ISSN 1682-296X (Print)
ISSN 1682-2978 (Online)

Bio Technology

ANSInet
Asian Network for Scientific Information
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

Biotechnology 11 (3): 127-132, 2012
ISSN 1682-296X / DOI: 10.3923/biotech.2012.127.132

Microalgal Culture Systems: An Insight into their Designs, Operation and Applications

Charles U. Ugwu and Hideki Aoyagi
Graduate School of Life and Environmental Sciences, University of Tsukuba,
1-1-1 Tennoudai, Tsukuba, Ibaraki 305-8572, Japan

Abstract: Mixing is of paramount importance to microalgal cultures. There are various methods of inducing mixing in microalgal cultures; however, the type of mixing to be adopted would depend on various factors such as, the type of microalgal strain, type of culture system (i.e., open ponds or photobioreactors), scale of culture systems (i.e., small or large-scale cultures), as well as, on the environment where the culture is operated (i.e., indoor or outdoor type). In any case, mixing is mainly done to improve the mass transfer efficiency in the culture broth and to maintain efficient distribution of gases (air, oxygen, carbon dioxide, etc.) and nutrients. Furthermore, efficient mixing would improve the light utilization by the microalgal cells and thus, enhance biomass productivities. It is therefore, important to implement efficient mixing to maximize the potentials of microalgae during cultivation. This paper reviews some strategies to achieve mixing in microalgal culture systems, with more emphasis on photobioreactor designs, operation and applications.

Key words: Mass transfer, microalgae, mixing, photobioreactor, open ponds

INTRODUCTION

Microalgae are cultivated for a variety of purposes such as in, providing food supplements and bioactive compounds (Borowitzka, 1999; Ugwu *et al*., 2008; Akpolat and Eristurk, 2008), space research (Yoshihara *et al*., 1996; Ai *et al*., 2008) and for biofuel production (Chisti, 2007; Azmat *et al*., 2007). Microalgae can be grown in open ponds or photobioreactors. However, open ponds are currently, the preferred option for commercial production of algae. Despite that commercial production of microalgae is done in open ponds, efforts to prevent contamination and control the culture conditions have been very challenging. Photobioreactor, on the other hand, has attracted much interest in recent years given their potential uses in growing microalgae under culture-controlled conditions. Photobioreactor (PBRs) are used for growing algae mainly for production of high-value compounds (e.g., biomass, vitamins, amino acids, colorants etc.). Most microalgal cultures are aerated with air and carbon dioxide; however, their ability to give desirable yields of algal biomass and products would depend on their designs, hydrodynamics, mixing conditions, as well as on their mass transfer efficiencies. Studies have indicated that oxygen transfer efficiency is one of the most important criteria for assessing the performance of bioreactors (Rubio *et al*., 1999; Ugwu *et al*., 2002; Sauid and Murthy, 2010). Mixing is known to play some important roles in microalgal cultures (Thomas and Gibson, 1990; Ugwu *et al*., 2008; Eriksen, 2008).

This study reviews the potentials and drawbacks of some microalgal cultures when they are operated in photobioreactors and open ponds and then highlights on how their performances can be improved by applying principles of mixing, hydrodynamics and mass transfer.

OPEN PONDS

Raceway ponds equipped with paddle wheels are the most commonly used systems for mass cultivation of algae (Oswald, 1988; Boussiba *et al*., 1988; Hase *et al*., 2000). One of the problems encountered in open ponds is contamination by protozoa, ciliates and bacteria etc. Most microalgae that have their optimum growth at neutral or lower pH cannot be operated for long time in open ponds. Contamination can be avoided by cultivating some strains at high pH since only few contaminants can survive under this condition. One algal strain which survives under high pH is Spirulina (Vonshak and Richmond, 1988; Oncel and Akpolat, 2006). Another commonest strain which is commercially grown in open ponds is *Dunaliella* sp. (Abd El-Baky *et al*., 2004). *Dunaliella* sp. is commercially cultivated in ponds because it can grow under high

Corresponding Author: Charles U. Ugwu, Graduate School of Life and Environmental Sciences, University of Tsukuba, 1-1-1 Tennoudai, Tsukuba, Ibaraki 305-8572, Japan Tel: +81-29-853-7212

salinity. It should be noted that contamination is inevitable in open ponds. However, traces of contaminants can be overlooked in outdoor cultures as long as they do not cause significant reduction in the number and quality of algal biomass. Microalgae that can grow under extreme conditions of pH (e.g., Spirulina) or salinity (e.g., Dunaliella) are able to withstand contamination even in low density cultures. However, contamination can occur in those cultures due to pH gradients and this could be attributed to inefficient mixing system.

PHOTOBIOREACTORS

There are several types of PBRs that have been proposed till date; however, tubular PBR, bubble-column, flat plate PBR are the major ones that are considered for mass cultivation of microalgae. Tubular PBR has been well used for outdoor microalgae cultures (Lee and Low, 1991; Molina *et al*., 2001; Ugwu *et al*., 2002; Ferreira *et al*., 2012), one of the major reasons being attributed to the large illumination surface area. Although, there are lots of potential advantages of using tubular PBRs, the limitation in scaling them up has restricted its application in commercial production (Ugwu *et al*., 2008).

Generally, mixing in PBRs is done by bubbling directly with air pump or indirectly, by airlift system. Bubbling of air (aeration) at high aeration can be used in inclined tubular PBRs whereby tremendous amount of force is required to circulate cultures from the riser to the downcomer sections. For instance, in inclined tubular PBRs (Fig. 1), pumps are used to move the cultures from the aeration port along the riser to the downcomer sections. On the other hand, some airlift PBRs are built in such a way that air is pushed up to a certain height through a different pipe before it is released downwards to the tubes along the riser section. A typical horizontal plane tubular PBRs which utilizes airlift system is shown in Fig. 2. Airlift system is generally recommended in algal cultures that are fragile and sensitive to shear stress. Efficient mixing can be done by increasing the aeration

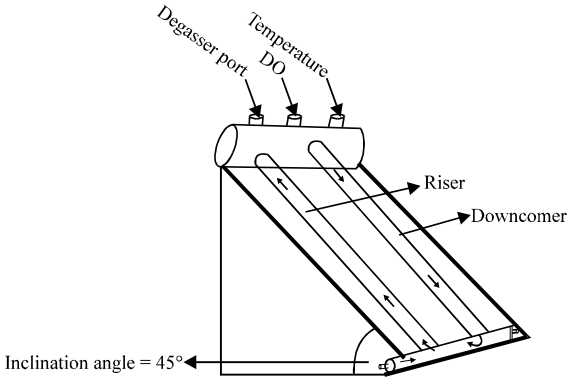

Fig. 1: A tubular photobioreactor inclined at 45° to the horizontal plane and consisting of riser and downcomer tubes (Ugwu *et al*., 2002)

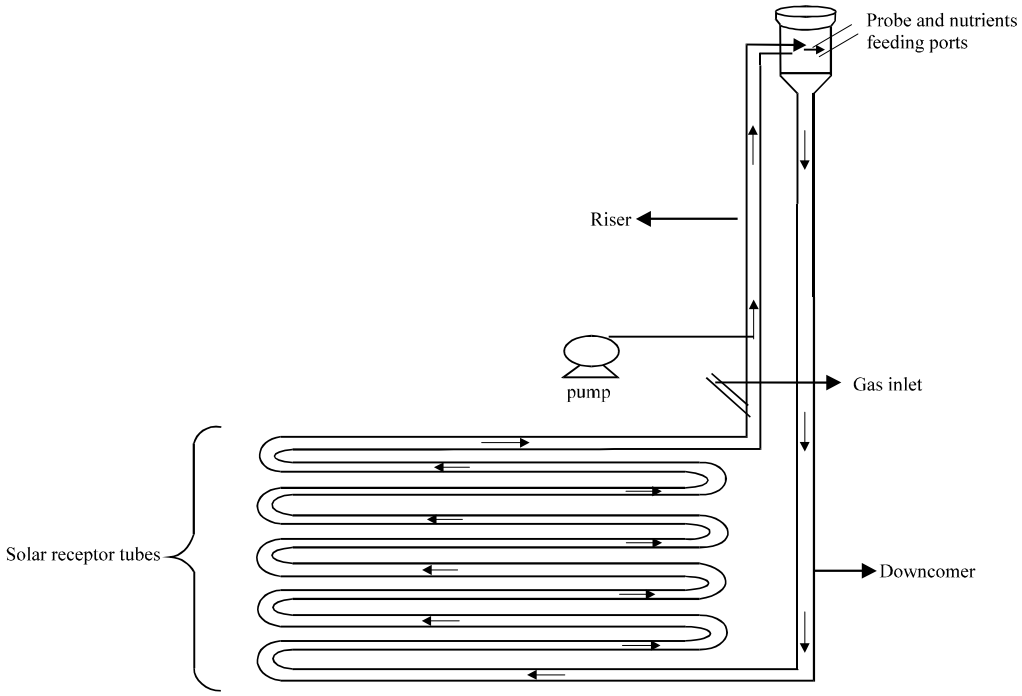

Fig. 2: A typical horizontal serpentine tubular photobioreactor equipped with an airlift system

rates; however, it has to be as moderate as possible to avoid some damages to the algal cells. One way of improving mixing in tubular PBR is by introduction of vertical mixing (Grima *et al*., 1999) or installation of static mixers (Ugwu *et al*., 2002; Ugwu and Aoyagi, 2011). Static mixers can ensure that cells are circulated between the upper and lower sections of the tubes, thereby resulting in high mass transfer and efficient distribution of light and nutrients in the tubular PBR. Although some designs of tubular PBRs have been tested, the most challenging issue in most part is their ability to maintain efficient mass transfer.

Different types of mixing in microalgal cultures

Orbital shaking in test tubes and flasks: Generally used culture vessels in laboratories include test tubes, flasks and smaller reactors. Pre-culture of microalgae are normally prepared in test tubes and flasks before they are inoculated to larger reactors. Horizontal mixing is applied in test tube cultures whereas flask cultures are mixed by orbital shaking under shakers equipped with artificial light.

Mixing by direct bubbling of air in tubular PBRs: In most tubular PBRs, air is bubbled to the cultures using air pumps and the direction of airflow is normally in a plug flow, depending on the aeration rate. However, back-mixing can occur if the flow becomes turbulent either due to installation of static mixers or by increased aeration rate (Fig. 3). Also depending on other factors (e.g., algal strain, static mixers, diameter of tubes, etc.), a vertical mixing can be introduced in tubular PBRs. Algal cultures are swirled from the point of aeration to the static mixers which in turn, pushes the broth up toward the upper section of the tubes and then returns them to the lower parts before passing them to the next mixers and to the downcomer section.

Mixing in pneumatically-agitated PBRs: Unlike tubular PBR, mixing in bubble column PBRs can be random and erratic mixing when gas is sparged from the bottom of the PBR (Miron *et al*., 1999; Halim *et al*., 2011). Figure 5 shows two different types of bubble column PBRs. In a conventional type bubble PBR, external illumination is done using some fluorescent lamps (Fig. 4a). Gases are bubbled from the bottom of the reactor and bubbles can be seen inside the column in a non-defined movement. On the other hand, a bubble column PBR equipped with draft tube would maintain a well-defined air flow along the riser and downcomer sections. Although, draft tube types are

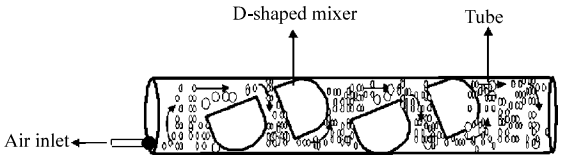

Fig. 3: A riser tube installed with four D-shaped static mixers. The static mixers were alternately arranged to induce back-mixing (Ugwu *et al*., 2003)

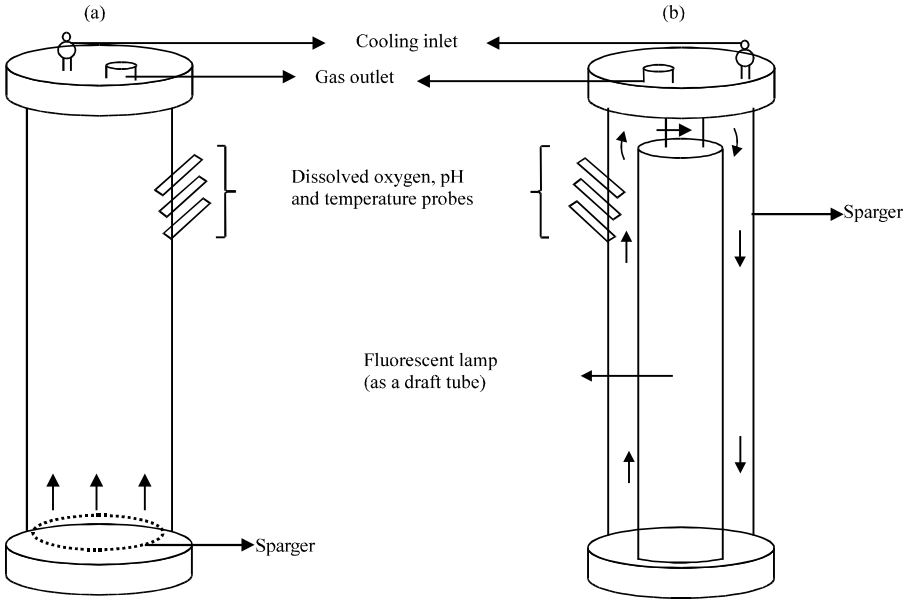

Fig. 4(a-b): Bubble column photobioreactors (a) Without a draft tube and (b) With a draft tube

very promising in column PBRs, they can cause light stratification problems, especially the opaque types. One way of solving this problem is by constructing a draft tube that can provide internal illumination to the column PBR (Fig. 4b). In this case, gas is supplied from the top of the riser section (left side) and then allowed to flow downward through the lower part of the draft tube and then upwards again in an anti-clockwise direction.

Raceway-mixing in ponds: Raceway pond is one of the oldest culture systems for cultivation of microalgae (Vonshak and Richmond, 1988; Boussiba *et al*., 1988; Hase *et al*., 2000). To some extent, a defined circulation can be maintained in a raceway pond when paddle wheels are continuously used to move the cultures from one side of the pond to the other part of it. Paddle wheels usually consist of a few blades that are arranged axially and connected by a rotor which circulates the cultures in a raceway manner. Unlike PBR systems, mixing speed in ponds is slow and not turbulent. Poor mixing of microalgae in ponds would result in flocculation of the cells. A combination of paddling (using paddle wheels) and stumping (installing blocks in the ponds) can provide better mixing of cultures.

Mixing pattern in flat-plate photobioreactors: Flat plate PBRs for outdoor cultivation of microalgae are usually characterized by random mixing (Zhang *et al*., 2001; Feng *et al*., 2011). As in the case of bubble column PBRs, gases are sparged from the lower part and bubbles could be seen moving randomly or erratically across the entire PBR (Fig. 5a). Flat-plate PBR can also be in compact form (split-plate or baffled type), similar to airlift system (Fig. 5b). This design will ensure a well-defined flow of cultures along the plates, thereby improving the mass transfer in the PBR. In one study, it was reported that an airlift-type vertical column PBR reactor equipped with baffles showed higher mass transfer efficiencies compared to that without baffles (Degen *et al*., 2001).

Parameters for studying mixing characteristics of fluids in PBRs: Mixing systems in PBRs are generally studied using either tracer dyes or pH indicators. Tracer technique using pH meters are commonly used in tubular PBR (Rubio *et al*., 1999; Ugwu *et al*., 2008). Thus, the time taken for a known concentration of alkali or acid to completely undergo complete mixing (evidenced by constant pH) with the liquid inside the reactor is considered as the mixing time. In long tubular PBRs, liquid velocity would give an insight of the movement and mixing behavior of algal cultures across the tubes.

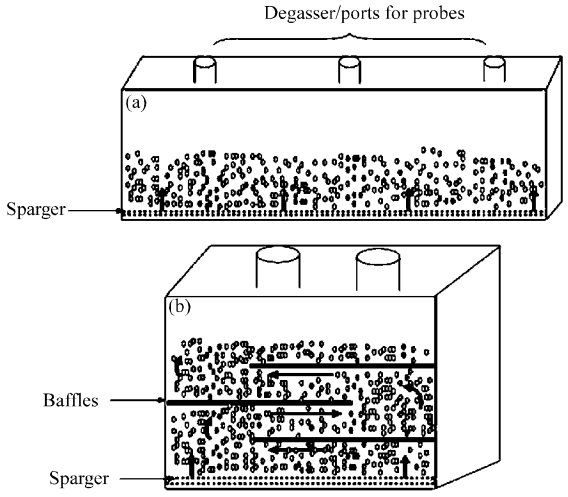

Fig. 5(a-b): Flat plate photobioreactors showing the movement of bubbles (a) Without baffles and (b) With baffles

Furthermore, despite that the velocity of gas bubbles can be useful in evaluating the mixing characteristics in tubular PBR, it does not specifically justify the extent of dispersion of gas bubbles in the liquid or culture broth. This is due to the fact that even when tiny bubbles are bubbled to the tubular photobioreactors, they tend to coalesce to form interface along the tubes, resulting in poor gas-liquid transfer. We have previously shown that static mixers reduced the interfacial area and enhanced gas-liquid transfer efficiency in the tubular PBRs (Ugwu *et al*., 2003, 2008).

Interrelationship between superficial gas velocity and mass transfer efficiency: The overall volumetric mass transfer coefficient (k_La) and gas hold up are very useful parameters for studying the performance of a PBR. As a model, 4 pieces of D-shaped mixers in alternate position (Fig. 3) were installed in the riser section of an inclined tubular PBR (Fig. 1). Results indicated that by increasing the superficial velocity from 0.01-0.05 m sec^{-1} resulted in about 6.8 fold increase in k_La in tubular PBR with D-shaped static mixers (Fig. 6a). However, without D-shaped static mixers, the k_La increased by 4.8 fold at the same range of superficial gas velocity. At superficial velocity of 0.05 m sec^{-1}, the k_La was 2.2 fold higher than that without D-shaped static mixers. On the other hand, the mixing time was longer when the static mixers were used. However, the mixing time became shorter as the superficial gas velocity was increased (Fig. 6b). At superficial velocity of 0.05 m sec^{-1}, the mixing time in the PBR with 4 G-shaped static mixers were about 70% (i.e., 0.70 fold) higher than that without static mixers.

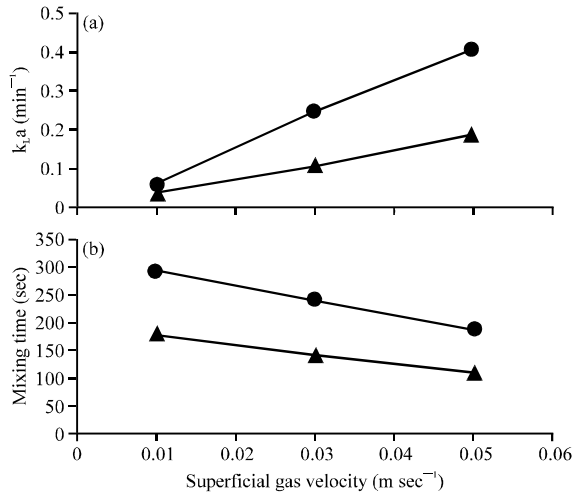

Fig. 6(a-b): Effect of superficial gas velocity on (a) k_La and (b) Mixing time (Ugwu *et al*., 2003)

Based on these data, it can be deduced that despite that static mixers can prolong the mixing time, k_La values can be much higher compared to those in PBRs without static mixers. It also implies that the longer mixing time could result in longer residence time which will invariably improve the gas-liquid transfer efficiency in the tubular PBR. In view of this, static mixers should be designed in such a way they should not stagnate dense algal cultures.

Effect of mixing on the light utilization efficiency by the microalgal cells: Mixing ensures uniform distribution of light in the PBR and thus improves the light utilization by the algal cells. For instance, a well mixed algal culture will ensure that cells at the top of the PBR (highly illuminated surfaces) is circulated to the low part of the PBR (very low illuminated surfaces), resulting in a flashing light effect. On the other hand, if algal cultures are not well mixed, the cells closer to the highly illuminated regions are photoinhibited by the high illumination whereas those at the lower part of the PBR are light-starved (Ogbonna *et al*., 1995). Although light supply to the PBR is very important, the ability of the algal cells to maximize the light available to them is another issue that has been shown to determine algal productivity.

Future prospects and conclusion: The use of fossil fuels as the main industrial and transportation energy source has increased so much recently, resulting to global warming and other environmental issues. Given that these fossil fuels are not renewable, there is an urgent need to develop some alternative energy sources. One of the most promising ways of generating renewable energy is through microalgae production. Some microalgae are capable of accumulating oils in their cells and the prospects of this biofuel from microalgae for sustainable development are being investigated (Chisti, 2007; Singh *et al*., 2011). In order to realize these objectives, open ponds and PBRs are being given serious considerations. Improvement in the mixing system is required in any of the culture systems as it would help a lot in increasing the algae biomass productivity, resulting in high efficiency in the use of microalgal products and process operation.

REFERENCES

Abd El-Baky, H.H., F.K. El-Baz and G.S. El-Baroty, 2004. Production of lipids rich in omega 3 fatty acids from the halotolerant alga *Dunaliella salina*. Biotechnology, 3: 102-108.

Ai, W., S. Guo, L. Qin and Y. Tang, 2008. Development of a ground-based space micro-algae photo-bioreactor. Adv. Space Res., 41: 742-747.

Akpolat, O. and S. Eristurk, 2008. Some physical parameters to effect the production of *Heamatococcus pluvialis*. J. Applied Sci., 8: 763-771.

Azmat, R., Uzma and F. Uddin, 2007. Biosorption of toxic metals from solid sewage sludge by marine green algae. Asian J. Plant Sci., 6: 42-45.

Borowitzka, M.A., 1999. Pharmaceuticals and Agrochemicals from Microalgae. In: Chemicals From Microalgae, Cohen, Z. (Ed.). Taylor and Francis, London, ISBN-13:9780748405152, pp: 313-352.

Boussiba, S., E. Sandbank, G. Shelef, Z. Cohen and A. Vonshak *et al*., 1988. Outdoor cultivation of the marine microalga *Isochrysis galbana* in open reactors. Aquaculture, 72: 247-253.

Chisti, Y., 2007. Biodiesel from microalgae. Biotechnol. Adv., 25: 294-306.

Degen, J., A. Uebele, A. Retze, U. Schmid-Staiger and W. Trosch, 2001. A novel airlift photobioreactor with baffles for improved light utilization through the flashing light effect. J. Biotechnol., 92: 89-94.

Eriksen, N.T., 2008. The technology of microalgal culturing. Biotechnol., Lett., 30: 1525-1536.

Feng, P., Z. Deng, Z. Hu and L. Fan, 2011. Lipid accumulation and growth of *Chlorella zofingiensis* in flat plate photobioreactors outdoors. Bioresour. Technol., 102: 10577-10584.

Ferreira, L.S., M.S. Rodrigues, A. Converti, S. Sato and J.C.M. Carvalho, 2012. *Arthrospira* (*Spirulina*) *platensis* cultivation in tubular photobioreactor: Use of no-cost CO_2 from ethanol fermentation. Applied Energy, 92: 379-385.

Grima, E.M., F.G.A. Fernandez, F.G. Camacho and Y. Chisti, 1999. Photobioreactors: Light regime, mass transfer and scale up. J. Biotechnol., 70: 231-247.

Halim, R., B. Gladman, M.K. Danquah and P.A. Webley, 2011. Oil extraction from microalgae for biodiesel production. Bioresour. Technol., 102: 178-185.

Hase, R., H. Oikawa, C. Sasao, M. Morita and Y. Watanabe, 2000. Photosynthetic production of microalgal biomass in a raceway system under greenhouse conditions in Sendai City. J. Biosci. Bioeng., 89: 157-163.

Lee, Y.K. and C.S. Low, 1991. Effect of photobioreactor inclination on the biomass productivity of an outdoor algal culture. Biotechnol. Bioeng., 38: 995-1000.

Miron, A.S., A.C. Gomez, F.G. Camacho, E.M. Grima and Y. Chisti, 1999. Comparative evaluation of compact photobioreactors for large-scale monoculture of microalgae. J. Biotechnol., 70: 249-270.

Molina, E., J. Fernandez, F.G. Acien and Y. Chisti, 2001. Tubular photobioreactor design for algal cultures. J. Biotechnol., 92: 113-131.

Ogbonna, J.C., H. Yada and H. Tanaka, 1995. Light supply coefficient: A new engineering parameter for photobioreactor design. J. Ferment. Bioeng., 2: 369-376.

Oncel, S.S. and O. Akpolat, 2006. An integrated photobioreactor system for the production of *Spirulina platensis*. Biotechnology, 5: 365-372.

Oswald, W.J., 1988. Large-Scale Algal Culture Systems (Engineering Aspects). In: Micro-Algal Biotechnology, Borowitzka, M.A. and L.J. Borowitzka (Eds.). Cambridge University Press, Cambridge, pp: 357-394.

Rubio, F.C., F.G. Fernandez, J.A. Perez, F.G. Camacho and E.M. Grima, 1999. Prediction of dissolved oxygen and carbon dioxide concentration profiles in tubular photobioreactors for microalgal culture. Biotechnol. Bioeng., 62: 71-86.

Sauid, S.M. and V.V.P.S. Murthy, 2010. Effect of palm oil on oxygen transfer in a stirred tank bioreactor. J. Applied Sci., 10: 2745-2747.

Singh, A., P.S. Nigam and J.D. Murphy, 2011. Mechanism and Challenges in Commercialisation of Algal Biofuels. Bioresource Technol., 102: 26-34.

Thomas, W.H. and C.H. Gibson, 1990. Effect of small-scale turbulence on microalgae. J. Applied Phycol., 2: 71-77.

Ugwu, C.U., J.C. Ogbonna and H. Tanaka, 2002. Improvement of mass transfer characteristics and productivities of inclined tubular photobioreactors by installation of internal static mixers. Applied Microbiol. Biotechnol., 58: 600-607.

Ugwu, C.U., J.C. Ogbonna and H. Tanaka, 2003. Design of static mixers for inclined tubular photobioreactors. J. Applied Phycol., 15: 217-223.

Ugwu, C.U., H. Aoyagi and H. Uchiyama, 2008. Photobioreactors for mass cultivation of algae. Bioresour. Technol., 99: 4021-4028.

Ugwu, C.U. and H. Aoyagi, 2011. Evaluation of the mass transfer capacity of a long tubular photobioreactor with static mixer and its outdoor performance with microalgal cultures. Trends Applied Sci. Res., 6: 1222-1229.

Vonshak, A. and A. Richmond, 1988. Mass production of the blue-green alga *Spirulina*: An overview. Biomass, 15: 233-247.

Yoshihara, K., H. Nagase, K. Eguchi, K. Hirata and K. Miyamoto, 1996. Biological elimination of nitric oxide and carbon dioxide from flue gas by marine microalga NOA-113 cultivated in a long tubular photobioreactor. J. Ferment. Bioeng., 82: 351-354.

Zhang, K., S. Miyachi and N. Kurano, 2001. Evaluation of a vertical flat-plate photobioreactor for outdoor biomass production and carbon dioxide bio-fixation: Effects of reactor dimensions, irradiation and cell concentration on the biomass productivity and irradiation utilization efficiency. Applied Microbiol. Biotechnol., 55: 428-433.