Nigerian Research Journal of Chemical Sciences **(ISSN: 2682-6054)**
Vol. 7, 2019

Microalgae for Biofuels - A Review

Haruna Ibrahim
Ibrahim ShehuShema Centre for Renewable Energy Research,
Umaru Musa Yar'Adua University, Katsina-Nigeria.
Email: ibrahimhauna@gmail.com

ABSTRACT

The ability of biofuels produced from algae to reduce environmental pollution significantly is driving the research for the development of algae-biofuel technologies. Two major methods are used for the cultivation of microalgae: the open pond and closed pond (photobioreactor). These micro-organisms utilize CO_2 from industrial pollution and convert it into biomass, fatty acids and lipids during photosynthesis. Hence the growth of algae cleans environment as well as provides feedstocks for fuels. From microalgae, biofuels such as biodiesel, bioethanol, methane and hydrogen, and food supplements, carotenes, are produced. However, the commercial development of microalgae is impeded by the cost of its harvest. Harvesting microalgae can be done by techniques such as centrifugation, filtration, sedimentation, flocculation, flotation, ultrasonic vibration, and screening. Flocculation followed by centrifuge or filtration, a two-step method, is cost effective for harvesting of microalgae.

Keywords: Biofuels, cultivation, development, harvest, microalgae

INTRODUCTION

With ever growing demand for fossil fuels and environmental degradation caused by burning of fossil fuels, research for development of a sustainable alternative fuel is carried out world over. The triglycerides from current seed crops cannot meet the demand for diesel. The potential of photosynthetic microorganisms as an alternative to biofuel crops together with their potential as a promising technology for CO_2 utilization form another area of research interest [1]. The ability of biofuels produced from algae to reduce environmental pollution significantly is, in turn, driving the research for the development of algae-biofuel technologies [2]. Benemann [3] reported that the concept of microalgae for CO_2 utilization and conversion to fuels dates back to 1960s and it has been developed experimentally and conceptually. The biodiesel produced worldwide is about 13 billion gallons per year of which USA consumes over 60 billion gallons

per year [4]. The present world biodiesel output is far less than quantity of diesel US consumes. Recently, microalgae have received much attention as a renewable energy resource because the photoautotrophic mechanism can convert atmospheric carbon dioxide into biomass, fatty acids, and lipids [5]. Development of microalgae for biofuels will go a long way to providing energy security and curb climate change.

This study describes the utilization of microalgae for CO_2 fixation, species of algae, cultivation, harvesting and the products derivable from the micro-plants.

Microalgae

Microalgae are microscopic photosynthetic organisms found in both marine and freshwater environments [6]. The potential of photosynthetic microorganisms as an alternative to biofuel crops, higher oil yield, growth on a non-agricultural land and as a promising technology for CO_2 fixation is a subject of strong interest [1]. These organisms use energy naturally from the sun to combine water with carbon dioxide (CO_2) to create biomass in aquatic environments [6]. Algae are a large and diverse group of simple plant like organisms, ranging from unicellar to multicellar forms [7]. Algae are non-food resources that can be grown on non-productive land for its reach oil for biodiesel. It has higher productivity than its cousin food crop oil sources [4]. Microalgae have high biomass productivity, high lipid content, with low cultivation cost [8]. Microalgae have high growth rate, short growth time, and low land usage compared to sister crops biofuel feedstocks [9].

Extensive investigations have been carried out on the nutritional and toxicological safety of the algal biomass [10]. It can utilize waste CO_2 that could cause environmental hazard [4].

Pokoo-Aikin *et al*, [7] classified microalgae into nine major groups which are cyanobacteria (cyanophyceae), green algae (chlorophyceae), diatoms (bacillariophyceae), yellow-green algae (xantophyceae), golden algae (chrysophyceae), red algae (rhodophyceae), brown algae (phaeophyceae), dinoflagellates (dinophyceae) and 'pico-plankton' (prasinophyceae and eustigmatophyceae). The diatoms (bacillariophyceae) dominate the phytoplankton of the oceans, but are also found in fresh and brackish water with approximately 100,000 species are known to exist. The green algae (chlorophyceae) are also found abundant in freshwater, even swimming pool, stored mainly as starch and oils. The blue-green algae (cyanophyceae) much closer to bacteria in structure, is good in fixing nitrogen from the atmosphere and there are about 2,000 known species found in a variety of habitats. The golden algae (chrysophyceae) are similar to the

diatoms with more complex pigment systems, and can appear yellow, brown or orange in color. About 1,000 species are known to exist, primarily in freshwater systems, store natural oils and carbohydrates. The following strains of microalgae are found in India: t*olypothrix*, *pithophora, spirogyra, hydrodictyon*, *rhizoclonium* and c*ladophora* [11]. Sialve *et* al [1] listed the following microalgae: euglena gracillis, chlamydomonas reinhadtii, chlorella pyrenoidosa, chlorella vulgaris, and scenedesmus obliquus. Unicellular structure of microalgae help them to utilize solar energy more efficiently in the manufacturing food, lipid, oil, etc, than higher plants [6].

Microalgae, as plants, store energy as carbohydrates and lipids, and these lipids are similar to those produced by row crops such as soybean, sunflower, Jatropha, etc [12]. Microalgae have much more oil than macroalgae and it is much faster and easier to grow and harvest [11]. Microalgae can produce more biomass per unit land area than agricultural crops depending on the technology used and the local climate [13].

Cultivation

Algae cultivation is usually achieved by two methods: open ponds and closed pond, called photobioreactors (PBR). The standard open pond commercial technology for microalgae biomass production uses raceway type, paddlewheel mixed pond [3]. Much progress has been made in increasing the yield through photo bioreactor design, selection of strains and genetic engineering of metabolic pathways [14]. Production of microalgae requires land, light, water, CO_2 and nutrients (macro and micro nutrients) [4]. One of the major interests attracting the cultivation of microalgae is the reduction of industrial CO_2 waste. CO_2 is a common industrial pollutant. Thus microalgae can contribute to reducing atmospheric CO_2 by consuming CO_2 wastes from industrial sources such as power plants [7]. The idea of producing methane gas from algae was proposed in the early 1950s. During that period, early researchers visualized a process in which wastewater could be used as a medium and source of nutrients for algae production [6]. Microalgae are highest CO_2 fixation and O_2 production. Growing in liquid medium, which can be handled easily, make algae to stand high in front of other oil seed crops [11]. Survey conducted by Loubie`re et al [15] at six main hatcheries in the French West Coast, of microalgae production presented in 2002, showed that for flagellate microalgae, from 400 to 6,000 billion cells per day were produced, and for diatoms, from 900 to 20,000 billion cells per day were produced. Previous studies have demonstrated that lipid content in some microalgae could be

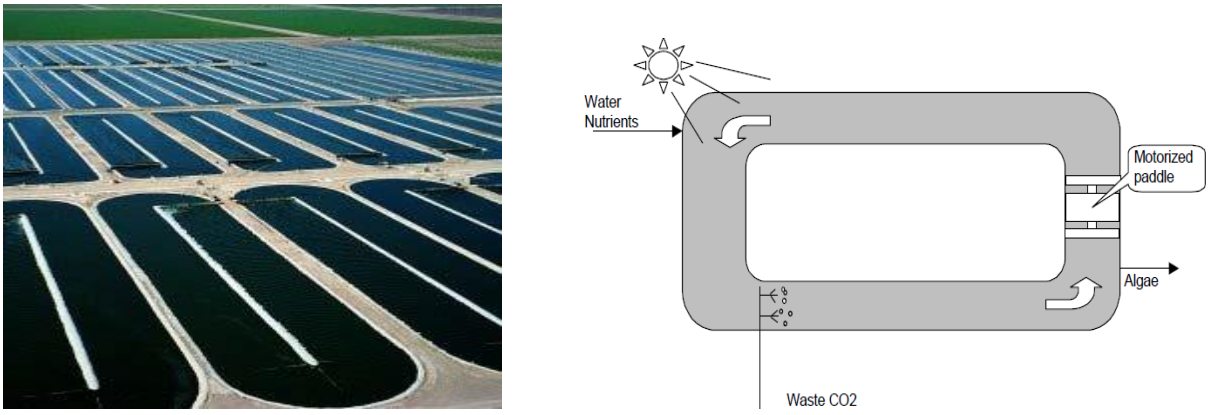
increased by various cultivation conditions such as nitrogen deprivation, high light intensity, low temperature, high salt concentration and high iron concentration [5].

Opened ponds

Open ponds are the most widely used systems for commercial cultivation of microalgae because they are considered more energy-efficient and require less investment for construction and operation than closed reactors [16]. Open pond culturing system is simpler and cheaper to construct, easier to operate and maintain, but easily contaminated, difficult to control temperature and light utilization and water. Culture systems can be carried out using shallow (10 cm deep) ponds stirred with paddle wheels in areas of high solar insolation autotrophically with simple inorganic media [4].

The microalgae may also be contaminated with heterotrophic bacteria even when it is grown [10]. It was reported by Loubie`re et al [15] that currently microalgal cultivation in French hatcheries is mainly performed by means of batch culture systems consisting of vertical aerated column reactors. This method has some drawbacks: a partial control of biomass quality and quantity, low productivities, contamination, requires frequent handling and cleaning operations, biofouling.

Figure 1 (a), depicts an algae farm with many ponds. Algae, water and nutrients are charged into the pond. The ponds are equipped with paddlewheels which keep algae flowing round the racetrack as indicated in figure 1(b). The depth of open ponds in most cases about 10 cm. This keeps algae exposed to sunlight [6]. The system is flexible, relatively easily scalable and of low cost which is being used in U.S. to produce high value food supplement; beta carotene [3].

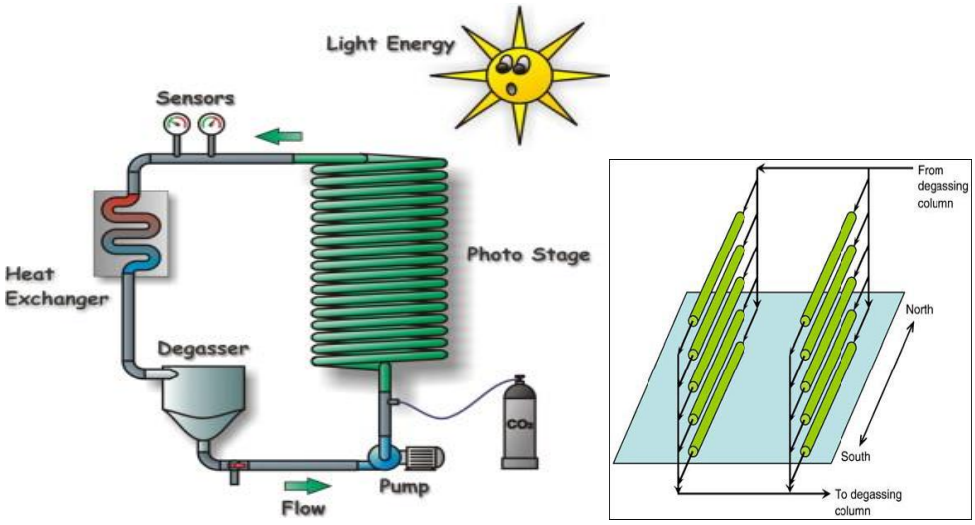


a) A farm with multiple ponds b) A unit pond

Figure 1: Opened ponds for microalgae production

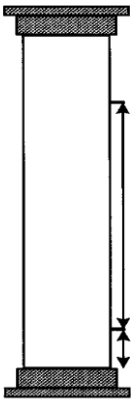
Closed ponds

A photobioreactor are closed equipment working either outdoors or indoors, in which a single species is inoculated to keep a clean-culture operation [17] which provides a controlled environment and enables high productivity of algae. It consists of straight, coiled, or looped transparent tubing laid out in a specific geometric arrangement designed to maximize light capture [18]. In closed ponds, microalgae are cultured in closed vessels equipped with light to catalyze the photosynthetic reaction. The closed vessels equipped with light are therefore called photobioreactors. Closed cultivation systems offer better control of contamination and cell physiology than open systems, leading to higher growth and quality of the harvested product, but increased manufacturing costs [17]. Being a closed system, all growth requirements of algae are introduced into the system and controlled according to the requirements. They could be glass tubes or acrylic tubes that allow light penetration to be utilized by the microalgae for manufacturing of lipid triglycerides and others. Advantages of closed pond culturing system includes: prevention of loss of water due to evaporation and suitable for sensitive strains. The closed pond culturing also prevents degradation of soil and underground water. Some of the popular photobioreactors used for culturing microalgae include: bubble column, flat panel and coil.



a) Coil b) Flat panel

b) Inclined bubble column



d) Bubble Colum

Figure 2: Photobioreactors

Culture Media

Different media have been used to culture microalgae at different culturing conditions. Maji et al., [13] cultured *Chlorella vulgaris, Scenedesmus obliquus* and *Cholorococcum sp* in Bristol's medium in a 250 mL conical flasks under controlled conditions (20±1°C temperature; 31±1% relative humidity; 140 µmol photons/m^2/s; 12:12 h light/dark cycle). Cultures were aerated for 8 hours daily with atmospheric air (0.05 L.s-1). Rinanti and Purwadi [19] cultured *Ankistrodesmus*sp and C. *vulgaris Provasoli Haematococcus m*edia in controlled incubators with a rotational speed of 100 rpm, 25 °C, fed with 2% pure CO_2 with a flow rate of 5 $Lmin^{-1}$, illuminated by fluorescent lamps (50 µmolphotons m^{-2} s^{-1}) with 16/8 hour light/dark cycle. Salim et al [14] cultured Chlorella vulgaris and Scenedesmus obliquus marine medium contained NaCl (27.00 g L^{-1}), $MgSO_4 \cdot 7H_2O$ (6.60 g L^{-1}), $MgCl_2 \cdot 6H_2O$ (5.60 g L^{-1}), $CaCl_2 \cdot 2H_2O$ (1.50 g L^{-1}), KNO_3 (1.45 g L^{-1}), $NaHCO_3$ (0.04 g L^{-1}), TRIS (hydroxymethyl) aminomethane (3.94 g L^{-1}), EDTA-Na_2 (95 µg L^{-1}), $ZnSO_4 \cdot 7H_2O$ (11 µg L^{-1}), $CoCl_2 \cdot 6H_2O$ (5 µg L^{-1}), $MnCl_2 \cdot 4H_2O$ (90 µg L^{-1}), $Na_2MoO_4 \cdot 2H_2O$ (30 µg L^{-1}) and $CuSO_4 \cdot 5H_2O$ (5 µg L^{-1}) dissolved in demineralized water. For the freshwater medium KNO_3 (3 g L^{-1}), $NaH_2PO_4 \cdot 2H_2O$ (0.26 g L^{-1}), KH_2PO_4 (0.74 g L^{-1}), HEPES (2.38 g L^{-1}), H_3BO_3 (61.80 µg L^{-1}), EDTA-Fe (III)-Na, (0.11 g L^{-1}), EDTA-Na_2 (37 mg L^{-1}), $ZnSO_4 \cdot 7H_2O$ (3.20 mg L^{-1}), $MnCl_2 \cdot 4H_2O$ (13 mg L^{-1}) and $CuSO_4 \cdot 5H_2O$ (1.83 mg L^{-1}) in demineralized water. The lipid content of a number of microalgal species has been found profoundly to be high with Nitrogen limitation/deficiency [2].

Harvest

Harvesting process involves separation of microalgae from the growth medium. Due to their small size, negative surface charge and similar densities to that of water, harvesting is a difficult task [13]. Harvesting microalgae can be done by techniques such as centrifugation, filtration, sedimentation, flocculation, flotation, ultrasonic vibration, and screening [13, 19]. All these methods require high energy inputs for harvesting of the algal cells which makes large scale production of microalgae for target products uneconomical [13]. Salim et al [14] reported that studies have showed that for open ponds, the contribution of the costs for harvesting is more than 30% of the total cost of algal production. Branyikova et al [20] reported that a two-step harvesting process canbe cost-effective: a low-cost technique such as flocculation and then followed by the energy consuming physical cell separation processes (centrifugation or filtration) that require expensive equipment. Current harvesting methods include biological, chemical, mechanical, and to a lesser extent, electrical operations [20]. Salim et al [14] claimed that evaluation of several harvesting methods showed that flocculation combined with flotation or sedimentation and subsequent further dewatering by centrifugation or filtration is the most promising cost and energy efficient alternative. Flocculation or sedimentation is a most promising technique due to its low cost and energy requirements [13]. Chemical flocculation can be performed using Zn^{2+}, Al^{3+}, Fe^{3+} or other chemical flocculants easily but expensive and not environmentally friendly [19]. The flocculants need to be removed first before the residual water is discharged into water bodies, hence additional cost. Chemical methods of flocculation using inorganic and organic flocculants are in practice but have contributed to high cost and maintenance [13]. Bioflocculation involves flocculation induced by polysaccharidesand proteins derived from microorganisms [13] with bacteria, fungi, or microalgae as flocculants [19].

Rinanti and Purwadi, [19] harvested microalgae by biofloculation using *S. obliquus* microalgae as floculant and was found to be very effective and promising. Maji et al., [13] harvested microalgae in different P^H media and found that S. *obliquus* and *Chlorococcum*sp flocculated higher at lower P^H while *C. vulgaris* flocculate at higher P^H. Salim et al., [14] harvested Chlorella vulgaris and Scenedesmus obliquus by autoflocculating and found that autofloculating microalgae induced faster sedimentation of non-flocculating microalgae and also increased the harvesting efficiency. The term autoflocculation is flocculation caused by secreted extracellular biopolymers (EPS) whereas bioflocculation involves the use of other microorganisms [20].

Product Derivable from Microalgae

From microalgae, different types of biofuels can be derived which includes: methane produced by anaerobic digestion of algal biomass, biodiesel derived from microalgal oil and photo-biologically produced bio-hydrogen [11]. An algal biorefinery could produce oils, protein, and carbohydrates [6]. Sialve *et al* [1] reported that apart from the lipid, which can be converted to biodiesel, biomass can be transformed into bioethanol, anaerobic digestion of microalgae can also produce methane. They also claimed that ammonia is produced along with methane during anaerobic digestion of microalgae. Many algae are exceedingly rich in oil, which can be converted to biodiesel [21]. It is understood that microalgae can be utilized to produce three different biofuels namely, biodiesel, bioethanol and methane which can be further processed to biomethanol. As emphasis switched to production of natural oils for biodiesel, microalgae became the one of the most suitable source because they produce more of the right kinds of natural oils needed for biodiesel.

Table 1 provides the bio-molecular composition of some species of microalgae [1]. With their protein and carbohydrate contents, microalgae are good source of feed for animals. Loubie`re et al., [15] claimed that the following microalgae are commonly cultivated for feeding mollusks; *Isochrysis affinis galbana, Pavlova lutheri, Chaetoceros muelleri, Chaetoceros calcitrans, Thalassiosira pseudonana, Skeletonema marino.* Implantation of microalgal biorefineries contributes in the area of CO_2 fixation from combustion gases, wastewater treatment, and recycling of nutrients [16]. It was also reported by Kiman et al., [8], that microalgae is extensively used today in various businesses and industries involved in supplementary health products, cosmetics, medicine, aquaculture and bioenergy. Above all algal biodiesel is one of the only avenues available for high-volume re-use of CO_2 generated in power plants [6]. Microalgae growth requires a lot of CO_2 hence, mitigation of CO_2 waste from industries that is a major source of greenhouse gas (GHG).

Table 1: Molecular composition of some strain of microalgae [1]

Microalgae strain	**Protein (%)**	**Lipid (%)**	**Carbohydrate (%)**
Euglena gracillis	39-61	14-20	14-18
Chlamydomonas, reinhadtii	48	21	17
Chlorella pyrenoidosa	57	2	16

Chlorella vulgaris	51-58	14-22	12-17
Dunaliellasalunia	57	6	32
Spirullinamaxaima	60-71	6-7	13-16
Spyrullinaplatenis	46-63	4-9	8-14
Scenedesmusobliquus	50-56	12-14	10-17

CONCLUSION

Large areas of the globe that are not suitable for high level terrestrial agricultures can be used for growing microalgae. From microalgae, biofuels such as bioethanol, biodiesel, biohydrogen, and methane and food supplement carotene can be produced. Large scale commercialization of microalgae cultivation is hampered by cost of harvest. A two-step harvesting process can be cost-effective: a low-cost technique such as flocculation and then followed by the energy consuming physical cell separation processes (e.g., centrifugation or filtration) that require expensive equipment.

REFERENCES

1. Sialve B., Bernet, N. & Bernard O. (2009). Anerobic digestion of microalgae as a necessary step to make microalgal biodiesel sustainable. *Biotechnology Advances*, 27, 409-416.
2. Mallick, N., Bagchi, S. K., Koley, S. & Singh, A. K. (2016). Progress and Challenges in Microalgal Biodiesel Production. Review. *Frontiers in Microbiology* doi: 10.3389/fmicb.2016.01019
3. Benemann, J. R. (1997). CO_2 Mitigation with Microalgae Systems. *Energy Convers. Mgmt*. Vol. 38 Suppl. ppS475-S479.
.4. Pienkos, P. T. (2007). The Potential for Biofuels from Algae, Algae Biomass Summit San Francisco, National Renewable Energy Laboratorym, CA.
5. Hsieh, C., Wu, W. (2009). Cultivation of microalgae for oil production with a cultivation strategy of urea limitation. *Bioresource Technology*, 100(17), 3921–3926.
6. Sheehan, J., Dunahay, T., Benemann, J. & Roessler, P. (1998). A Look Back at the U.S. Department of Energy's Aquatic Species Program: Biodiesel from Algae, National Renewable Energy Laboratory
7. Pokoo-Aikins, G., Nadim, A., El-Halwagi, M. M. & Mahalec, V. (2009). Design and analysis of biodiesel production from algae grown through carbon sequestration.

Clean Technique Environmental Policy (2010) 12:239–254 DOI 10.1007/s10098-009-0215-6

8. Kiman, S., Highina, B. K., Hamza, U. & Hala, F. (2016). Extraction of Biodiesel from Microalgae by Direct *In Situ* Method. University of Maiduguri Faculty of Engineering Seminar Series, Volume 7, 7-11.
9. Lee, J., Yoo, C, Jun, S., Ahn, C. & Oh, H. (2009). Comparison of several methods for effective lipid extraction from microalgae. *Bioresource Technology*, Elsevier doi:10.1016/j.biortech.03.058 1-3
10. Mahadevaswamy, M. & Venkataraman, L. V. (1981). Microbial load in mass cultures of green algae *Scenedes musacutus* and its processed powder. *J. Biosci.*, 3 (4), 439-448.
11. Kumar, P., Suseela, M.R. & Toppo, K. (2011). Physico-Chemical Characterization of Algal oil: a Potential Biofuel. *Society of Applied Science, Asian Journal Experimental Biology, 2(3) 493-497*
12. Putt, R. (2007). Algae as a Biodiesel Feedstock: A Feasibility Assessment. Center for Microfibrous Materials Manufacturing (CM^3) Department of Chemical Engineering Auburn University, Alabama 1-45.
13. Maji, G. K., Choudhury, S., Hamid, S., Prashanth, R., Arjunan, S. & G1, S. (2018). Microalgae Harvesting via Flocculation: Impact of P^H, Algae Species and Biomass Concentration. *Methods of Microbiology and Molecular Biology*, Volume 1 Issue 2, 1-5, ISSN 2517-7435.
14. Salim, S., Bosma, R., Vermuë, M. H. & Wijffels, R. H. (2010). Harvesting of microalgae by bio-flocculation. *Journal of Applied Phycol*, 23, 849–855. DOI 10.1007/s10811-010-9591-x
15. Loubie`re, K., Olivo, E., Bougaran, G., Pruvost, J., Robert, R, & Legrand, J. (2009). A New Photobioreactor for Continuous Microalgal Production in Hatcheries Based on External-Loop Airlift and Swirling Flow, *Biotechnol Bioeng.* 102(1):132-47. doi: 10.1002/bit.22035.
16. Costa, J. A. V., Catarina, B., Freitas, B, Santos, T. D., Mitchell, B. G. & Morais, M. G. (2019). Chapter-9 Open pond systems for microalgal culture. Sustainable Platform for Fuels, Chemicals and Remediation, Biofuel from Algae (2^{nd} Edition) 199-223
17. García-Garibay, L M., Gómez-Ruiz, A.E., Cruz-Guerrero, E.B. (2014). Single Cell Protein: The Algae**.** Encyclopedia of FoodMicrobiology **(**Second Edition**)**
18. Burgess, S. J., Tamburic, B., Zemichael, F., Hellgardt, K. & Nixon, P. (2011). Solar-Driven Hydrogen Production in Green Algae. *Advances in Applied Microbiology*, 75, 71-110

19. Rinanti, A. & Purwadi, R. (2018). Harvesting of freshwater microalgae biomass by *Scenedesmus* sp. as bioflocculant. The 4th International Seminar on Sustainable Urban Development: IOP Conference Series, Earth and Environmental Science, Volume 106 Conference 1, 012087 doi: 10.1088/1755-1315/106/1/012087.
20. Branyikova, I., Prochazkova, G., Potocar, T., Jezkova, Z. & Branyik, T. (2018). Review: Harvesting of Microalgae by Flocculation. *Fermentation, 4*, 93; doi:10.3390/fermentation4040093 Available at: www.mdpi.com/journal/fermentation
21. Patil, V., Tran, K. & Giselrød, H. R. (2008). Towards Sustainable Production of Biofuels from Microalgae *Review*. *International Journal of Molecular Sciences*, ISSN 1422-0067 www.mdpi.org/ijms/, *9*, 1188-1195; DOI: 10.3390/ijms9071188.