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Short Communication

## Unraveling characteristics of simultaneous nitrification, denitrification and phosphorus removal (SNDPR) in an aerobic granular sequencing batch reactor

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### HIGHLIGHTS

- An aerobic granular AOA SBR was constructed for effective SNDPR.
- Fate of carbon, nitrogen and phosphorus was explored.
- Characteristics and mechanisms of SNDPR was investigated.
- Perspectives on optimization of operation was revealed.

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### ABSTRACT

An aerobic granular sequencing batch reactor (SBR) on an aerobic/oxic/anoxic (AOA) mode was operated for 50 days with acetate sodium as the sole carbon source for simultaneous carbon, nitrogen and phosphorus removal. Excellent removal efficiencies for chemical oxygen demand (COD) ( $94.46 \pm 3.59\%$ ), nitrogen ( $96.56 \pm 3.44\%$  for ammonia nitrogen ( $\text{NH}_4\text{-N}$ ) and  $93.88 \pm 6.78\%$  for total inorganic nitrogen (TIN)) and phosphorus ( $97.71 \pm 3.63\%$ ) were obtained over operation. Mechanisms for simultaneous nutrients removal were explored and the results indicated that simultaneous nitrification, denitrification and phosphorus removal (SNDPR) under aerobic conditions was mainly responsible for most of nitrogen and phosphorus removal. Identification and quantification of the granular AOA SBR revealed that higher rates of nutrients removal and more potentials were to be exploited by optimizing the operating conditions including time durations for AOA mode and the feeding compositions.

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### 1. Introduction

In conventional biological nutrients removal processes, nitrogen is removed by nitrification process under aerobic conditions and the following denitrification process under anoxic conditions (Tsuneda et al., 2006), while phosphorus removal is achieved by anaerobic phosphorus release and aerobic phosphorus (He et al., 2016a). Influent ammonia nitrogen ( $\text{NH}_4\text{-N}$ ) is oxidized to nitrite ( $\text{NO}_2\text{-N}$ ) and/or nitrate ( $\text{NO}_3\text{-N}$ ) by autotrophic nitrifying microbes via nitrification way, then the produced  $\text{NO}_2\text{-N}$  and/or  $\text{NO}_3\text{-N}$  is reduced to nitrogen gas by heterotrophic denitrifying organisms via denitrification way (Kuba et al., 1993; Morling, 2001). The organisms responsible for phosphorus removal is referred to polyphosphate accumulating organisms (PAOs), which conduct

phosphorus release by absorbing carbon sources and storing them as polyhydroxybutyrate (PHB), and phosphorus uptake luxuriously by using the stored PHB as the energy source (Bassin et al., 2012; Kim et al., 2002). Therefore, three separate organisms including nitrifying and denitrifying bacteria and PAOs are necessary for simultaneous nitrogen and phosphorus removal, which are resident to different conditions including aerobic, anoxic and anaerobic conditions (Asadi et al., 2016; de Kreuk et al., 2005a).

Aerobic granular sludge is the biomass aggregates grown under aerobic conditions without a carrier material (de Kreuk et al., 2005b; Lochmatter et al., 2013), which has been recognized as an emerging and promising technology for wastewater treatment due to its advantages on high biomass retention, impact microbial structure, rapid settling velocity and so on (de Kreuk et al., 2005a; He et al., 2016b). Moreover, the layered structure of aerobic granules with aerobic zone, anoxic zone and anaerobic zone along the direction of mass transfer is favorable for growth of facultative

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and aerobic bacteria such as ammonia oxidizing bacteria, denitrifying organisms and PAOs (Wang et al., 2009). Therefore, aerobic granules can remove carbon, nitrogen and phosphorus simultaneously (He et al., 2016a,b; Yilmaz et al., 2008).

Previous researches efforts have proven that aerobic granules are beneficial for SNDPR in SBR (de Kreuk et al., 2005a; He et al., 2016b). However, there are multiple mechanisms proposed for simultaneous carbon, nitrogen and phosphorus removal, such as nitrification, denitrification (de Kreuk et al., 2005a), simultaneous nitrification and denitrification (SND) (Zeng et al., 2003), enhanced biological phosphorus removal (EBPR) (Lu et al., 2016), denitrifying phosphorus removal (DNPR) (He et al., 2016b), and some processes coupled by one or more of the above ones (He et al., 2016b; Lu et al., 2016; Semerci and Hasilci, 2016). The mechanism for SNDPR depends on several factors and both the characteristics of reactor and the microbes have significant effects on the transformation of nutrients (Lu et al., 2016). Therefore, different reactor configuration and setup may lead to different ways of nutrients transformation, which requires more efforts to discover the inner associations. However, little work has been done to study the mechanism of SNDPR in the aerobic granular SBR for synthetic domestic wastewater treatment.

Therefore, an aerobic granular sequencing batch reactor operated on anaerobic/oxic/anoxic (AOA) mode was configured to examine the capacity for simultaneous carbon, nitrogen and phosphorus removal. The major aim of the present work was to carefully explore and assess the removal capacity of the system. In view of the removal performance of the configured granular AOA SBR, optimization strategies were also proposed.

## 2. Materials and methods

### 2.1. Set up and operation

A plexiglass sequencing batch reactor (SBR) was configured with a diameter of 100 mm and a height of 500 mm, giving an effective working volume of 3.6 L and a ratio of height to diameter (H/D) of 5.0 (Fig. 1). 1.8 L of synthetic wastewater was pumped into the reactor at the beginning of each cycle with an exchange ratio of 50%. During the oxic period, air was introduced into the reactor from the bottom at a constant airflow rate of about 2.5 L/min, thus providing a dissolved oxygen (DO) concentration of about 5.0 mg/L at the end of the oxic phase. The reactor was subject to mechanical stirring (250 rpm) through the anaerobic, oxic and anoxic phases to prevent settling. Temperature was not controlled during the treatment period and the water temperature was  $19 \pm 2$  °C.

The SBR was operated under sequencing AOA conditions for 50 days on a 6-h-cycle. Each cycle consisted of 2 min of feeding, 120 min of anaerobic phase, 90 min of oxic phase, 144 min of anoxic phase, 2 min of settling and 2 min of discharge periods.

### 2.2. Seed sludge and wastewater

Prior to the present study, mature aerobic granules was inoculated into the reactor, which were formed in our previous work (He et al., 2016b). The granules were with an average diameter of  $1.5 \pm 0.5$  mm and a sludge volume index at 5 min ( $SVI_5$ ) of  $22.58 \pm 0.69$  mL/g. The concentration of the mixed liquor suspended solids (MLSS) in each reactor was  $4.4 \pm 0.5$  g/L.

The system was fed with synthetic wastewater as follows (per liter): NaAc 256.27 mg,  $NH_4Cl$  76.4 mg,  $KH_2PO_4$  14.6 mg,  $CaCl_2$  10.6 mg and  $MgSO_4 \cdot 7H_2O$  90 mg and 1 mL of trace solution as described by He et al. (2016a). The pH of influent wastewater was adjusted to about 7.5 using HCl or NaOH solutions without

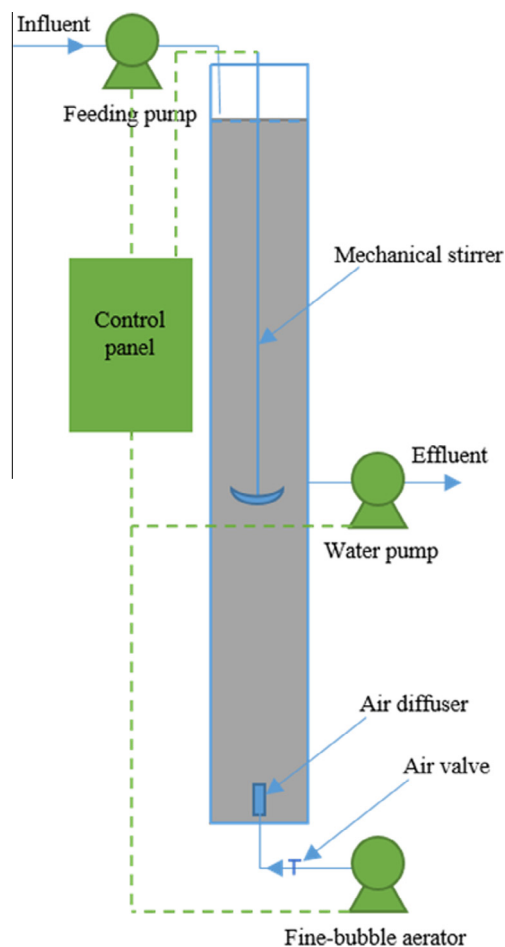


Fig. 1. Schematic diagram of the sequencing batch reactor.

control during the operation. No discharge of sludge was detected over operation.

### 2.3. Analytical methods

The COD, nitrogen (including  $NH_4^+-N$ , nitrate ( $NO_3^- -N$ ), nitrite ( $NO_2^- -N$ )), TP, MLSS, sludge volume index at 5 min ( $SVI_5$ ) were measured according to the standard methods (APHA, 2005). Total inorganic nitrogen (TIN) was regarded as the sum of  $NH_4^+-N$ ,  $NO_3^- -N$ ,  $NO_2^- -N$  (Long et al., 2014). The pH and DO were measured using a pHs-25m and YSI5000 meter. EPS were extracted with a modified heat extraction method by Yang et al. (2014). Protein (PN) content was determined by a modified Lowry method and polysaccharides (PS) content was analyzed using a phenol-sulfuric acid method (Long et al., 2014). EPS was regarded as the sum of PN and PS.

## 3. Results and discussion

### 3.1. Reactor performance

The time course of carbon, nitrogen and phosphorus concentrations over operation was present in Fig. 2. However, the whole process could be manually divided into two phases according to the removal performance for nitrogen including  $NH_4^+-N$ ,  $NO_2^- -N$  and  $NO_3^- -N$ , namely phase I (day 1–4) and phase II (day 5–50), though minimal effects on COD and phosphorus removal were found within both phases (Table 1).

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