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Aerobic Granules in Lab-Scale Bioreactor: A Focus on Size Distribution and Settling Velocity Properties

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ABSTRACT

Biogranulation technology using aerobic granules is one of the most important discoveries in the field of wastewater treatment. This advanced technology is better than conventional wastewater treatment method because it is cost-effective and space-reduced. The evolution of aerobic granules from seed sludge to granules in sequencing batch reactor system was studied in this study. The purposes of this study are to measure the size of aerobic granules over time and to determine the settling velocity of the aerobic granules against time. The sequencing batch reactor system was operated in four processes involved feeding, aeration, settling and withdraw. The bioreactor was inoculated with seed sludge sample taken from wastewater treatment plant in Arau, Perlis and run at room temperature. Synthetic wastewater containing acetate and other elements were used to feed the biomass in the bioreactor system. The aerobic granules started to be detected in the sequencing batch reactor after two weeks of operation. The aerobic granules in size categories (>0.2<0.4, >0.4<0.6 and >0.6 mm) had increased over the time. An increase of settling velocity (29.8 m/h) was observed in this present study. These findings demonstrate that the increase of aerobic granules size and settling velocity were important in determining the evolution of aerobic granules study.

Keywords: *bioreactor; aerobic granules; granules size; settling velocity; biogranulation*

1. INTRODUCTION

Biogranulation technology especially using anaerobic process has been commercialized widely in treating various types of wastewater. However, aerobic granulation technology is the latest technique discovered for treating wastewater. This technique involved the role of microorganisms in the granules form to treat varieties of wastewater such as industrial and pharmaceutical [1-4]. This technology using microorganisms to treat pollutant in wastewater. Microorganisms have a high potential to treat pollutant in liquid and solid medium [5].

The aerobic granulation process owns some good benefits, for instance, the increase of settling velocity and thicker biomass structure as compared to activated sludge process in treating wastewater [6]. The granules started to form when microorganisms attach to each other and become immobile. The aggregation of microorganisms were caused by the certain conditions such as food source and reactor operation. The aerobic granules consist of various type of bacteria species due to different zone exist in one granule such as aerobic and anoxic zones. In addition, high removal effeciency of nutrients and organic chemicals in wastewater

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was due to the existence of various types of bacteria in aerobic granules such as *Pseudomonas sp*. and *Bacillus sp*. [7].

In contrast, there are some negative effects of anaerobic granulation. Anaerobic granulation technology is inappropriate to remove nutrients from wastewater, needs a high temperature during the operation period and requires an elongated start-up phase [8].

To make it work, aerobic granules is the most important element to be considered. Therefore, the aim of this study is to demonstrate the evolution of aerobic granules. There are three objectives involved in this study: (i) to measure the percentage of size of aerobic granules over time. (ii) to observe the settling velocity of the aerobic granules against time and (iii) to investigate the relationship between the size of aerobic granules and settling velocity.

2. METHODOLOGY

2.1 Bioreactor set-up

The bioreactor column with 2500 mL working volume was fabricated for this experiment. A cylindrical shape column was made from acrylic material with 10 cm diameter and 35 cm in height was used to set-up the bioreactor in the laboratory. All appliances were connected to the reactor by tubing. Two sets of peristaltic pumps were needed to pump the influent and effluent of this bioreactor system. The bioreactor consists of five stopcocks as four sampling points for the outlet (a, b, c, and d) and one for the inlet (e) as depicted in Figure 1.

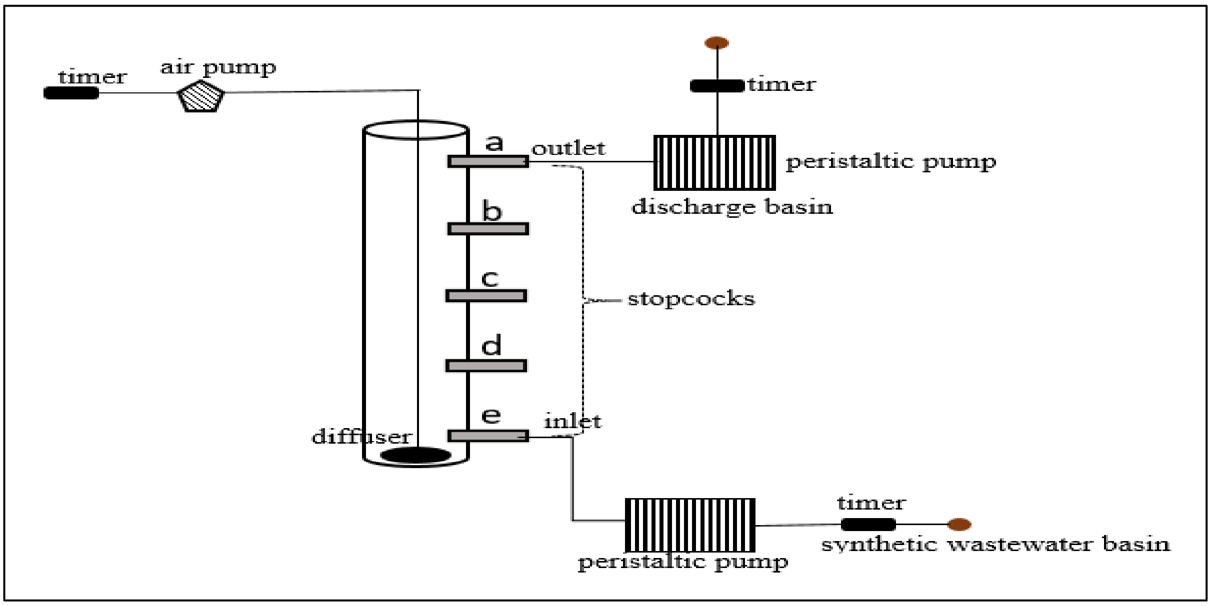

Figure 1: The schematic Drawing for Lab-scale Bioreactor Set-up

An air pump was required for the oxygen supply to the bioreactor. The air pump was located higher than reactor to make sure a good flow of oxygen supply. All appliances including peristaltic pumps and air pump used in the bioreactor system were controlled by digital timers. Two types of the basin were required in this experiment for synthetic wastewater and discharge basins. In addition, a plastic diffuser was placed at the bottom of the reactor. Fine

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air bubbles from the air pump was provided through the tubing into the reactor. The fine bubble is needed to enhance the granulation process.

2.2 Bioreactor operation

The lab-scale bioreactor system functioned continuously at room temperature with 24 hours of hydraulic retention time (HRT). There were four important cycles in this process that started off with the feeding of synthetic wastewater using a peristaltic pump, aeration using an air pump, settling and discharge using a peristaltic pump. Each cycle was controlled by a digital timer set at a certain time as shown in Table 1.

The biomass in the bioreactor was fed with synthetic wastewater in each cycle followed the recipe by [9]. The synthetic wastewater recipe used in this study consists of three main mediums (P, Q, and R) and are depicted in Table 2. The main ingredient in this recipe is acetate as the carbon source for the biomass in the reactor. During aeration process, oxygen was supplied from the air pump at the bottom of the bioreactor through a plastic diffuser to enhance the granulation process. The settling cycle was needed to let the biomass settled at the bottom of the bioreactor before the discharge process took place.

Table 1: The Operation Cycle of The Reactor

Cycle	Time (minutes)
Feeding	30
Aeration	120
Settling	4.8
Discharge	25.2

Table 2: Synthetic Wastewater Recipe

Medium P	Medium Q	Medium R
CH_3COONa	NH_4Cl	Trace elements
$MgSO_4.7H_2O$	K_2HPO_4	
KCl	KH_2PO_4	
Distilled water	Distilled water	

2.3 Activated sludge

About 900 mL of activated sludge was used to start the granulation process in the bioreactor. The activated sludge was collected from the wastewater treatment plant nearby. The activated

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sludge was filtered to take out the debris. The morphology of sludge was observed using the light microscope. The activated sludge was brown in color with fluffy texture was shown in Figure 2. The biomass concentration of the activated sludge such as mixed liquor suspended solid (MLSS) and mixed liquor volatile suspended solid (MLVSS) were determined by the standard method of American Public Health Association [10].

Figure 2: Activated Sludge

2.4 Size of granules analysis

Inverted microscope attached with a digital camera was used to monitor the morphology and size of the aerobic granules in two to three times a week. Besides that, the morphological study was done by examining the granules samples with the naked eye and a ruler was used to measure the diameter of the granules. This manual measurement was held if the granules were big enough to be seen by naked eye.

2.5 Percentage of the aerobic granules size

Three different sizes (0.2, 0.4, and 0.6 mm) of mesh sieves were used to determine the distribution size of the AG in this study as depicted in Figure 3. To start the experiment, the mesh sieves were arranged in decreasing size-order from the top down with a basin at the base as shown in Figure 4 [11].

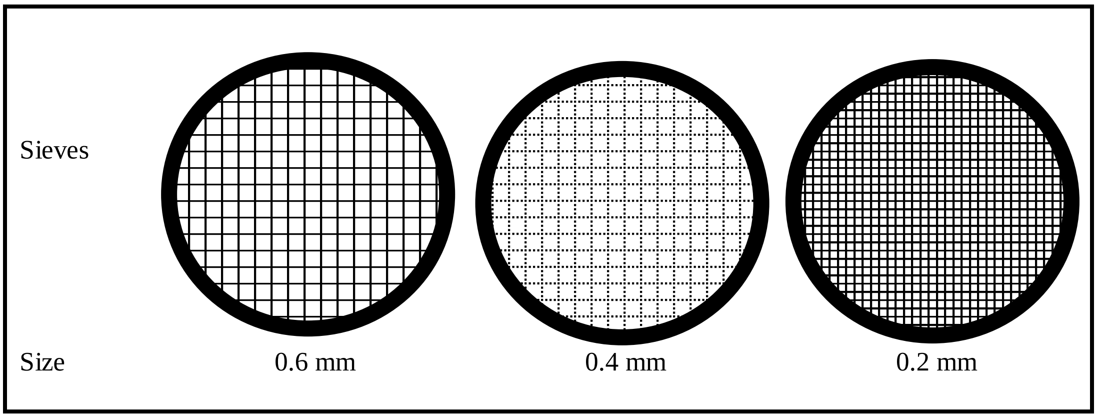

Figure 3: Mesh Sieves Sizes Used in This Study

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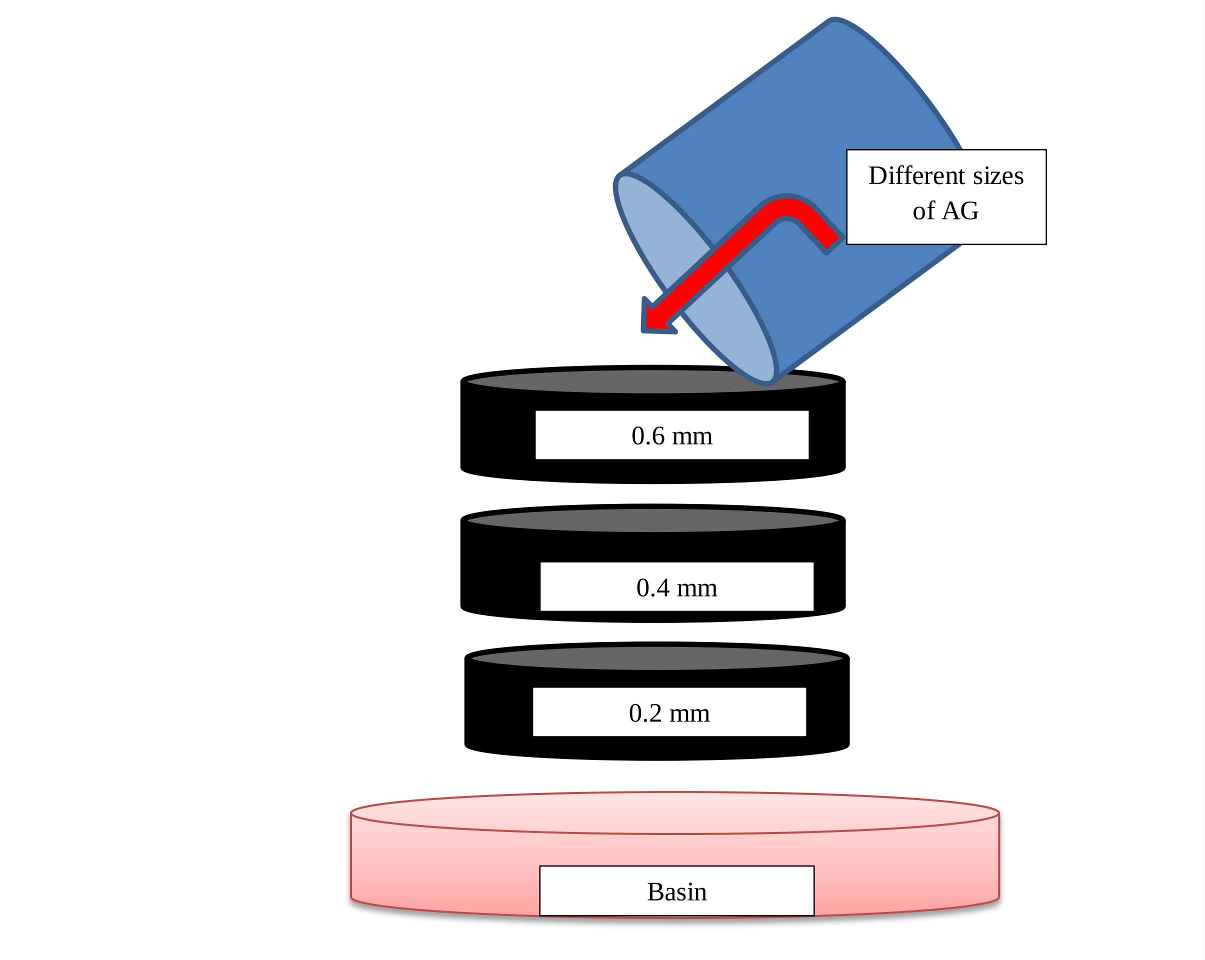

Figure 4: The Arrangement of Mesh Sieves

2.5 Settling velocity (SV)

The settling velocity of the aerobic granules was determined using a 1 Litre measuring cylinder with a known height and a stopwatch. The measuring cylinder was filled with tap water in which certain granule was released into and the time for it to reach the bottom taken. The formula to calculate settling velocity was given in Equation (1).

$$Settling\ Velocity\ (m/h) = \frac{Distance}{Time} \qquad (1)$$

3. RESULTS AND DISCUSSION

3.1 Biomass concentrations

The initial concentration of biomass in this study was 5.8 g MLSS/L. The MLSS and MLVSS values fluctuated for the first 14 days of the granulation in the reactor as shown in Figure 5. However, the biomass concentrations started to increase steadily from the 14 days since

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aerobic granules started to appear in the reactor. The biomass concentrations increase sharply from day 27 to day 30 since matured granules were developed.

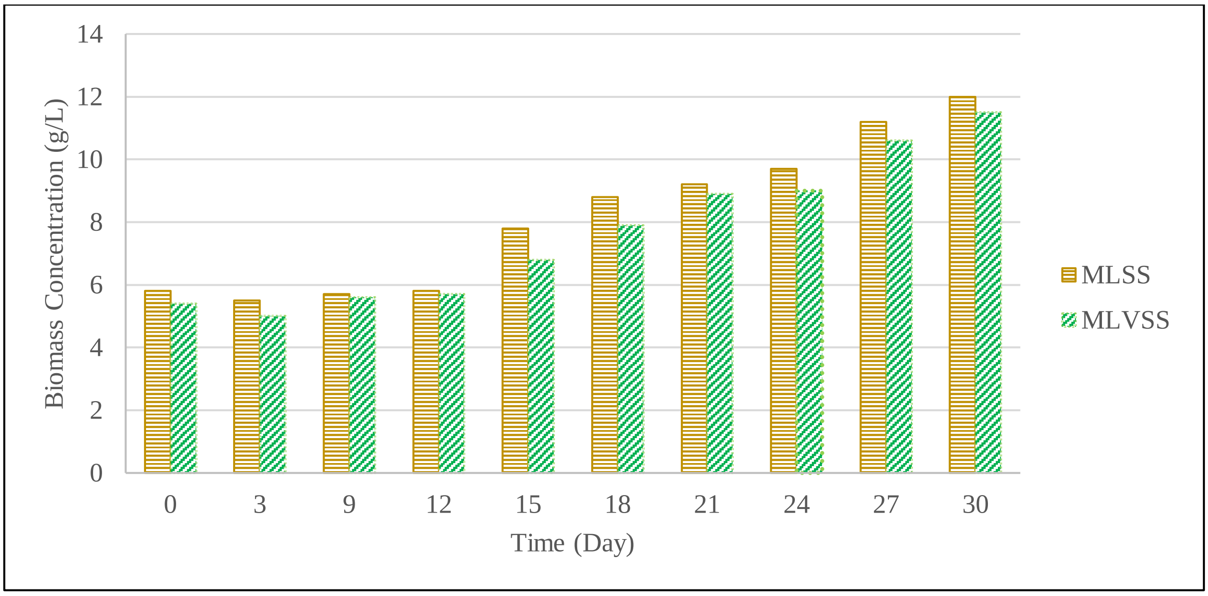

Figure 5: Biomass Concentration

3.2 Size distribution of aerobic granules

In this study, size distribution of aerobic granules was monitored from the beginning of the experiment as shown in Figure 6. The size distribution of aerobic granular sludge (AGS) was determined on different days throughout the experiment. The size of activated sludge was mostly less than 0.2 mm (approximately 85%) at the early operation of granulation process. Only 16% of biomass with diameter more than 0.2 mm was detected.

As reported by [12], at the early stage granules dominating the bioreactor was less than 1 mm diameter size approximately 70 % of the biomass. The decrease in the trend of aerobic granules which diameter <0.2 mm was shown throughout the experiment. However, other size categories (>0.2<0.4, >0.4<0.6 and >0.6 mm) indicating their increase are recorded over the time. The domination of aerobic granules >0.4 and >0.6 mm is shown at the end of the experiment. The growth and aging process of biomass organisations in aerobic granules was determined by the granule size. [10-12]. Granule size is a very important tool in determining the physical performance and features of aerobic granules [13]. As time increased, the biomass that attached together in each granule also increased. This is because the biomass effectively adapted with the reactor condition. This condition contributed to the increase in size of each aerobic granule.

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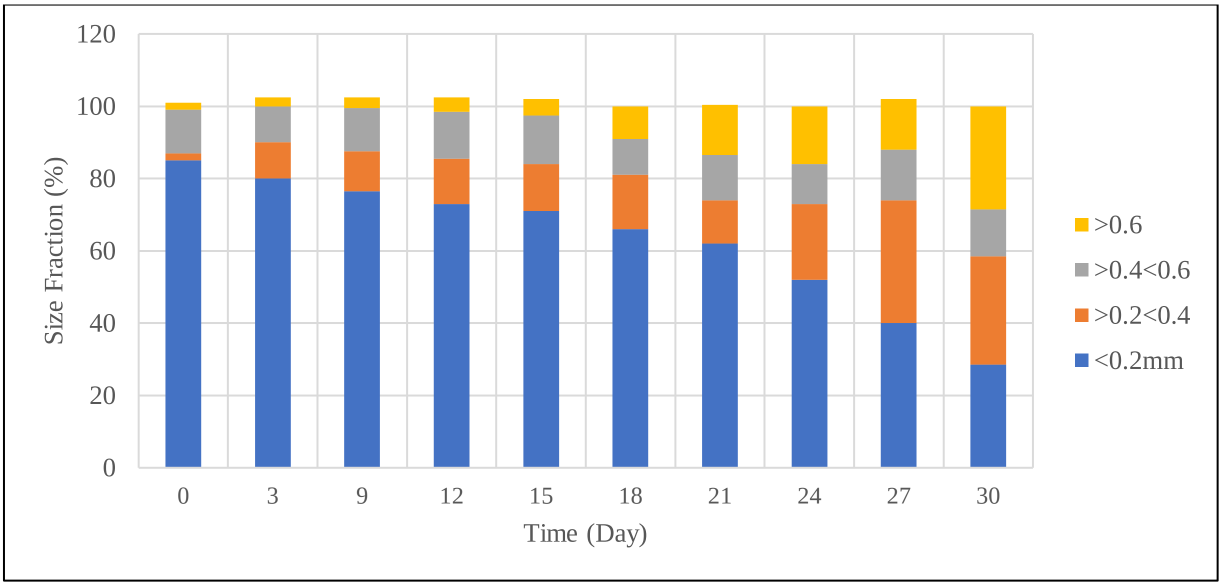

Figure 6: Size Distribution of Aerobic Granules

3.3 Settling velocity

The results of settling velocity of AGS is presented in Figure 7. The settling velocity of AGS began to increase as it increased in granule size over the granulation time. The AGS shows excellent settling properties at the end of the experiment with the highest settling time was 29.8 m/h. As reported by [14] that the successful of granulation process was indicated by the the settling ability and compactness of the granules. A study by [15] mentioned that there is positive correlation between granule size and other parameters such as the settling velocity, total and biomass densities. In addition, the settling velocity of AGS cultivated in this study was in range with previous studies by [11,16,17].

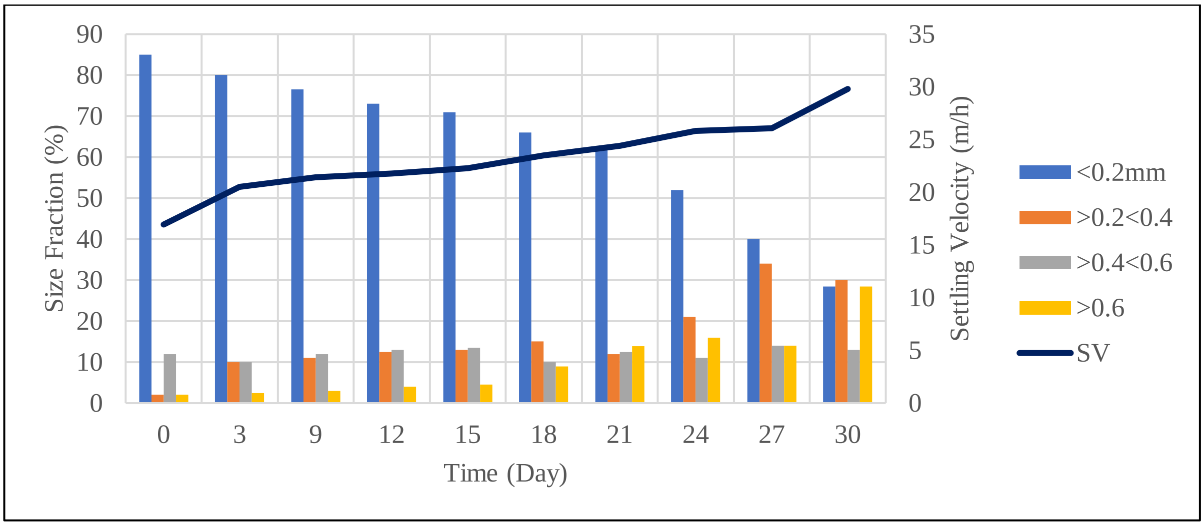

Figure 7: Size Distribution and Settling Velocity of Aerobic Granules

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4.0 CONCLUSION

This study demonstrates that the size of aerobic granular sludge (AGS) did increase over the time. The aerobic granules with size >0.4 and >0.6 mm is dominated in the reactor at the end of the experiment. Other than that, the good settling velocity properties were shown by AGS against time. The positive relationship was observed between granule size and settling velocity. In the future, other parameters, for example the sludge volume index (SVI) can be investigated thoroughly to see the relationship between the granules size and settling velocity.

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