International Journal of Applied Science and Engineering
2011. 9, 3: 161-168

Analyzing Algae Growth and Oil Production in a Batch Reactor under high Nitrogen and Phosphorus Conditions

Tzu-Yi Pai [a,*] and Wei-Jia Lai [b]

[a.] *Department of Science Application and Dissemination and Master Program of Environmental Education and Management, National Taichung University of Education, Taichung, Taiwan, ROC.*
[b.] *Department of Environmental Engineering and Management, Chaoyang University of Technology, Taichung, Taiwan, ROC.*

Abstract: The feasibility for simultaneous local fresh water oleaginous algae cultivation and wastewater nitrogen removal were discussed in this study. The operation factors including illumination intensities, carbon source, initial nitrogen source, and retention time were fixed. The results indicated that the algae biomass increased from 28.3 mg/L to 254 mg/L at the 7th day. The oil weight increased from 5.75 mg/L to 85.34 mg/L at the 7th day. The percentage of algae oil increased from 20.4 % to 33.6 %, or by 13.2 %. The first-order equation was found to be the excellent fit model for describing the growth of algae biomass and production of oil content. The values of growth rate constant of algae and production rate constant were 0.3095 1/day and 0.3738 1/day, respectively. The value of production rate constant was about 1.2 times as that of the value of growth rate constant of algae, indicating an unbalance growth of algae biomass and oil content. The removal efficiency for ammonia nitrogen and phosphate was 84.8 % and 36.2 %, respectively.

Keywords: Algae biodiesel; wastewater nitrogen removal; wastewater phosphorus removal; carbon fixation.

1. Introduction

Algae are sunlight-driven cells that can be applied in bioremediation and as fertilizers for fixing nitrogen. Additionally, these photosynthetic microorganisms can transform carbon dioxide to potential foods and fuels.

Microalgae can provide renewable biofuels in several different ways including photobiological biohydrogen production [1-5]; methane production derived from anaerobic digestion of the algal biomass [6]; and biodiesel produced from microalgal oil [7-10].

Microalgae grow extremely rapidly and commonly double their biomass within 24 hours. For some species, biomass doubling times during exponential growth are shorter than 3.5 hours. Many microalgae are rich in oil. Depending on species, oil content in biomass can exceed 80% by weight of dry biomass [11, 6]. Oil levels of 20–50% are quite common [12]. The algal growth rate and the oil content of the biomass determine the oil productivity. Microalgae with high oil productivities are preferred for producing biodiesel.

* Corresponding author; e-mail: bai@ms6.hinet.net
Accepted for Publication: May 16, 2011

Various microalgae produce many different types of lipids, hydrocarbons and other complex oils [9, 13-14]. Not all algal oils are suitable for manufacturing biodiesel, but suitable oils exist in many microalgae species. Adopting microalgae to make biodiesel will not compromise production of food, fodder and other products derived from crops.

Because the shortage of petroleum and, more seriously, the emerging concern about global warming that is associated with burning fossil fuels [10], utilizing microalgae as fuel source becomes noticeably [15, 7]. But most microalgae must be cultivated using ocean water, i.e. the cultivation reactor must be established near seashore.

Wastewater treatment becomes more important in many countries due to the amount of wastewater from households and industry is steadily increasing every year with the explosion of population and development of industries [16-23]. The nitrogen and phosphorus in wastewater are regulated strictly meanwhile the nutrients including nitrogen and phosphorus are necessary for algae cultivation. If the problems including fixation carbon, carbon dioxide reduction, wastewater treatment, and biomass energy usage are taken into account, the algae are the important way for biodiesel production and nutrient removal.

Therefore the feasibility for simultaneous local fresh water oleaginous algae cultivation and wastewater nitrogen removal were discussed in this study.

2. Materials and Methods

2.1. Batch reactor

The cultivation system used to carry out the batch tests for culturing algae biomass is shown in Figure 1. It consists of circular chambers with the volume of 1 liter, a magnetic stirrer for stirring, and light source beside the chamber. The illumination intensities and temperature were controlled at 2300 Lux and 30 centigrade, respectively. There was no extra carbon source for local fresh water oleaginous algae cultivation. In addition, the initial nitrogen and phosphorus concentrations were varied.

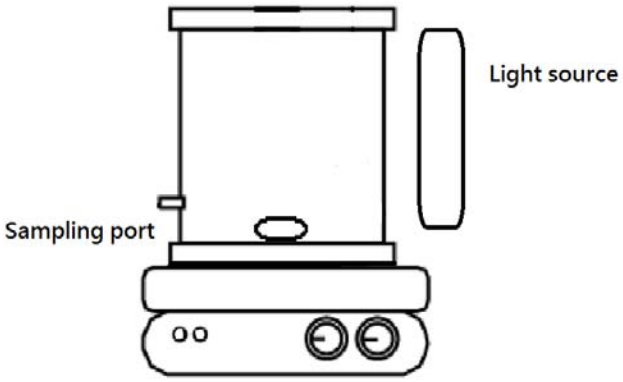

Figure 1. The oleaginous algae cultivation batch reactor

2.2. Analytical methods

Soxhlet extraction method was adopted to determine the oil content in algae biomass. In this procedure, the dry algae biomass was placed into the Soxhlet extraction apparatus for extracting the lipid. The analytical methods for algae biomass, pH, nitrogen, and phosphorus were performed according to Standard Methods [24]. The suspended solids were used to represent the algae biomass.

3. Results and Discussion

3.2. Algae biomass and oil content

Figure 2 shows the growth of algae biomass. The algae biomass increased from 28.3 mg/L to 254 mg/L at the 7th day. The concentration at the 7th day was about 9 times as that at initial time. The oil content in algae biomass revealed an unbalanced growth when comparing to the growth of algae biomass. The oil content increased from 5.75 mg/L to 85.34 mg/L at the 7th day as shown in Figure 3. The concentration at the 7th day was about 15 times as that at initial time. The percentage of algae oil increased from 20.4 % to 33.6 %, or by 13.2 % as shown in Figure 4. In accordance with Table 1, the oil content of algae in this study was close to the values of *Botryococcus braunii, Chlorella sp., Cylindrotheca sp., Isochrysis sp.,* and *Nannochloris sp.* [12].

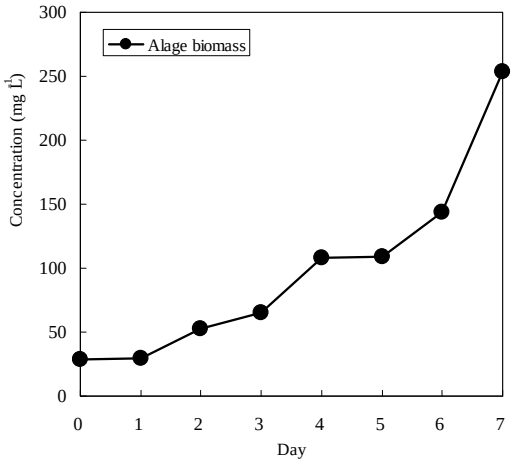

Figure 2. Growth of algae biomass

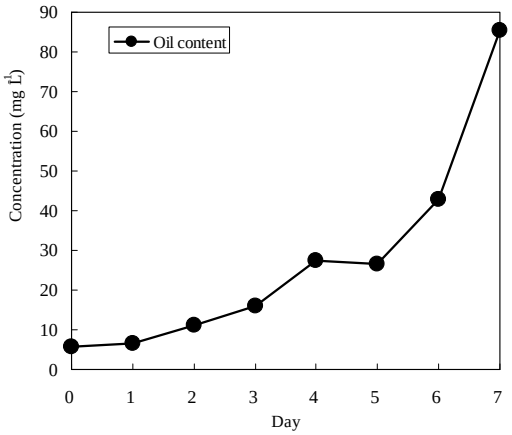

Figure 3. The concentration of algae oil content

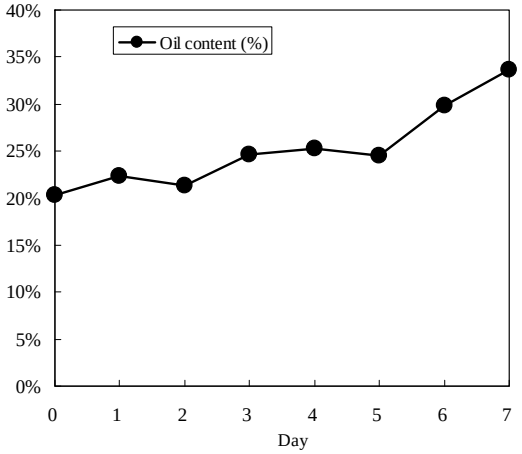

Figure 4. The percentage of algae oil content

Table 1. Oil content of some microalgae [12]

Microalga	Oil content (% dry wt)
Botryococcus braunii	25–75
Chlorella sp.	28–32
Crypthecodinium cohnii	20
Cylindrotheca sp.	16–37
Dunaliella primolecta	23
Isochrysis sp.	25–33
Monallanthus salina N	20
Nannochloris sp.	20–35
Nannochloropsis sp.	31–68
Neochloris oleoabundans	35–54
Nitzschia sp.	45–47
Phaeodactylum tricornutum	20–30
Schizochytrium sp.	50–77
Tetraselmis sueica	15–23
This study	20-37

Two first-order equations were employed to analyze the growth kinetic of algae and oil content as follows.

$$\frac{dC_{algae}}{dt} = k_{algae} C_{algae} \quad (1)$$

$$\frac{dC_{oil}}{dt} = k_{oil} C_{oil} \quad (2)$$

where C_{algae} is the algae concentration at time t (mg/L); k_{algae} is the growth rate constant of algae (1/day); t is time (day); C_{oil} is the oil weight at time t (mg/L); k_{oil} is the production rate constant of algae (1/day).

The experimental data of algae concentration and oil content were fitted to these two first-order equations by non-linear regression method. According to Figure 5 and Figure 6, first-order equation was found to be the excellent fit model for describing the growth of algae biomass and production of oil content. The R-squared values (R^2) in the first-order equations for describing the growth of algae biomass and production of oil content were 0.9679 and 0.973, respectively.

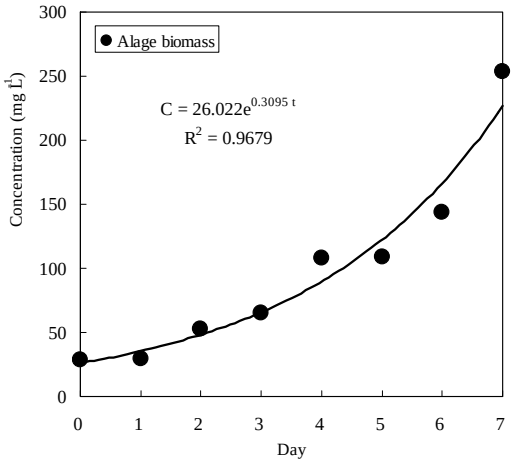

Figure 5. Curve fitting of algae biomass

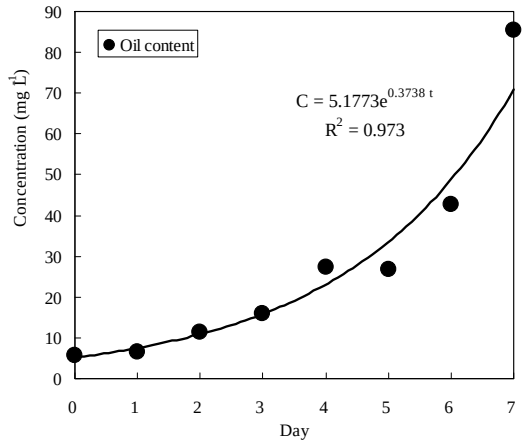

Figure 6. Curve fitting of algae oil content

These two equations were described as follows.

$$C_{algae} = C_{algae,0} e^{0.3095t} \quad (3)$$

$$C_{oil} = C_{oil,0} e^{0.3738t} \quad (4)$$

where $C_{algae,0}$ and $C_{oil,0}$ is the algae concentration and oil weight at initial time, respectively (mg/L).

Equation (3) and (4) showed that the values of k_{algae} and k_{oil} were 0.3095 1/day and 0.3738 1/day, respectively. The values of k_{oil} was about 1.2 times as that of the values of k_{algae}, indicating an unbalance growth of algae biomass and oil content. Because of the unbalance growth of algae biomass and oil content, some cultivation strategies including hydraulic retention time, nutrient dosage, or withdrawing algae biomass should be investigated to gain higher oil content in the further study.

3.2. Nutrient removal

The NH_3-N concentration was reduced from 33.0 mg/L to 5.0 mg/L as shown in Figure 7. Meanwhile the phosphate was reduced from 9.4 mg/L to 6.0 mg/L as shown in Figure 8. The removal efficiency for NH_3-N and phosphate was 84.8 % and 36.2 %, respectively.

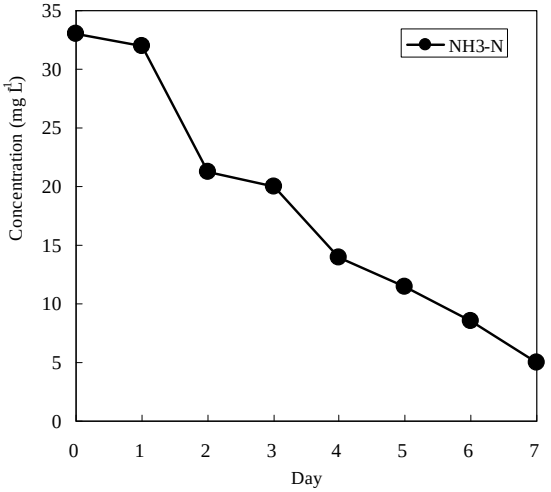

Figure 7. The reduction of NH_3-N concentration.

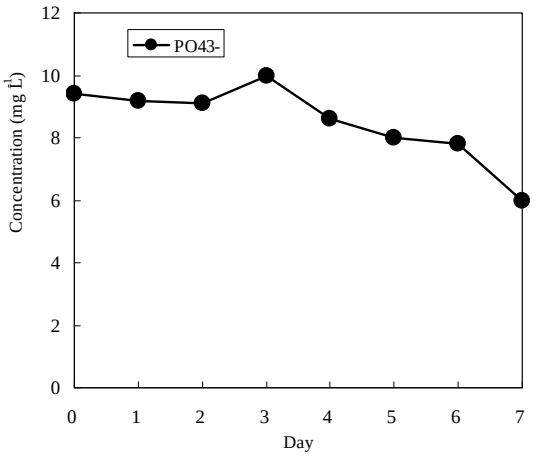

Figure 8. The reduction of phosphate concentration.

4. Conclusions

The feasibility for simultaneous local fresh water oleaginous algae cultivation and wastewater nitrogen removal were discussed in this study.

The results indicated that the algae biomass increased from 28.3 mg/L to 254 mg/L at the 7th day. The oil weight increased from 5.75 mg/L to 85.34 mg/L at the 7th day. The percentage of algae oil increased from 20.4 % to 33.6 %, or by 13.2 %.

The first-order equation was found to be the excellent fit model for describing the growth of algae biomass and production of oil content. The values of k_{algae} and k_{oil} were 0.3095 1/day and 0.3738 1/day, respectively. The value of k_{oil} was about 1.2 times as that of the values of k_{algae}, indicating an unbalance growth of algae biomass and oil content. The removal efficiency for ammonia nitrogen and phosphate was 84.8 % and 36.2 %, respectively. The local fresh water oleaginous algae are potential for oil production.

References

[1] Ghirardi M. L., Zhang L., Lee J. W., Flynn T., Seibert M., Greenbaum E., and Melis A. 2000. Microalgae: a green source of renewable H2. *Trends in Biotechnology*, 18, 12: 506-511.

[2] Akkermana I., Janssenb M., Rochac J. and Wij1elsd R. H. 2002. Photobiological hydrogen production: photochemical efficiency and bioreactor design. *International Journal of Hydrogen Energy*, 27(11-12): 1195-208.

[3] Melis A. 2002. Green alga hydrogen production: progress, challenges and prospects. *International Journal of Hydrogen Energy*, 27(11-12): 1217-1228.

[4] Fedorov A. S., Kosourov S., Ghirardi M.L. and Seibert M. 2005. Continuous H2 photoproduction by Chlamydomonas reinhardtii using a novel two-stage, sulfate-limited chemostat system. *Applied Biochemistry and Biotechnology*, 121 (1-3): 403-412.

[5] Kapdan I. K. and Kargi F. 2006. Bio-hydrogen production from waste materials. *Enzyme and Microbial Technology*, 38, 5: 569-582.

[6] Spolaore, P., Joannis-Cassan, C., Duran, E., and Isambert, A. 2006. Commercial applications of microalgae. *Journal of Bioscience and Bioengineering*, 101, 2: 87-96.

[7] Sawayama, S., Inoue, S., Dote, Y., and Yokoyama, S.-Y. 1995. CO2 fixation and oil production through microalga. *Energy Conversion and Management*, 36(6-9): 729-731.

[8] Dunahay T. G., Jarvis E. E., Dais S. S., and Roessler P. G. 1996. Manipulation of microalgal lipid production using genetic engineering. *Applied Biochemistry and Biotechnology*, 57-58: 223-231.

[9] Banerjee A., Sharma R., Chisti Y., and Banerjee U. C. 2002. Botryococcus braunii: a renewable source of hydrocarbons and other chemicals. *Critical Reviews in Biotechnology*, 22, 3: 245-279.

[10] Gavrilescu M. and Chisti Y. 2005. Biotechnology—a sustainable alternative for chemical industry. *Biotechnology Advances*, 23(7-8): 471-499.

[11] Metting F. B. 1996. Biodiversity and application of microalgae. *Journal of Industrial Microbiology & Biotechnology*, 17(5-6): 477-489.

[12] Chisti Y. 2007. Biodiesel from microalgae. *Biotechnology Advances*, 25: 294-306.

[13] Metzger P. and Largeau C. 2006. Botryococcus braunii: a rich source for hydrocarbons and related ether lipids. *Applied Microbiology and Biotechnology*, 66, 5: 486-496.

[14] Guschina I. A. and Harwood J. L. 2006. Lipids and lipid metabolism in eukaryotic algae. *Progress in Lipid Research*, 45, 2: 160-186.

[15] Nagle N. and Lemke P. 1990. Production of methyl-ester fuel from microalgae. *Applied Biochemistry and Biotechnology*, 24-25, 1: 355-361.

[16] Pai T. Y., Wang S. C., Lo H. M., Chiang C. F., Liu M. H., Chiou R. J., Chen W. Y., Hung P. S., Liao W. C., and Leu H. G. 2009a. Novel modeling concept for evaluating the effects of cadmium and copper on heterotrophic growth and lysis rates in activated sludge process. *Journal of Hazardous Materials*, 166, 1: 200-206.

[17] Pai T. Y., Wan T. J., Hsu S. T., Chang T. C., Tsai Y. P., Lin C.Y., Su H. C., and Yu L. F. 2009b. Using fuzzy inference system to improve neural network for predicting hospital wastewater treatment plant effluent. *Computers & Chemical Engineering*, 33, 7: 1272-1278.

[18] Pai T. Y., Wang S. C., Chiang C. F., Su H. C., Yu L. F., Sung P. J., Lin C. Y., and Hu H. C. 2009c. Improving neural network prediction of effluent from biological wastewater treatment plant of industrial park using fuzzy learning approach. *Bioprocess and Biosystems Engineering*, 32, 6: 781-790.

[19] Pai T. Y., Chang H. Y., Wan T. J., Chuang S. H., and Tsai Y. P. 2009d. Using an extended activated sludge model to simulate nitrite and nitrate variations in

TNCU2 process. *Applied Mathematical Modelling*, 33, 11: 4259-4268.

[20] Pai T. Y., Wang S. C., Lin C. Y., Liao W. C., Chu H. H., Lin T. S., Liu C. C., and Lin S. W. 2009e. Two types of organophosphate pesticides and their combined effects on heterotrophic growth rates in activated sludge process. *Journal of Chemical Technology and Biotechnology*, 84, 12: 1773-1779.

[21] Pai, T. Y., Wan, T. J., Tsai Y. P., Tzeng, C. J., Chu, H. H., Tsai, Y. S., and Lin, C. Y. 2010a. Effect of sludge retention time on biomass and kinetic parameter of two nitrifying species in anaerobic/oxic process. *CLEAN-Soil Air Water*, 38, 2: 167-172.

[22] Pai, T. Y., Chiou, R. J., Tzeng, C. J., Lin, T. S., Yeh, S. C., Sung, P. J., Tseng, C. H., Tsai, C.H., Tsai, Y. S., Hsu W. J., and Wei, Y. L. 2010b. Variation of biomass and kinetic parameter for nitrifying species in TNCU3 process at different aerobic hydraulic retention time. *World Journal of Microbiology and Biotechnology*, 26, 4: 589-597.

[23] Pai, T. Y., Chen, C. L., Chung, H., Ho, H. H., and Shiu, T. W. 2010c. Monitoring and assessing variation of sewage quality and microbial functional groups in a trunk sewer line. *Environmental Monitoring and Assessment*, 171(1-4): 551-560.

[24] APHA, AWWA, and WEF 1998. *"Standard Methods for the Examination of Water and Wastewater"*, 20th ed., American Public Health Association/American Water Works Association/Water Environment Federation, Washington, DC.