

Article

## Potential of Palm Oil Mill Effluent (POME) as Medium Growth of *Chlorella sp* for Bioenergy Production

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**Abstract:** Energy is essential and vital aspect for development in Indonesia. Biodiesel has received much attention as renewable energy in recent years according to lack of petroleum energy. One of potential biodiesel is produced from microalgae. Earlier research was reported that palm oil mill effluent (POME) is potential medium to be used as microalgae to grow because contain high nutrient source. The objective of the research is to determine growth rate and biomass productivity in *Chlorella sp* cultured in POME concentration using urea as nitrogen source. *Chlorella sp* was cultured in 20-70% POME using additional urea with concentration of 0.1 g/L and 1 g/L at a flask disk, pH 6.8-7.2; aerated using aquarium pump and fluorescence lamp 3000-6000 lux as light source. The biomass concentration was measured using spectrophotometer Optima Sp-300 OD at 680 nm wavelength within 15 days cultivation to calculate specific growth rate. At the end of cultivation, *Chlorella sp* was filtered and measured as dry weight. Result indicated that *Chlorella sp* at 50% POME and 1g/L urea showed a higher specific growth rate (0.066/day).

**Keywords:** *Chlorella sp*; POME; microalgae; biodiesel; biomass.

### 1. Introduction

Indonesia is a largest producer of crude palm oil (CPO) in the world. In 2008, Indonesia produced 44% of shared demand of CPO from around the world (Rupani et al., 2010) and from 2005 to 2008 the production rose up to 8.88%. Moreover, the production is still increasing as predicted from 2010 to 2014, it will grow at about 5.22% per annum (Table 1). About 1 ton fresh fruit bunch (FFB) can be converted to 0.2 ton CPO, while 0.66 ton will be released as palm oil mill effluent (POME). This enormous amount of POME was produced due to high amount of water used.

**Table 1.** Commodities of Indonesia agriculture 2010-2014

Commodities	Year			Growth/annum
	2011	2012	2013	
Coconut Palm	24.429	25.046	27.046	5.22%
Rubber	2.711	2.741	2.771	1.10%
Coconut	3.290	3.317	3.348	0.86%

Source: Deptan, 2009.

Almost of POME in Indonesia is treated by using open anaerobic ponds to reduce COD and BOD content. The raw POME originated from palm mill has high COD 50000 mg/L, BOD 25000 mg/L, while the pond is only able to reduce up to 1400 mg/L and 700 mg/L for COD and BOD, respectively.

**Table 2.** POME and treated POME characteristic

Parameter*	POME	Treated POME
pH	3.91-4.9	4-6
COD	83356	1400
TSS	49233.57	700
Total N	1494.66	456
NH <sub>3</sub> N	50.42	200
PO <sub>4</sub> P	315.36	18

Note: \*all in ppm except pH (Habib et al., 1998 and 2003).

The characteristic of POME before and after treated using anaerobic pond is listed in Table 2. From the composition, it is clearly seen that POME still contained high content of nutrient such as nitrogen and phosphorus even after treatment. This nutrient is highly potential for nutrient of microalgae growth especially to support the photosynthetic reaction to produce biomass. The biomass which depends on composition can be converted to other valuable product such as lipid for biofuel, protein for feed supplement or carbohydrate for bioethanol.

The need of renewable energy in Indonesia can not be avoided since within 15 years, the oil reserve will be diminished. Therefore, finding new source of renewable energy is really essential and vital aspect for Indonesia. One of the potential renewable energy resources is microalgae. The productivity of microalgae as compared to terrestrial plant is higher (Chisti, 2007) while the lipid content varies between 30-60%. Therefore, the objective of this paper is to utilize POME as growth medium of microalgae especially *Chlorella* sp.

## 2. Experimental Method

### 2.1. Cultivation Medium

Medium for cultivation, POME was collected from PTPN VII, Lampung. To be used as medium, POME was pretreated to eliminate solid materials and bacterial content.

### 2.2. Culture *Chlorella* sp

*Chlorella* sp was provided by BBPAP Jepara and cultivated in a modified medium with consists of 40 ppm urea, 30 ppm TSP, 10 ppm ZA, 1 ppm FeCl<sub>3</sub> and 25 µg/L vitamin B12.

*Chlorella* sp was cultivated in different POME concentration (20, 50 and 70%) and different low-high urea concentration (0.1 and 1 g/L). Light intensity was maintained in 3000 lux, pH 6.8-7.2, 28 °C temperature, and aerated using aquarium air pump to mix the medium in 1 L glass flask disk.

### 2.3. Measurement

The concentration of biomass was measured using spectrophotometer Optima sp-300, 680 nm wavelength for 15 days in every day. The optical density was plotted in biomass to make regression between optical density and biomass. Specific growth rate was calculated using equation from exponential growth (Eq 1):

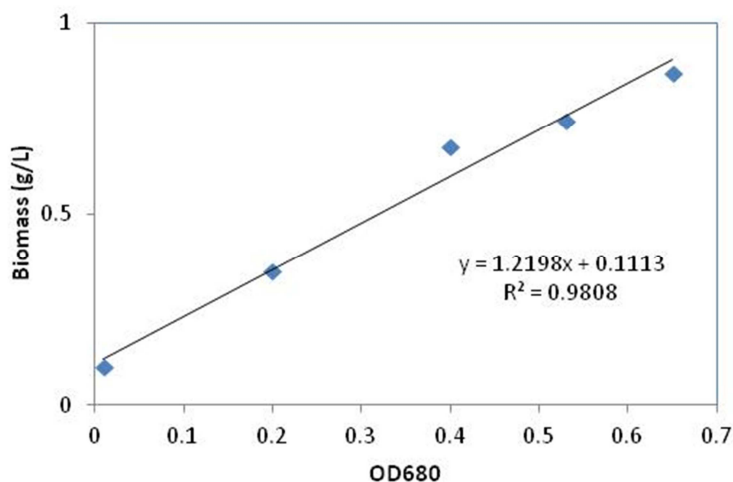
$$\mu = \frac{\ln(x_t) - \ln(x_0)}{t - t_0} \quad (1)$$

Biomass was harvested in the end of cultivation using control pH 11 (Oh et al., 2001). Biomass was dried at 55 °C tray dryer for 2 h and the productivity (x) (mg/L/d) was calculated using produced biomass divided by cultivation time.

## 3. Results and Discussion

### 3.1. Biomass vs Optical Density

Fig. 1 shows that the linear correlation between biomass and optical density was found. It was confirmed that OD 1 = 1.22 g/L. This is slightly different with the one obtained by Puangbut & Leasing (2012) with  $y = 1.5343x$ , ( $R^2 = 0.977$ ). The specific growth rate was determined by Eq 1 and the result is depicted in Table 3. The result indicates that 50% POME and 1 g/L urea has the highest specific growth rate ( $\mu$ ) than other variables.



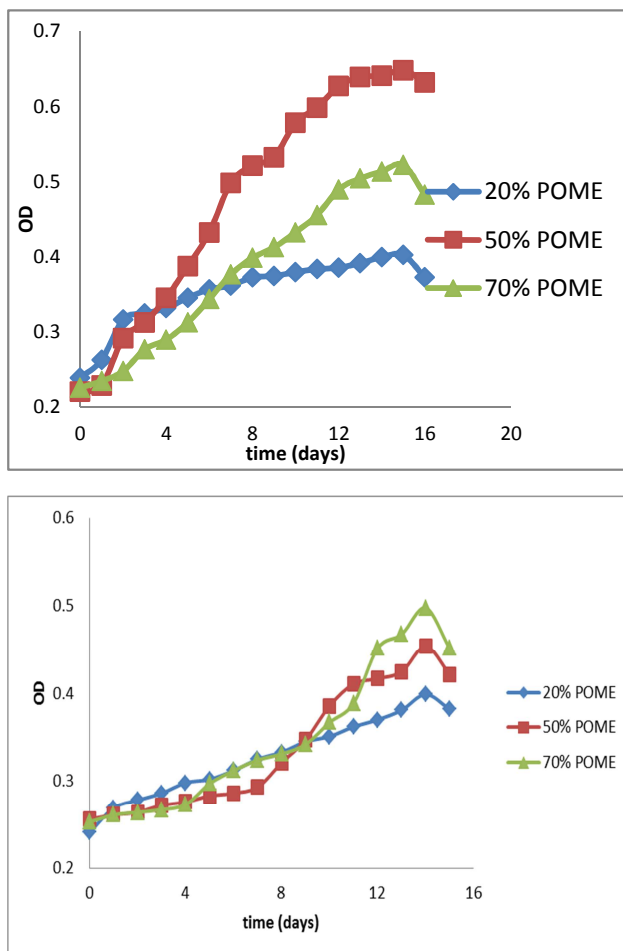
**Figure 1.** Correlation between OD and dry mass *Chlorella* sp cultivated in POME at 680 nm.

### 3.2. Effect of Urea Addition in POME to Algae Growth

The growth of algae in POME was evaluated by addition of external nutrient i.e. urea with concentration of 0.1 and 1 g/L. The result (Table 3) shows that *Chlorella* sp in this experiment has lower growth rate compared to the one obtained by Putri et al. (2011) with  $\mu = 0.084/\text{day}$ . The alga cell growth and the effect of urea in the medium are shown in Fig. 2. The use of 1 g/L urea as micro-nutrient enhanced the growth of algae both in achieving maximum optical density and shorter lag phases. This is an evidence that for completion of photosynthetic reaction, POME need additional nutrients.

**Table 3.** Specific growth rate ( $\text{day}^{-1}$ ) *Chlorella* sp in different POME and addition of urea concentration

Urea	POME concentration		
	20%	50%	70%
0.1 g/L	0.020	0.057	0.058
1 g/L	0.036	0.066	0.059



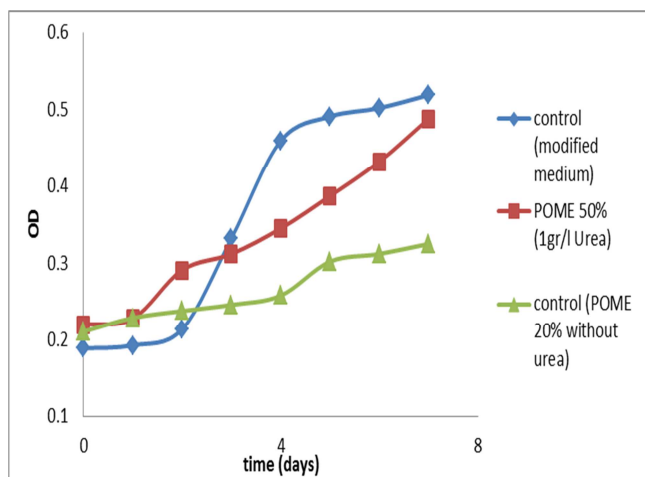
**Figure 2.** Growth phase *Chlorella* sp in different POME medium: (a) growth *Chlorella* sp in 1 g/L urea; (b) growth of *Chlorella* sp in 0.1 g/L urea.

### 3.3. Effect of Different Medium

The comparison of *Chlorella*'s growth both in control and POME medium were shown in Fig. 3. The control modified medium is optimum medium with composition of 40 ppm urea, 30 ppm TSP, 10 ppm ZA, 1 ppm FeCl<sub>3</sub> and 25 µg/L vitamin B12. In this medium, *Chlorella* can adapt and grow well achieving OD = 0.52 with growth rate of 0.85/day. The utilization of POME 50% and 1 g/L urea could achieve high density biomass of OD = 0.5, however, the growth rate is only 0.43/day, while without addition of urea shows even lower optical density.

### 3.4. Productivity

The productivity of *Chlorella* cultivated in POME medium is shown in Table 4. Algae cultivated in POME medium shows the highest productivity in 50% POME and 1 g/L urea achieving 58 mg/L/d. For 50-70% POME and 1 g/L urea show that the C:N:P ratio in the medium is closest to recommended CNP ratio for photosynthetic according to Edwards et al. (1980) i.e. 56:9:1 of weight ratio.



**Figure 3.** The comparison growth of *Chlorella* sp in different POME medium and control medium.

At higher POME concentration, the productivity is lower, due to tannin acid. This is supported by Habib et al. (2003) who stated that higher POME concentration influences dark brown color in medium which may come from tannic acid. Tannic acid can inhibit through shading effect in light intensity and photosynthetic reaction.

As compared to 50% POME (1 g/L urea), it has higher nitrogen ratio, but *Chlorella* still grow in higher specific growth rate. Lower tannic acid in 50% POME influence growth of microalgae, although nitrogen source is high but *Chlorella* still can tolerant to his condition. *Chlorella* sp has lower specific growth rate in POME 20% although the medium has lower tannic acid and more light intensities. This lower growth rate is caused by high nitrogen content. The excessive nitrogen content can be an inhibitor if it can not be utilized to form biomass.

**Table 4.** Biomass productivity (mg/L/d) in different concentration of POME and urea

Urea	POME concentration		
	20%	50%	70%
0.1gr/l	37.8	38.2	42
1gr/l	46.3	58.4	48.93

#### 4. Conclusions

The growth of *Chlorella* has been evaluated in POME medium by varying urea concentration. The highest specific growth rate and biomass productivity were recorded from 50% POME and 1 g/L urea. The high use of POME will be not visible due to the present of tannic acid and bacterial contents which may inhibit the growth of algae cell. This research gives a prospect of the use of POME as algae

medium, while the other benefits would be reduction of external nutrients such as  $\text{NaHCO}_3$ , urea and micronutrients. The advantages are the clean treated POME after the harvesting of algae biomass.

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