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Characteristics of aerobic granulation at mesophilic temperatures in wastewater treatment

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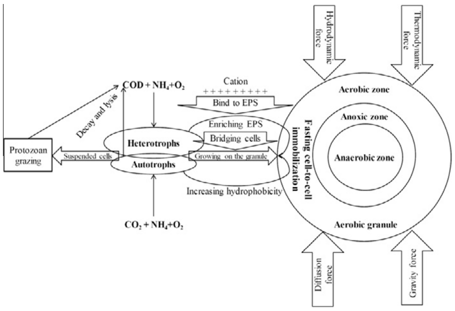
HIGHLIGHTS

- Aerobic granulation was carried out at mesophilic temperatures (35 °C).
- Significant organic and ammonia were simultaneously removed.
- Diversified species could be developed through mesophilic aerobic granulation.
- The growth kinetics of heterotrophs and autotrophs were estimated.
- Biochemical and physical interactivities create a protective granule's shell.

GRAPHICAL ABSTRACT



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ABSTRACT

Compact and structurally stable aerobic granules were developed in a sequencing batch reactor (SBR) at mesophilic temperatures (35 °C). The morphological, biological and chemical characteristics of the aerobic granulation were investigated and a theoretical granulation mechanism was proposed according to the results of the investigation. The mature aerobic granules had compact structure, small size (mean diameter of 0.24 mm), excellent settleability and diverse microbial structures, and were effective for the removal of organics and nitrification. The growth kinetics demonstrated that the biomass growth depended on coexistence and interactions between heterotrophs and autotrophs in the granules. The functions of heterotrophs and autotrophs created a compact and secure layer on the outside of the granules, protecting the inside sludge containing environmentally sensitive and slow growing microorganisms. The mechanism and the reactor performance may promise feasibility and efficiency for treating industry effluents at mesophilic temperatures using aerobic granulation.

1. Introduction

Aerobic granulation is a process of microbial self-immobilization without the support of a carrier to form a multicellular association and compact structure (Beun et al., 1999; de Kreuk et al., 2005). The formation of granules in an aerobic sequencing batch reactor (SBR) has been extensively studied for high strength wastewater treatment (Adav et al., 2008; Abdullah et al., 2013). Wastewaters that require biological treatment at a high strength, like pulp and paper effluents, many food processing effluents and anaerobic digested effluents, are often discharged at high temperatures (Pokhrel and Viraraghavan, 2004; Lefebvre and Moletta, 2006; Cui et al., 2011). The aerobic granulation appears to be a promising technology to treat these types of wastewaters, but has never been tested for simultaneous organics removal and nitrification at mesophilic temperatures (35 °C).

Temperature can play a vital role in bacteria growth by influencing the rates of enzymatically catalyzed reactions and by affecting the rate of diffusion of substrate to the cells (Feller and Gerday, 2003). It has been reported that the applications of aerobic granulation at a thermophilic temperature (55 °C) have potential advantages over other applications, including low waste biomass

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production, higher degradation rates, elimination of cooling requirements for high temperature wastes, enhanced solubility and degradation of low-solubility substrates and rapid inactivation of pathogens (Zitomer et al., 2007). Despite this, thermophilic aerobic granulation may not be a practical approach due to the poor solubility of oxygen at such high temperatures. Detailed information of the mesophilic temperatures (30–40 °C) effect on aerobic granulation is limited. Furthermore, the formation of aerobic granules is a complicated ecological process, in which many factors need to be further investigated.

The aerobic growth of heterotrophs and autotrophs could coexist and interact in aerobic granules (Yang et al., 2004). The heterotrophs are responsible for the oxidation of biodegradable organic carbon, whereas the autotrophs are responsible for the oxidation of ammonia to nitrite and nitrate through nitrification. In a conceptual microbial structure of the granules, a granule could simultaneously contain heterotrophs on the outside and autotrophs in the middle (Ni et al., 2008). In general, the aerobic granulation was carried out with the presence of high strength organic matters that creates competition between the autotrophs and heterotrophs for DO (Liu et al., 2003). The interspecies competition could result in a decrease in nitrification efficiency because the autotrophic growth rate is much lower than the heterotrophic growth rate (Nogueira et al., 2002). At mesophilic temperatures, the activated sludge appeared to have a negative impact on the performance of organics removal, whereas the nitrification proceeded better (Fdz-Polanco et al., 1994; Vogelaar et al., 2002). Therefore, the understanding of the growth and activity of autotrophs and heterotrophs in aerobic granules is important for optimizing the performance of wastewater treatment.

This study demonstrated that a mesophilic (35 °C) aerobic granular biomass could be developed using the synthetic organic and ammonium wastewater. The process of mesophilic aerobic granulation was evaluated to understand the morphological, biological and chemical characteristics of granules. The roles of heterotrophs and autotrophs at mesophilic temperature were discussed based on experimental results and kinetic analysis. Furthermore, the theoretical mechanism for the formation of aerobic granules at a mesophilic temperature was discussed.

2. Methods

2.1. Operation of reactor

The laboratory SBR was a cylindrical acrylic glass vessel (100 cm in height and 6 cm in diameter), with a working volume of 2.85 L. Air was introduced through the bottom by a fine bubble aerator at 3 L min^{-1}. The temperature was controlled at around 35–37 °C by a water jacket. The reactor was operated in a successive cycle of 6 h, including 6 min of influent filling, 320–330 min of aeration, 20–30 min of settling and 4 min of effluent discharge. The minimum DO concentration detected in the reactor was above 2 mg L^{-1}, while the pH fell in the range of 7.5–8.5. It was operated more than 70 days without excess sludge discharge; hence the effluent was the only passage for biomass wasting.

2.2. Media

The reactor was inoculated with activated sludge taken from an aeration tank of a wastewater treatment plant in Ansan City, Korea. The amount of inoculum was about 2.5 L, with a mixed liquor suspended solids (MLSS) concentration of 2500 mg L^{-1}. Synthetic wastewater prepared with tap water was used with glucose as a carbon source (450 ± 24 mg L^{-1} as COD). The chemical addition to the synthetic wastewater included $(NH_4)_2SO_4$ (150 ± 25 mg L^{-1} as NH_4–N), KH_2PO_4 (70 mg L^{-1}), $MgSO_4$ (20 mg L^{-1}), $CaCl_2$ (100 mg L^{-1}), trace solution 1 (1 mg L^{-1}) and trace solution 2 (1 mg L^{-1}). Trace solution 1 contained (per liter deionized water) ethylenediaminetetraacetic acid (EDTA) (5 g) and $FeSO_4{\cdot}7H_2O$ (9.144 g). Trace solution 2 contained (per liter deionized water) EDTA (15 g), $ZnSO_4{\cdot}7H_2O$ (0.43 g), $CoCl_2{\cdot}6H_2O$ (0.24 g), $MnCl_2{\cdot}H_2O$ (0.66 g), $CuSO_4{\cdot}5H_2O$ (0.25 g), $NaMoO_4{\cdot}2H_2O$ (0.22 g), $NiCl_2{\cdot}6H_2O$ (0.19 g), $Na_2SeO_4{\cdot}H_2O$ (0.21 g) and H_3BO_4 (0.014 g).

2.3. Analytical methods

Chemical oxygen demand (COD), MLSS, mixed liquor volatile suspended solids (MLVSS) and sludge volume index (SVI) were determined according to Standard Methods (APHA, 2005). NH_4–N was measured using a spectrophotometer (DR/2500, Method 8038, Nessler Method, Hach Co., USA). NO_2–N and NO_3–N were measured using an ion chromatography (790 Personal IC, Metrohm Ltd., Switzerland). The samples of effluent water were filtered by a 0.45 μm nylon syringe filter (Whatman International Ltd.) for the analysis of COD, NH_4–N, NO_2–N and NO_3–N. Particle size distribution was analyzed by an electrophoretic light scattering system (Zeta-potential & Particle Size Analyzer ELSZ-2, Otsuka Electronic Co., Ltd., Japan). According to the methods proposed by Avcioglu et al. the oxygen uptake rate (OUR) was calculated by monitoring DO concentration (Avcioglu et al., 2003). The microstructure of mature granules was examined with a scanning electron microscope (SEM) (MIRA3, TESCAN Inc., USA). The chemical compositions of sludge were analyzed by an energy dispersive spectrometry system for the SEM (TEAMTM EDS, EDAX Inc., USA).

2.4. Determination of heterotrophs and autotrophs

The biomass was removed from the SBR and placed into the batch reactors where it was aerated more than a week. During this period, oxygen uptake rate (OUR) and nitrate production rate (NPR) were periodically tested for determining the active biomass of heterotrophs (X_H, mg L^{-1}) and autotrophs (X_A, mg L^{-1}). In the OUR tests 20 mg L^{-1} of thiourea was added to inhibit nitrification. The values of heterotrophic decay (b_H, d^{-1}) and autotrophic decay (b_A, d^{-1}) are obtained from the data on the change in OUR and NPR over time.

$$\ln \mathrm{OUR} = \ln(f_d \cdot b_H \cdot X_H) - b_H \cdot t \tag{1}$$

$$-\ln \mathrm{NPR} = \ln(f_d \cdot b_A \cdot X_A) - b_A \cdot t \tag{2}$$

where f_d is the biodegradable fraction of 0.8 (McCarty, 1975).

The initial concentrations of active biomass in the batch reactors are determined by using the baseline endogenous OUR and NPR. Therefore, the initial X_H and X_A represent the active biomass of heterotrophs and autotrophs when the sludge is taken out of the SBR.

$$\mathrm{OUR}_{\mathrm{Initial}} = f_d \cdot b_H \cdot X_{H,\mathrm{Initial}} \tag{3}$$

$$\mathrm{NPR}_{\mathrm{Initial}} = -f_d \cdot b_A \cdot X_{A,\mathrm{Initial}} \tag{4}$$

2.5. Estimation of growth kinetics

The Gompertz (Eq. (5)) was adopted to describe the biomass growth of the aerobic granulation. The Gompertz model is regarded as one of the most appropriate models to describe bacteria growth data (Zwietering et al., 1990).

$$X_{H,A} = A \cdot \exp(-\exp(R_{\max} \cdot e \cdot (\lambda - t)/A + 1) \tag{5}$$