Effect of sludge discharge positions on steady-state aerobic granules in sequencing batch reactor (SBR)

Lin Liu, Da-Wen Gao and Hong Liang

ABSTRACT

We have investigated the effect of sludge discharge location on the steady-state aerobic granules in sequencing batch reactors (SBRs). Two SBRs were operated concurrently with the same sludge retention time using sludge discharge ports at: (a) the reactor bottom in R1; and (b) the reactor middle-lower level in R2. Results indicate that both reactors could maintain sludge granulation and stable operation, but the two different sludge discharge methods resulted in significantly different aerobic granule characteristics. Over 30 days, the chemical oxygen demand (COD) removal of the two reactors was maintained at similar levels (above 96%), and typical bioflocs were not observed. The average aerobic granule size in R2 was twice that in R1, as settling velocity increased in proportion to size increment. Meanwhile, the production yields of polysaccharide and protein content in R2 were always higher than those in R1. However, due to mass transfer limitations and the presence of anaerobes in the aerobic granule cores, larger granules had a tendency to disintegrate in R2. Thus, we conclude that a sludge discharge port situated at the reactor bottom is beneficial for aerobic granule stability, and enhances the potential for long-term aerobic granule SBR operation.

Key words | aerobic granules, sequencing batch reactor, sludge discharge, wastewater treatment

INTRODUCTION

Aerobic granulation is a new and promising method for overcoming the principal shortcomings of the activated sludge process. Aerobic granules have been shown to possess a number of advantages over conventional activated sludge, such as possessing a compact and strong microbial structure, having good settling properties and high biomass retention (Liu & Tay 2004; Zheng et al. 2005; Liu et al. 2009).

At the present time, the majority of lab- and pilot-scale aerobic granulation research programs place the sludge discharge port in the middle part of the reactor (Beun et al. 2002; Ni et al. 2009). According to selection pressure theory, at the end of one cycle, sludge particles with better settling ability may be selected by the port, and flocculent sludge that cannot settle in the chosen settling time is washed out of the reactor (Qin et al. 2004,a, b). This aerobic granulation process is accelerated and enhanced by the higher volume exchange ratio obtainable in sequencing batch reactors (SBRs) (Wang et al. 2006). Thus, the positioning of the discharge port at the middle-lower part of the reactor is regarded as a key component in the design of SBRs for aerobic granulation. However, in steady-state granular sludge systems, the effect of the sludge discharge method on aerobic granules is not clear. Additionally, in full-scale systems, a discharge port at the middle-lower level would lead to the accumulation of impurities and fine sand at the reactor bottom, with a negative impact on the stability of the reactor.

Therefore, it is important to study the effect that different methods of sludge discharge may have on the properties of mature aerobic granules, and to find the most suitable sludge discharge location for SBR granular sludge systems. In this study, two granular sludge reactors were concurrently operated with the same sludge retention time using: (a) a sludge discharge port at the reactor bottom in R1; and (b) a port at the reactor middle-lower level in R2. It was expected that the results derived from this study would give further insight into the operation of steady-state aerobic granular systems.

MATERIALS AND METHODS

Experimental set-up and SBR operation

Experiments were performed in two SBR reactors (55 cm in working height and 25 cm in diameter) with a working volume of 12 L. The influent was added from the top of the reactor, and the effluent was discharged at 20 cm above the reactor bottom with a volumetric exchange ratio of 60%. The cycle time in both reactors was approximately 4 h. Each cycle consisted of 1 min of influent addition, 230 min of aeration, 1 min of settling time and 5 min of discharging. In both reactors, the sludge retention time was 10 days, and the temperature was maintained at 24 ± 1°C using a ribbon heater and temperature controller. Air was introduced by air pump at a flow rate of 0.4 m³/h through a diffuser at the bottom of the reactor. Figure 1 shows a schematic of the two SBRs; 'a' and 'b' indicate the sludge discharge ports used in R1 and R2, respectively.

Synthetic wastewater was used throughout the experiment. The influent glucose concentration in the wastewater was measured as chemical oxygen demand (COD), which was 540–560 mg/L. Other components were NH₄⁺-N (NH₄Cl) 32–36 mg/L, PO₄³⁻-P (KH₂PO₄) 5–7 mg/L, NaHCO₃ 250 mg/L, and trace element solution 1.0 mL/L. The composition of the trace element solution was CaCl₂ 50 mg/L, MgSO₄ 50 mg/L, FeCl₃ 20 mg/L, CuSO₄ 50 mg/L, MnSO₄·H₂O 50 mg/L, KCl 18 mg/L, AlCl₃ 15 mg/L, ZnSO₄·7H₂O 30 mg/L, H₃BO₃ 40 mg/L. Mature aerobic granules, obtained from an SBR, were used as seed sludge, with an initial concentration of 4,000 mg/L in mixed liquor suspended solids (MLSS).

Analytical methods

COD, NH₄⁺-N, moisture content and sludge volume index (SVI) were measured periodically using Standard Methods (APHA 1998). Extracellular polymeric substances (EPS) in the granular sludge were extracted by the heating centrifugation extraction method (Adav & Lee 2008). The polysaccharide (PS) content of the EPS was measured using the phenol–sulfuric acid method, and the protein (PN) content was quantified by the modified Lowry method (Lowry et al. 1951; Dubois et al. 1956). The granule samples were taken for size distribution analysis, which was conducted based on the dry weight of aerobic granules passed through different sized wet sieves (Laguna et al. 1999). Physical strength of the aerobic granules could be expressed as the integrity coefficient (%), which has been previously described (Tay et al. 2002). Specific gravity of the sludge was measured using the method described by Zheng et al. (2005).

RESULTS AND DISCUSSION

Morphology observation

The observation clearly shows that both sludge discharge methods could support aerobic granulation, but the granules in R1 were of a smaller size and had a more compact structure than those in R2. Also, anaerobes could be detected in the core part of aerobic granules in R2. These results are similar to those found in other research. Compared with the floccular sludge, the special structure (large size and compactness) of aerobic granules limits the diffusion of substrate and dissolved oxygen, allowing biodegradation of cells located in the core part of the aerobic granule under the oxygen-limited conditions (Li & Liu 2005; Wang et al. 2005; Li et al. 2008). Although the sludge discharging method in R1 could not remove sludge that had disintegrated and thus had poor settling ability, only typical floccular sludge was found in the SBR. This result implies that floc sludge could not dominate following aerobic granulation, and smaller size granules would instead form in the SBR.

Size distribution

The size distribution of aerobic granules was related to the reactor sludge discharge method, as shown in Figure 2.
Initially, approximately 70% of seed granules were in the range of 1.0–2.0 mm, and only 5% were below 0.5 mm. The granule sizes in R1 and R2 showed different trends over the 30-day operation, but granules in R2 were consistently larger than in R1. For R1, the highest granule fraction decreased to a range of 0.8–1.0 mm, and granules with a size smaller than 0.5 mm increased to approximately 9% of the total after 20 days. After that time, the aerobic granule size distribution in R1 remained relatively unchanged. In a column SBR, the aerobic granule sizes have a positive correlation with settling velocity, so the larger aerobic granules settle to the bottom of the reactor first (Toh et al. 2003). Thus, in R2, the larger granules remained in the reactor, resulting in a higher portion of bigger granules in general. In R1, the larger granules were quickly washed out through the discharge port at the bottom of the reactor, and the size distribution of aerobic granules was maintained at a lower and more stable level. However, owing to disintegration of very large granules in R2, from which sludge was withdrawn from the middle-lower port, the size of granules did not follow a steady-state trend, but fluctuated (Figure 2). For R2, the proportion of granules in the range of 0–0.5 mm was always less than 5%, but the highest volume percentage of aerobic granules fluctuated, increasing from 1.3–1.8 mm to 2.0–2.3 mm by day 10, then changing to 1.5–1.8 mm and 1.8–2.0 mm by days 20 and 30, respectively.

**Extracellular polymeric substances**

EPS are secreted largely by microorganisms (mainly bacteria) in harsh environments, and play an important role in building and maintaining the structural stability of aerobic granules through the cohesion and adhesion of microbial cells (Wang et al. 2005; Adav et al. 2008). The main components of EPS are proteins and polysaccharides (Horan & Eccles 1986). Our results illustrated in Figure 3 indicate that the PS and PN content in EPS showed a similar dependence on granule size in both reactors. For R1, we found that the PN content in EPS decreased from 48.2 mg/g MLVSS (mixed liquor volatile suspended solids) to 38.37 mg/g MLVSS, but the PS content of granules remained largely unchanged from the initial seed granules (about 15 mg/g MLVSS) over the 30 days. However, in the granular system with the discharge sludge port at the middle-lower part of the reactor, PS and PN content
increased to 27.14 mg/g MLVSS and 63.23 mg/g MLVSS, respectively, over 10 days, but the EPS also had a larger rebound after 10 days, and it did not achieve a stable level in this study. Meanwhile, as shown in Figure 3, the PS and PN contents in R2 were always higher than those in R1. This finding implied that the granule size and size distribution in the SBRs were related to the EPS content of the biomass in the systems.

Physical characteristics

As the diameter of aerobic granules changes, so do the physical properties, which can affect reactor operation. The physical characteristics of aerobic granules in the two reactors were analyzed in terms of specific gravity, moisture content, physical strength, volatile suspended solids/suspended solids (VSS/SS) and settling ability. These data are shown in Table 1 and Figure 4. Moisture content and specific gravity are important indicators of the physical characteristics of activated sludge. As illustrated in Table 1, the moisture content and specific gravity of aerobic granules in R1 and R2 had decreased after 30 days, but still exhibited higher levels than floc sludge (Zheng et al. 2005; Shi et al. 2009). Moisture content and specific gravity of aerobic granules in R2 underwent larger changes than those in R1. This result indicated that sludge discharge from the bottom of the reactor was beneficial, leading to the development of mature aerobic granules that maintained highly dense and compact structures. This is advantageous to the operation of a steady-state granular system. In this study, physical strength was expressed as the integrity coefficient (%), which is defined as the ratio of residual granules to the total weight of granule sludge after 5 min of shaking at 200 rpm on a platform shaker. The integrity coefficient was 97.4% for R1 and 84.3% for R2. Towards the end of the experiment, the strength of the aerobic granules showed a decreasing trend in both reactors, but with larger amplitude for R2. Thus, aerobic granules in the reactor with discharge sludge from the bottom of the reactor had better ability to withstand frequent collision and attrition among aerobic granules, and stronger friction between granules and liquid.

The MLVSS to MLSS (VSS/SS) ratio measured in this study was larger than that found in full-scale reactors. It is possible that the synthetic wastewater used had lower levels of impurities than typical, real wastewater. The VSS/SS ratio of granules in R2 was approximately 84%, which was less than the value in R1. However, as both SBRs operated under identical conditions over the 30-day experiment, the different VSS/SS ratios can be attributed to the different sludge discharge methods employed in the reactors. This finding indicates that the sludge contains more biomass in R1, because the non-biological impurities with better settling ability and lower moisture content (e.g., dead and mineralized granules, crystallization of calcium and magnesium) would be discharged with waste sludge. In addition, diffusion limitations of the substrate could occur from aerobic granules with a size of more than 0.4 mm, and the substrate removal rate of the granules with a size of 0.5 mm was almost three times that of the granules of 1 mm size (Gao et al. 2011). Thus, granule size and bioactivity might have a corresponding relationship. As seen in Figure 4, aerobic granules in R2 showed better settling ability in the middle stages of the experiment, but the SVI₃₀ of sludge in R2 rose to a higher value than that

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Seed sludge</th>
<th>R1</th>
<th>R2</th>
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<td>VSS/SS (%)</td>
<td>94.2</td>
<td>94</td>
<td>88.7</td>
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Figure 3 | Changes in PS and PN content in EPS over 30 days.

Figure 4 | SVI₃₀ values of aerobic granules in R1 and R2 over 30 days.
in R1 by the end of the study. On day 3, the SVI30 value of aerobic granules in R1 began to increase, attaining a level of 30–33 ml/g after 10 days, compared with a seed granule value of 25 ml/g. In R2, the SVI30 value initially decreased to 20 ml/g, but then increased to 36 ml/g after 23 days. This result indicates that sludge discharge from the bottom of the reactor could have a positive effect on the sludge settling ability.

The performance of COD and nitrogen removal

The changes to COD and nitrogen removal during a typical cycle of the two reactors were investigated, and the removal efficiency is shown in Table 2. In this study, we maintained similar levels of total biomass in R1 and R2, and operated the two reactors under identical conditions. Therefore, the observed differences in COD and nitrogen removal were directly related to the aerobic granule characteristics. COD removal was highly efficient in both R1 and R2, with values of 97.2 and 96.8%, respectively, using similar influents. However, the nitrogen efficiency clearly differed between the two reactors. It was found that the NH₄⁺-N removal in R1 (98.2%) was a little higher than that in R2 (96.6%), but the total nitrogen (TN) removal efficiency in R2 was approximately 4% higher than in R1. This finding demonstrated that the aerobic granules retained good organic carbon degradation performance under different sludge discharge methods, but the sludge properties had an important effect on nitrogen transformation and removal. A dissolved oxygen gradient exists along the granule structure, and the granule can be divided into an aerobic zone, a micro-oxygen zone and an anoxic zone, with a wide range of microenvironments (Gao et al. 2014). The microenvironments promote growth of different trophic bacteria within the granules, all with different metabolic functions including nitrifiers, denitrifiers and others. Thus, compared with the granules in R1, the bigger size aerobic granules in R2 had a larger anoxic volume at the core available for denitrification.

| Table 2 | COD and nitrogen removal in two reactors during the aerobic phase |
|---------|------------------------|------------------------|------------------------|------------------------|------------------------|
|         | COD (mg/l) | NH₄⁺-N (mg/l) | NO₂⁻-N (mg/l) | NO₃⁻-N (mg/l) | TN (mg/l) | COD | NH₄⁺-N | TN |
| R1      | Influent | 546         | 34.3          | 0.25         | 1.54      | 36.09 | 97.2 | 98.2 | 89.7 |
|         | Effluent | 15          | 0.6           | 0.73         | 2.38      | 3.71  |      |      |     |
| R2      | Influent | 540         | 35.8          | 0.18         | 0.96      | 36.94 | 96.8 | 96.6 | 93.6 |
|         | Effluent | 21          | 1.2           | 0.58         | 0.56      | 2.34  |      |      |     |

CONCLUSION

The steady-state granular sludge system could retain sludge granulation and stable operation under different sludge discharge methods. However, the sludge discharge method affected the size distribution of aerobic granules, which further affects the performance and characteristics of aerobic granules in the SBR. The granular sludge system with a discharge port at the bottom of the reactor kept the maximum particle size under 1 mm in diameter, thereby increasing the bioactivity and improving the physical characteristics of aerobic granules in the reactor. Thus, it can be concluded that sludge discharge port set-up at the reactor bottom is beneficial for stability of aerobic granules, and enhances the potential for long-term stable operation of aerobic granular biomass systems.

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