ISSN 2176-5480

22nd International Congress of Mechanical Engineering (COBEM 2013)
November 3-7, 2013, Ribeirão Preto, SP, Brazil

SEMICONTINUOUS CULTIVATION OF MICROALGAE IN COMPACT PHOTOBIOREACTOR

Diego de Oliveira Corrêa
Beatriz Santos
Universidade Federal do Paraná, Caixa Postal 19011, Curitiba, PR, Brasil, CEP: 81531-980
diego.biodoc@gmail.com,
biologia_bia@yahoo.com.br

Bruno Miyawaki
Universidade Federal do Paraná, Caixa Postal 19011, Curitiba, PR, Brasil, CEP: 81531-980
bruno_miyawaki@hotmail.com

Nelson Fernando Herculano Selesu
Universidade Federal do Paraná, Caixa Postal 19011, Curitiba, PR, Brasil, CEP: 81531-980
nefeherse@hotmail.com

Andre Bellin Mariano
Universidade Federal do Paraná, Caixa Postal 19011, Curitiba, PR, Brasil, CEP: 81531-980
andrebmariano@gmail.com

José Viriato Coelho Vargas
Universidade Federal do Paraná, Caixa Postal 19011, Curitiba, PR, Brasil, CEP: 81531-980
vargasjvcv@gmail.com

Abstract. *Microalgae are recognized as a promising alternative for the generation of renewable fuels and replacing the use of fossil fuels, however, the maintenance of cultives on a large scale is one of the main limiting factors for the production of biodiesel from microalgae. In industrial scale production, cultivation may be performed in open systems such as ponds and tanks, or in closed systems, such as the photobioreactor used in this study. To characterize the potential for microalgae biomass production in the system, two independent experiments were performed with a microalgae of the genus Scenedesmus in the compact photobioreactor with area equal to10 m^2 and operating volume of 12 m^3. Considering that the cultures were performed in the external environment, parameters such as light and temperature could not be controlled. The results showed that photobioreactor used has the ability to maintain cultures of microalgae for a long period. The first experiment lasted 100 days and presented daily output equal to 0.0029 $g.L^{-1}.dia^{-1}$ of dry biomass, while the second experiment lasted 54 days and presented daily output of 0.0063 $g.L^{-1}.dia^{-1}$ of dry biomass. Considering a production area equal to one hectare, the data point to the superior productivity of some crops traditionally used in the production of oil for generation of biodiesel, such as soybeans or sunflower, for example. The analyzes conducted to determine the oil content in the microalgae used indicated that approximately 13% of its dry weight is composed of lipids. The present study allowed the understanding of the processes involved in growing microalgae on an industrial scale and contributed to the evaluation of the production potential of the system used, indicating new ways and possibilities to be explored.*

Keywords: *Microalgae, Photobioreactor, Biodiesel, Biomass.*

1. INTRODUCTION

The cultivation of microalgae are fundamental tools for the elucidation of many processes, ecologicals, ontogenetics, physiologicals, biochemicals and behavioral, among others. In addition to its use for obtaining information about microalgae also have many other economically important applications such as the generation of biomass for animal and human nutrition and production of substances of industrial interest (Lourenço, 2006).

An important feature of the systems of cultivation of microalgae is its versatility, enabling to relate different applications in the same process as the wastewater treatment, the production of food supplements, animal feed, pharmaceuticals and chemicals. Another attractive feature of the use of microalgae as compared to the use of other micro-organisms, is their photosynthetic capacity to convert solar energy into biomass with chemical composition attractive from the point of view of energy (De La Noue and De Pauw, 1988).

Cultures of microalgae can be carried out in open systems, such as lakes or ponds or in closed systems such as photobioreactors. A photobioreactor is a reactor in which photosynthetic organisms are grown or used to carry out a biological reaction (Mata *et al.*, 2010).

ISSN 2176-5480

1.1 Open systems

The systems for cultivation of microalgae most used currently are tanks type race, that consists of a shallow artificial lake, in order to create opportunities largest exposure to sunlight, and a blade system, which is responsible for agitation and homogenization of the medium in cultivation. Despite being widely diffused in the large-scale production, the model has some deficiences, such as low control over the growing conditions, the possibility of contamination by external agents and, in particular, the need for a large area for its construction and operation.

1.2 Closed systems

Due to the features of the crops, the use of photobioreactors for the cultivation of microalgae has some advantages, among which we can point out the most control of the conditions as the system is closed and there is no contact with the outside environment, reducing the likelihood contamination of different organisms cultivated.

The use of photobioreactors also has the advantage of occupying a much smaller area than that of open systems and hence no competition for the use of agricultural areas that could be used for food production. Photobioreactors also have high efficiency in biomass production when used CO_2 injection in the middle, increasing the growth of microalgae (Suali and Sarbatly, 2012).

1.3 Biodiesel from microalgae

The increased production of oil plants for generation of biofuels on arable land may have consequences for the global food supply. In contrast, the production of biodiesel from microalgae is widely regarded as one of the most efficient ways to produce biofuels. Vegetable oils are a renewable and potentially inexhaustible biodiesel production, on the other hand its use can worsen the picture of hunger in developing countriesas it there may be competition for use of available areas for food production (Demirbas, 2011).

Microalgae are the only matrix for production of biodiesel can replace the use of fossil fuels and, unlike other oilseeds, have rapid growth, in addition to having high lipid content, commonly presenting content 20-50%. They have also rapid growth: microalgae commonly doubling the biomass over a period of 24 hours, although during the exponential growth phase this time may be up to 3.5 h (Chisti, 2007).

Inserted in this context, and based on promising and use of microalgae, it is necessary scientific research and development of its cultivation for biodiesel production. Thus, this paper aims to contribute to the generation of new data about the cultures performed in the photobioreactors of the Nucleus for Research and Development of Self-sustainable Energy.

2. MATERIAL AND METHODS

With the purpose of performing the comparison of the yield obtained, were performed two independent cultures with the same microalgae using photobioreactor in the external environment. For a better understanding of the methodologies used and the results obtained in this project, from that time the first crop cultivation will be called A, while the second crop cultivation will be called B.

2.1 Semicontinuous cultivation system

Both crops in compact tubular photobioreactor were performed in semicontinuous mode, which means that the point of maximum growth dilutions were made of the system, removing an aliquot for flocculation and addition of fresh medium, aiming to maintain the cultures for as long possible. Figure 1 shows a brief diagram of the process of diluting the photobioreactor.

ISSN 2176-5480

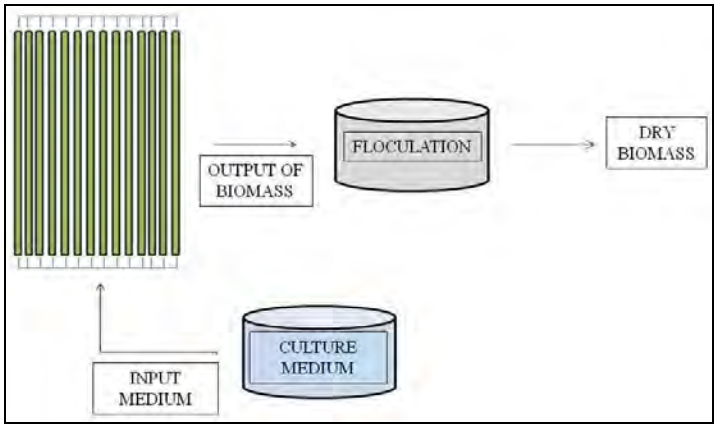

Figure 1. Dilution scheme of the photobioreactor

As can be seen in the diagram, the dilutions carried out consisting in withdrawing a portion of cultivation to flocculate and obtain dry biomass and subsequent addition of culture medium and water equal to the volume removed.

2.2 Tubular compact photobioreactor

The system used in the cultivation of microalgae consists of a compact tubular photobioreactor which occupies an area of 10 m^2 with operating volume of approximately 12 m^3 and is installed in the outer area of the Nucleus for Research and Development of Self-sustainable Energy (NPDEAS). The FBR has 3.5 kilometers of transparent PVC tubes arranged alternately in order to maximize the use of space. In addition to the structure itself, the photobioreactor is composed of two pumps with ¼ HP power working in parallel, being responsibles for the circulation of the fluid within the system, and a compressed air line which provides atmospheric air to the photobioreactor. Figure 2 shows the appearance of the photobioreactor used in the experiments.

Figure 2. Tubular compact photobioreactor

Is important show that the system is modular, allowing its use with different operation volumes. This factor is important, especially in the process of inoculation photobioreactor.

2.3 Inoculum production

The operation volume of the photobioreactor is approximately 12 m^3 of liquid. Therefore, it was necessary to the growth of previous culture which served as inoculum for the main experiments. The production of inoculum consists of three preliminary steps as follows:

- Growth of culture in 2 L Erlenmeyer flasks;

ISSN 2176-5480

- Cultivation in 20 L gallons;
- Growth in rectangular tank with a volume of 2 m^3 for subsequent inoculation photobioreactor.

The volume produced at each stage described previously served as inoculum for the subsequent step. Figure 3 shows a flowchart for the inoculation of the photobioreactor.

Figure 3. Flowchart for inoculation of the photobioreactor

Considering from laboratory scale to production in the early inoculation photobioreactor, the process has a total duration of approximately 63 days.

2.4 Determination of growth and cultivation parameters

Determination of growth of microalgae was performed daily using the parameters of cell concentration, optical density measured by spectrophotometry, pH variation, temperature and dry biomass. These data were used for preparation of the growth curves and choice of points which included the dilution system. All collected data were recorded and analyzed in spreadsheets specifically designed for this purpose. All parameters analyzed were collected in triplicate.

2.5 Content of total lipids

In order to determine the potential of microalgae used in biodiesel production, the quantification of the total lipid content present in the dry biomass produced during one of the points of maximum crop growth A. Therefore, we used the methodology Bligh-Dyer, adapted from Soares (2010).

2.6 Flocculation and filtration

The biomass obtained during the performance of crops was flocculated using ferrous sulfate heptahydrate ($FeSO_4.7H_2O$) at a concentration of 0.2 $\mu mol.L^{-1}$ as flocculating agent, filtered to remove moisture and stored in a freezer for later extraction of lipids and conversion into biodiesel.

3. RESULTS AND DISCUSION

Since inoculation photobioreactor until the end of the cultive A took place 100 days, a period corresponding to 28 March 2012 to 06 July 2012. In this period, were realized six dilutions of the system, which consisted in the removal of 3 m3 of liquid and adding fresh culture medium for the same volume removed. Therefore, the culture remained viable for a longer period of time and semicontinuous mode of operation has proved to be a method applicable to the system of biomass production on an industrial scale using photobioreactors.

During the cultivation, it was possible to observe the formation of algal biofilm fouling with the inner walls of pipes, which results in reducing the amount of light that enters the cultivation, thus causing damage to growing cells. At the end of the 100 days of cultivation was performed withdrawal of the total volume of the reactor and cleaning tubes for the removal of biofilm, both materials were processed by flocculation and had a biomass filtered and stored.

The cultive B lasted 54 days, the period between the day on September 5 and October 29, 2012. During this period were performed five dilutions of the system, with the removal of 1 m^3 each and addition of fresh culture medium and sufficient water to 1 m^3. Despite having less than the total length of the first experiment, the second crop also demonstrated the ability of the system to maintain a viable culture for extended period of time.

ISSN 2176-5480

Molds to the first crop, in cultive B were also biofilm formation on the inner walls of the pipes of the system, which was predominant factor for the completion of the experiment. With the end of cultivation, the total volume of the reactor was processed by flocculation and the resulting biomass was stored in a freezerThe amount of biofilm formed was used to carry out other experiments unrelated to this project.

3.1 Temperature variation

As the photobioreactor is built in the outdoor area of the NPDEAS, crops occurred in conditions of the external environment, thus, suffered temperature variations resulting from environmental changes of the respective periods. Figure 4 shows the values of minimum and maximum temperature recorded during the performance of crops A and B.

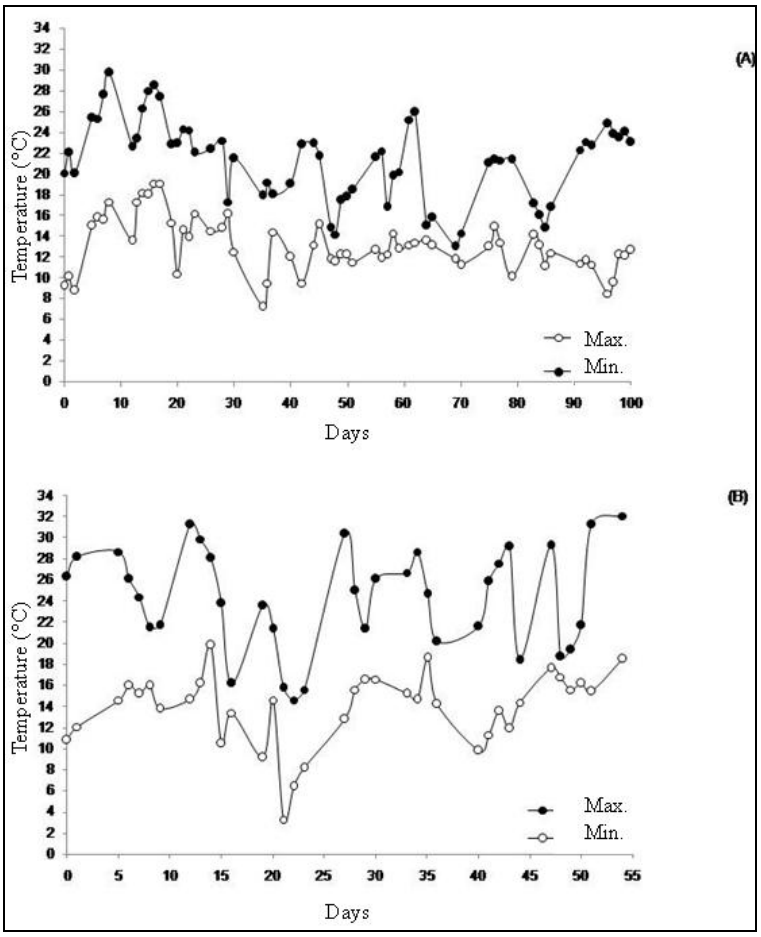

Figure 4. Temperature variation in the culture A and B

During the period of the cultive A, the system has experienced significant variations in temperature, with the temperature range of 22.5 ° C and the minimum temperature recorded was 7.2 ° C and a maximum of 29.7 ° C.

The cultive B has been carried out over year period with higher temperatures, the system also experienced significant temperature variations. The temperature range for the period was 29.9 ° C, with a minimum recorded 3.3 ° C and maximum 33.2 ° C.

Although we have recorded temperatures as low as those mentioned, microalgae in culture remained viable, demonstrating extreme resilience to environmental variations arising from differences in seasons crossed during this period. This feature enhances the ability of the microalgae used as biomass production in industrial production scale, since it was effective in growth even in situations not favorable.

3.2 Cellular growth

Using data from the counts in optical microscopy, we determined the growth curves of the microalgae in two cultures performed. For each point used for making the bend, three counts were made and the mean employed in making the graph and the associated standard deviation. Figure 5 shows the growth curve constructed using the data obtained from cultures of A and B and the volume of reactor operation.

ISSN 2176-5480

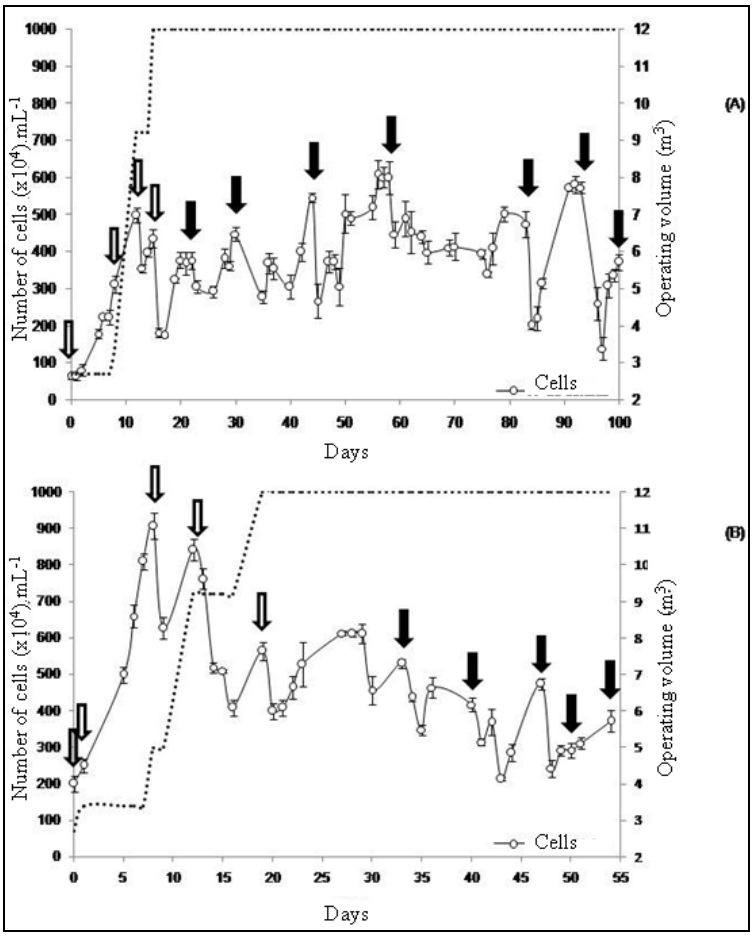

Figure 5. Cellular growth in the culture A and B

The above figure details the cell growth, expressed in the number of cells per milliliter culture, and the points at which the opening maneuvers were performed during the extension of inoculation (white arrows), the moments in which dilutions were made (black arrows), with removal and replacement of the middle rate for the same volume removed and the operating volume of the reactor (dotted line).

The figure shows also bars the standard deviation calculated from the average of the data obtained. To achieve a confidence interval of 95% was used twice the standard deviation of the average calculated for each of the registered points. For the cultive A, the peak cell density were obtained on days 58 and 93, while lower values were observed on day one after inoculation and on day 97 of culture.

The cultive B began with average 200(x10^4)cells.mL^{-1}, reaching peak concentration in cell 908(x10^4)cells.mL^{-1}, on the eighth day of culture, when it was held maneuver opening new branches. Generally speaking, higher values of cell concentration observed for the cultive B were obtained in the early stages of the experiment, which is in part a reflection of the intense process of biofilm formation, which causes a decrease in the amount of light inside the tubes and reduced growth of algae.

It is imperative to note that the maximum concentration values observed in cultive B were higher than those recorded for the cultive A, this result is a reflection of a change in the system of aeration photobioreactor held between one experiment and another.

The modification mentioned refers to the placing of a membrane on the diffusive compressed air inlet, resulting in production of air bubbles with smaller diameter, increasing the efficiency of the gaseous exchange in the system.

It can be seen that the openings of extensions and dilutions occurred at times when the cell concentration recorded was high in both experiments, this methodology allowed the system could provide the greatest possible amount of biomass for the period, ensuring continued growth and prolonged . It is worth noting that, in general, the points in which were recorded the lowest values of cell concentration coincide with the moments after the completion of dilutions of the system, where they were aliquots for flocculation.

ISSN 2176-5480

3.3 Optical density

As with other analyzes, the optical density measurements were performed daily for the two experiments, in order to provide a general profile of growth and multiplication of microalgae in culture. The choice of wave length at 540 nm was due to a band not to be absorbed by the chlorophyll, in this manner, the absorption data obtained reflect the concentration of cells in the medium due to light scattering.

Figure 6 shows the variation of optical density at 540 nm during the period of the crop A and B in the photobioreactor. Are represented as points were performed maneuvers opening stations during inoculation (white arrows) as well as the times in which dilutions were made (black arrows) with rate of removal and replacement of medium for the same volume removed.

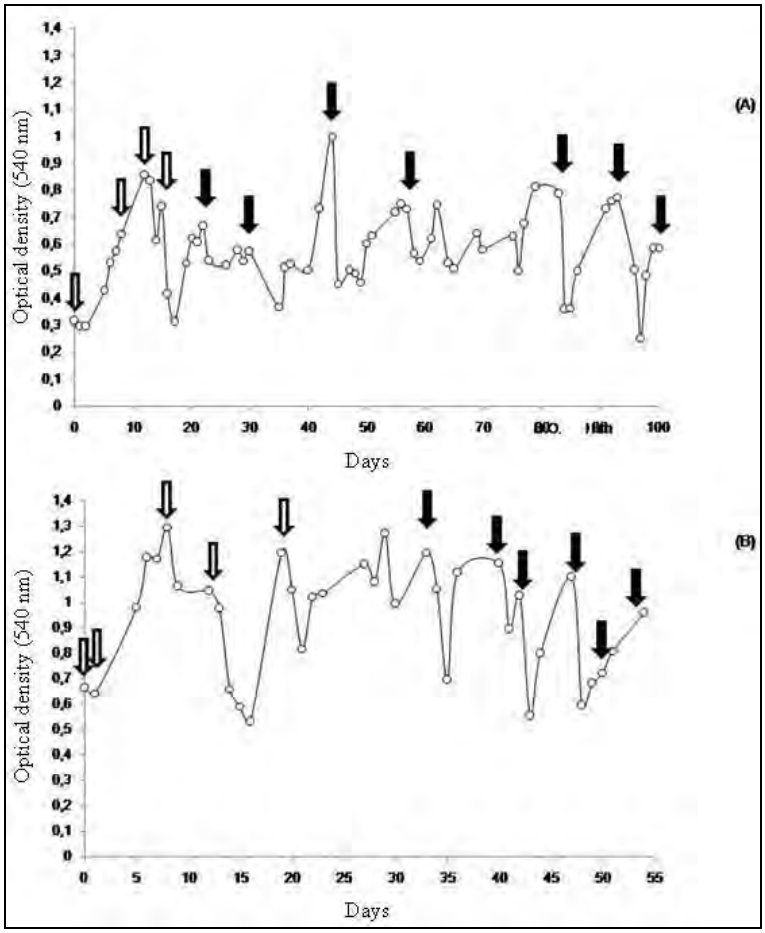

Figure 6. Optical density in the culture A and B

In general, monitoring the change in optical density can be visualized as an alternative to monitoring the cultivation of microalgae, as well as being valid for this purpose, presents the advantage of being more practical and faster than counting by taking less time the person performing the analysis.

3.4 Biomass produced

The estimation of dry biomass was performed by correlation between the optical density at 540 nm and dry biomass measured in five different samples of crops. Table 1 presents the amount of dried biomass obtained in each of the six dilutions made in the cultive A, increased biomass recovered in the process of biofilm formed inside the tubes.

The values stated in the table are for the volumes processed for each maneuver dilution system, except the value of the biomass obtained from the recovery of the biofilm. For each dilution were taken 3 m^3 performed only at the end of 100 days were processed is 12 m^3 related to the total volume of the reactor.

It is possible to highlight that the minimum production obtained during the period was 422 g dry biomass, when they were processed 3 m^3 cultivation on the 30th day of the experiment. With the exception of biomass recovered from the biofilm, the highest production obtained was recorded on the 44th day of culture, when they were recovered 736 g of dry biomass.

ISSN 2176-5480

Table 1. Biomass produced in the culture A

Period (days)	Dry biomass ($g.L^{-1}$)	Volume (m^3)	Total dry biomass (g)
22	0.164	3	493
30	0.141	3	422
44	0.245	3	736
62	0.183	3	550
83	0.193	3	580
93	0.191	3	573
100	0.144	12	1,734
Biofilm	1.800	2.5	4,500
Total	0.2950	32.5	9,588

For cultivation B, the biomass production was measured at each dilution held in the photobioreactor. Table 8 shows the yield achieved in each of the five dilutions made during the cultivation B plus the amount obtained from the processing of the total volume of the reactor at the end of the experiment.

Table 2. Biomass produced in the culture B

Period (days)	Dry biomass ($g.L^{-1}$)	Volume (m^3)	Total dry biomass (g)
33	0.6999	1	700
40	0.4456	1	446
42	1.2819	1	1,282
47	0.4047	1	405
50	0.1945	1	194
54	0.2355	12	2,826
Total	0.5437	17	5,853

As previously mentioned, were five dilutions of culture B to obtain biomass. The highest yield observed was recorded in the third dilution, performed at 42 days of cultivation, with approximately 1,282 g of dry biomass, considering the processed volume of 1 m^3. The lower production was recorded on the 50^{th} day, with an average of 194 g of dry biomass considering the same volume processed previously.

The formation of biofilm was essential for the closing of cultures, since the deposition of algae on the internal walls of the pipes resulted in decreased light penetration and reducing the flow velocity of the fluid.

Although harmful, biofilm formation was the reason why the methodology was developed for cleaning the reactor, which consists of passing sponges displaced by compressed air from inside the tubes by removing the adhered cells. This practice has the advantage of not using chemicals, allowing the tubes are cleaned in parallel with the completion of the crops, and may further extend the duration of the experiments.

3.5 Content of total lipids

The content of lipids present in the biomass obtained for the two crops do not directly reflect the amount of fatty acids susceptible to conversion to biodiesel, since it considers all substances of lipidic matrix of cells, such as cell membranes, energy storage, photosynthetic pigments, among others. Although the figures do not reflect the amount of fatty acids present in the biomass, the quantification of the total lipid content was performed in order to define, in general, the potential of microalgae used in generating biodiesel.

After the analysis of three samples obtained from the photobioreactor biomass were measured values of total lipids, with a mean of 13.1 ± 0.2% of the dry biomass of microalgae obtained. These results are similar to the values found for total lipid Scenedesmus sp., Obtained in experiments conducted by other researchers NPDEAS.

Literature data indicate that microalgae of the genus Scenedesmus may present levels of lipids that comprise a range of 1.9% to 55% by weight of dry biomass. Table 3 shows the levels of lipids present in different freshwater microalgae according to the literature.

ISSN 2176-5480

Table 3. Content of lipids in different microalgae

Specie	Lipid content (%)
Botryococcus sp.	25 – 75
Chaetoceros muelleri	33.6
Chaetoceros calcitrans	14.6 – 39.8
Chlorella emersonii	25 – 63
Chlorella protothecoides	14.6 – 57.8
Chlorella sorokiniana	19 – 22
Chlorella vulgaris	5 – 58
Chlorella sp.	10 – 48
Chlorella pyrenoidosa	2
Chlorococcum sp.	19.3
Ellipsoidion sp.	27.4
Haematococcus pluvialis	25
Scenedesmus obliquus	11 – 55
Scenedesmus quadricauda	1.9 – 18.4
Scenedesmus sp.	19.6 – 21.1

The results coincide with the figures for other species of the same genus, although these data are lower than the values recorded for species with high lipid content, as the species Chlorella emersonii, which has 25% to 63% by weight of the biomass dry the corresponding lipids.

3.6 System productivity

Through the yield data obtained values were estimated on the photobioreactor productivity during the duration of each crop. Table 4 shows values related to the operational characteristics of the system as well as the figures for the production of microalgae biomass in cultures A and B. To estimate the annual production of oil per hectare, we used the density of soybean oil (0.93 $g.L^{-1}$), and the lipid content of microalgae, around 13%.

Table 4. Productivity in the cultures A and B

Characteristics	A	B
Area (m^2)	10	10
Processed volume (L)	32,500	17,000
Period (days)	100	54
Total biomass produced (g)	9,588	5,853
Concentration of biomass ($g.L^{-1}$)	0.29	0.34
Daily productivity of biomassa ($g.day^{-1}$)	95.88	108.39
Volumetric productivity ($g.L^{-1}.day^{-1}$)	0.0030	0.0064
Estimated annual productivity of biomass ($Kg.year^{-1}$)	35	39,5
Estimated annual productivity of biomass per hectare ($Kg.ha^{-1}.year^{-1}$)	35,000	39,500
Estimated annual productivity of oil per hectare ($L.ha^{-1}.year^{-1}$)	4,231	4,775

Considering the amounts of biomass achieved, it is possible to estimate the productivity of the reactor under different perspectives.

The total biomass produced in the culture A was approximately 9.6 kg of dry biomass of microalgae during the 100 days of cultivation. Calculating the average daily productivity, it can be said that the production was 96 g dry biomass per day. Extrapolating these results over a period of 365 days of cultivation, it can be said that the expected output corresponds to approximately 35 kg of dry biomass of microalgae for one year. It should be remembered that productivity is related to an area of 10 m^2 in order to estimate what biomass productivity in large scale production, can be considered one hectare of usable area, which would result in an annual production of approximately 35 tons of biomass dried microalgae per hectare per year.

Using data relating to the cultivation B, it is possible to conduct an analysis of productivity in the same way. The total dry matter produced in the duration of cultivation B was 5.85 kg with a daily production of 108.39 g of biomass. Considering a period equal to one year of cultivation, it is possible to estimate that the production would be approximately 39.5 kg of dry biomass from microalgae. Again, the productivity values are calculated based on the area occupied by the photobioreactor, however, it is possible to extrapolate the data and calculate the annual yield of one hectare of cultivation of microalgae, which results in the production of approximately 39.5 tons of dry biomass.

ISSN 2176-5480

When comparing the levels of productivity of the two crops, the result is similar, indicating that the methodology used in the production of biomass, yet has slight variations, has proved effective.

Comparing the values of daily productivity for the two crops was concluded that the overall productivity of the cultivation B of approximately would be 10.8 kg of dry biomass of microalgae production period in the cultivation equal to A. Disregarding the influence of biofilm biomass in the cultivation A, it is clear that the productivity of culture B would almost double the productivity recorded in cultivation A.

In order to achieve a more accurate comparison, values were calculated productivity for crops, excluding the biofilm biomass obtained in cultivation A. Table 5 presents the values of productivity of both crops.

Table 5. Productivity in the cultures A and B (without biofilme)

Características	A	B
Total biomass produced (g)	5,088	5,853
Concentration of biomass ($g.L^{-1}$)	0.16	0.34
Daily productivity of biomassa ($g.day^{-1}$)	50.88	108.39
Volumetric productivity ($g.L^{-1}.day^{-1}$)	0.0016	0.0064
Estimated annual productivity of biomass ($Kg.year^{-1}$)	18.6	39.5
Estimated annual productivity of biomass per hectare ($Kg.ha^{-1}.year^{-1}$)	18,600	39,500
Estimated annual productivity of oil per hectare ($L.ha^{-1}.year^{-1}$)	2,248	4,775

The above table shows that the productivity of the cultivation B was twice the productivity registered in the cultivation A, when it is considered the recovered biomass of biofilm formed in the first crop.

Again, it is possible to make comparisons with data available in the literature on biomass productivity and lipid content in different cultivars traditionally employed in the production of biodiesel. Table 6 shows the productivity data for different kinds of oil used to produce biodiesel, as well as the oil yield data estimated for the photobioreactor.

Table 6. Productivity in the traditional cultures

Specie	Lipid content (%)	Annual productivity ($L.ha^{-1}.ano^{-1}$)
Corn (*Zea mays*)	44	172
Soybean (*Glycine max*)	18	636
Canola (*Brassica napus*)	41	974
Sunflower (*Helianthus annuus*)	40	1,070
Palma (*Elaeis guineensis*)	36	5,366
Scenedesmus sp. (Photobioreactor)	13	4,503

Considering the data obtained in productivity and a production area equal to one hectare, one can conclude that the productivity achieved in both cultures performed in photobioreactor is higher than the productivity of the four main crops for the production of biodiesel.

While productivity has sunflower oil equals 1,070 $L.ha^{-1}.year^{-1}$, the photobioreactor has the potential to produce about 4,503 $L.ha^{-1}.year^{-1}$, considering the average yields achieved in cultures A and B .

Also in comparison, considering the expected yield for the cultivation of sunflower equal to an area occupied by the photobioreactor, 10 m^2, the amount of oil produced was only about 1 $L.year^{-1}$, while the microalgae production is used estimated approximately 4.5 $L.year^{-1}$.

Comparing the microalgae cultivations performed in the FBR with the main crops used for biodiesel production, the one that is the most productive Palma, with annual oil production of 5,366 $L.year^{-1}$, higher than the 4,503 $L.year^{-1}$ estimated for the photobioreactor.

4. CONCLUSION

The primary purpose of this study consisted in realization in crops in semicontinuous mode of microalgae in photobioreactors developed by NPDEAS in order to understand the production potential of the system. Thus, the data collected during the execution of the experiments showed that the oil yield in both crops was higher when compared to other cultures that have the same purpose of biodiesel production, however, it is noteworthy that the species of microalgae has low potential for this purpose. These factors indicate that the system has the potential to improve the results, either by optimization of the process or the use of other species of microalgae with increased total lipid content.

ISSN 2176-5480

5. ACKNOWLEDGEMENTS

The authors acknowledge the National Council for scientific and technological development (CNPq) and Nilko Metalurgica Ltda for the financial support and the Federal University of the Parana for the infastructure.

6. REFERENCES

Chisti, Y., 2007. "Biodiesel from microalgae". *Biotechnology advances*, Vol. 25, p. 294 - 306.

Demirbas, A., 2011. "Biodiesel from oilgae, biofixation of carbon dioxide by microalgae: A solution to pollution problems". *Applied Energy*, Vol. 88, p. 3541 - 3547.

De La Noue, J.; De Pauw, N., 1988. "The potential of microalgal biotechnology: A review of production and uses of microalgae". *Biotechnology advances*, Vol. 6, p. 725 - 770.

Lourenço, S. O., 2006. *Cultivo de Microalgas Marinhas: Princípios e Aplicações*. São Carlos: RiMa.

Mata, T. M.; Martins, A. A.; Caetano, N. S., 2010. "Microalgae for biodiesel production and other applications: A review". *Renewable and Sustainable Energy Reviews*, Vol. 14, p. 217 - 232.

SOARES, D., 2010. *Avaliação do crescimento e celular e produtividade de lipídeos de microalgas marinhas em diferentes regimes de cultivo*. Master thesis, Universidade Federal do Paraná, Curitiba.

Suali, E.; Sarbatly, R., 2012. "Conversion of microalgae to biofuel". *Renewable and Sustainable Energy Reviews*, Vol. 16, p. 4316 – 4342.

7. RESPONSIBILITY NOTICE

The authors are the only responsible for the printed material included in this paper.