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Biosolids reduction by the oxic-settling-anoxic process: impact of sludge interchange rate

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Abstract

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Keywords

rate, sludge, impact, process, anoxic, settling, oxic, reduction, biosolids, interchange

Disciplines

Engineering | Science and Technology Studies

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Biosolids reduction by the oxic-settling-anoxic process: Impact of sludge interchange rate

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Abstract

The impact of sludge interchange rate (SIR) on sludge reduction by oxic-settling-anoxic (OSA) process was investigated. The sludge yield of an OSA system (a sequencing batch reactor, SBR, integrated with external anoxic reactors) was compared to that of a control (an SBR attached to a single-pass aerobic digester). SIR (%) is the percentage by volume of sludge returned from the external reactor into the main bioreactor of the OSA, and was varied from 0-22%. OSA achieved greater sludge reduction when fed with unsettled sewage (sCOD=113 mg/L) rather than settled sewage (sCOD=60 mg/L). The SIR of 11% resulted in the highest OSA performance. At the optimum SIR, higher volatile solids destruction and nitrification/denitrification (*i.e.*, conversion of destroyed volatile solids into inert forms) were observed in the external anoxic and intermittently aerated (*i.e.*, aerobic/anoxic) reactors, respectively. Denitrification in the aerobic/anoxic reactor was inefficient without SIR. Effluent quality and sludge settleability were unaffected by SIR.

Keywords: denitrification, oxic-settling-anoxic, sludge interchange rate, sludge/biosolids reduction.

1. Introduction

The worldwide trend of increasing environmental standards, particularly in sewer coverage, entails the production of excess sludge, an inevitable by-product of biological wastewater treatment. The EU, the USA, and China each produce 6-11 million tons of sludge as dry solids (DS) per year (Fytli and Zabaniotou, 2008; Semblante et al., 2014; Yang et al., 2015). Australia also produces a significant amount of DS (*ca.* 0.3 million tons) each year (Semblante et al., 2014). The production of excess sludge is problematic because sludge volume is notoriously

difficult to reduce due to its unique biological properties (Mowla et al., 2013; Ratanatamskul and Saleart, 2015), and there are only a few options for sludge disposal (landfilling, incineration, agricultural reuse) (Tchobanoglous et al., 2003). Landfilling is heavily restricted especially in countries with limited space. Incineration removes only 70% of DS and creates ash with high metal content (Fytili and Zabaniotou, 2008). The agricultural reuse of stabilised and dewatered sludge called “biosolids” has been encouraged to enable nutrient recycling. However, the transport of biosolids to end users is potentially costly (Semblante et al., 2014). Furthermore, biosolids may contain trace organic compounds, such as pharmaceuticals, pesticides, and industrial chemicals, which have long-term effect on the environment and human health (Clarke and Cummins, 2015; Semblante et al., 2015b). Therefore, to decrease the costs and risks associated with sludge treatment and disposal, it is imperative that sludge production is reduced. Research efforts have generated innovative strategies such as the use of advanced oxidation processes to destroy biomass (Wang et al., 2015; Yao et al., 2008), chemical addition to disrupt metabolic processes (Fang et al., 2015; Feng et al., 2014), and sludge cycling in alternating redox conditions through the oxic-settling-anoxic (OSA) process (Semblante et al., 2014). Although the use of advanced oxidation processes and chemical addition has shown potential to reduce sludge, they require high capital investment (Foladori et al., 2010) and/or may introduce undesired products in treated water (Mahmood and Elliott, 2006). In contrast, OSA offers a potentially cost-efficient and low-impact alternative to sludge reduction. OSA reduces sludge production by temporarily holding activated sludge in an external anoxic and substrate-deficient reactor, then recirculating it to the main bioreactor. Several mechanisms of sludge reduction in OSA have been hypothesized. They include enhanced cell lysis, extracellular polymeric substance (EPS) degradation, and selection of slow-growing bacteria (Navaratna et al., 2014; Semblante et al., 2014). A few OSA configurations including those that

involve external anaerobic (Chon et al., 2011; Chudoba et al., 1992) or anoxic (Coma et al., 2013; Saby et al., 2003) reactors have been discussed in the literature. Laboratory-scale OSA fed with synthetic wastewater have shown promising sludge reduction (e.g. 50-80%) (Chon et al., 2011; Saby et al., 2003; Sun et al., 2010). However, these high sludge reduction values have not been realized in full-scale systems (Coma et al., 2013). In a previous study, 30% reduction in the sludge yield was achieved by a laboratory scale OSA system using real sewage (Semblante et al., 2015a). OSA performance is influenced by different operation conditions, such as oxidation reduction potential (ORP), sludge retention time (SRT), and sludge loading rate of the external reactor (Coma et al., 2013; Saby et al., 2003; Ye et al., 2008). To date, the manipulation of these parameters have only resulted in variable success (Coma et al., 2013; Saby et al., 2003; Ye et al., 2008).

To improve OSA performance and ensure reliable performance for the water industry, it is essential to elucidate the impact of operation conditions such as sludge interchange rate (SIR) and influent chemical oxygen demand (COD) on sludge reduction. Changing SIR varies the residence time of sludge in aerobic/anoxic regimes and may have important implications on sludge reduction mechanisms. However, current information in the literature is inadequate to pin-point the optimum SIR value or range for sludge reduction.

Khursheed et al. (2015) observed that increasing the ratio of sludge exposed to anaerobic and aerobic conditions ($0-8.24 \text{ g MLVSS}_{\text{anaerobic}}/\text{g MLVSS}_{\text{aerobic}}$) in OSA enhanced sludge reduction (0-39.8%). Saby et al. (2003) investigated the impact of sludge retention time (SRT) in the external anoxic reactor of OSA over a range of 11-17 days and observed 23-58% reduction in biosolids production under longer SRTs or smaller SIRs. The SRT of the anoxic reactor in the study of Saby et al. (2003) was significantly longer than that of Ye et al. (2008) (5.5-11.5 h), but similar sludge reduction has been achieved by both studies. On the other hand, Sun et al. (2010)

were able to achieve an enhanced sludge reduction (from 53 to 77%) by increasing the frequency of return from once per day to four times per day while maintaining the SIR between an SBR and an external anaerobic reactor at 10%. Given the inconsistent trends reported in the literature, it is worthwhile to systematically investigate the impact of SIR on OSA performance. Additionally, influent COD concentration affects biomass growth and substrate consumption (Gómez et al., 2006), but its impact on OSA remains to be evaluated. Thus, a systematic investigation under different influent COD concentrations is essential to assess the performance of OSA in plants with and without primary sedimentation.

To address the aforementioned research gaps, this study aims to systematically investigate the impact of SIR on sludge reduction by OSA at different influent strengths, *i.e.*, using real sewage before and after primary settling. Volatile solids content and a range of water quality parameters including COD and nutrient concentrations of the reactors were monitored during continuous operation of the reactors over a period of 475 days to elucidate the impact of SIR on sludge reduction.

2. Materials and methods

2.1 Wastewater characteristics

Unsettled and settled sewage (Table 1) were collected from the Wollongong WWTP fortnightly and stored at 4 °C. The former was collected at the beginning while the latter was collected at the outlet of the sedimentation channel. It is noted that due to rapid hydrolysis of readily biodegradable solid particles and the higher soluble ammonia concentration in the unsettled sewage, the soluble COD (sCOD) of the unsettled sewage was significantly higher than that of the settled sewage (113 ± 87 , $n=33$ vs. 60 ± 32 , $n=48$) as can be seen in Table 1.

[Table 1]

2.2 Reactor configuration and operation

Two systems were operated in parallel: the first consists of SBR_{control} (5 L) attached to a single-pass aerobic digester (2 L) forming the control system (Figure 1a), and the second consists of SBR_{OSA} (5 L) attached to a sequential aerobic/anoxic reactor (2 L) and anoxic reactor (2 L) to form the OSA system (Figure 1b).

[Figure 1]

SBR_{control} and SBR_{OSA} were fed with real wastewater (Section 2.1), and operated at 4 cycles/day and a HRT of 12 hours. Each cycle comprised of 15 min of filling, 5 hours and 30 min of aeration, 1 hour of settling, and 15 min of decanting. The SRT of both SBRs was maintained at 10 days by regular sludge wastage (W) (Figure 1).

The aerobic digester of the control system (Figure 1a) was continuously aerated using an air diffuser. The SRT of this digester was maintained at 20 days by regular sludge wastage (Q_{out}).

The aerobic digester was fed with sludge from SBR_{control} thickened to 5-10 g/L by centrifugation for 10 min at 3,267xg (Q_{in}). The supernatant produced by the thickening step was discarded.

In the OSA system (Figure 1b), the aerobic/anoxic reactor was intermittently aerated (*i.e.*, 8/16 hours aeration on/off) using an air diffuser placed at the bottom of the reactor, while the anoxic reactor was kept airtight using a silicone-lined cap with inlet and outlet ports. The aerobic/anoxic reactor was fed with sludge from SBR_{OSA} thickened to 5-10 g/L (q_1).

Thirty-three percent (33%) of sludge from the aerobic/anoxic reactor was transferred to the anoxic reactor (q_2), and 67% was discharged to achieve a total SRT of 20 days (q_3). The sludge discharged from the aerobic/anoxic reactor was thickened to 16-24 g/L by centrifugation for 10 min at 3,267xg. The supernatant was returned to SBR_{OSA}, and the pellet was discarded. Sludge from the anoxic reactor was returned to the aerobic/anoxic reactor (q_4) and SBR_{OSA} (q_5).

The control and OSA systems were initially operated for 151 days using settled sewage as feed, during which steady-state was achieved. Then, to observe the impact of SIR on OSA

performance, the SIR between the anoxic reactor and SBR_{OSA} (q_5) was adjusted to 11, 16.5, and 22% by volume (Figure 1b) while using settled sewage as feed to the SBRs (Supplementary Table S1). The best OSA performance, *i.e.*, the highest reduction in sludge yield of SBR_{OSA} relative to SBR_{control} was achieved at 11%, and thus this condition was also evaluated using unsettled sewage as feed to the SBRs (Section 3.2). To confirm the observed trends with unsettled sewage, the interchange of sludge between SBR_{OSA} and the external anoxic reactor was suspended (*i.e.*, there was no SIR), and then resumed at 11% (Supplementary Table S1).

2.3 Calculation of sludge yield

Sludge reduction was determined by comparing the sludge yield of the SBRs. The observed sludge yield (Y_{obs}) was defined as:

$$Y_{obs} = \frac{P}{C} = \frac{g \text{ MLVSS}}{g \text{ sCOD}} \quad \text{Equation 1}$$

wherein P is the sludge produced in terms of mixed liquor volatile suspended solids (MLVSS) and C is the substrate consumed in terms of sCOD. The experimental Y_{obs} was derived from the slope of the linear regression of the cumulative sludge produced versus the cumulative substrate consumed (Supplementary Table S2). Cumulative values were obtained by incrementing the variations in sludge production and substrate consumption in previous sampling intervals (Chon et al., 2011).

The reduction in sludge yield due to OSA was estimated using Equation 2:

Sludge yield reduction (%)

$$= \frac{\text{Sludge yield}_{\text{SBR}_{\text{control}}} - \text{Sludge yield}_{\text{SBR}_{\text{OSA}}}}{\text{Sludge yield}_{\text{control}}} \times 100 \quad \text{Equation 2}$$

2.4 Analytical techniques

Total and volatile suspended solids (TSS and VSS) of influent and effluent and MLSS and MLVSS of sludge were measured using the APHA Standard Method 2540 (Eaton et al., 2005). The sludge volume index (SVI) was measured using 1000 mL of sludge according to APHA Standard Method 2710-D (Eaton et al., 2005). The sCOD of influent and effluent was measured by passing samples through 1 µm filter paper followed by measurement using a Hach DBR200 COD Reactor and a Hach DR/2000 spectrophotometer (program number 430 COD LR) according to US-EPA Standard Method 5220. Total organic carbon (TOC) and total nitrogen (TN) were analyzed using a TOC/TN-VCSH analyzer (Shimadzu, Japan). Ammonia (NH₃) and orthophosphate (PO₄³⁻) were measured using flow injection analysis (Lachat instruments, Milwaukee, USA) following the APHA Standard Method 4500 (Eaton et al., 2005). Nitrite and nitrate were measured using ion chromatography (Shimadzu, Japan) with Ionpac AS23 anion-exchange column.

3. Results and discussion

3.1 Impact of sludge interchange rate

3.1.1 Reactor performance

OSA performance was initially investigated using low strength (settled) sewage (Supplementary Table S1). During this period, the TOC removal efficiency of SBR_{control} (58.4±31.1%; n=48) and SBR_{OSA} (59.3±34.5%; n=48) was almost identical (Supplementary Figure S3). Moreover, the

effluent quality of $SBR_{control}$ and SBR_{OSA} were similar to each other in terms of COD and ammonia concentration (Figure 2). The results indicate that the effluent quality from the main bioreactor (*i.e.*, SBR_{OSA}) was unaffected by any variation in SIR.

[Figure 2]

$SBR_{control}$ and SBR_{OSA} did not exhibit any discernible orthophosphate removal. The amount of anoxic sludge (orthophosphate concentration= 48.4 ± 23.0 mg/L; $n=22$) interchanged with SBR_{OSA} was relatively low (0.033-0.067 L/day), therefore such interchange did not affect orthophosphate removal by SBR_{OSA} . The SVI of $SBR_{control}$ and SBR_{OSA} at all SIR was below 100 mL/g (Supplementary Figure S4), confirming that SIR had negligible impact on sludge settleability.

3.1.2 Sludge reduction

Discernible variation in MLVSS of $SBR_{control}$ and SBR_{OSA} occurred due to temporal fluctuations in the sCOD of the settled sewage (Figure 3). Nonetheless, $SBR_{control}$ maintained a slightly lower MLVSS than that of SBR_{OSA} when SIR was 16.5 and 22%. The opposite occurred when SIR was changed to 11% (Figure 3). This is an indication that sludge reduction by SBR_{OSA} was enhanced at an SIR of 11%, and the sludge yield data (Table 2) further demonstrated this trend.

[Figure 3]

[Table 2]

An SIR of 11, 16.5, and 22 % was equivalent to a residence time of 17.2, 16, and 15 days, respectively, in the external reactors. Comparison of the sludge yield (Y_{obs}) of $SBR_{control}$ and SBR_{OSA} (Supplementary Figure S5) shows that the highest sludge yield reduction (*i.e.*, 53%) was attained at an SIR of 11% (Table 2). Although a systematic investigation of SIR is not available in the literature, the findings of this study agree with the general trend reported in a few available studies. Saby et al. (2003) and Coma et al. (2013) have suggested that longer treatment of sludge

under substrate- and oxygen-deficient conditions may lead to higher sludge reduction in OSA. However, they did not investigate the processes and reactions that were impacted by residence time. These are critically discussed in Section 3.3.

Sun et al. (2010) achieved a 30% enhancement in sludge reduction by increasing the sludge exchange frequency, while maintaining an SIR of 10%. The mechanism behind this trend was not elucidated. Nevertheless, it is possible that faster degradation of the returned sCOD occurred in the SBR at higher return rate (Sun et al., 2010). Further investigation of the combined effect of SIR and sludge interchange frequency is recommended, but that is beyond the scope of this study.

3.2 Impact of wastewater strength

3.2.1 Reactor performance

OSA performance was further investigated at higher feed sCOD by changing the feed provided to the SBRs from settled (sCOD = 60 ± 32 mg/L; n=48) to unsettled sewage (sCOD = 113 ± 32 mg/L; n=32) (Table 1 and Section 2.1). During this period, the OSA was operated at an SIR of 11% and without SIR (Supplementary Table S1). Changing the feed did not impact the wastewater treatment performance of the SBRs in that the effluent TOC, sCOD and ammonia concentrations of SBR_{control} and SBR_{OSA} were comparable (Figure 2 and Supplementary Figure S3) at both regimes. However, as noted in Section 3.2.2, this had implications on the extent of sludge yield reductions achieved.

3.2.2 Sludge reduction

The sludge yield of both SBR_{control} and SBR_{OSA} decreased when feed was changed from settled to unsettled sewage (Table 2). This is probably a response of microorganisms to the sudden change in wastewater characteristics (sCOD and composition). Nonetheless, at SIR of 11%, the

sludge yield of SBR_{OSA} decreased further to nearly zero and therefore the calculated sludge reduction was 100% (Table 2). This result suggests that OSA is most effective for treatment plants being fed with relatively high strength (*e.g.*, unsettled) sewage.

Higher influent sCOD concentration significantly increased the MLVSS/MLSS ratio of SBR_{OSA} from 0.53 ± 0.10 ($n=43$) to 0.66 ± 0.11 ($n=33$) (two sample *t*-test; $t(65)=4.15$, $p=1.99$, $\alpha=0.05$).

There is minimal study on the effect of MLVSS loading on OSA external reactors. However, a previous study also showed that higher MLVSS loading on the external reactors can reduce the MLVSS of the waste activated sludge (Semblante et al., 2015a). Higher MLVSS loading has been also reported to improve the performance of anaerobic digesters possibly by influencing the activity of hydrolysing bacterial groups (Mao et al., 2015).

To determine the performance of OSA at an SIR approaching zero, the interchange of sludge between SBR_{OSA} and external reactors was stopped (*i.e.*, there was no SIR). At this stage, SBR_{control} and SBR_{OSA} were essentially under the same operation conditions, and therefore they eventually exhibited a similar sludge yield (Table 2).

To determine the repeatability of the findings, SIR was resumed at 11% with unsettled sewage (Day 442-475). Sludge yield reduction by SBR_{OSA} was again evident, but it was lower than that achieved in the previous trial (Day 347-400). However, this can be attributed to the fact that sCOD concentration of unsettled sewage during Day 442-475 (74 ± 36 mg/L; $n=8$) was significantly lower compared to that at earlier periods (128 ± 96 mg/L; $n=24$), which reaffirms our recommendation of feeding higher strength sewage to OSA plants.

Unlike the anecdotal use of an SIR of around 10% in the previous studies (Chon et al., 2011; Novak et al., 2007), this study systematically studied the impact of SIR over a range of 0 – 22% and showed the greatest sludge yield reduction at an SIR of 11%. This study furthermore demonstrates that the sludge yield reduction can only be ascertained as a range (*e.g.*, between 30

and 100% sludge reduction) depending on wastewater strength, and the beneficial effect of OSA is derived better at higher feed strength as it leads to greater amount of volatile solids undergoing treatment in the external reactors.

Endogenous MLVSS production may occur in the aerobic digester and aerobic/anoxic and anoxic reactors when biomass consumes products of cell lysis (Hao et al., 2010). Therefore, the sludge yield of the control (combined SBR_{control} and aerobic digester) and OSA (combined SBR_{OSA} and external aerobic/anoxic and anoxic reactors) systems were also calculated (Supplementary Table S6). Similarly, the greatest sludge reduction was observed at SIR of 11% with either settled or unsettled sewage.

3.3 Analysis of sludge reduction mechanisms

To understand the impact of SIR on the sludge reduction mechanism of OSA, the volatile solids, organic, and nutrient concentrations in the aerobic/anoxic and anoxic reactors were analysed. Results show that an intermediate SIR (11%) promoted (a) volatile solids destruction in the anoxic reactor (evident in the release of ammonia and phosphate) and (b) nitrification/denitrification in the aerobic/anoxic reactor. This process ensures that destroyed volatile solids are converted into inert products.

3.3.1 Settled sewage

The external anoxic reactor was responsible for volatile solids destruction in OSA. The MLVSS/MLSS ratio of the external anoxic reactor (0.45 ± 0.13 , $n=37$) was generally lower than that of the aerobic/anoxic reactor (0.51 ± 0.10 , $n=37$) and SBR_{OSA} (0.52 ± 0.10 , $n=37$) when settled sewage was used as the feed (Figure 4). Volatile solids destruction was indicative of cell lysis. Saby et al. (2003) noted that cell lysis in OSA was greater when the external reactor was anoxic

(ORP < -150 mV) than when it was aerobic (ORP = 100 mV). In this study, volatile solids destruction at lower SIR was corroborated by the increase of orthophosphate concentration in the sludge supernatant. The increase in orthophosphate, a product of cell lysis, has also been observed or expected in OSA external reactors in previous studies (Goel and Noguera, 2006; Saby et al., 2003). Here, orthophosphate concentration in the external anoxic reactor was 1.75 times higher at the SIR of 11% (52.9 ± 21.5 mg/L; n=16) than at 22% (30.1 ± 2.2 mg/L; n=4) (Figure 5). Notably, volatile solids destruction and the increase in orthophosphate concentration was more evident when the MLVSS loading to the external reactors was high (*i.e.*, when the MLVSS/MLSS ratio of SBR_{OSA} increased as a result of feeding unsettled sewage), and therefore discussed in more detail in Section 3.3.2.

[Figure 4]

[Figure 5]

In addition to volatile solids destruction in the external anoxic reactor, results also revealed the occurrence of nitrification/denitrification in the external aerobic/anoxic reactor. It has been hypothesized that OSA reduces sludge by selecting certain enriched bacteria that readily consumes the biodegradable products of cell lysis. These bacteria convert lysed materials into inert forms (*e.g.*, H₂O, CO₂, and N₂) and consequently decrease organic load during a continuous cycle of sludge interchange (Semblante et al., 2014). According to Zhou et al. (2014), these bacteria possibly include slow-growing nitrifiers (*e.g.*, *Nitrospira*), which are enriched in OSA. In the current study, a relationship between SIR and nitrification has been observed: a net increase of ammonia occurred in the aerobic/anoxic reactor when SIR was 22% but not when it was 11% (Figure 5), indicating that the former condition did not favour nitrification. The low residence time of sludge (15 days) in the external aerobic/anoxic reactor at the SIR of 22% could be responsible for the lack of ammonia removal. Nitrification is generally improved by longer

sludge residence time. For instance, Li and Wu (2014) found that nitrifiers were enriched in SBRs when SRT was increased from 5-40 days. Likewise, Chuang et al. (1997) found that increasing SRT from 10 to 15 days enhanced ammonia removal in an activated sludge-biofilm reactor. More significant removal of ammonia in the external aerobic/anoxic reactor (62-74%) was observed when feed was unsettled sewage, and this is discussed in greater detail in Section 3.3.2.

The SIR of 11% and 22% both resulted in an accumulation of nitrate in the aerobic/anoxic reactor, indicating poor denitrification. This was due to insufficient COD. The theoretical COD/N ratio for biological denitrification is 3.74 (Chiu and Chung, 2003), but the actual COD/N loading ratio into the aerobic/anoxic reactor during this period (*i.e.*, when feed was settled sewage) was 1.49-1.70 only. The enhancement of denitrification efficiency at the SIR of 11% was clearly observed when the feed was unsettled sewage (Section 3.3.2).

3.3.2 Unsettled sewage

Similar to the observation when feed was settled sewage (Section 3.3.1), the average MLVSS/MLSS ratio of the external anoxic reactor was lower than that of the aerobic/anoxic reactor and SBR_{OSA} (Figure 4). This low MLVSS/MLSS ratio suggests that the external anoxic reactor was primarily responsible for volatile solids destruction in the OSA system. Furthermore, the levels of orthophosphate and ammonia in the external anoxic reactor increased by 1.5 times and 7-10 times, respectively (Figure 5). Notably, nutrients accumulated in the external anoxic reactor (Figure 5), but not COD (Supplementary Figure S7). This was because the COD released by volatile solids destruction was consumed during denitrification reaction. The findings here are different from that of Saby et al. (2003), which observed an accumulation of COD in the external

anoxic reactor. This is probably because the additional aerobic/anoxic reactor used in this study enriched denitrifying organisms, which consumed nitrate and COD (Figure 5).

The highest removals of ammonia (62-74%) and nitrate (17-21%) in the external aerobic/anoxic reactor were observed when SIR was 11% and the feed was unsettled sewage. In contrast, no removal but rather an accumulation of ammonia and nitrate occurred when SIR was 22% (Section 3.3.1). This confirms that lower SIR increased the residence time of sludge in the external aerobic/anoxic reactor and facilitated nitrification/denitrification (Ye et al., 2008), consequently converting by-products of volatile solids destruction into inert species (Semblante et al., 2014).

When low strength (settled) sewage was used as the feed, denitrification in the external aerobic/anoxic reactor was negligible at an SIR of 11% probably because of low COD/N loading ratio in the reactor (Section 3.3.1). The COD/N loading ratio during the experimental run with unsettled sludge (1.73-2.24) was higher than that of the previous run (1.49-1.70) due to lower nitrate concentration in the feed sludge (Figure 5). The increased availability of COD potentially contributed to the enhancement of denitrification in the aerobic/anoxic reactor during this period.

3.4 Role of sludge interchange in OSA

In the absence of SIR, there was no influence of the external reactors on the main bioreactor, and, hence, no sludge reduction in SBR_{OSA} . On the other hand, suspending the recirculation of sludge between SBR_{OSA} and the external reactors had minimal effect on volatile solids reduction in the external anoxic reactor (Figure 4) and nitrification in the aerobic/anoxic reactor (Figure 5). However, during this period, the concentration of nitrite increased by 5-10 times in the

aerobic/anoxic and anoxic reactors (Figure 5), which, according to Cortez et al. (Cortez et al., 2009), can be taken as an indicator of inefficient denitrification. This decline in denitrification efficiency in the external aerobic/anoxic reactor was possibly due to insufficient COD in absence of sludge interchange.

The relevance of interchanging sludge in the OSA process is further emphasized when OSA performance is compared with single-pass aerobic or anaerobic digesters. The MLVSS/MLSS ratio of the control aerobic digester (0.55 ± 0.08 , $n=37$ and 0.68 ± 0.08 , $n=31$ when feed was settled and unsettled sewage, respectively) was slightly higher than that of $SBR_{control}$ (0.52 ± 0.22 , $n=37$ and 0.69 ± 0.22 , $n=31$ when feed was settled and unsettled sewage, respectively), indicating that aerobic digestion was unable to induce significant volatile solids destruction (Supplementary Figure S8). Furthermore, when the circulation between SBR_{OSA} and the external reactors were disconnected (i.e., when there was no SIR and the external reactors virtually functioned as single-pass digesters), the sludge yield of SBR_{OSA} became comparable to that of $SBR_{control}$. The results of this study reinforce previous findings demonstrating that SBRs in OSA systems have lower sludge yield than SBRs attached to single-pass aerobic or anaerobic digesters (Chon et al., 2011; Ye et al., 2008). This suggests that in the absence of sludge interchange between the main bioreactor and external reactors, the mechanism responsible of reducing sludge yield in the main bioreactor is switched off.

4. Conclusion

An intermediate SIR (11%) increased sludge residence time in the external reactors and maximized OSA performance through two mechanisms: (a) providing optimum environment for volatile solids destruction as evidenced by the increase in orthophosphate under anoxic

conditions; (b) facilitating the conversion of lysed materials into inert forms as evidenced by the decrease in ammonia and nitrate under aerobic/anoxic conditions. SIRs over 11% showed lower OSA performance, whereas without SIR sludge reduction in the main bioreactor cannot take place. Better OSA performance occurred at higher volatile solids loading to the external reactors. Effluent quality and sludge settleability were unaffected by SIR.

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Figure 1. Schematic diagram of the control system composed of $SBR_{control}$ and a single-pass aerobic digester system (a) and the OSA system composed of SBR_{OSA} and external aerobic/anoxic and anoxic reactors (b). The SIR between the anoxic reactor and SBR_{OSA} (q_5) was adjusted to 0, 11, 16.5, or 22%. Consequently, the transfer rate of sludge from the anoxic reactor to the aerobic/anoxic reactor (q_4) was 33, 22, 16.5, or 11%, respectively.

Figure 2. sCOD (a), ammonia (b), and orthophosphate (c) concentrations in $SBR_{control}$ and SBR_{OSA} at different SIR (0-22%). The dotted line indicates the change of feed from settled to unsettled sewage. Days 0-151 served as the reactor acclimatization period.

Figure 3. Influent COD and MLVSS of $SBR_{control}$ and SBR_{OSA} at different SIR (0-22%). The dotted line indicates the change of feed from settled to unsettled sewage. Days 0-151 served as the reactor acclimatization period.

Figure 4. MLVSS/MLSS ratio of SBR_{OSA} and external aerobic/anoxic and anoxic reactors at different SIR (0-22%). The dotted line indicates the change of feed from settled to unsettled sewage. Days 0-151 served as the reactor acclimatization period.

Figure 5. Average phosphate (a), ammonia (b), nitrate (c), and nitrite (d) concentrations of the feed sludge, supernatant of the aerobic/anoxic reactor, and supernatant of the anoxic reactor at different SIR. Error bars indicate standard deviation of 'n' samples as follows: n=4 and 17 for SIR of 22% and 11%, respectively (settled sewage); n=12, 8 and 9, for SIR of 11, 0, and 11%, respectively (unsettled sewage). The dotted line indicates the change of feed from settled to unsettled sewage.

Figure 1

Figure 2

Figure 3

Figure 4

Figure 5

List of Tables

Table 1. Average properties of settled and unsettled sewage

Property	Settled sewage	Unsettled sewage
sCOD (mg/L)	60±32 (n=48)	113±87 (n=33)
TOC (mg/L)	50.6±21.9 (n=48)	49.8±24.2 (n=33)
NH ₃ -N (mg/L)	31.2±7.5 (n=48)	68.1±31.7 (n=33)
PO ₄ ⁻³ -P (mg/L)	26.0±12.0 (n=48)	46.7±48.2 (n=33)
pH	5.9 (n=42)	6.9 (n=32)
TSS (g/L)	0.60±0.12 (n=48)	0.67±0.08 (n=33)
VSS (g/L)	0.17±0.09 (n=48)	0.19±0.07 (n=33)
VSS/TSS	0.28 (n=48)	0.28 (n=48)

Table 2. Sludge yield (Y_{obs}) of SBR_{control} and SBR_{OSA} at different operation conditions (arranged chronologically). Days 0-151 served as the reactor acclimatization period.

Time (day)	Feed type	Feed sCOD concentration (mg/L)	SIR of OSA (%)	Sludge yield (Y_{obs})				
				SBR _{control}	R ²	SBR _{OSA}	R ²	Reduction (%)
152-195	Settled sewage	60±43 (n=11)	16.5	10.54	0.85	7.87	0.92	25
196-274	Settled sewage	58±40 (n=20)	22	3.50	0.85	4.01	0.93	None
275-346	Settled sewage	59±18 (n=18)	11	1.54	0.93	0.73	0.78	53
347-400	Unsettled sewage	105±68 (n=14)	11	0.50	0.60	0.00	0.85	100 ^b
401-441	Unsettled sewage	162±121 (n=10)	0	0.14	0.70	0.14	0.78	None
442-475	Unsettled sewage	74±36 (n=9) ^a	11	1.96	0.93	1.40	0.93	29

^a sCOD of the unsettled sludge during this run was comparable to that of settled sludge at earlier periods because of wet weather

^b No excess sludge yield

Biosolids reduction by the oxic-settling-anoxic process: Impact of sludge interchange rate

Supplementary data

Bioresource Technology

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Table S1. Summary of operation conditions

Time (day)	Feed	SIR (%)
0-151	Settled sewage	Stabilisation period
152-195	Settled sewage	16.5
196-274	Settled sewage	22
275-346	Settled sewage	11
347-400	Unsettled sewage	11
401-441	Unsettled sewage	None (sludge interchange between SBR _{OSA} and external reactors is suspended)
442-475	Unsettled sewage	11

The MLVSS produced by $SBR_{control}$ ($P_{SBR_{control}}$) and SBR_{OSA} ($P_{SBR_{OSA}}$) at a given time interval were quantified using a mass balance of biomass and shown in Equation S.1 and S.2, respectively:

$$P_{SBR_{control}} = \Delta MLVSS_{SBR_{control}} \times V_{SBR_{control}} \times (VSS_{e1} \times X_{out} + MLVSS_{SBR_{control}} \times W - VSS_i \times X_{in}) \times \Delta t \quad \text{Equation S.1}$$

$$P_{SBR_{PSA}} = \Delta MLVSS_{SBR_{PSA}} \times V_{SBR_{PSA}} + (VSS_{e2} \times X_{out} + MLVSS_{SBR_{PSA}} \times W - VSS_i \times X_{in} - MLVSS_{ANX} \times q_5) \times \Delta t \quad \text{Equation S.2}$$

wherein $MLVSS_{SBR_{control}}$, SBR_{OSA} or ANX is the biomass concentration of the SBRs and the anoxic reactor, $V_{SBR_{control}}$ or SBR_{OSA} is the effective reactor volume, VSS_i is the volatile suspended solids concentration of the influent, VSS_{e1} or $e2$ is volatile suspended solids concentration of the effluent, X_{in} or out is flow rate of the influent or effluent, W is the flow rate of sludge wasted from the SBRs, q_5 is the flow rate of sludge returned from the anoxic reactor to SBR_{OSA} (Section 2.2), and t is time. Notably, VSS_i is deducted from the calculation of $P_{SBR_{control}}$ and $P_{SBR_{OSA}}$ to discount the significant amount of volatile solids carried by real wastewater (e.g. 0.1-0.5 g/L), and $MLVSS_{ANX}$ is deducted from the calculation of $P_{SBR_{OSA}}$ to discount the biomass that was only recycled back to SBR_{OSA} from the external anoxic reactor.

The Y_{obs} of the control and OSA systems were determined. The synthesis of cells in the aerobic digester and OSA may occur even under limited substrate conditions when microorganisms consume products of cell lysis, so those reactors may also contribute to MLVSS production of the whole system. The MLVSS production of the control ($P_{control}$) and OSA (P_{OSA}) systems were calculated using Equations S.3 and S.4, respectively:

$$P_{control} = \Delta MLVSS_{SBR_{control}} \times V_{SBR_{control}} + \Delta MLVSS_{AE} \times V_{AE} + (VSS_{e1} \times X_{out} + MLVSS_{AE} \times Q_{out} - VSS_i \times X_{in}) \times \Delta t \quad \text{Equation S.3}$$

$$P_{OSA} = \Delta MLVSS_{SBR_{OSA}} \times V_{SBR_{OSA}} + \Delta MLVSS_{AE/ANX} \times V_{AE/ANX} + \Delta MLVSS_{ANX} \times V_{ANX} + (VSS_{e2} \times X_{out} + MLVSS_{AE/ANX} \times q_3 - VSS_i \times X_{in}) \times \Delta t \quad \text{Equation S.4}$$

wherein $MLVSS_{AE/ANX}$ or ANX is the biomass concentration of the aerobic/anoxic and anoxic reactors, V_{AE} , AE/ANX or ANX is the effective digester or reactor volume, Q_{out} is the flow rate of sludge wasted from the aerobic digester, and q_3 is flow rate of sludge wasted from the aerobic/anoxic reactor. Notably, the sludge interchanged within the external reactors and between SBR_{OSA} and external reactors were retained in the system hence it is not necessary to deduct those sludge flows from the calculation of P_{OSA} .

The amount of substrate consumed C by individual SBRs, and the control and OSA systems was calculated according to Equation S.5:

$$C = (COD_i - COD_{e1/e2}) \times X_{in} \quad \text{Equation S.5}$$

wherein COD_i and $COD_{e1/e2}$ are the soluble COD of the influent and effluent, respectively.

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597 **Figure S3.** TOC concentration and removal efficiency of $SBR_{control}$ and SBR_{OSA} at different
 598 sludge interchange rates (0-22%). The dotted line indicates the change of feed from settled to
 599 unsettled sewage. Days 0-151 served as the reactor acclimatization period.

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Figure S4. SVI of SBR_{control} and SBR_{OSA} at different sludge interchange rates (0-22%). The dotted line indicates the change of feed from settled to unsettled sewage. Days 0-151 served as the reactor acclimatization period.

Figure S5. Sludge yield of SBR_{control} and SBR_{OSA} at different sludge interchange rates (0-22%) with settled sewage (a) and unsettled sewage (b) as feed.

Table S6. Sludge yield (Y_{obs}) of control (*e.g.*, combined $SBR_{control}$ and aerobic digester) and OSA (*e.g.*, combined SBR_{OSA} and external aerobic/anoxic and anoxic reactors) systems at different operation conditions

Feed	SIR of OSA (%)	Y_{obs} (g MLVSS/g COD)			
		Control system	R^2	OSA	R^2
Settled sewage	16.5	8.75	0.87	6.69	0.96
Settled sewage	22	0.96	0.65	1.95	0.57
Settled sewage	11	1.23	0.88	~0	-
Unsettled sewage	11	1.06	0.75	-0.58	0.92
Unsettled sewage	11	1.92	0.90	1.31	0.92

Figure S7. COD and TOC concentrations in the aerobic/anoxic and anoxic reactors. Feed sludge is the combined mixed liquors from SBR_{OSA} and anoxic reactor that was fed to the aerobic/anoxic reactor. The dotted line indicates the change of feed from settled to unsettled sewage.

Figure S8. MLVSS/MLSS ratio of SBR_{control} and the aerobic digester. The dotted line indicates the change of feed from settled to unsettled sewage. Days 0-151 served as the reactor acclimatization period.