STUDY OF PALM OIL MILL EFFLUENTS TREATMENT USING BIOENGINEERED STRUCTURE BIOMEDIA

A. A. M. Azoddein^{1*}, A. B. Bustary², F. A. Azli³, N. H. Z. Abidin ⁴, I. M. Ali ⁵, M. S. M. Desa⁶,

^{1,2,3,4,5,6} Faculty of Chemical and Natural Recourses Engineering, University Malaysia Pahang, Lebuhraya Tun Razak, 26300 Gambang, Pahang, Malaysia

ABSTRACT

Palm Oil Mill Effluent (POME) is waste produced by the palm oil processing mills in Malaysia and also generates large quantities of liquid waste. This research is aim to remove high content of suspended solids and oil in POME by using Bio Engineered Structure (BES) bio media. Besides that, it also to investigate condition of BES biomedia for POME treatment in bioreactor system and also the main factors affecting BES biomedia performance in POME treatment. The BES biomedia for POME wastewater treatment system is a compact biological purification system and enhanced the conditions for the microbiology to breakdown the pollution. The nutrient broth mixed with Pseudomonas putida (P. putida) was injected to BES biomedia tank with the ratio of 1:9 P. putida to POME. The growth of P. putida was compared between in shake flask and BES biomedia reactor. In shake flask, the P. putida take a shorter time to encounter with the environment while in BES biomedia reactor it take more period to encounter with a new environment. From the aeration rate, it shows that at rate of one L/min have the highest removal efficiency of Chemical Oxygen Demand (COD), 46.73%, Biochemical Oxygen Demand (BOD), 99.03% and Total Suspended Solids (TSS), 71.88%. As conclusion, BES biomedia is potentially to be a good method to treat POME that will reduce the uncontrolled waste and environmental pollution to our country.

KEYWORDS: P. putida; BES biomedia; palm oil mill effluent

1.0 INTRODUCTION

Malaysia is the largest producer and exporter of palm oil. In 2001, the total oil palm planted area registered with Malaysian Palm Oil Board (MPOB) was 3,449 012 hectares with 405 approved mills with a total capacity of 75.99 million tonnes of fresh fruit bunches (FFB). Meanwhile, 46 palm oil refineries were in operation with 15.48 million tonnes capacity of crude palm oil per year. Palm Oil Mill Effluent (POME) contains of about 95% to 96% water, 0.6% to 0.7% oil and 4% to 5% total solid including 2% to 4% suspended solids which are mainly debris from mesocarp. Pollution problem from the POME can be solve if changes are made into a sellable product. In addition, it also creates a business opportunity for the industry. Oil palm currently occupies the largest

^{*}Corresponding Email: aaziz@ump.edu.my

acreage of farmed land in Malaysia (Arif et al., 2001). The lifespan for oil palm is over 200 years and after 32 to 38 months planting, the plant can be first harvest. Besides that, the peak yield of palm oil plant can be reach after 5 to 10 years (Kittikun et al., 2000).

Pseudomonas putida, (P. putida) is a rod-shaped, non-spore forming, gram-negative bacteria that utilizes aerobic metabolism. *P. putida* also has a multiple polar flagella for motility where it usually has two or three wavelength long. Flagella rotation always changes the direction because *P. putida* sensitive to the environment and sensing chemo attractants (Harwood, et al., 1989). The characteristics of *P. putida* are higher degree of saturation of fatty acid and membrane fluidity, survive deadly toxins and allow it to thrive in contaminated areas. Metabolism from *P. putida* transforms the harmful organic solvent to nontoxic composites (Härtig, 2005).

Over the last four decades, the Malaysian palm oil industry has developed to become a main agriculture-based industry. Malaysian palm oil contributed for about 39% of the world palm oil production and 44% of world exports (Malaysia Palm Oil Council, 2010). The production of palm oil produces large amounts of polluted waste water known as POME. Raw POME usually has high in BOD (25,000 mg/L), COD (53,630 mg/L), oil and grease (O&G) (8370 mg/L) and total solids (TS) (43,635 mg/L) which can cause significant environmental impact if it left untreated (Chan et al., 2011). Table 1 shows the palm oil effluent discharge standards.

Parameter	Standard	Standard	Standard	Standard	Standard	Standard
	Α	В	С	D	Ε	\mathbf{F}
pH	5-9	5-9	5-9	5-9	5-9	5-9
BOD	5000	2000	1000	500	250	100
COD	10000	4000	2000	1000	-	-
TS	4000	2500	2000	1500	-	-
NH ₃ -N	25	15	15	10	150	100
Total N ₂	200	100	75	50	-	-

 Table 1. Palm oil effluent discharge standards (Malaysia Palm Oil Board, 2012)

*Units in mg/L except pH** No discharge standard after 1984

The BES biomedia treatment system is designed for treating the POME wastewater where the water is aerated. Then, it will produce oxygen and the microorganism, which live on stones, and plants will used the oxygen to enhance the conditions for the microbiology to break down the pollution as shown in Figure 1.

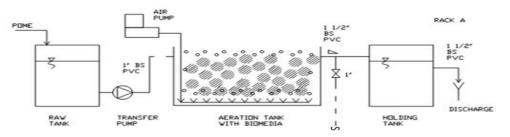


Figure 1. BES Biomedia Bioreactor Process Flow

ISSN: 2180-3811

Vol. 6 No. 2 July-December 2015

2.0 MATERIALS AND METHOD

2.1 Wastewater preparation

Hot and acidic fresh POME (pH 7 to 9) was obtained from Felda Palm Industries Sdn. Bhd., Felda Lepar Hilir 3, Gambang, Pahang and the color of POME is brownish. It has a colloidal suspension and containing high concentration of organic matter. It was preserved at temperature 4°C, but above its freezing point. The required volume was thawed to room temperature before feeding into the bio media compartment.

2.2 Mechanism and optimizing procedure

Two-liter bioreactor was used for the treatment of wastewater. The process is a batch process and POME was filled and released for every 24 hours. The bio media was put inside the BES biomedia bioreactor and the oxygen is injected and the aerated water will be used by the microorganisms. The initial pH, COD, BOD and TSS of POME was measured before filled into the bioreactor. For the aeration rate, the treatment began with aeration rate at 0.5 L/min and increased to 1.0 L/min and 1.5 L/min.

2.3 BES Biomedia Bioreactor

The biomedia was tied in the net and placed inside the BES bioreactor. The initial pH, COD, BOD and TSS of raw POME were measured by using USEPA method. Then, the raw tank was filled with the POME and the nutrient broth mixed with *P. putida* was injected to the BES biomedia tank with the ratio of 1:9 *P. putida*. The sample was treated for 24 hours as the optimum growth of *P. putida* in the tank. The aeration rate were varies at 0.5 L/min, 1.0 L/min and 1.5 L/min for every treatment. The temperature was set to constant at 37°C and the volume of sample is two-litter. After 24 hours, the treated POME was then released and tested for its COD, BOD and TSS.

2.4 Analysis

2.4.1 BOD

BOD is a measurement of the microorganisms' ability to digest organic matter, usually in 5 days incubation at 20°C by analysing the depletion of oxygen. BOD is the most commonly used parameter for determining the oxygen demand on the receiving water of a municipal or industrial discharge. BOD can also be used to evaluate the efficiency of treatment processes, and is an indirect measure of biodegradable organic compounds in water. Usually BOD₅ will be examined by dilution method (Andrew et al., 2005).

2.4.2 COD

COD is a measurement of oxygen requirement of a sample by adding strong chemical oxidant. In this procedure, the sample is heated for two hours with a strong oxidizing agent, potassium dichromate. Oxidizable organic compounds react, reducing the dichromate ion $(Cr^{2O7^{2-}})$ to green chromic ion (Cr^{3+}) . When the 3 to 150 mg/L

ISSN: 2180-3811	Vol. 6 No. 2 July-December 2015
15511. 2100 5011	Vol. 0110. 2 July December 2013

colorimetric or titrimetric method is used, the amount of Cr^{6+} remaining is determined. When the 20-1,500 mg/L or 200-15,000 mg/L colorimetric method is used, the amount of Cr^{3+} produced is determined. The COD reagent also contains silver and mercury ions. Silver is a catalyst, and mercury is used to complex chloride interferences. Test results for the 3 to 150 mg/L range are measured at 420 nm and test results for the 20 to 1,5000mg/L and the 200 to 15,000 mg/L range are measured at 620 nm (Andrew et al., 2005).

2.4.3 TSS

Suspended solids in water may consist of inorganic and organic particles or of immiscible liquids. Inorganic solids such as clay, silt, and other soil constituents are common in surface water. Environmental Protection Agency (EPA) has set a maximum suspended solids standard of 30 mg/L for most treated wastewater discharges. A well-mixed measured sample is filtered through a weighed standard glass fibre filter and the residue retained on the filter is dried to a constant weight at 103°C to 105°C. The increase in weight of the filter represents the total suspended solids. If the suspended material clogs the filter and prolongs filtration, it may be necessary to increase the diameter of the filter or decrease the sample volume (Andrew et al., 2005).

2.5 Testing

The treated water will be test for few parameters which are BOD, COD and TSS. The equipment that will be used for each test are:

- i. BOD: BOD Incubator/ Dissolved Oxygen Meter
- ii. COD: COD Digestion Reactor /Spectrophotometer, HACH DR/2400
- iii. TSS: Buchner flask and funnel/vacuum pump

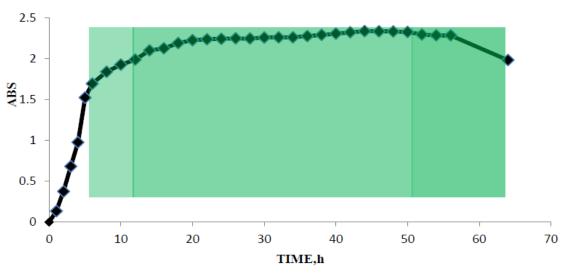
To estimate the pollutants removal efficiency, equations in Table 2 were used in this study.

No	Symbol	Unit	Description	Equation
1	%COD	%	Overall COD removal efficiency	(CODin – CODout)x 100 CODin
2	%BOD	%	Overall BOD removal efficiency	(BODin – BODout)x 100 BODin
3	%TSS	%	Overall TSS removal efficiency	(TSSin – TSSout)x 100 TSSin

Table 2. Calculation for main parameters, Chan Y.J et al. (2012)

3.0 **RESULT AND DISCUSSION**

3.1 Growth of *P. putida* in shake flask



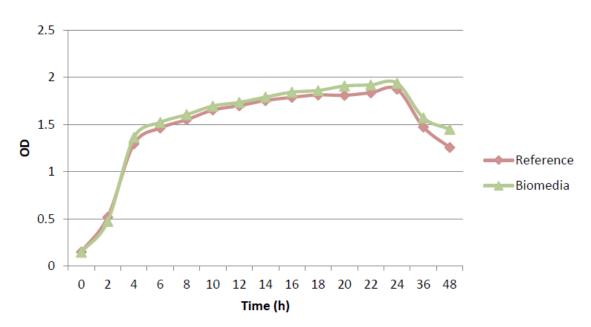
Pseudomonas Putida Growth Curve

Figure 2. P. putida growth curve in shake flask

The growth curve of bacteria commonly determine by the simply condition of batch culture. The suitable condition for *P. putida* in incubator shaker is 37°C and shaking speed of 180 rpm. The bacteria growth curve basically contains of four phase which are lag, exponential growth, stationary and death phase. From Figure 2, it shows that the graph of *P. putida* against time. The ABS (absorbance) is referring to the viable or living bacteria at certain time. The growth of *P. putida* obtain is quite similar to the standard growth of bacteria that claim from Widdel (2007).

Lag phage occur in the period of time 0 to 1 hour. This phase happen in very short of period. It illustrates that *P. putida* take a shorter time to encounter with the new environment that supply to them. The second phase is exponential phase where in this stage the living bacteria is rapidly increased within time. From the graph, it shows that the exponential phase occurs in the period of 1 to 7 hours. During this phase the bacteria is comfortable with the environment supply where the optimum condition for the *P. putida* to growth up and create as much as colonial possible by degrade the substrate. The stationary phase occur in the period of 7 to 48 hours where in this phase the growth rate becomes slower because the bacteria is death. Since the process occurs in batch culture, the substrate supply will decrease and cause degradation that occur through the exponential phase. Besides that, *P. putida* is the living thing that will produce waste and secondary metabolic products in this phase. The death phase occur in the period of 48 to 64 hours where the number of living bacteria will decrease within time.

ISSN: 2180-3811



3.2 Growth of *P. putida* in BES biomedia bioreactor

Figure 3. Graph for P. putida growth in BES Biomedia Bioreactor

Figure 3 shows the growth pattern of *P. putida* in BES biomedia bioreactor. There are two types of growth which the first type was the sample without biomedia as a reference and the second type with the biomedia. This experiment was run to identify the effect of biomedia of growth pattern *P. putida*. The growth of *P. putida* obtained is similarly with to the standard growth of bacteria and *P. putida* growth in conical flask by controlling temperature and rotation speed.

In lag phase of *P. putida* at 0 to 2 hours, it is slightly different with the growth in conical flash which requires more time. Thus, the growth in BES biomedia take a longer time to encounter with a new environment that supply to them. The figure also shows that the bacteria were rapidly growth without biomedia. In second stage, the exponential phase show that the *P.putida* growth with biomedia was higher without biomedia and occurs in the period of 2 to 5 hours. During this phase, the bacteria are comfortable with the new environment and known as optimum condition for the *P.putida* to growth and multiply the colonial as much as possible.

At period from 5 to 24 hours, the bacteria growth is slowly increase and this phase is called a stationary phase where growth rate is equal to death rate. The death phase occur in the period of 24 to 48 hours. During this phase, the bacteria did not multiply more due to lack of carbon source as a feed stock to bacteria and the rate of bacteria death is faster than it multiply.

ISSN: 2180-3811

3.3 Effect of aeration rate

Table 3 shows the effect on BOD, COD, and TSS removal efficiency due to *P. putida* growth at certain aeration rate. At rate of 0.5 L/min, it shows that, the lowest removal efficiency of BOD is 70.67%, COD is 31.82% and TSS is 59.35%. On the other hand, at 1.5 rpm, there is an increment on the removal efficiency of BOD which at 89.72%, COD at 41.24% and TSS at 67.86 %. Meanwhile, at one L/min, it shows the highest removal efficiency of BOD at 99.03%, COD at 46.73% and TSS at 71.88%. Therefore, the optimum condition for *P. putida* growth is when the aeration rate is at 1.0 L/min.

Aeration Rate (L/min)	0.5	1.0	1.5	
BOD (mg/L)				
Before (DO)	468	369	374	
After (DO)	137.26	3.57	38.45	
Removal Efficiency (%)	70.67	99.03	89.72	
COD (mg/L)				
Before	5500	4600	4850	
After	3750	2450	2850	
Removal Efficiency (%)	31.82	46.73	41.24	
TSS (mg/L)				
Before	2140	1280	1960	
After	870	360	630	
Removal Efficiency (%)	59.35	71.88	67.86	

Table 3. Data for different rate of aeration in biomedia bioreactor

The bacterium growth is increasing with the aeration rate that was been set up. From the figures, it shows that at a rate of 1.0 L/min, the system contributes to the highest removal efficiency. This is due to the bacterium begins to die because of lack of nutrients and high metabolisms of bacterium (Cascaval, 2003)

4.0 CONCLUSION

The removal of organic matter in POME was successfully obtained by using the *P. putida* in BES biomedia bioreactor. It used the formulation ratio of 1:9 of *P. putida* to POME with a different aeration rate. The optimum conditions of aeration rate at 1.0 L/min shows a high removal efficiency of COD, BOD and TSS which are at 46.73%, 99.03% and 71.88% respectively.

ACKNOWLEDGEMENT

The authors would like to acknowledge the financial support from University Malaysia Pahang (Grant RDU1103102) and Felda Palm Industries Sdn. Bhd. Felda Lepar Hilir 3, Gambang, Pahang for supporting this research.

Vol. 6 No. 2 July-December 2015

REFERENCES

- Andrew, D.E., Lenore, S.C., Eugene, W.R. and Arnold, E.G. (2005). Standard Methods for the Examination of Water and Wastewater. American Public Health Association, Washington, D.C.
- Arif, S., Tengku Mohd Ariff, T.A., (2001). The case study on the Malaysian palm oil. UNCTAD/ESCAP Regional Workshop on Commodity Export Diversification and Poverty Reduction in South and Southe-East Asia, Bangkok.
- Borja, R., Banks, C. J., (1994). Treatment of palm oil mill effluent by up-flow anaerobic filtration. *Journal of Chemical Technology and Biotechnology*, 61: 103-109.
- Borja, R., Banks, C. J.,(1995). Comparison of anaerobic filter and an anaerobic fluidized bed reactor treating palm oil mill effluent. *Process Biochemistry*, 30: 511-521.
- Cascaval, D. (2003). Studies on Oxygen Mass Transfer in Stirred Bioreactors: Suspensions of bacteria, yeasts and fungis, *Journal of Chem. Ind.*, 57 (6): 276-287.
- Chan, Y. J., Chong, M. F., Law, C. L., (2010). Biological treatment of anaerobically digested palm oil mill effluent (POME) using a Lab Scale Sequencing Batch Reactor (SBR). *Journal of Environmental Management*. 91:1738-1746.
- Härtig, C., Loffhagen, N., Harms, H. (2005). Formation of trans Fatty Acids Is Not Involved in Growth-Linked Membrane Adaptation of Pseudomonas putida. *Applied and Environmental Microbiology*. 71: 1915-1922
- Harwood, C.S., Fosnaugh K., Dispensa M. (1989). Flagellation of Pseudomonas putida and analysis of its motile behavior. *Journal of Bacteriology*. 171: 4063-4066.
- Kittikun, A. H., Prasertsan. P, Srisuwan. G & Krause, A. (2000). Environmental Management of Palm Oil Mill. Retrieved from http://www.globetree.org/jackyfoo/ic-mfa/kittikun/index.html
- MPOC, Malaysian Palm Oil Council, (2010). Retrieved from http://www.mpoc.org.my
- Widdel, F. (2007). Theory and Measurement of Bacterial Growth. Grundpraktikum Mikrobiologie, 4. Sem. (B.Sc.), Universität Bremen.
- Warden, W. B., Biomedia (2012). Filter media. Retrieved from http://wardenbiomedia.com/process/filter.html