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Is polymeric substrate in influent an indirect impetus for the nitrification process in an activated sludge system?

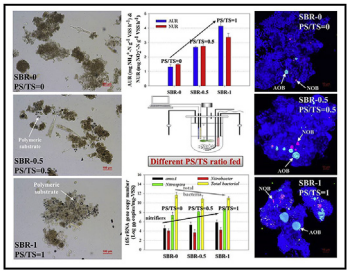
Bin-Bin Wang [a], Ya-Wei Gu [a], Jian-Meng Chen [a], Qian Yao [b], Hui-Juan Li [b], Dang-Cong Peng [b], Feng He [a, *]

[a] College of Environment, Zhejiang University of Technology, Hangzhou, Zhejiang, China
[b] School of Environmental and Municipal Engineering, Xi'an University of Architecture and Technology, Xi'an, Shaanxi, China

HIGHLIGHTS

- Links between PS/TS ratio in influent and nitrification process were explored.
- Nitrification rate was enhanced with the raise of PS/TS ratio.
- Abundance of nitrifiers in mixed culture increased with the raise of PS/TS ratio.
- Proliferation of nitrifiers' microcolony was enhanced with the raise of PS/TS ratio.
- PS was an indirect impetus for nitrification process in mixed culture.

GRAPHICAL ABSTRACT



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ABSTRACT

Different from monomeric substrate, polymeric substrate (PS) needs to undergo slow hydrolysis process before becoming available for consumption by bacteria. Hydrolysis products will be available for the heterotrophs in low concentration, which will reduce competitive advantages of heterotrophs to nitrifiers in mixed culture. Therefore, some links between PS and nitrification process can be expected. In this study, three lab-scale sequencing batch reactors with different PS/total substrate (TS) ratio (0, 0.5 or 1) in influent were performed in parallel to investigate the influence of PS on nitrification process in activated sludge system. The results showed that with the increase of PS/TS ratio, apparent sludge yields decreased, while NO_3^--N concentration in effluent increased. The change of PS/TS ratio in influent also altered the cycle behaviors of activated sludge. With the increase of PS/TS ratio from 0 to 0.5 and 1, the ammonium and nitrite utilization rate increased ~2 and 3 times, respectively. The q-PCR results showed that the abundance of nitrifiers in activated sludge for PS/TS ratio of 0.5 and 1 were 0.7–0.8 and 1.4–1.5 orders of magnitude higher than that for PS/TS ratio of 0. However, the abundance of total bacteria decreased about 0.5 orders of magnitude from the former two to the latter. The FISH observation confirmed that the nitrifiers' microcolony became bigger and more robust with the increase of PS/TS ratio. This paper paves a path to understand the role of PS/TS in affecting the nitrification process in biological wastewater treatment systems.

1. Introduction

The organic contaminants that must be removed from wastewater in biological wastewater treatment systems are complex

* Corresponding author. 18 Chaowang Road, Hangzhou, Zhejiang Province, 310014, China.
E-mail address: fenghe@zjut.edu.cn (F. He).

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mixtures of polymeric and monomeric components (Levine et al., 1991). Polymeric substrate (PS), such as polysaccharides, proteins, lipids, etc. has been reported to be an important fraction of total COD present in wastewater and can account for about 50–90% of the organic load in wastewater treatment systems (Levine et al., 1991; Kappeler and Gujer, 1994; Eliosov and Argaman, 1995; Roeleveld and van Loosdrecht, 2002; Martins et al., 2011). Different from monomeric substrate (MS), PS cannot directly cross the bacterial membrane via specific active transports. Thus, these high molecular weight substrates need to undergo a slow hydrolysis process before becoming available for growth and storage of bacteria (Gujer et al., 1999).

PS is supposed to have significant impacts on a variety of biological processes in wastewater treatment systems owing to its high content and slow metabolic process in wastewater. Puigagut et al. (2007) found that there were different microfauna population structures between PS and MS fed activated sludge systems. PS, such as potato starch and sweet potato starch, was also reported to have a positive impact on sludge settleability via influencing the growth behavior of filamentous bacteria in activated sludge (de Kreuk et al., 2010; Martins et al., 2011; Wang et al., 2013). The content of PS in wastewater was also found to affect denitrification, phosphate release and phosphate uptake rates in biological nutrient removal systems, as well as the carbon and energy footprint of wastewater treatment processes (Drewnowski and Makinia, 2011; Gori et al., 2011). The effects of PS on productions and characteristics of extracellular polymeric substances in activated sludge were also investigated (Wang et al., 2014).

In spite of these research efforts, the relationship between PS and nitrification process in biological wastewater treatment systems has been scarcely studied and the results from limited literatures are even inconsistent. Figueroa and Silverstein (1992) investigated the effects of PS and MS on nitrification in a lab-scale rotating biological contactor and concluded that PS had the same inhibition impacts as MS on nitrification. Michaud et al. (2006) evaluated the effect of the PS/nitrogen ratio on the heterotrophic bacterial communities and nitrification efficiency of a submerged biological filter. They found that the increase of PS/nitrogen ratio had positive impacts on heterotrophic bacterial abundance and consequently had negative effects on nitrification efficiency in the biological filter. However, their study did not involve the comparison between PS and MS. Different from these studies, Puigagut et al. (2007) found that nitrification activity in PS fed activated sludge system was higher than that in MS fed system, indicating the different impacts of PS and MS on nitrification. Nevertheless, Puigagut et al. (2007) did not report the mechanism underlying this difference and only related the different nitrification activity of these two systems to the specific predatory activity of metazoa on nitrifiers. Hence, the precise role of PS in affecting nitrification process in biological wastewater treatment system still needs to be clarified.

Hydrolysis is widely considered as the rate-limiting step for PS degradation and consumption. It means that hydrolysis products will be available for the heterotrophs in a low (growth rate limiting) concentration, which will then reduce the competitive advantages of heterotrophs to nitrifiers in mixed culture. The present work aims to investigate the influences of PS on the competitive balance between heterotrophs and nitrifiers and consequently on the nitrification activity in mixed culture. Filling this research gaps will allow us to understand the significance of PS in affecting the nitrification process in biological wastewater treatment systems. To this end, quantitative polymerase chain reaction (q-PCR) and fluorescent in-situ hybridization (FISH) were also applied to compare community abundance and to observe in situ spatial organization of nitrifiers in activated sludge.

2. Materials and methods

2.1. Experimental setup

The experiments were performed in three parallel aerobic sequencing batch reactors (SBRs) (some detailed operation parameters can refer to Table 1), which were inoculated using activated sludge obtained from a municipal wastewater treatment plant (WWTP) in Hangzhou, China. The SBR had a working volume of 5 L with 30 cm in height and 18 cm in diameter (Fig. S1). The detailed configurations of SBR can be found in Wang et al. (2014). In brief, peristaltic pumps were employed for influent feeding and effluent withdrawing. The liquor mixing, oxygen supply and temperature maintenance in SBRs were achieved by vertical mixer, air pump and temperature controller, respectively. Programmable logic controller was used to automatically control the SBRs operated in cycles of 6 h with 2 min of filling time, 268 min of reacting time, 60 min of settling time, 5 min of withdrawing time and 25 min of idling time. A synthetic wastewater (the detailed components can refer to Tables S1 and S2) was served as the feed medium of SBRs with an initial total substrate (TS, the sum of the PS and MS) of 400 mg-COD L^{-1} and ammonia nitrogen (NH_3-N) of 30 mg L^{-1}. The influent was stored in a refrigerator at 4 °C to prevent the hydrolysis of PS. Three PS/TS ratio (0, 0.5 or 1) were used in the corresponding SBR to evaluate the influence of PS on nitrification process in activated sludge. Sweet potato starch and soluble glucose were selected as model substrates to simulate PS and MS, respectively. The exchange ratio of SBRs was controlled at 0.5 which corresponded to the influent volume of 2.5 L in each cycle. The sludge retention time was kept in 10 d through discharging 500 mL

Table 1
Operation parameters of SBR-0, SBR-0.5 and SBR-1.

SBR system	Organic substrate	PS[c]/TS[d] ratio in substrate (COD basis)	Aeration intensity (L air min^{-1})	DO in the famine time (mg O_2 L^{-1})	Temperature (°C)	pH	SRT (day)	HRT (h)	Sludge content in steady state (mg MLSS L^{-1})	Sludge loading rate in steady state (mg COD mg^{-1} MLSS day^{-1})
SBR-0	SolG[a]	0	0.5	>2	25	6.5 −7.2	10	12	3379 ± 406	0.24
SBR-0.5	SolG + SPS	0.5	0.5	>2	25	6.5 −7.2	10	12	2542 ± 267	0.31
SBR-1	SPS[b]	1	0.5	>2	25	6.5 −7.2	10	12	1642 ± 281	0.49

[a] SolG = soluble glucose.
[b] SPS = sweet potato starch.
[c] PS = polymeric substrate.
[d] TS = total substrate.