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CULTIVATION AND CHARACTERISTICS OF AEROBIC GRANULAR SLUDGE FOR SIMULTANEOUS ORGANICS AND NUTRIENTS REMOVAL PERFORMANCES AT HIGH TEMPERATURE

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Abstract: With inoculum sludge from a conventional activated sludge wastewater treatment plant, a sequencing batch reactor fed with synthetic wastewater was operated at 50 ± 1 °C to study the formation of aerobic granular sludge (AGS) for simultaneous organics and nutrients removal with a complete cycle time of 3 h. The AGS were successfully cultivated with excellent settling ability and demonstrated exceptional performance in the organics and nutrients removal with influent loading rate and COD/N ratio of 1.6 kg COD $(L.d)^{-1}$ and 8.3, respectively. Stable, regular, dense and fast settling granule (average diameter, 2.0 mm and settling velocity, 26 m h^{-1}) were developed in a single reactor. In addition, 76% COD removal efficiency was observed in the system at the maturation stage of the granulation, while its ammonia and total phosphorus removal efficiencies were up to 88% and 69%, respectively. The study demonstrated the capabilities of AGS formation in a single, high and slender column type-bioreactor at high temperature which is suitable to be applied for hot climate and low humidity condition (e.g. Saudi Arabia).

Keywords: *Aerobic granular sludge; Sequencing batch reactor; High temperature*

1.0 Introduction

Treatment of domestic wastewater in urban areas is one of the essential aspects to be count in the development of a country in order to sustain individual's health and welfare. Wastewater that is not being treated can cause the spread of disease in the form of several types of endemic and epidemic illnesse (Al-Rehaili, 1997). There are many methods to treat wastewater ranging from modest, low priced, and less efficient processes to very advanced, highly efficient and pricey operations. The selection among these processes should consider local conditions such as weather, social characteristics, economy, availability of enforceable standards, availability of land and power, required

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operation skills and its availability, monitoring activities, effluent disposal options, effluent reuse practices and requirements (Al-Rehaili, 1997). Some of the most regularly used wastewater treatment technologies are activated sludge process (ASP), sequencing batch reactor (SBR), up-flow anaerobic sludge blanket reactors followed by facultative aerobic lagoon (UASB–FAL) and constructed wetlands (CWs) (Kalbar et al., 2012).

Aerobic granular sludge (AGS) has been widely studied in these recent years. AGS is a dense community made up of symbiotic organisms, possessing good biological activity and excellent mass transfer efficiency. Aerobic granular sludge-based reactors represent an attractive alternative to conventional activated sludge systems due to their small footprint and low excess sludge production (de Bruin et al., 2004). The sludge developed in such systems has high biomass concentration, good settling properties, high chemical oxygen demand (COD) removal efficiency, and high phosphorus removal capacity (de Kreuk et al., 2005a). In addition, simultaneous nitrification–denitrification can occur in granules influenced by the bulk oxygen concentration and granule size (Beun et al. 2001; Mosquera-Corral et al. 2005). Aerobic granular sludge has mainly been developed in sequencing batch reactor (SBR) systems, some using airlift or bubble column reactors. Several laboratory scale studies were extensively carried out to identify the most important factors influencing the aerobic granular sludge formation such as organic loading rate, settling time, hydrodynamic shear force and substrate composition (Adav et al., 2008). However, the cultivation of aerobic granular sludge is a complicated ecological process, in which many factors need to be further explored.

In most studies on aerobic granular sludge, SBRs (AGS-SBR) have been operated at ambient temperature, e.g., 20–25 °C (Morgenroth et al., 1997; de Kreuk and van Loosdrecht, 2004; Whang and Park, 2006) or lower (de Kreuk et al., 2005b). Because detailed information of the high temperature effects on aerobic granulation is limited, it becomes main purpose of the present study to investigate the granulation process, stability, density and performances of aerobic granules at 50 °C.

In this article, aerobic granulation was cultivated in SBR. The morphology of granular sludge, their settling properties and treatment efficiencies were also discussed. The aim of this study is to enrich the knowledge of cultivation procedure, and to encourage the application of aerobic granular sludge in wastewater treatment.

2. Materials and methods

2.1 Experimental procedure and rector operation

Experiments was carried out in a cylindrical column bioreactor (internal diameter of 6.5 cm and total height of 100 cm) with a working volume of 3 L. During the start-up period, 1500 mL of activated sludge from a municipal sewage treatment plan was added into the bioreactor system as inoculums in the bioreactor. The bioreactor was operated in SBR mode at a cycle of 3 h: 5 min of feeding, 40 min of circulation, 110 min of aeration, 15 min of settling, 5 min of effluent withdrawal and 5 min idling. A set of two peristaltic pumps were used to feed and to discharge the wastewater in the reactor system. During the filling phase, wastewater was introduced through ports located at the bottom of the bioreactor. While fine air bubbles for aeration were supplied by means of air bubble diffusers were placed at the bottom. The effluent was withdrawn through the outlet ports positioned at medium height in the column bioreactor which had a volumetric exchange ratio of 50%. The sludge retention time was set by the discharge of suspended solids with the effluent. The bioreactor was operated at the temperature of 50 ± 1 °C, using water bath sleeves and a thermostat.

Measurements of the parameters such as mixed liquor suspended solid (MLSS), mixed liquor volatile suspended solid (MLVSS), COD, NH_3-N and total phosphorus (TP) were carried out according to Standard Methods for the Examination of Water and Wastewater (APHA, 2005).

2.2 Synthetic wastewater characteristics and seed sludge

The reactors were fed with the same medium as the one used by de Kreuk et al. (2005). It was prepared as two stock solutions that were mixed with distilled water prior to feeding. Solution A contained sodium acetate (63 mM), $MgSO_4 \cdot 7H2O$ (3.6 mM) and KCl (4.8 mM). Solution B contained NH_4Cl (35.4 mM), K_2HPO4 (4.2 mM), KH_2PO4 (2.1 mM), and 10 mL L^{-1} trace element solution. The trace element stock solution contained: EDTA 342.2 mmol L^{-1}, $ZnSO_4 \cdot 7H_2O$ 15.3 mmol L^{-1}, $CaCl_2 \cdot 2H_2O$ 111.3 mmol L^{-1}, $MnCl_2 \cdot 4H_2O$ 51.1 mmol L^{-1}, $FeSO_4 \cdot 7H_2O$ 35.9 mmol L^{-1}, $Na_2Mo_7O_{24} \cdot 2H_2O$ 2.7 mmol L^{-1}, $CuSO_4 \cdot 5H_2O$ 12.6 mmol L^{-1}, and $CoCl_2 \cdot 6H_2O$ 13.5 mmol L^{-1}. Per cycle, 150 mL of both solutions were added to the reactor together with 1200 mL of tap water.

Activated sludge was used as inoculums, which were taken from Al Madinah Sewage Treatment Plant in Saudi Arabia. The amount of inoculum was about 1.5 L, with a mixed liquor suspended solid (MLSS) concentration 13.7 g L^{-1} and a mixed liquor volatile suspended solid (MLVSS) concentration 9 g L^{-1}. The seed sludge appeared fluffy, irregular and loose structure with brown color.

3. Results and discussion

3.1 Aerobic granular sludge formation and morphology observation

During the early stage, seed sludge appeared as a fluffy, irregular, loose-structure morphology and rich with filamentous organisms under microscopic examination as shown in Figure 1. The sludge color progressively turned from dark brown to yellowish brown at the final stage of the experimental period. In the initial stage of granulation, the loose flocs were easily broke up into small pieces if placed under vigorous shaking.

After few days, the flocs-like sludge started to disappear and changed to small granules with average diameter 0.9 mm. Under high shear force, the flocs became denser causing the aggregation of the biomass to secrete more exopolysaccharides (EPS) (Dulekgurgen et al., 2008). Interactions between inter-particle bridging process among EPS, microbial cells and ion contributes to the evolution of seed sludge from flocs to granules (Sheng et al., 2010). EPS can promote the cell hydrophobicity and change the surface charges on the microorganisms (Zhu et al., 2012), which can increase microbial cell adhesion and granulation.

In the following weeks, the small granules became more regular in shape and progressively increased in size, while more flocculent sludge washed out from the bioreactor, resulting in the accumulation of the aerobic granules with high settling velocity. Finally, mature granules formed after 30 days of inoculation with average diameter is 2.0 mm. The compact mature granules were smooth with a solid surface and contributes to a stable operation of the bioreactor.

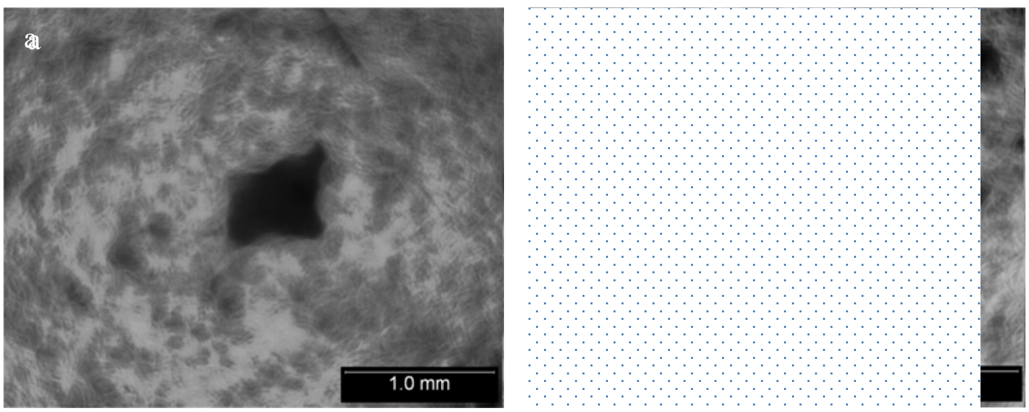
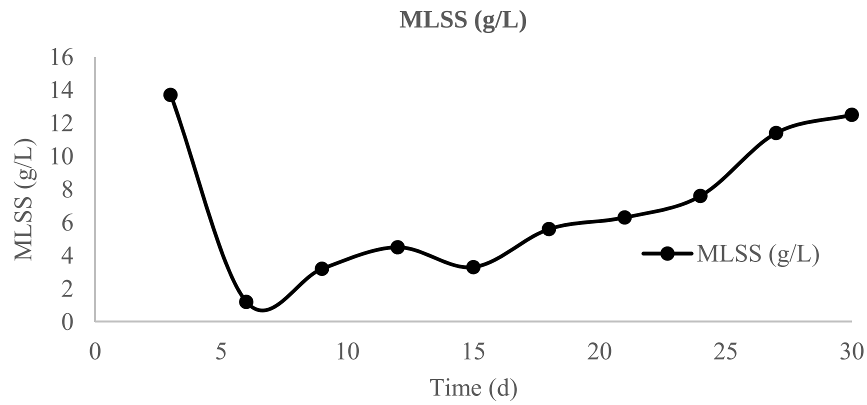


Figure 1: Images of granules at 50°C after startup at (a) 15 d and (b) 30 d

3.2 Biomass profile of granules

Figure 2 displays the MLSS and MLVSS variation in the SBR system from the first day until day-30 of the experiment. During the initial stage of the experiment, most of the sludge was washed-out from the bioreactor leading a significant decline in the biomass concentration and increased the effluent solids concentration. Figure 2 also reveals a rapid decrease of MLSS from 13.7 g L^{-1} to 1.2 g L^{-1} in the first 6 days, probably due to the short settling time applied in the cycle system. During the days between 9 and 24, both MLSS and MLVSS concentration kept rising but with occasional decrease perhaps due to some microorganisms in the bioreactor were adapting themselves to degrade non-active microorganisms and the seed microorganisms were to adapt to the synthetic wastewater. The concentration of the biomass was improving as the small granules started to appear in the bioreactor on the day-15. Subsequently, both MLSS and MLVSS concentrations increased uniformly and achieved steady state at about 12.5 g L^{-1} and 10.5 g L^{-1}, respectively during days 30. The same trend was also spotted for the MLVSS content which ranging from initial concentration of 9 g L^{-1} to 10 g L^{-1} at the end of the experiment. The MLVSS to MLSS ratio is about 0.84 and a stable condition of biomass concentration indicates a good accumulation of biomass in the bioreactor.



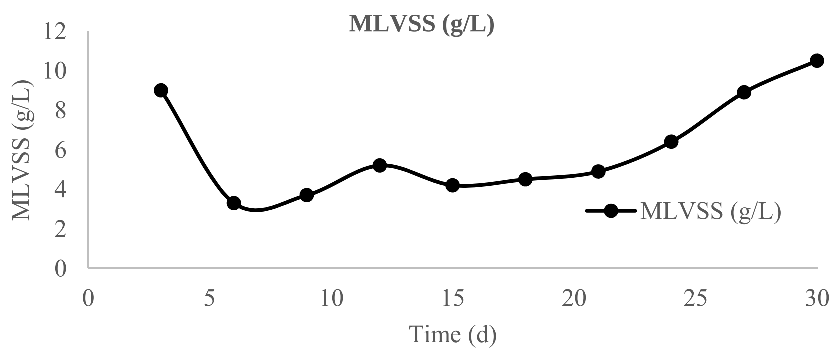


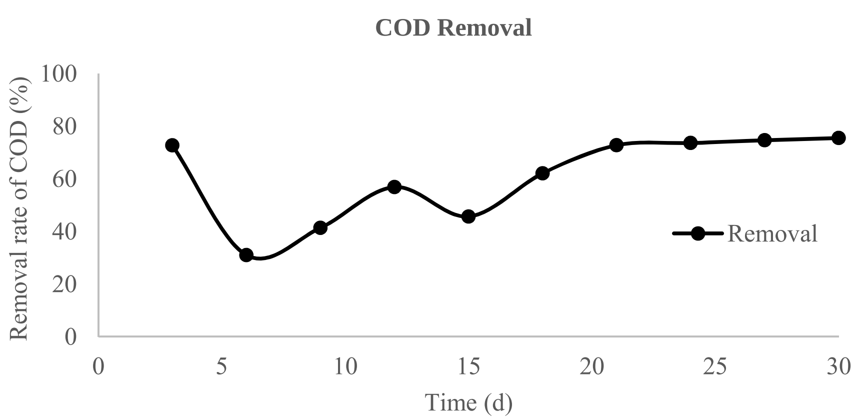
Figure 2: Variation of biomass concentrations and sludge volume index in SBR within 30 days for (a) MLSS concentration and (b) MLVSS concentration

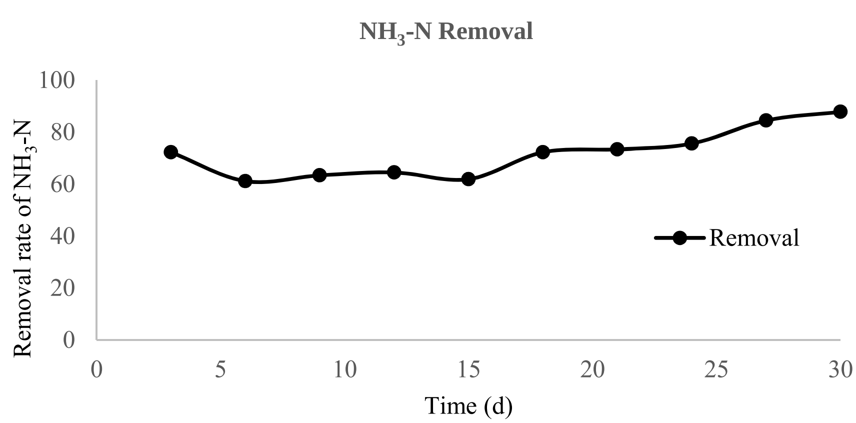
3.3 Organics and Nutrients Removal Efficiencies of granules

Figure 3 illustrates the removal rate of organics and nutrients in the SBR system with respect to COD, ammonia nitrogen and total nitrogen from beginning until the end of granules development period. At the beginning of the bioreactor operation, the removal rate of COD dropped from 72% to 31%, probably due to the adapting process of the sludge with synthetic wastewater. In the first 18 days, the removal rate of COD kept increasing but with sporadic decrease. Thereafter, the COD removal efficiency improved uniformly and became stable for the remaining period. When the granular sludge started to evolve from flocculent sludge in the bioreactor system, it enhanced the degradation ability for COD removal efficiency up to 76%, which is comparable to previous work done by Abdullah et al. (2011) in treating palm oil mill effluent (POME) and Rosman et al. (2013) in treating rubber wastewater using aerobic granular sludge.

This result indicates the high biological activity occurred during microbial aerobic degradation process of synthetic wastewater.

The ammonia removal efficiency was 72% at the beginning of the bioreactor operation and then dropped significantly to 61%. Subsequently, the removal rate of ammonia improved when granules started to form. Figure 3 reveals the removal efficiencies for ammonia and total phosphorus kept increasing but with occasional decrease. Afterwards, the removal efficiencies for ammonia and total phosphorus increased gradually and achieved steady state at about 88% and 69%, respectively during days 30. The ammonia concentrations in the effluent shows a significant better quality and maintained below 10 mg L^{-1} upon the formation of aerobic granular sludge which indicates an effective ammonia removal efficiency. Nitrifying bacteria population within the aerobic granules became predominant after the biodegradation of organics which help in nitrification process. Belmonte et al. (2009) stated that nitrification process could be improved by promoting the development of granules that enhance the retention of large amounts of nitrifying bacteria in the bioreactor system leading a higher removal efficiency for ammonia of above than 94%. The sufficient oxygen level supplied in the bioreactor system enabled a good oxidation for ammonia and more than 80% of ammonia being removed during the aerobic reaction phase which shows a stable and excellent nitrification process happened in the system.





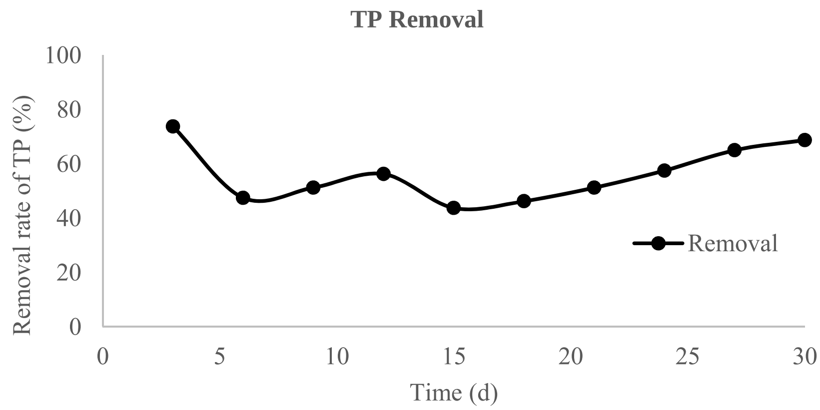


Figure 3: Profile of removal performances in the SBR system within 90 days for (a) COD, (b) ammonia nitrogen and (c) TP.

4. Conclusions

Compact and stable aerobic granular sludge with an excellent settling ability and an average diameter size of 2.0 mm were successfully developed in an SBR system fed with synthetic wastewater. A good COD removal rate of 76% was achieved after 30 days of operation with influent loading rate and COD/N ratio of 1.6 kg COD $(L.d)^{-1}$ and 8.3, respectively. Furthermore, the removal rate of ammonia was 88% while total phosphorus was 69% were observed in the single bioreactor system in treating the synthetic wastewater. Therefore, the study presented herein suggested feasibility of the developed aerobic granular sludge for the treatment of domestic wastewater at high temperature of 50°C which is suitable to be applied for hot climate and low humidity condition (e.g. Saudi Arabia).

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