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Simultaneous phosphorus and nitrogen removal in a continuous-flow two-sludge system

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Abstract: The ability of simultaneous biological phosphorus and nitrogen removal was investigated in a lab-scale continuous-flow two-sludge system. Alternating anaerobic and anoxic conditions were combined with contact oxidation stage for treating raw municipal wastewater. Long-term experiments showed that the contradiction of competing for the organic substrate between denitrifying bacteria and PAOs (phosphorus accumulating organisms) in traditional phosphorus and nitrogen removal system has been resolved. The system can adapt to low influent COD/TN ratio (C/N). Furthermore the SRT (sludge retention time) of nitrifying sludge and denitrifying phosphorus removal sludge can be controlled at optimal conditions respectively. The removal efficiency of COD, TP, TN, and NH_4-N was 81.78%, 92.51%, 75.75%, and 84.47% respectively. It was also found that the appropriate influent C/N should be controlled at the range of 3.8—6, while the optimal C/N to the system ranged between 4—5, and the BFR (bypass sludge flow rate) should be controlled at 0.35 around.

Keywords: continue-flow two-sludge system; denitrification phosphorus removal; nitrification; contact oxidation

Introduction

In biological phosphorus removal(BPR) activated sludge systems, heterotrophic microorganisms referred to phosphorus accumulating organisms(PAOs) take up readily biodegradable organic substrate under anaerobic conditions. The organic matter storage occurs in a form of PHA (predominantly poly-β-hydroxybutyrate (PHB) and poly-β-hydroxyvaler-aterate (PHV)). The energy required for PHA storage by PAOs is obtained from decomposition of intracellular polyphosphates, which results in a phosphate release. Simultaneously, a decrease in the glycogen content of cells occurs to keep the redox balance in the cell. In aerobic environment PAOs use organic storage products (predominantly PHB) to generate energy, which is utilized for bacterial growth, replenishment of the glycogen and phosphate storage in a form of intracellular polyphosphate(Kirsten and Anneli, 1994; Sidat *et al.*, 1995; Drysdate *et al.*, 1999; Lee *et al.*, 2001).

However, in a wastewater treatment plant, combining nitrogen and phosphorus removal will decrease denitrification capacity because of the competition between denitrification bacteria and phosphorus accumulating bacteria for organic substrate. Alternatively, denitrifying phosphorus removal bacteria (DPB), which are capable of using nitrate as electron acceptor for simultaneously removal phosphorus and nitrogen from wastewater, has been introduced in the last two decades (Hascoet and Flurentz, 1985; Comeau *et al.*, 1987; Johwan *et al.*, 2002; Hu *et al.*, 2003; Meinhold *et al.*, 1999; Carta *et al.*, 2001). For these bacteria, nitrate is utilized for oxidation of stored PHB and is removed as nitrogen gas (N_2) from wastewater. It is reported that DPB has not only the similar potential for phosphorus removal but also the similar biological metabolism on phosphorus and intracellular organic substance like PHB and glycogen to the conventional A/O phosphorus- removing organism.

The concept of biological phosphorus removal under alternating anaerobic and anoxic condition combined with fixed-film nitrification was applied for the first time by Wanner *et al.* (1991) and further studied by Jeniček *et al.*(Jeniček *et al.*,1993; Peng *et al.*, 2004; Ahn *et al.*, 2003). Kuba *et al.*(1996) studied the ability of phosphorus and nitrogen removal in a SBR tow-sludge system, which they called A_2N process (Anaerobic-Anoxic SBR and Nitrification SBR).

The purpose of this study was to investigate the denitrifying phosphorus removing capacity and the important affecting factors on continuous-flow two-sludge system for simultaneous phosphorus and nitrogen removal.

1 Materials and methods

1.1 Laboratory-scale plant description

The lab-scale continuous flow two-sludge system used for experiments was arranged according to Fig.1. Raw municipal wastewater is introduced into anaerobic tank (1) where phosphate is released and most of organic substrate is absorbed and mainly stored as PHB in bacterial cells. The first settler(2)

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separates activated sludge (DPB) from ammonia and phosphorus-rich supernatant. The liquid stream then goes to contact oxidation reactor(3) where nitrification occurs. Nitrification sludge in the second settler (4) is flowed back to reactor (3). And organic-substrate-rich sludge in settler(2) bypassed nitrification and is mixed with the nitrified effluent from (3) in anoxic stage(5). Here nitrates are removed mainly by DPB as electron acceptor for anoxic P-uptake. Post-aeration step (6) allows nitrogen gas stripping from the sludge before final settler(7).

The operational parameters of the operated activated sludge system are summarized in Table 1.

Table 1 Operational parameters of the continuous-flow two-sludge system

Reactor	Volume, L	Parameter	Value
Anaerobic reactor	9	Influent flow rate(Q)	3 L/h
Settler 1,2,3	3	Bypass sludge flow ratio(r)	0.38
Nitrification reactor	24	Return sludge flow ratio(R)	0.42
Anoxic reactor	16	HRT total	19.7 h
Post-aeration reactor	1	SRT(DPB)	16 d

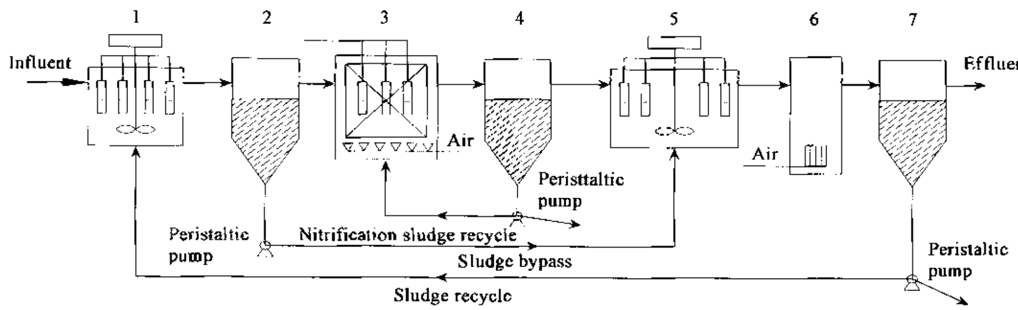


Fig.1 Configuration of continuous-flow two-sludge process
1. anaerobic stage; 2. settler 1; 3. contact oxidation stage (nitrifying with suspended stuffing); 4. settler 2; 5. simultaneously denitrification with P-uptake; 6. post-aeration; 7. final settler

1.2 Wastewater and sludge

1.2.1 Wastewater

Raw wastewater used for the laboratory plant was regularly collected from the fresh domestic sewage. The major characteristics of influent wastewater are shown in Table 2.

Table 2 The major characteristic of influent wastewater Unit: mg/L

Term	pH	T, ℃	COD_{Cr}	BOD_5	NH_4^+	TN	TP	Alkalinity
Max/min	7.1/7.6	25	325/115	180/44	46/28	50/30	6.7/2.1	426/322
Average	7.3	25	187	100	38	44	5.5	360

1.2.2 Sludge

The inoculated activated sludge for the system was taken from EBPR process in Harbin Wenchang Wastewater Treatment Plant.

1.2.3 Sludge domestication

The whole experiment was divided into two main periods according to the operational conditions. Firstly, nitrifying and denitrifying phosphorus removal sludge were domesticated respectively (around two months). Secondly, the lab scale plant was connected and operated continuously until the combined system reach steady removal effect (almost one month). Then the system was run for one year and a half.

In order to domesticate DPB sludge, sequencing anaerobic/aerobic/anoxic conditions are employed in the initial 30 d. In the following 30 d, sequencing anaerobic/anoxic conditions were controlled by introducing NO_3^- into the anoxic tank until denitrifying phosphorus uptake was induced obviously.

1.3 Analytical methods

The samples from the lab-scale plant were taken with a volume of 50 ml and immediately filtered. COD_{Cr}, BOD_5, alkalinity, and MLSS were measured according to APHA Standard Methods(APHA, 1985), and TN, TP, NH_4^+ and NO_3^- were analyzed by spectro-photometer (UV-2550, Japan). The dissolved oxygen (DO) and temperature were measured continuously with oxygen and temperature meter (WTW level2). ORP and PH were measured with ORP and pH meter (WTW level2).

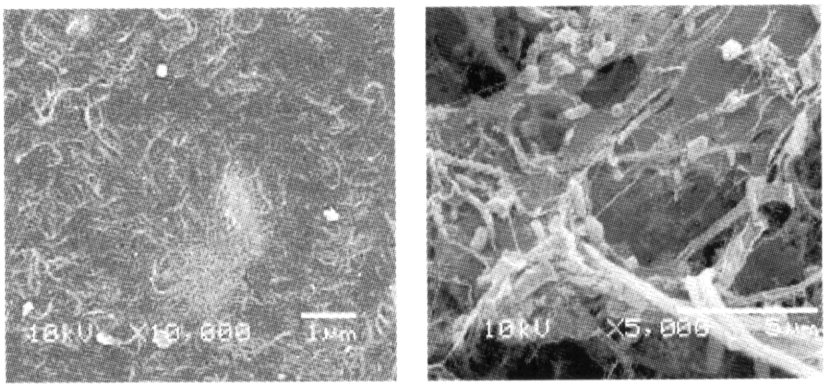
Sudan Black and Methylene Blue staining for the detection of PHA and polyphosphate in sludge cells were carried out according to Serafim *et al.*(2002).

2 Results

2.1 Microorganism culture

2.1.1 Nitrification bacteria

Fig.2 clearly shows the surface of contact medium from contact oxidation tank. Fig.2a is the photograph of the contact medium surface before operation. After 60 d operation, biofilm can be seen on the surface of the medium, and around 80% NH_4^+ in influent was removed. Then the whole system was connected. Another one month later, almost 100% NH_4^+ can be removed. Fig.2b clearly shows nitrifying bacteria attached to the surface of contact medium.

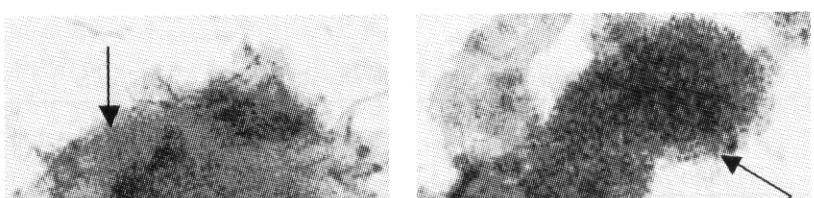


a. Before operation b. 100 days later

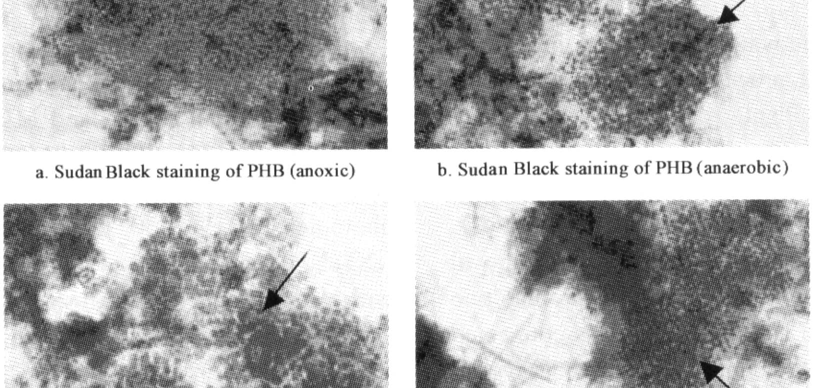
Fig.2 Microphotograph of the contact medium surface

2.1.2 DPB in anaerobic/anoxic tank

After around 60 d domestication, the sludge presented optimistic P-uptake phenomenon under anoxic condition and P-release phenomenon under anaerobic condition. The straining microphotograph of PHB and polyphosphate are shown in Fig.3. Under anoxic condition, PHA is oxidized and poly-P is compounded in DPB, which can be seen clearly from Fig.3a and Fig.3c. On the other way, under anaerobic conditions, PHA is compounded and poly-P is hydrolyzed (Fig.3b and Fig.3d). Arrows show the typical cluster of denitrifying phosphate removal bacteria in the microphotograph.



a. Sudan Black staining of PHB (anoxic) b. Sudan Black staining of PHB (anaerobic)



c. Methylene Blue staining of poly-P (anoxic) d. Methylene Blue staining of poly-P (anaerobic)

Fig.3 Sludge of DPB(bright field), arrows show a typical cluster of DPB

2.2 Long-term operation

The long-term operation results are shown in Fig. 4. COD removal efficiency can be seen from Fig.4a, although the influent COD ranged between 115 and 325 mg/L, the effluent COD ranged between 25 and 45 mg/L with average value of 32.22 mg/L, and the removal efficiency is 81.78%. The performance of the system as to the COD removal was quite stable in spite of a great variation of COD value in the influent, because all of the tanks were able to removal COD at different degrees.

The removal efficiency of NH_4^+ is given in Fig.

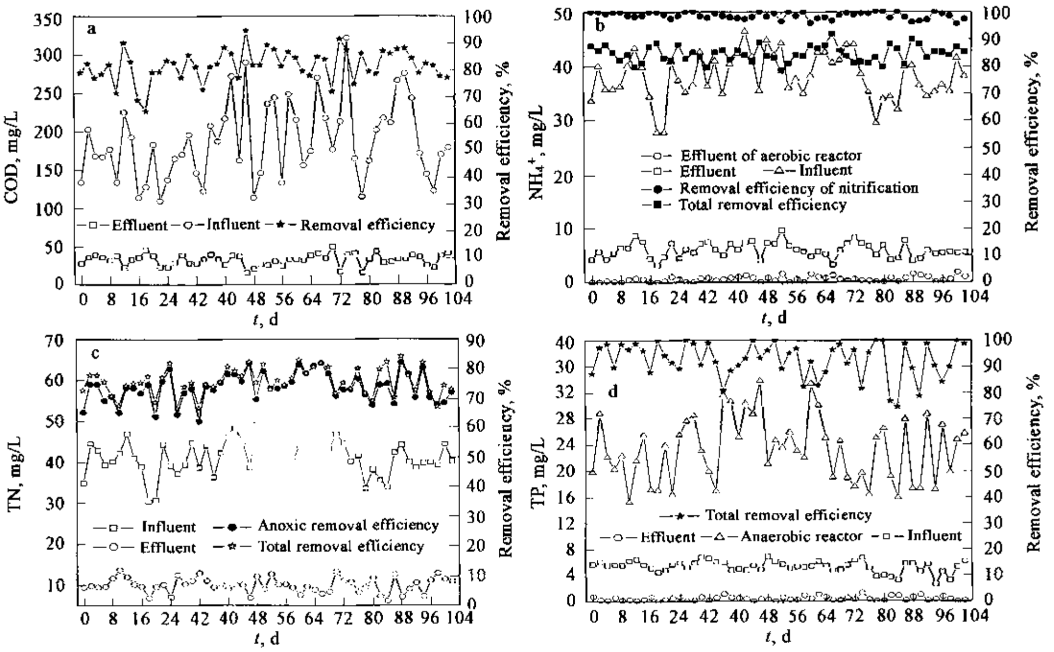


Fig.4 Removal efficiencies of COD, NH_4^+-N, TN and TP

4b. The concentration of NH_4^+ in influent ranged between 28 and 42 mg/L, while the effluent ranged between 4 and 8 mg/L, which means the removal efficiency of NH_4^+ is 84.47%. In the contact oxidation reactor, the removal efficiency of NH_4^+ is almost 100%.

The performance of the system as to TN removal is shown in Fig.4c. TN in the system is mainly removed in anoxic reactor, where nitrate was utilized as electron acceptor for DPB to oxidizing PHB to took up phosphate, and nitrate was simultaneously converged to N_2. When the concentration of TN in influent ranged between 33 and 47 mg/L, the average of TN in effluent is 8.85 mg/L.

Phosphorus removal in the continuous-flow two-sludge system was stable and the efficiency is high, which are more effective than other enhanced biological phosphorus removal(EBPR) system. As can be seen from the Fig.4d, most effluent points are lower than 0.5 mg/L and few are in the range of 0.5—1 mg/L. The average value is 0.42 mg/L. When the average concentration of TP in influent is 5.07 mg/L, the removal efficiency is more than 90%. TP is took up predominately in anoxic tank, while nitrifying tanks has little phosphorus uptake ability.

Fig.5 shows the average concentrations of TP, TN, NO_3^-, NH_4^+ and COD along the system. In the system TP and TN are removed predominantly in anoxic stage, while COD is mainly removed in anaerobic stage.

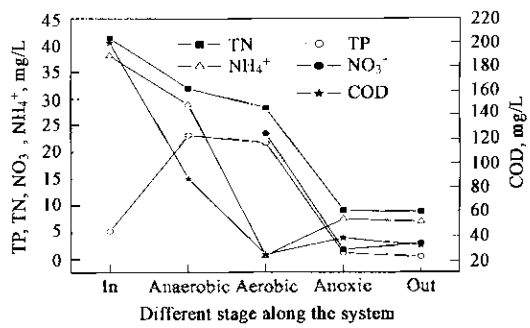


Fig.5 Average concentration of TP, TN, NO_3^-, NH_4^+ and COD along the system

3 Discussion

3.1 Influence of COD/TN ratio(C/N)

It is noted from Fig.6a, when influent C/N ranged between 2.5 and 3, only about 50% TN could be removed by DPB for the surplus of the produced NO_3^- from nitrifying stage. This led to the poor removal of phosphorus, since the excess NO_3^- from the return sludge restrained the release of phosphorus in the anaerobic stage by consuming the limited organic in the influent. With the increase of controlled C/N ratio to 3.8—6, the removal efficiencies of TN, TP and COD were also improved to around 83%, 92% and 86% , respectively. From Fig.6b, it is therefore confirmed that the perfect C/N for the system lies between 4 and 5. If influent C/N ratio was higher than 8, the removal efficiency of TN increased to the highest level, while TP decreased to 30%—40%. It was the result of the competition between DPB and denitrifying bacteria for limited NO_3^- in anoxic reactor, where NO_3^- was more easily used by denitrifying bacteria, and it led to the lack of NO_3^- for denitrifying phosphorus uptake.

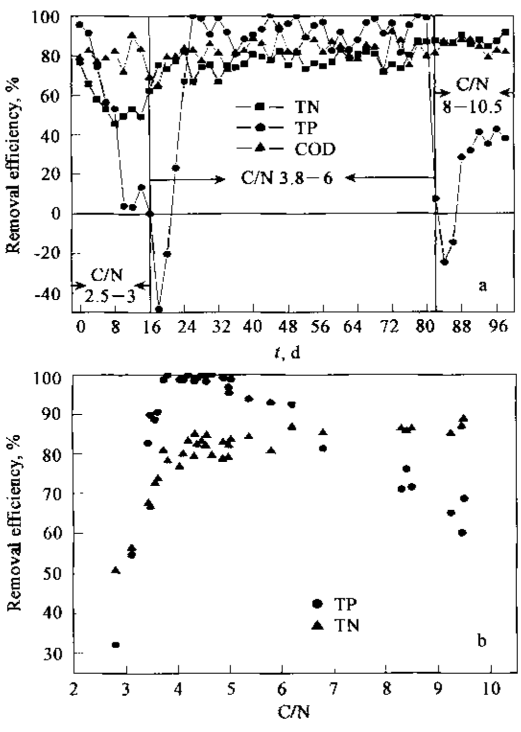


Fig.6 The effect of influent C/N ratios on the system

3.2 Influence of bypass sludge flow rate(BFR)

The amount of ammonia and nitrate in anoxic reactor was mainly determined by BFR, as shown in Fig.7.

When BFR was higher than 0.48, on one hand it elevated the concentration of ammonia near to 10 mg/L, on the other hand nitrate was lack and further led to the decrease of P-uptake amount in anoxic reactor. When the flow rate was lower than 0.28, although the concentration of ammonia decreased to the lowest level(around 4—6 mg/L), the concentration of nitrate and TP were both increased for the DPB

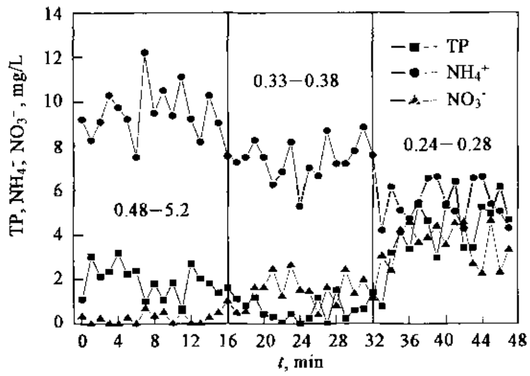


Fig.7 Concentration of TP, NH_4^+ and NO_3^- in anoxic stage at different BFR

sludge were not able to accumulate in anoxic reactor. The optimal BFR should be controlled at 0.35 around.

4 Conclusions

The continuous-flow two-sludge system fed with municipal wastewater was operated for around one and a half year at the experimental scale. The main conclusions could be draw from the presented study: (1) Nitrifying bacteria and DPB were separated completely, which enabled the system to treat wastewater with low influent C/N. (2) In the contact oxidation reactor, complete nitrification was achieved. (3) Efficient phosphorus uptake with simultaneous denitrification occurred in the anoxic reactor. The removal efficiency of COD, TP, TN and NH_4^+-N were 81.78%, 92.51%, 75.75% and 84.47% respectively. (4) It was found that the appropriate influent C/N should be controlled at the range of 3.8—6, while the optimal C/N to the system raged between 4—5. Furthermore BFR should be controlled at 0.35 around.

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