

## Carbon Dioxide Sequestration by Locally Isolated and Commercial Strain of Microalgae

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The main objective of this research is to investigate the growth rates and CO<sub>2</sub> sequestration potential of local and commercial strains of microalgae. Two local strains used were *Chlorella Sorokiniana* (UKM 2) and *Characium sp.* (UKM 1), both isolated from Palm Oil Mill Effluent (POME) and two commercial strains used were *Chlorella Vulgaris* and *Ankistrodesmus sp.*. The highest performance showed by *C. Vulgaris* where its growth rates was  $\mu_m = 0.47d^{-1}$  followed by *Ankistrodesmus sp.*  $\mu_m = 0.43d^{-1}$ , *C. sorokiniana*  $\mu_m = 0.4d^{-1}$  and *Characium sp.*  $\mu_m = 0.37d^{-1}$ . Based on the yield, *Ankistrodesmus sp.* had highest yield which is 3g/L, followed by *C. Vulgaris* 2.72g/L, *C. Sorokiniana* 2.52g/L and *Characium sp.* 2.27g/L. CO<sub>2</sub> sequestration rate was calculated based on the microalgae's yield where *Ankistrodesmus sp.* showed highest CO<sub>2</sub> sequestration, at 5.64g/L.

**Key words:** Locally isolated strain, commercial strain, growth profile, carbon dioxide sequestration.

Originally, the concentration of CO<sub>2</sub> in the atmosphere is 0.03%, however, due to an increased in human population, industrial activities, burning of fossil fuels and others the CO<sub>2</sub> concentration has reached up to 0.041% in July 2015 (Tan & Keeling, 2015). CO<sub>2</sub> is a part of greenhouse gases (Chiu *et al.*, 2011; Tebbani, Lopes, & Becerra, 2015) which potentially could cause dangerous climate changes. Furthermore, CO<sub>2</sub> tends to remain in the air longer than any other gases such as NO<sub>x</sub> and SO<sub>x</sub> (Rahaman *et al.*, 2011), and makes it very crucial to be eliminated first.

Studies by many researchers has shown that if the concentration of the CO<sub>2</sub> continue to increase, it would become a hazard to human life where Hansen (2007) stated in his report that if the

concentration of CO<sub>2</sub> reach 450ppm (0.045%), it would become dangerous and could be highly destructive to the world's global climate. This should be taken into serious consideration since the CO<sub>2</sub> emissions is proportional to the world's population growth rate which is estimated to increase exponentially by 1.198 every year (World Population Prospects, 2012).

Many techniques have been implemented to reduce the massive amount of CO<sub>2</sub> in the atmosphere. For example photosynthesis process (using tree to absorb CO<sub>2</sub>), geological formation (storage of CO<sub>2</sub> in deep ocean floor) and chemical techniques. However, these techniques had their certain drawbacks; trees has low growth rates and causing a very low absorption of CO<sub>2</sub> (Wang, Li, Wu, & Lan, 2008), geological formation require high cost (Bilanovic, Andargatchew, Kroeger, & Shelef, 2009) and chemical technique is very expensive and consumed a lot of energy (Rahaman *et al.*, 2011). Therefore, the proposed method was to use

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biological approach to fix CO<sub>2</sub> without damaging the environment.

Microalgae is a promising technique to absorb CO<sub>2</sub> due to its advantages. Microalgae has a very high growth rates which is 10 times better than trees (Cheng *et al.* 2013; Anjos *et al.* 2013) and has a very large surface area of absorption due to its small size (Cheng *et al.*, 2013; Wang *et al.*, 2008; Yusuf, 2007). In addition, culturing microalgae only takes 3% of the total land required to plant trees (Chisti, 2008). Furthermore, the biomass that produced from the cultivation can be used to produce pharmaceutical products (Ho, Chen, & Chang, 2012; Ho, Chen, Lee, & Chang, 2011), feed stocks, and other high value added products such as biofuels and biogas (Cheng *et al.* 2012; Kondili, & Kaldellis 2007; Rahaman *et al.* 2011; Yusuf 2007). Apart from that, microalgae have been proven to treat wastewater (effluent from industry) where the wastewater content nutrient that will render the microalgae's growth (Blair *et al.* 2014; Darzins *et al.* 2010; Wang *et al.* 2008).

Objective of this research was to study the growth rates and CO<sub>2</sub> sequestration potential of local strains and commercial strains. Two local strains, namely *C.Sorokiniana* and *Characium sp.*, and two commercial strains, namely *C.Vulgaris* and *Ankistrodesmus sp.* were used in this research. The growth rates of microalgae were determined using optical density (OD) for every 24 hour until it reaches the 10<sup>th</sup> day and the biomass was used to identify the amount of CO<sub>2</sub> sequestration by the microalgae.

## MATERIALS AND METHODS

### Microalgae strains culture condition

*C.vulgaris* was obtained from Algae Technology Malaysia, *Ankistrodesmus sp.* was obtain from Culture Collection of Algae at the University of Texas (UTEX), *C.Sorokiniana* (UKM 2) and *Characium sp.* (UKM 1) was isolated from Palm Oil Mill Effluent (POME). The microalgae were cultured in a conical flask with 1L of sterilized Bold Basal Medium (BBM)

The culture were sparged with CO<sub>2</sub> gas from air by using a pump (SB-988, China) through sterilized silicon tube with a 3mm internal diameter (ID). The flasks were covered using silicon tube stoppers. The flow rate used was 1L/min per flask.

Flow rates were maintained by installing a flow meter (Cole Parmer, USA) in each flasks. Light was continuously supplied by using a white fluorescent lamp at intensity 495  $\mu\text{mol m}^{-2} \text{s}^{-1}$  fluorescent (450 sunlight) (Apogee Instrument). The intensity was measured using lux meter (TES Digital Lux Meter, China) and it was control by having an enclosed system that cover the whole flask where the light was attached inside the enclosed system to ensure it is focusing on the microalgae only. Temperature was controlled at 30 and the pH was maintained at 7. Medium used was 1BBM.

### Determination of growth profile

Growth profiles of the strains were study. Comparisons had been made between local and commercial strains. All sets were duplicated to ensure the exactitude of the experiment. The microalgae's growth was determined using the optical density (OD) for every 24 h and the reading was taken using spectrophotometer (HACH DR 2800, USA). Each sample was diluted with a factor 10<sup>-1</sup> to give an absorbance in the range of 0.1-1.0. The equation used for growth rates of microalgae can be obtain from (Yang *et al.*, 2011) where;

Growth rate equation: , (1)

y is the OD at time t, y<sub>m</sub> is the maximum OD, y<sub>o</sub> is initial OD and is the maximum specific growth rates of microalgae.

### Determination of Biomass and CO<sub>2</sub> Sequestration Rate

Biomass was collected at the 10<sup>th</sup> day using filter paper (Whatman 0.45) with diameter 47mm and dried overnight in an oven (DHG 9146-A, China) at 105°C. It was then measured using an analytical balance (XB 220A, Switzerland). Later, the biomass was used to determine the CO<sub>2</sub> sequestration rate using equation developed by (Yusuf, 2007); 1.88 biomass productivity (yield). This equation was derived based on microalgal biomass CO<sub>0.48</sub>H<sub>1.83</sub>N<sub>0.11</sub>P<sub>0.01</sub>.

## RESULTS AND DISCUSSION

### Determination of Growth Rates

Growth rates of microalgae is determined more easily using the OD method (Rocha, Garcia, & Henriques, 2003). Growth rates of each strains were obtained by using the growth rates equation as shown in equation (1). For commercial strains

of microalgae, *C.Vulgaris* showed a better performance compared to *Ankistrodesmus sp.* where *C.Vulgaris* had and *Ankistrodesmus sp.* had only . Growth rate equation for *C.Vulgaris* was shown in equation (2) while growth rate equation for *Ankistrodesmus sp.* was shown in equation (3). Growth profile for *C.Vulgaris* and *Ankistrodesmus sp.* were shown in Figure 1 where the growth always led by *C.Vulgaris*.

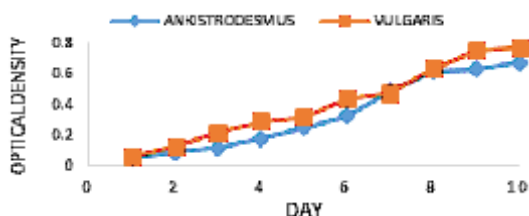
Growth rate equation of *C.Vulgaris*;  
 ... (2)

Growth rate equation of *Ankistrodesmus sp.*;  
 ... (3)

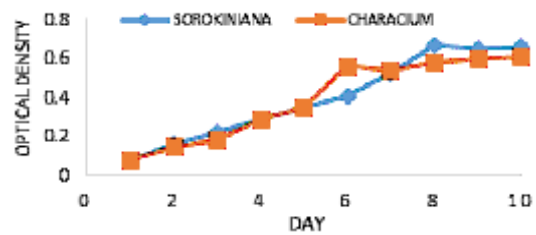
For local strains of microalgae, *C.Sorokiniana* showed a better performance compared to *Characium sp* where the specific growth rates ( of *C.Sorokiniana* was slightly higher than *Characium sp.* where its value was  $0.4d^{-1}$  while *Characium sp.* was only  $0.37d^{-1}$ . Same goes with the maximum optical density, *C.Sorokiniana* obtained 6.6 for the maximum optical density while *Characium sp.* obtained only 6. However, *Characium sp.* showed a better performance at an early phase of the growth as shown in Figure 2 where its OD value almost the same as *C.Sorokiniana* and at day 6, the value was slightly higher by 1.54 OD. It showed that, at the beginning of growth, *Characium sp.* can growth very well until all the nutrient is used up, then the growth started to decrease and slower than *C.Sorokiniana*. The overall equation for growth rates of *C.Sorokiniana* and *Characium sp.* can be obtain in equation (4) and (5).

Equation for growth rates of *C.Sorokiniana* ;  
 ... (4)

Equation for growth rates of *Characium sp* ;  
 ... (5)



**Fig. 1.** Growth profile of commercial strains of microalgae, *C.vulgaris* and *Ankistrodesmus sp.*. Both cultures were inoculated at 10% of the total medium prepared in duplicated

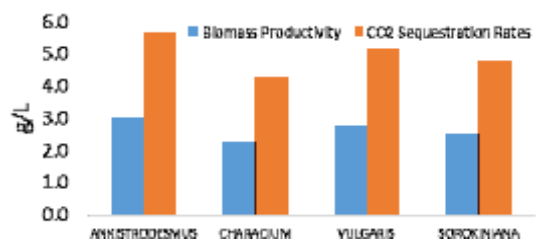


**Fig. 2.** Growth profile of local strains of microalgae, *C. sorokiniana* and *Characium sp.*. Both cultures were inoculated at 10% of the total medium prepared in duplicated

**Determination of Biomass and CO<sub>2</sub> Sequestration Rate**

The weight of the biomass was obtained and was analysed. Based on Figure 3, *Ankistrodesmus sp.* showed a high value of yield compared to other types of microalgae, which is 3g/L. Even though from previous section it was established that the maximum growth was demonstrated by *C.Vulgaris*, however for yield, *C.Vulgaris* showed a lower value compared to *Ankistrodesmus sp.*, which is only 2.72g/L.

Both commercial strains had higher growth rates and produced a larger amount of yield compared to local strain where *Ankistrodesmus sp.* yield was 3g/L yield and *C.Vulgaris* yield was 2.72g/L while *C.Sorokiniana* yield was 2.52g/L and *Characium sp.* was 2.27g/L only. The same result can be seen for CO<sub>2</sub> sequestration where again *Ankistrodesmus sp.* had the highest rate of CO<sub>2</sub> sequestration (5.64g/L). Hence, the commercial strains are better than the locally isolated strains in terms of both growth rates and CO<sub>2</sub> sequestration potential. This is mainly because isolated strains was extracted from the POME causing it to contain a lot of bacteria and other foreign substances which will reduce the



**Fig. 3.** Biomass productivity (yield) of 4 strains of microalgae and its potential in the CO<sub>2</sub> sequestration rate

effectiveness of the growth and ability to capture CO<sub>2</sub>. Nevertheless, both types of microalgae have proven in their capability and potential to be an agent for CO<sub>2</sub> sequestration.

### CONCLUSION

The commercial strains of microalgae (both *C. Vulgaris* and *Ankistrodesmus sp.*) gave a better performance compared to locally isolated strains of microalgae in terms of growth rates, yields, and CO<sub>2</sub> sequestration rate. Commercial strains had better performance due to its strain purities' where local strain was isolated and yet to be optimized for growth in BBM medium. The results of the CO<sub>2</sub> sequestration rate was consistent with previous studies where it is proportional to the yield obtained.

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