# Thermophilic Palm Oil Mill Effluent (POME) treatment using a mixed culture cultivated from POME

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In this study, a mixed culture specifically for thermophilic anaerobic treatment of Palm Oil Mill Effluent (POME) has been cultivated from a mesophilic seed sludge using a batch fermenter. The performance of the cultivated mixed culture on the treatment of POME was also evaluated with the batch fermenter. The cultivation was successful as the cultivated thermophilic mixed culture managed to reduce at least 90 % of Chemical Oxygen Demand (COD) in POME with after 6 days of operation from POME treatment with a Mixed Liquor Suspended Solid (MLSS) concentration of 14000 mg/L producing biogas with 66% of methane.

# 1. Introduction

POME is a wastewater generated from palm oil milling activities and it is conventionally treated with ponding systems or open digesting tanks (Ma et al., 2003). The application of high-rate anaerobic bioreactors to replace conventional treatment methods for POME treatment has escalated as these high-rate anaerobic bioreactors had smaller foot prints, producing better treated effluent quality and greater biogas volume with higher purity of methane which can be utilized for energy generation purposes.

Operation of anaerobic treatment of POME under thermophilic conditions was proven to produce effluent of better quality and also having higher biogas production rate (Poh and Chong, 2009). However, anaerobic thermophilic sludge for POME treatment is not readily available in the market. Thermophilic systems requires a longer time for start-up to allow the mesophilic sludge to acclimatize with the substrate and temperature shift. To reduce the start-up period required by the anaerobic thermophilic POME treatment system and minimize the effect of temperature change to the mesophilic sludge (i.e.: bioreactor upset), it is necessary to cultivate a thermophilic mixed culture tailored for thermophilic anaerobic POME treatment.

Several studies showed that a cultivated mixed microbial consortium for the treatment of a targeted wastewater can be obtained by acclimatizing the existing seed sludge from any biological sludge basin with the targeted wastewater as the substrate. Sreekanth et al. (2009) obtained an inoculum specifically for pharmaceutical wastewater treatment by utilizing slaughterhouse wastewater sludge as the seed source and allowed the sludge

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to be acclimatized to the system by feeding pharmaceutical wastewater as the substrate. Similarly, Tan and Ji (2010) also utilized sludge from oil-contaminated wastewater treatment as the seed source to obtain a carbazol-degrading microbial consortium by feeding carbazol-containing wastewater as the substrate.

This paper elaborates on the cultivation of an anaerobic thermophilic mixed culture for POME treatment. The performance of the cultivated mixed culture was also assessed based on the treatment efficiency of POME under thermophilic condition.

## 2. Material and Methods

### 2.1 Batch fermenter for bacteria cultivation

Minifors (Infors HT, Switzerland) batch fermenter was used for the cultivation of the mixed culture for POME treatment at thermophilic condition. The working volume of the fermenter for bacteria cultivation was 2 litres. The bioreactor was inoculated with 0.5 L of seed sludge and 1.5 L of POME. The increase of organic loading rate (OLR) in the batch fermenter was shown in Figure 1. pH of the fermenter was adjusted to 6.8 with 1M sodium hydroxide. Sodium bicarbonate was further added to provide alkalinity to the system and to maintain the mixture at a pH of 7.0. Nitrogen gas was used to purge oxygen from the fermenter. Stirring speed of the fermenter was fixed at 100 rpm for complete mixing in the system.



Figure 1: Influent COD concentration and OLR profile during the cultivation period

The initial temperature of the bioreactor was set at 35 °C for 4 h before a single step increase of temperature to 55 °C to allow the adaptation of mesophilic seed sludge. This strategy was employed based on the study conducted by Bouškouvá et al. (2005) which found that a one-step temperature increase provided a shorter start-up period as compared to a stepwise increase approach. An adapted population of mixed culture is obtained when the effluent Chemical Oxygen Demand (COD) remained constant with less than 5 % variation (Ji et al., 2009).

#### 2.2 Seed material

The seed sludge for the cultivation of mixed culture was taken from a mesophilic anaerobic wastewater treatment system of an oleo-chemical manufacturing plant (Pan Century Edible Oils Sdn. Bhd., Johor, Malaysia).

#### 2.3 Substrate for mixed culture cultivation

Raw POME was used as a substrate for the cultivation of mixed culture. POME was collected from Golconda Palm Oil Mill (Klang, Selangor) and was preserved at 4 °C if not used immediately. Table 1 shows the average characteristics of raw POME.

*Table 1: Average characteristics of raw POME used for the cultivation of thermophilic mixed culture* 

Parameters	COD	sCOD	SS	$pH^*$
mg/L	30500	16990	11150	4.5
* * * 11				

\*no units for pH

## 2.4 Steady-state fermenter operation

The treated POME effluent in the fermenter was withdrawn and replaced with new feed when at least 80% of COD is being degraded to prevent POME from becoming a limiting substrate in the system. COD, soluble COD (sCOD) and suspended solids (SS) concentrations of the treated POME were measured according to AHPA Standard Methods (Eaton et al., 2005). Biogas volume was measured with a water displacement system while the biogas composition was measured with Gas Data GFM 416 series biogas analyzer.

## **3. Results and Discussion**

#### 3.1 Cultivation of thermophilic mixed culture

Figure 2 shows the profiles for pH and COD removal efficiency in the batch fermenter during the cultivation period. The COD removal efficiency of the system was highly dependent on the pH of the system during start-up. During the first 6 days of start-up, the removal of COD from the system increased steadily to 44 % until the pH of the system reduced from 7.68 to 7.0 where the COD removal efficiency was found to decline thereafter. NaHCO<sub>3</sub> was dosed into the batch fermenter when pH of the fermenter dropped to less than 7 to maintain it at a pH of 7. A COD removal efficiency of 54% was achieved when pH in the batch fermenter was restored to 7 and above. Similar observation was obtained for Run 2. Dosing of buffer was required in the initial stage as the methanogens were not quick enough to convert acetic acid to methane. In subsequent runs (3, 4, 5), pH of the fermenter was maintained within the optimum operating pH range (6.8-7.2) without the dosing of alkaline buffer. The pH rise in the system indicates that the methanogens have adapted to the system (Gerardi, 2003) and is able to proceed to the steady-state evaluation of the mixed culture.

The batch fermenter required a period of 120 days for cultivation. A COD removal efficiency of 84% was achieved with the batch fermenter with an OLR of 0.64 g COD/L day when the steady-state was achieved. The highest and average methane concentration recorded during the cultivation period was 72 % and 64.5 % respectively. The average methane concentration was similar to the value recorded from the study on thermophilic anaerobic contact digestion for POME treatment (65 %) (Ibrahim et al., 1984). When the COD removal efficiency of the batch fermenter has maintained constant around 84 %  $\pm$  0.5 for 3 consecutive days, the batch fermenter was fed with

undiluted raw POME to evaluate the performance of the mixed culture on the treatment of POME under thermophilic conditions.



*Figure 2: Profiles of COD removal efficiency and pH of the batch fermenter for POME treatment at thermophilic condition* 

#### 3.2 Mixed culture performance under steady-state conditions

Table 2 lists the feed conditions, operating conditions and the quality of the treated effluent during steady-state runs. The average methane concentration in the biogas produced from the batch fermenter for POME treatment at thermophilic condition was 66%, which was higher than conventional treatment methods (Yacob et al., 2006; Yacob et al., 2005) and falls within the range of methane concentration produced from high-rate anaerobic bioreactor (62.0-84.0%) (Najafpour et al., 2006). Furthermore, the methane concentration was also comparable to the 65 % obtained by Ibrabim et al. (1984) on thermophilic anaerobic contact digestion of POME.

The effect of MLSS concentration on POME treatment under thermophilic condition was evaluated simultaneously under Run 6-9. Seed sludge was added into the system and the feed volume of POME was reduced to increase the MLSS concentration. The volume of seed sludge and POME in the batch fermenter for each steady-state runs were indicated in Table 2. The HRT required to reduce more than 80 % of COD of POME was reduced from 16 days (Run 5) during cultivation period to  $\leq$ 7 days (Run 6-9) during steady-state period. This suggests that a mixed culture consisting of thermophilic microbes was successfully cultivated as the culture showed great improvements in terms of COD removal efficiency and methane composition in the biogas.

Figure 3 shows the relationship of the concentration of MLSS in the batch fermenter and the COD removal efficiency attainable after 6 days of operation. The COD removal

efficiency significantly improved from an 80 % to 90 % when the MLSS concentration of the fermenter increased from 5600 mg/L to 14000 mg/L. Nevertheless, no significant increase in the COD removal efficiency was observed when the MLSS concentration was increased to 15000 mg/L. This indicated that the MLSS concentration should be maintained at 14000 mg/L in the system for at least 90% COD removal.

			F	eed Condi	tions						
Run	HRT (days)		pH	COD	)	sCOI	)	SS (mg/L)			
				( <i>mg/</i>	L)	(mg/1	L)				
6	7		5.28	2460	0	1535	0	8550			
7	6		4.29	2980	0	1660	0	11750			
8	6		4.33	3780	0	1990	0	13550			
9	7		4.20	4085	0	1990	0	12950			
Operating Conditions											
Run	Temperatur	re	OLR		Feed	volum	e Seed	sludge volume			
	(°C)		(kg COL	D/m³day)	(mL)		(mL)				
6	55		1.41		800		700				
7	55		1.74		700		800				
8	55		1.89		600		900				
9	55		0.94		900		900				
			Treated	l Effluent <b>(</b>	Conditie	ons					
Run	pH	COD	removal	sCOD		SS r	emoval	Methane			
		efficier	ncy (%)	removal		efficien	ncy (%)	concentration			
				efficiency	v (%)			(%)			
6	7.44	82.9		79.2		70.2		70.1			
7	7.19	85.1		82.5		75.2		64.9			
8	7.15	90.4		83.2		69.0		69.5			
9	7.25	82.9		81.2		53.3		66.7			
		00									
		90 - 80 -	•	+ + +			-				
	5	70 -									
	effici	60	X								
	noval	50 -									
	D ret	40 -	/								
	al CO	30 -									
		20 -									
					,						
			2730 4730			730 1273	30 14730				
				MLSS Concent	ration (mg/	L)					

*Table 2 Average characteristics of raw POME used for the cultivation of thermophilic mixed culture* 

Figure 3: MLSS concentration against the attainable COD removal efficiency after 6 days of operation

## 4. Conclusion

A mixed culture specifically targeted for anaerobic treatment of POME at thermophilic condition was successfully cultivated. The mixed culture was able reduce up to 90 % of COD in POME after 6 days of operation and MLSS concentration of 14000 mg/L. Average methane concentration in the biogas produced was 65 %.

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