# Formation and characteristics of microbial granules containing photosynthetic bacteria using palm oil mill effluent (pome)

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Abstract—The aim of this study was to investigate the possibility of applying bacterial photosynthesis using granulation technology method in minimizing carbon dioxide (CO<sub>2</sub>) gas emitted within a treatment system. By using a sequencing batch reactor (SBR) system with palm oil mill effluent (POME) as feeding, the photosynthetic microbial granules were developed after 100 days at an organic loading rate (OLR) of 1.53 kg COD/m<sup>3</sup>. day and hydraulic retention time (HRT) of 8 hours (cycle time = 4 h). In terms of the morphological structure, the sludge evolved from dispersed loose shaped into a more stable and smoother compact granular form with good settling properties. The sludge settleability also improved from 0.50 to 2.88 cm/s periodically due to the increased of biomass concentrations (6.90 - 8.25 g/L) with a maximum granule size of 3.2 mm obtained. Other than that, the physical characteristics of the granules displayed great strength and stability as they attained low integrity coefficient at 2.22%. Based on the pigment analysis result using the UV-Vis spectrophotometer, the bacteriochlorophyll and carotenoids were found to correspond to that of purple non sulphur bacteria within the ranges of 801nm to 865nm (the existence of bacteriochlorophyll b) and 400nm to 500nm respectively

Keywords- CO2 gas, SBR system, POME, photosynthetic microbial granule, bacteriochlorophyll b

#### I. INTRODUCTION

In the 1980s, carbon dioxide (CO<sub>2</sub>) gas was proven to be one of the most hazardous greenhouse gases (GHG) as it increases from 57 to 80% of the current GHG contribution to global warming (Lahoff and Lahuja, 1990). In Malaysia, one of the major sources of GHG is from industrial wastewater treatment such as the ponding system to treat palm oil mill effluent (POME). Malaysia's palm oil industry had generated approximately 80 million dry tonnes of solid biomass per annum as the volume is expected to increase to 85-110 million dry tonnes by 2020. (Agensi Inovasi Malaysia, 2013).

The conventional ponding system, especially the anaerobic process produces harmful and odorous gases such as sulphur dioxide (SO<sub>2</sub>), methane (CH<sub>4</sub>) and CO<sub>2</sub> gases (Daelman et al., 2012). Methane and carbon dioxide gases are the gases that contribute to the greenhouse effect causing global warming synergism (WRI, 2014). The role of photosynthetic bacteria is important for the CO<sub>2</sub> sequestration within the microsystem of wastewater as it utilizes the CO<sub>2</sub> in the environment. Microbial communities in aerobic granules have been shown to be highly distinct from activated sludge, even within a single reactor system. Recent studies implied the importance of gaining an understanding of the functions of microbial communities (Zak et al., 2011) as population diversity alone may not be adequate in determining the microbial characteristics. Thus, this study intended to investigate the characteristics of photosynthetic microbial granules developed from POME. The main parameters included were the change in term of morphological structure, biomass concentration and settling properties. Also, during the cultivation of photosynthetic bacteria, the presences of the bacteria were identified.

### MATERIAL AND METHODS

The experiment was operated using a 3L doublejacketed cylindrical column (6.4 cm in diameter and 90 cm in height) which was used as an SBR system with a working volume of 1.2 L at an OLR of 2.75 kg COD/m<sup>3</sup>. day. It was run for 4 h cycle per day consisted of 5 min for filling up the influent, 80 min of anoxic condition, 130 min of aeration, 15 min for settling of sludge, and 5 min of effluent withdrawal with a volume exchange ratio (VER) of 50% at room temperature (between 20-27 ± 2°C). In addition, the reactor was equipped with two light sources providing illumination of 3600 lux each (12 h light/ 12 h dark regime) for creating the photosynthetic condition.

For the morphological observations, the photosynthetic microbial granules were examined using the light microscope with  $PAX-IT^{TM}$  image analyzing system. The biomass concentration and the sludge volume index (SVI) were tested based on the Standard Methods for the Examination of Water and Wastewater (APHA, 1998). Also, the other parameters such as the settling velocity and the granular strength were conducted according to Linlin et al. (2005) and Ghangrekar et al. (1996). To detect bacteriochrophyll and carotenoids, the whole cell spectrum of the selected sample was analyzed within the range of 300-1100 nm using the DR 5000 spectrophotometer.

## III. RESULTS AND CONCLUSION

Based on the morphological observation (Fig. 1), initially, the sludge (average diameter = 0.04 mm) consisted of dispersed flocs (Fig. 1A). Then, the small flocs disintegrated under aerobic condition tend to merge together slowly forming flocculated sludge (Fig. 1B). It continued to develop into small aggregates (Fig. 1C) with an average diameter of 1.8 mm. Shortly, Fig. 1D showed the flocculated sludge forming bigger aggregates. The bacteria

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tend to colonize into particles, which produced extra cellular polymer generally bind the flocs properties together and contributed to the formation of denser aggregates (Snidaro et al., 1997) until granules were obtained after 100 days (Fig. 1E,F). As shown in Fig. 2(a), through time, the sludge biomass in the reactor was inconsistently increased and decreased. The settleability on the first two months was inconsistent rising to 57.8 m/h (day 31) and simultaneously dropped after 10 days till day 70. This might be because of the aggregation process which is considered a balanced between the floc formation and floc breakage occurred (Wan et al., 2011). Eventually, advanced granulation took place between 70 to 110 days as the settling velocity began to display steady improvement from 43.5 to 103.0 m/h (Fig. 2(b)). Other than that, Fig 3 displayed great in strength and stability of the granules as they attained low integrity coefficient at 2.22%. Moreover, the bacteriochlorophyll and the carotenoids detected (Fig. 4) were found to correspond to that of purple non sulphur bacteria within the ranges of 801nm to 865nm and 400nm to 500nm respectively. Future studies will include optimization of the operational parameters such as the HRT and organic loading rate to produce granules that are more compact, of higher settling abilities and durability. It is also noteworthy to include immediate applications of the granules in wastewater treatment to minimize the emission of GHG

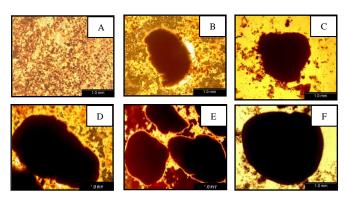


Fig 1. Microscopic physical observation using PAX-IT (magnification = 40x): (A) first day, (B) Two weeks, (C) 30 days, (D) Two months, (E) 90 days and (F) 100 days

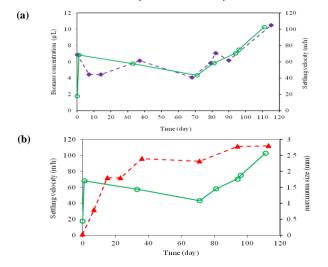


Fig. 2 (a) The changing trend of biomass concentration ( $-\Phi$ -), settling velocity ( $-\Phi$ -) and (b) the size of granules ( $-\Phi$ -) with time.

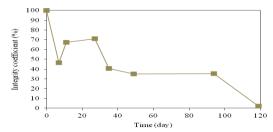


Fig 3. Granular strength profile of the granules

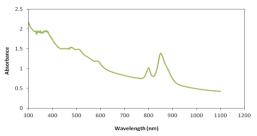


Fig. 4 Wavelength curve results for bacteriochlorophyll and carotenoids

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