of the existing treatment processes.

During the past few years, SBBRs have been extensively investigated for the removal of various wastewaters [8,9]. SBBRs have a potential advantage compared to the suspended growth processes because of less sludge loss and compact reactor design. Also, biofilm processes such as SBBRs offer the possibility of achieving simultaneous nitrification and denitrification due to the prevailing anoxic zone in the biofilm near the attached surface during the aeration phase. Therefore, SBBR has been developed for simultaneous removal of nitrogen and phosphorus [9,10]. As nitrification and P uptake both consume oxygen, organisms in such a system are potentially subjected to competition for oxygen. Also, denitrifiers and PAOs are in a competition relationship about organics. In SBBR, the mass transfer limitations for oxygen and organic substrate could be occurring and hampering the biological phosphorus removal in the biofilm [11]. However, the mass transfer limitation could be a minor effect on overall phosphorus removal because of the mainte-

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nance of thin biofilm by the frequent backwashing [12]. Gieseke et al. suggested that during the oxic period P uptake and nitrification occurred sequentially in time within the oxic surface layer of the biofilm and the sequential action was a result of the oxygen limitation of nitrifiers caused by the competition for oxygen with heterotrophic bacteria such as PAOs at the biofilm surface [10]. Therefore, SBBR may have an advantage in removing nitrogen because of simultaneous nitrification-denitrification but may have limitations for phosphorus removal.

We focused on the application of SBR and SBBR systems on small sewerage systems without pH control and with no extra addition of organics for denitrification. The EBPR in a small sewage system has been frequently deteriorated by a sudden addition of ammonium source such as livestock industry wastewater. In this research, SBBR is operated the same as SBR except for the support media used. With the increase of influent ammonium concentration, the N and P removals were evaluated and compared in order to investigate the inhibition effect of ammonium on N and P removal both in SBR and SBBR. And the total organic carbon (TOC), N and P removal characteristics were precisely investigated and discussed. The other purpose of this study was to investigate the effectiveness of pH, DO and oxidation reduction potential (ORP) profiles for indicating biological N and P removals.

## MATERIALS AND METHODS

### 1. Reactor System and Feed

The investigation was carried out by lab-scale SBR and SBBR (Fig. 1) maintained at a constant temperature of 20±1 ºC. Two 4 *l* laboratory scale reactors made of acryl sheet were used: SBBR was filled with polypropylene type media of 25% reactor volume and SBR served as a control. The support media were cubes (1.5×20×0.2 $cm^3$) of polypropylene (porosity, 93%; specific surface area, 4.0×$10^4$ $m^2/m^3$). A time controller was used for the system operating components such as influent pumps, effluent pumps, aerator and mixer according to the operating cycle. The feed was prepared daily from stock solutions and tap water. It had the following composition: glucose ($C_6H_{12}O_6$), (200 mg total organic carbon [TOC]/*l*); ammonium sulfate ($(NH_4)_2 \cdot SO_4$), 47.1 mg/*l*; dibasic potassium phosphate ($K_2HPO_4$), 84.2 mg/*l*; calcium chloride ($CaCl_2$), 3.76 mg/*l*; magnesium sulfate ($MgSO_4 \cdot 7H_2O$), 50 mg/*l*; manganese sulfate ($MnSO_4 \cdot H_2O$), 55 mg/*l*; ferrous sulfate ($FeSO_4 \cdot 7H_2O$), 2.22 mg/*l*; potassium chloride (KCl), 7.0 mg/*l*. And sodium bicarbonate ($NaHCO_3$), 300.0 mg/*l*, was added to prevent sudden pH drop. In order to investigate the effect of influent $NH_4^+$-N concentration, the ammonium sulfate concentration increased as 93 mg/*l*, 185 mg/*l* and 380 mg/*l*. And so the influent $NH_4^+$-N concentration was changed as about 20 mg/*l*, 40 mg/*l* and 80 mg/*l*.

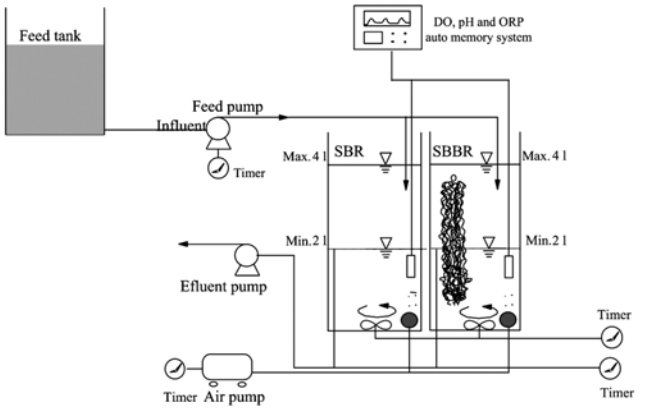


**Fig. 1. Schematic diagram of SBR and SBBR.**

### 2. Operation of SBR and SBBR

Seed sludge was obtained from a conventional municipal wastewater treatment plant for organic removal and acclimated to the synthetic wastewater in SBR and SBBR for about 3 months. The two reactors were operated with the same seeding sludge concentration (MLSS, 3,000 mg/*l*), influent synthetic wastewater concentration and operational strategy. The carbon to nitrogen (C/N) ratio in the influent was varied from 10 to 2.5 by increasing the influent ammonium concentration. The operating cycle was shown as follows: filling (F), 0.5 h; first non-aeration (NA), 3 h; aeration (A), 3.5 h; second non-aeration (NA), 3.5 h; settling and drawing (SD), 1.5 h. Mixing (150-200 rpm) was done by a magnetic stirrer during the filling, non-aeration and aeration phases. $N_2$ gas to remove the dissolved oxygen was not used. The pH in reactor was not controlled because pH control was not conducted in a real small sewerage system. The cycle began with the addition of 2.0 *l* of feed during filling phase. At the aeration phase, air was introduced to the reactors through the porous diffuser at a rate of 5.0 *l*/min, which provided a dissolved oxygen concentration of more than 6.0 mg/*l* and was controlled by a rotameter. At the settling and drawing phase, the mixing and aeration stopped and the supernatant (2 *l*) was drained by peristaltic pump after sludge settling phase (0.5 h), and the same amount of fresh synthetic wastewater was fed during the subsequent filling phase, resulting in a nominal hydraulic residence time (HRT) of 24 hours.

Twice a day, some sludge in reactors was wasted before the end of non-aeration period to keep constant MLSS and solid retention time (SRT) of 20-30 days. In a steady-state condition, reactors were operated for more than two weeks to collect the cycle test data.

### 3. Analytical Methods

The monitoring program consisted of two parts. A routine monitoring determined the overall performance characteristics, while a special cycle test provided data on the concentration of specific compounds over each operational condition in a cycle. Wastewater samples for dynamic studies were collected directly from the reactor. The influent samples were collected from the influent reservoir just after it was filled. When a pseudo-steady state was observed (i.e., repeated cyclic behavior), cyclic studies were performed with sampling intervals of 30 or 60 min. Samples were immediately filtered with 0.22 μm membrane filters (Advantec $\phi$47 mm) to remove all solids and microorganisms.

All anions ($NO_3^-$-N, $NO_2^-$-N, $PO_4^{3-}$-P) analyses were performed by ion chromatography (Metrohm, Ion Analysis Version 2.0). $NH_4^+$-N concentration and sludge parameters (SS, VSS) were measured according to Standard Methods [13]. TOC of sample was analyzed by a Shimazu TOC analyzer (TOC-5000).

The pH, DO concentration and ORP were monitored continuously. Electrodes of pH, DO and ORP were installed in a reactor and connected to the individual meter (Inolab Multi-Parameter Level 3) and computer. The reactor status was displayed on a computer

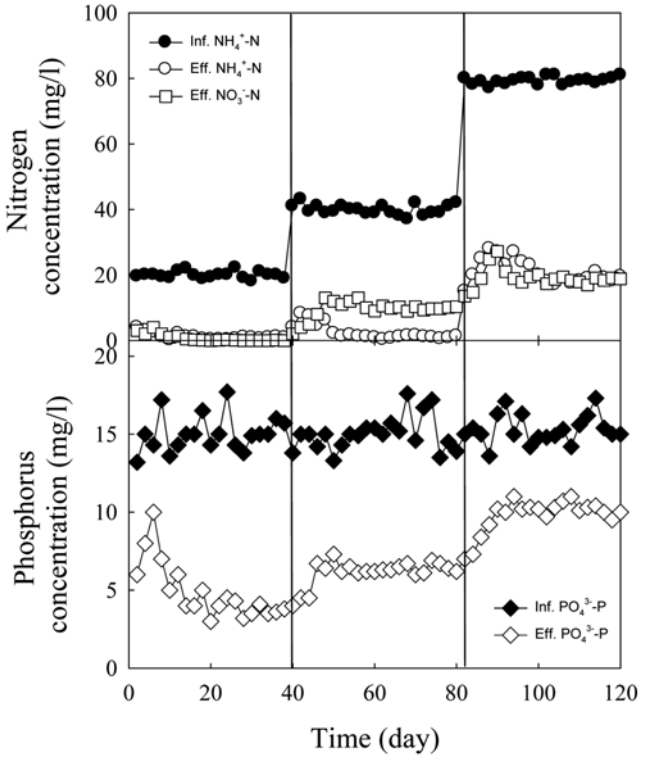


**Fig. 2. Nitrogen and phosphorus removal in SBR during 120 days operation.**

monitor, and the values of electrodes signal were stored every minute into the data file.

## RESULTS AND DISCUSSION

### 1. Effluent Nitrogen and Phosphorus Concentration in SBR and SBBR

The variation of nitrogen ($NH_4^+$-N, $NO_3^-$-N) and phosphorus ($PO_4^{3-}$-P) in SBR was observed according to the variation of influent $NH_4^+$-N concentration from 20 mg/*l* to 80 mg/*l* during 120 days (Fig. 2).

The settling ability of the sludge in SBR was very good throughout the entire period of operation, with the sludge volume index (SVI) between 70 and 110 ml/g (data not shown).

When the influent $NH_4^+$-N concentration was 20 mg/*l*, the pseudo-steady state reached within 10 days and the effluent $NH_4^+$-N and $NO_3^-$-N concentration were almost 0 mg/*l* except initial short unsteady periods (10 days) by the complete nitrification and denitrification. And so the total nitrogen removal efficiency was almost 100%.

When the influent $NH_4^+$-N concentration increased up to 40 mg/*l*, the effluent $NH_4^+$-N concentration was also maintained as almost 0 mg/*l* by the complete nitrification; however, the effluent $NO_3^-$-N concentration increased up to 9-11 mg/*l* because of the incomplete denitrification by the deficient organics. The total nitrogen removal efficiency was about 75%.

With the increase of influent $NH_4^+$-N concentration up to 80 mg/*l*, both incomplete nitrification and denitrification occurred. The effluent $NH_4^+$-N concentration increased to 17-21 mg/*l* by the incomplete nitrification and the effluent $NO_3^-$-N concentration was about 17-20 mg/*l* by the incomplete denitrification at pseudo-steady state. Therefore, the total nitrogen removal efficiency was not high as 53%.

Therefore, we could see that the denitrification was hindered at first and the nitrification was also hampered later by the increase of influent $NH_4^+$-N concentration.

Effluent phosphorus concentration was varied according to the increase of influent $NH_4^+$-N concentration irrespective of constant influent phosphorus concentration at about 15 mg/*l*.

When the influent $NH_4^+$-N concentration was low as 20 mg/*l*, the required time to reach a pseudo-steady state was about 20 days and the effluent phosphorus concentration was 3-4 mg/*l*. The phosphorus removal efficiency was 77% at that time. As the influent $NH_4^+$-N concentration increased to 40 mg/*l*, the effluent phosphorus concentration increased to 6-7 mg/*l* at pseudo-steady state and the phosphorus removal efficiency decreased to 56.7%. Also, the effluent phosphorus concentration increased to 10 mg/*l* as the influent $NH_4^+$-N concentration increased up to 80 mg/*l*. Thus the phosphorus removal efficiency was very low as 33.3%.

The nitrogen and phosphorus removal characteristics in SBBR (Fig. 3) were different from those in SBR. When the influent $NH_4^+$-N concentration was low as 20 mg/*l*, the effluent $NH_4^+$-N and $NO_3^-$-N concentrations were 2-3 mg/*l* and 0 mg/*l*, respectively, and the nitrogen removal efficiency was 87.5% which was a little lower than that in SBR. As the influent $NH_4^+$-N concentration increased to 40 mg/*l*, the effluent $NH_4^+$-N concentration was shown as 7-8 mg/*l* and the effluent $NO_3^-$-N concentration was still 0 mg/*l*. The total nitrogen removal efficiency was 81.3%. At the influent $NH_4^+$-N of 80 mg/*l* the effluent $NH_4^+$-N concentration increased up to 35-38 mg/*l* and the effluent $NO_3^-$-N concentration still remained at 0

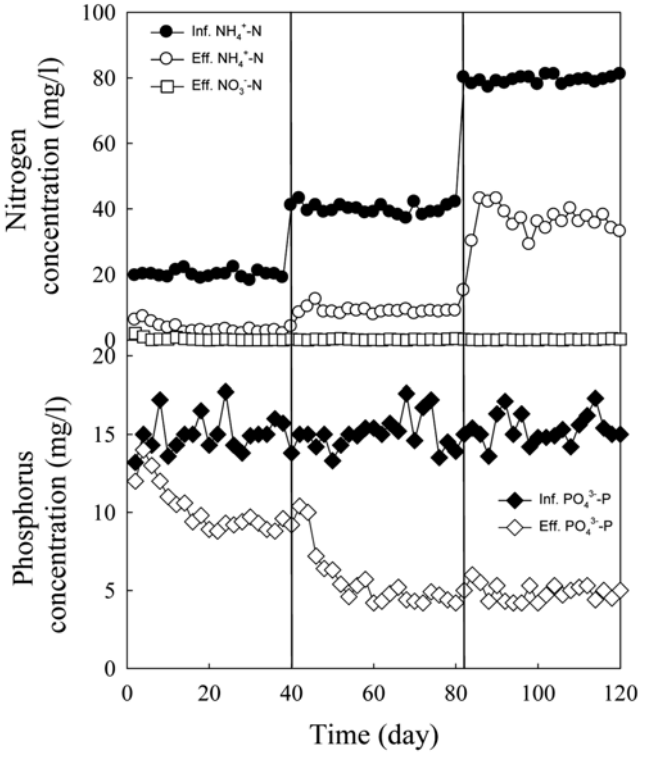


**Fig. 3. Nitrogen and phosphorus removal in SBBR during 120 days operation.**

mg/*l*. The total nitrogen removal efficiency decreased to 54.4%.

The effect of influent $NH_4^+$-N concentration on the effluent phosphorus concentration in SBBR is shown in Fig. 3(b). In this condition, the influent phosphorus concentration was maintained constant at 15 mg/*l*. The effluent phosphorus concentration was about 9.0 mg/*l* when the influent $NH_4^+$-N concentration was 20 mg/*l*, and the phosphorus removal efficiency was very low as 40% compared to 77% in SBR. However, when the influent $NH_4^+$-N concentration increased to 40 mg/*l*, the effluent phosphorus concentration decreased to about 4.5 mg/*l* and the phosphorus removal efficiency increased to 70%. The effluent phosphorus concentration and removal efficiency were unchanged irrespective of the increase of influent $NH_4^+$-N concentration to 80 mg/*l*.

In SBR, the total nitrogen removal efficiency decreased because of the untreated $NO_3^-$-N by the increase of influent $NH_4^+$-N concentration. The decrease of total nitrogen removal efficiency also occurred in SBBR because of the remaining $NH_4^+$-N concentration. The phosphorus removal efficiency in SBR decreased as the influent $NH_4^+$-N concentration increased; however, that in SBBR increased even though the reason is not clear at this time.

**2. Biological Nitrogen Removal in a Cycle**

Fig. 4 shows the typical profiles of $NH_4^+$-N, $NO_2^-$-N and $NO_3^-$-N concentrations in SBR and SBBR at a pseudo-steady state when the influent $NH_4^+$-N concentration was varied from 20 mg/*l* to 80 mg/*l*.

When the influent $NH_4^+$-N concentration was low as 20 mg/*l*, in SBR, the nitrification was completed within 2 h at the aeration state and the nitrification rate was 22.6 mg $NH_4^+$-$N_{removed}$/h. As the $NH_4^+$-N was removed a similar amount of $NO_3^-$-N was produced by nitrification. The produced $NO_3^-$-N was completely removed by denitrification at the second non-aeration phase. This complete denitrification was a very interesting phenomenon. At the conventional N removal system, external addition of carbon source such as methanol or acetate was required for denitrification. However, there were no additional organics and no TOC variation at the second non-aeration phase (Fig. 6(A)) in this system. Therefore, we expected that the denitrification was occurring by denitrifying phosphate accumulating organisms (DPAOs). Recently, the occurrence of DPAOs capable of utilizing nitrate instead of oxygen as an electron acceptor for phosphorus uptake has been reported [14,15]. However, the denitrification was not proved by DPAOs because phosphorus uptake was not occurring in this phase (Fig. 5(A)). Finally, we concluded that the nitrate was removed by denitrifying glycogen accumulating organisms (DGAOs). DGAOs are able to utilize nitrate instead of oxygen. By the first original report of Zeng et al., DGAOs were successfully enriched in a lab-scale SBR running with anaerobic/anoxic cycles and acetate feeding during the anaerobic period and the morphology of the sludge changed from floc structure to granular structure during the enrichment of DGAO [16]. However, in this study, the enrichment of DGAOs was possible using glucose as a sole car-

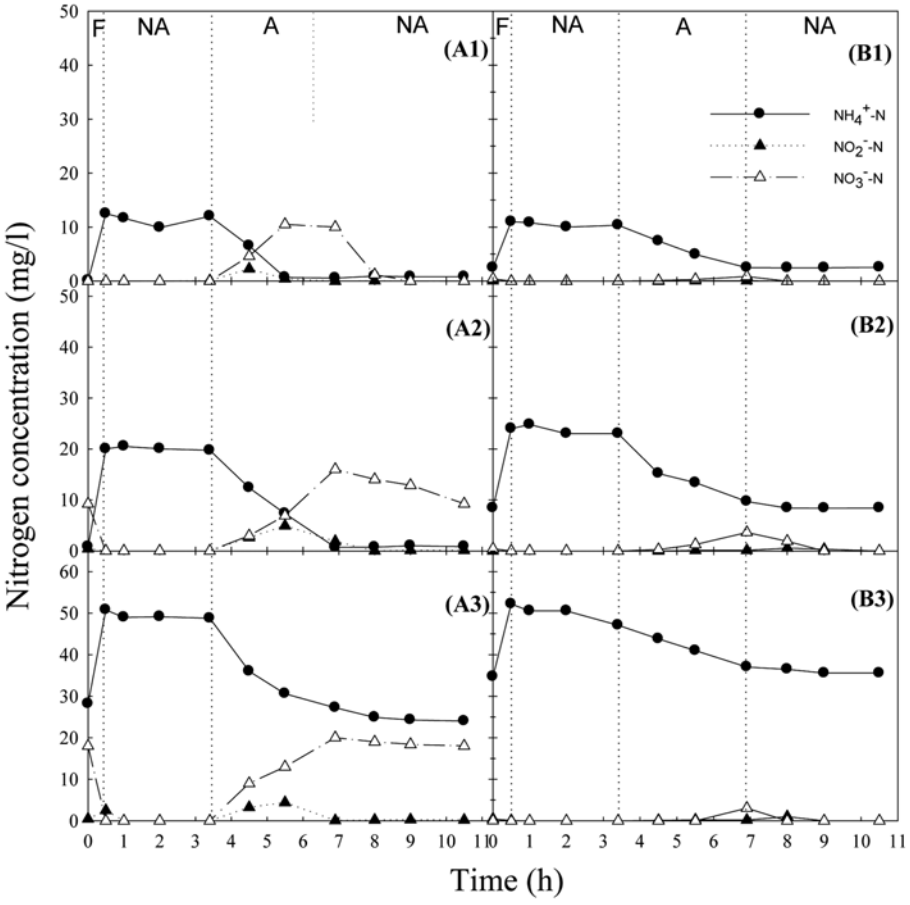


**Fig. 4. Typical profiles of $NH_4^+$-N, $NO_2$-N and $NO_3^-$-N concentrations in a cycle of SBR (A) and SBBR (B) during the initial ammonium concentration of 20 mg/*l* (1), 40 mg/*l* (2), 80 mg/*l* (3).**

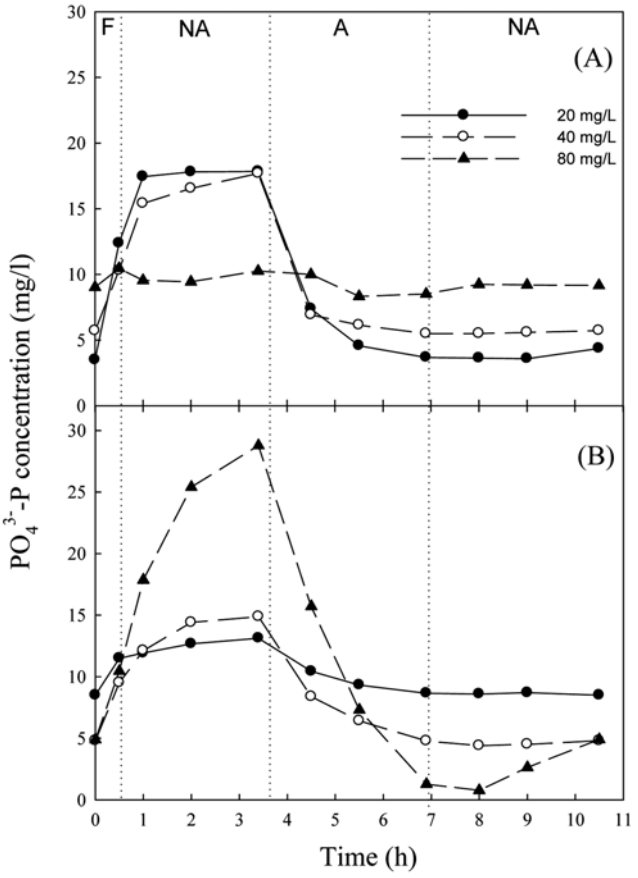


**Fig. 5. Typical profiles of $PO_4^{3-}$-P concentration in a cycle of SBR (A) and SBBR (B) during the initial ammonium concentration.**

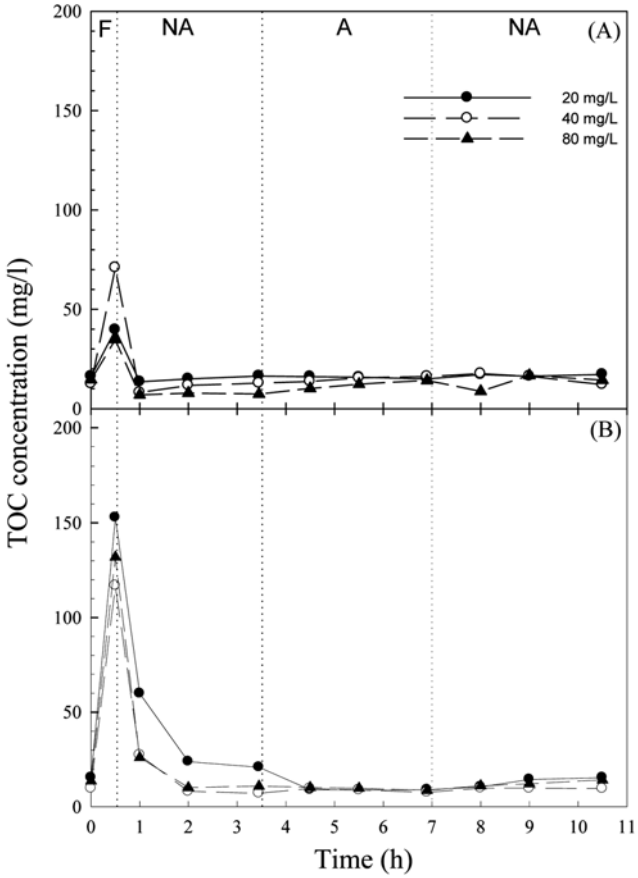


**Fig. 6. Typical profiles of TOC concentration $PO_4^{3-}$-P in a cycle of SBR (A) and SBBR (B) during the initial ammonium concentration.**

bon source instead of acetate and anaerobic-oxic-anoxic ($A_2O$) process, and the morphology was maintained as floc structure. In addition, it has been known that the presence of GAOs may indeed be responsible for the instability of some EBPR system and GAOs are a real competitor to PAOs for organics in EBPR system [17]. DGAOs compete with PAOs for volatile fatty acids (VFAs) as a competitor. However, in this study, if DGAOs were not working adequately at the second non-aeration (anoxic) phase, the $NO_3^-$ was not removed completely and the remaining $NO_3^-$ might hamper the release of P at the anaerobic phase in a next cycle. Consequently the EBPR might be deteriorated. Therefore, DGAOs and PAOs are both in a relationship between competitor and cooperator.

At the low influent $NH_4^+$-N concentration as 20 mg/*l*, in SBBR, 7.9 mg $NH_4^+$-N/*l* was removed for 3.5 h at the aeration phase; however, only 0.8 mg $NO_3^-$-N/*l* was produced. This result might be caused by the simultaneous nitrification/denitrification (SND) [18,19]. Even though the DO concentration in solution was maintained as 6-8 mg/*l* (Fig. 7 B2), the inner part of biofilm was kept at anoxic state because the oxygen diffusion limitation into biofilm was occurring. The external biofilm thickness was about 1 mm at this time. In this case the nitrifiers exist in external biofilm with high dissolved oxygen concentrations, whereas the denitrifiers are preferentially active in internal biofilm with very low dissolved oxygen concentrations.

As the influent $NH_4^+$-N concentration increased to 40 mg/*l*, in SBR, the nitrification was completed at the aeration phase and nitrification rate increased to 23.7 mg/h. However, the denitrification rate decreased and incomplete denitrification occurred at the second non-aeration state as the produced $NO_3^-$-N concentration increased. The remaining $NO_3^-$-N at the second non-aeration phase was transferred to the next cycle. The transferred $NO_3^-$-N was removed by ordinary heterotrophic organisms (OHOs) such as heterotrophic denitrifiers. At the first non-aeration phase, GAOs, PAOs and denitrifiers might be competing with each other for organic source. Denitrifiers might have a priority because of the $NO_3^-$-N at this phase. GAOs and PAOs would get less organics and accumulate less PHAs. Therefore, PAOs had less P release at the first non-aeration phase and had less P uptake at the aeration phase (Fig. 5A). Also, DGAOs had a less $NO_3^-$-N removal at the second non-aeration phase.

When the influent $NH_4^+$-N concentration was as high as 80 mg/*l*, the nitrification rate at the aeration state in SBR (Fig. 4 A3) increased to 34.5 mg/h, and the denitrification was hardly occurring at the second non-aeration phase. The remaining $NO_3^-$-N concentration was about 18 mg/*l*. The remaining $NO_3^-$-N at the first non-aeration phase in a next cycle would inhibit the growth and activity of the DGAOs and PAOs, which was similar to the case of influent $NH_4^+$-N concentration of 40 mg/*l*.

In SBBR, the nitrification rate in aerobic phase was not changed remarkably in spite of the increase of influent $NH_4^+$-N concentra-

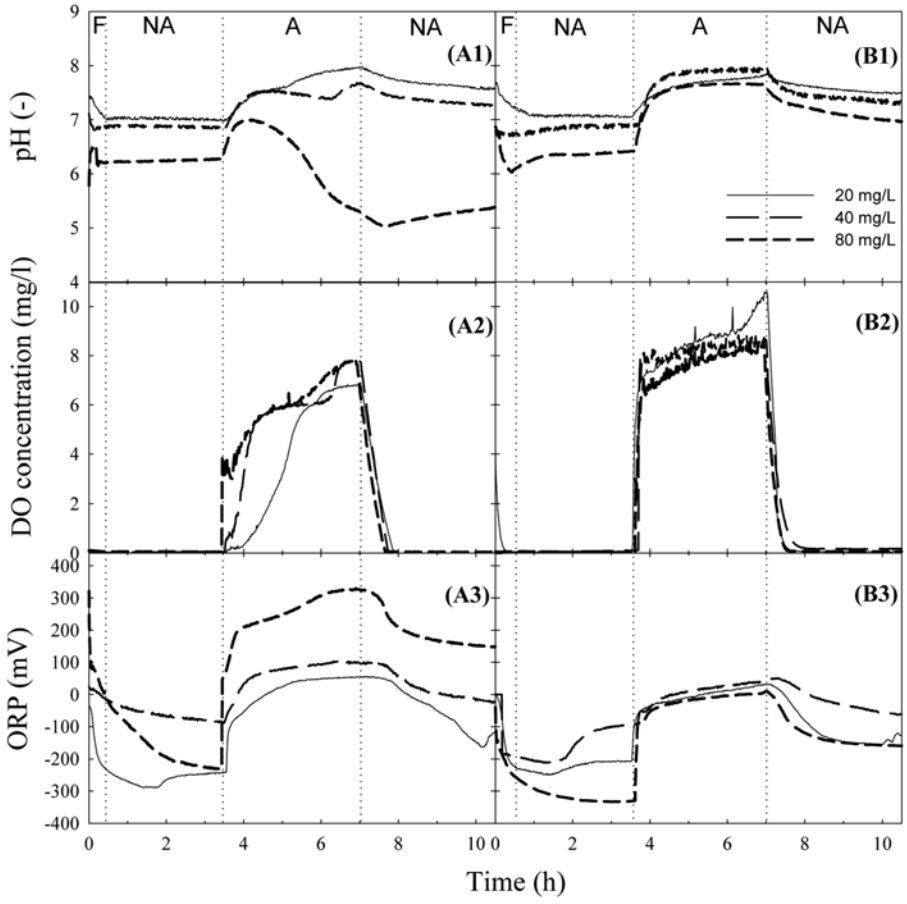


**Fig. 7. Typical profiles of pH (1), DO (2), ORP (3) in a cycle of SBR (A) and SBBR (B) during the initial ammonium concentration.**

tion to 40 mg/*l* and 80 mg/*l*, which was similar to the case of low influent $NH_4^+$-N concentration of 20 mg/*l*. By the SND, therefore, the produced $NO_3^-$-N concentration at the aerobic phase was very low. And the remaining $NH_4^+$-N concentration at the second non-aeration phase was increased with the increase of influent $NH_4^+$-N concentration because of the low nitrification activity in aerobic phase.

**3. Biological Phosphorus Removal in a Cycle**

The biological phosphorus removal characteristics in SBR and SBBR were compared with increasing influent $NH_4^+$-N concentration (Fig. 5). The remaining P concentration was combined with the influent P concentration during the filling period. Therefore, the P concentration after the filling period in SBR was estimated as 9.3 mg/*l*, 10.4 mg/*l* and 12.0 mg/*l* when the influent $NH_4^+$-N concentration was 20 mg/*l*, 40 mg/*l* and 80 mg/*l*, respectively.

When the influent $NH_4^+$-N concentration was low as 20 mg/*l*, in SBR, the P release occurred at the filling and non-aeration periods. The released P (8 mg/*l*) was taken up by PAOs at the aeration periods and no P uptake occurred at the second non-aeration phase. The ratio of P release to glucose uptake was 0.016 P-mol/C-mol, which is much less compared to the other reported data (0.28 P-mol/C-mol) with acetate as carbon source [20]. The lower ratio was caused by the fact that glucose metabolism requires less ATP (18 mmol/g) to be metabolized than acetate (158 mmol/g) [21]. However, even compared to other data using glucose as carbon source (0.035 P-mol/C-mol) the ratio in this study was low because carbon consumption was divided and accomplished by both PAOs and GAOs [22]. Zeng et al. reported that PAOs and GAOs each take up approximately half of the COD in the feed during the anaerobic period [20].

When the influent $NH_4^+$-N concentration increased to 40 mg/*l*, in SBR, the released P amount was a little lower than that at the influent $NH_4^+$-N concentration of 20 mg/*l*. Also, the P amount taken up was a little lower than that. This was caused by the remaining $NO_3^-$-N at the second non-aeration period. The heterotrophic denitrifiers during the feeding and 1$^{st}$ non-aeration phase consumed organics using $NO_3^-$-N as an electron acceptor, but PAOs had not taken up organics well. Therefore, P release and uptake amounts by PAOs were decreased.

Especially, the complete deterioration of P removal occurred when the influent $NH_4^+$-N concentration was increased to 80 mg/*l*. No P release and uptake occurred at this condition. This result could be caused by the remaining $NO_3^-$-N during the influent period by the increase of influent nitrogen concentration. However, this P deterioration could not be explained only by the inhibition effect of $NO_3^-$. Therefore, we investigated the pH variation in a cycle (Fig. 7 A1). When the influent $NH_4^+$-N concentration was high as 80 mg/*l*, the starting pH value at the feeding and first non-aeration phase was very low as pH 6.2 and pH decreased sharply to 5.3 by nitrification. By the report of Filipe et al., GAOs can be competitive in EBPR systems in which the pH of the anaerobic phase is low [23]. In addi-

tion, they studied the effect of pH on the aerobic metabolism of PAOs and GAOs at pH 6.5, 7.0 and 7.5, and suggested that the stability of EBPR was strongly dependent on the pH in the aerobic zone [24]. Therefore, if pH was low, the growth of PAOs would be inhibited, whereas the growth of GAOs would be only mildly affected. In this system, when the influent $NH_4^+$-N concentration was 40 mg/*l*, the pH range was 6.8-7.5 and the growth and activity of PAOs were not inhibited. However, when the influent $NH_4^+$-N concentration increased to 80 mg/*l*, the pH variation was severe at 5.2-7.0. Therefore, the growth and activity of PAOs were extremely inhibited by pH. The stable P removal could not be recovered after during 2 months when the influent $NH_4^+$-N concentration was lowered to back 20 mg/*l* because PAOs were washed out (data not shown).

At the low influent $NH_4^+$-N concentration of 20 mg/*l*, the released and taken up P amounts in SBBR were lower than those in SBR because the biofilm in SBBR might have diffusion limitation for P removal. At this condition, $NO_3^-$-N did not remain both in SBR and SBBR, and so the inhibition effect of $NO_3^-$-N on P release was not occurring. As the increase of influent $NH_4^+$-N concentration to 40 mg/*l*, P release and uptake in SBBR a little increased. Especially, when the influent nitrogen concentration increased to 80 mg/*l*, P concentration remarkably increased to 28 mg/*l* at the end of first aeration phase and P concentration decreased to 1.5 mg/*l* at the end of aeration phase. Therefore, we could see that PAOs in SBBR were not affected by the increase of influent $NH_4^+$-N concentration because $NO_3^-$ which could inhibit P removal was not remainiing at the first non-aeration phase and pH variation by the increase of influent $NH_4^+$-N concentration was small (Fig. 7B1). At that time, the biofilm thickness was increased to about 2 mm. This result was contrary to the suggestion of Falkentoft et al., who mentioned about the diffusion limitations hampering EBPR in the biofilm [11]. A second P release of about 4 mg/*l* occurred at the second non-aeration period. In this period, the TOC concentration was constant and low as 10 mg/*l* (Fig. 6B). Therefore, P release in this period was caused by the internal carbon source not by external carbon source. With the increase of biofilm thickness, the inner part of biofilm was maintained as an endogenous respiration state and internal carbon source could be used for the P release.

**4. Biological TOC Removal in a Cycle**

The variations of TOC concentration in SBR and SBBR are shown in Fig. 6. The influent TOC concentration was about 200 mg/*l*.

In SBR, the TOC concentration increased after the filling period; however, the TOC concentration was very low compared to the influent TOC concentration because the organics adsorbed to the suspended flocs and microorganisms instantaneously. The TOC concentration was sharply decreased by several type of microorganisms (GAOs, PAOs, OHOs etc.) within 30 min. Hereafter, the TOC concentration maintained low values as 10-20 mg/*l*. The variations of TOC by the increase of influent $NH_4^+$-N concentration in SBR were not occurring. When the influent $NH_4^+$-N concentration was 80 mg/*l*, EBPR and denitrfication were completely deteriorated (Fig. 4 and 5). Recently, Fang et al. reported that no substrate uptake was observed while the EBPR performance was in a deterioration state [25]. However, in this study, even though PAOs and DGAOs were washed out by deterioration, the TOC variation was not changed. Therefore, GAOs that remained in reactor might have been pH-resistant and consumed the whole organics. The stable activity of DGAOs was recovered after 2 months when the influent $NH_4^+$-N concentration was lowered back to 20 mg/*l* (data not shown). Therefore, we can assume that GAOs were not washed out irrespective of pH drop, and GAOs were changed to DGAOs at the denitrification condition because GAOs were same with DGAOs.

The TOC concentration at the end of filling period in SBBR was much higher than that in SBR because the absorption of organics by the attached biofilm was less activated than that by the suspended flocs. The TOC was consumed within the first non-aeration period. The TOC concentration was constant as 10-15 mg/*l* after the first non-aeration period irrespective of the variation of influent $NH_4^+$-N concentration.

**5. pH, DO, ORP Variation in a Cycle**

As the influent nitrogen concentration increased, the pH in SBR showed a remarkable change. At the low influent $NH_4^+$-N concentration of 20 mg/*l* (Fig. 7 A1), the inflection point (a) which was the end point of nitrification occurred during the aerobic period. However, the pH decline was not occurring because of the low nitrification and sufficient alkalinity in feed. When the influent $NH_4^+$-N concentration was 40 mg/*l*, the initial pH increment slope at the aerobic period increased, the inflection point (b) occurred later, and a little pH decline occurred at the latter aerobic period by the high nitrification and insufficient alkalinity. However, the pH variation band was not seriously different from that of 20 mg/*l*. As the influent nitrogen concentration increased to 80 mg/*l*, the initial pH increment slope was higher and the start point for pH decline was shown early. The pH inflection point was not shown at this period and pH decreased to 5.0 continuously because of the severe nitrification. And the pH variation band was great, which inhibited the action and growth of microorganisms, especially, PAOs.

In SBBR, however, the pH variation was stable and the pH inflection point that showed the ending point of nitrification was not occurring irrespective of the variation of influent $NH_4^+$-N concentration.

The variation of DO concentration in SBR was changed by the variation of influent nitrogen concentration. Especially, at the influent $NH_4^+$-N concentration of 40 mg/*l*, the inflection point of DO concentration that means the ending point of nitrification was clearly shown, and it coincided with the pH inflection point.

The whole DO concentration in SBBR was higher than that in SBR because the DO consumption was lower. The DO profile in SBBR was not changed by the increment of influent nitrogen concentration.

The ORP profile band in SBR was increased by the increment of influent nitrogen concentration. Also, a similar result was shown in SBBR. However, the ORP profile band in SBBR was lower than that in SBR.

## CONCLUSIONS

Even when SBR and SBBR were operated under the same condition, the nitrogen and phosphorus removal characteristics were different from each other during the operation period. The operation was divided into the three distinct phases by increasing the influent $NH_4^+$-N concentration to 20 mg/*l*, 40 mg/*l* and 80 mg/*l*. In SBR, even though complete nitrogen removal occurred by DGAOs at the low influent $NH_4^+$-N concentration of 20 mg/*l*, the total nitro-

gen removal efficiency decreased to 100%, 75%, 53%, respectively, as the influent $NH_4^+$-N concentration increased to 20 mg/*l*, 40 mg/*l*, 80 mg/*l*. However, in SBBR, the nitrification rate was not changed remarkably in spite of the increase of influent $NH_4^+$-N concentration. In SBR, at the high influent $NH_4^+$-N concentration of 80 mg/*l*, the complete deterioration of phosphorus removal occurred because of the abrupt pH drop. However, the phosphorus release and uptake in SBBR were increased at that state.

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