

PRELIMINARY ENVIRONMENTAL ASSESSMENT OF THE CULTIVATION OF MICROALGAE AGROINDUSTRIAL RESIDUE

Marisa Daniele Scherer Amanda Cristina Oliveira André Bellin Mariano José Viritato Coelho Vargas Universidade Federal do Paraná - Programa de Pós-Graduação em Ciência e Engenharia dos Materiais. marisa_bio_scherer@hotmail.com amandacrisliveira@gmail.com andrebmariano@gmail.com vargasjvcv@gmail.com

Cássia Maria Lie Ugaya

Universidade Tecnológica Federal do Paraná - Programa de Pós-Graduação em Engenharia Mecânica. cassiaugaya@utfpr.edu.br

Abstract. Microalgae are considered promising sources of raw material for biodiesel production. The cultivation of microalgae for obtaining biomass depends on several factors such as the amount of available nutrients, CO_2 , water, light, temperature and pH. The literature suggests the use of alternative nutrients for reducing the environmental impacts of biodiesel production from microalgae. This study aims to compare the environmental impact of microalgae cultivation with the synthetic nutrient Chu and effluent from cattle through the methodology of Life Cycle Assessment (LCA) via SimaPro® 7.0 software, with a reference flow of 10 g of dry biomass. Using this methodology, it was demonstrated that environmental impacts such as eutrophication can be reduced by 57.2 % using the nutrients from the cattle effluent on the microalgae culture. Further studies should be conducted in the future in a systematic way in order to avoid exchange of environmental impacts.

Keywords: Life Cycle Assessment [LCA], Microalgae Cultivation, Agroindustrial Residues.

1. INTRODUCTION

The major problem of environmental issue, along with fossil fuels, has directed the science to develop new alternatives for energy sources with the purpose of decentralization of products derived from fossil fuels, and thus diversifying the sources and the products obtained.

Microalgae have been considered as an important alternative source to diversify the raw materials used to produce biofuels, because they have interesting features like: use of areas unsuitable for agriculture; higher binding capacity of carbon dioxide (CO_2), increased synthesis of lipids, use of water unfit for human consumption (effluents and wastewater), depending on the region and cultivated species it can produce the whole year, and when grown in photobioreactors compact, it reduces areas of crops (Chisti, 2007; Rodolfi *et al.*, 2009).

One of the barriers of microalgae production process is the use of synthetic nutrients in the cultivation of microalgae, accounting for a significant portion of the environmental impacts and energy consumption (Lardon *et al.*, 2009). Some studies claim that if part of the nutrients used in the process were replaced by industrial and domestic effluents, the potential culture media for these organisms, there would be a reduction in the environmental impacts of this process and in energy demand (Lardon *et al.*, 2009; Clarens *et al.*, 2010; McGinn *et al.*, 2011).

Thus, the use of waste from agricultural sources, livestock or industrial is an alternative to minimize impacts caused by synthetic nutrients. The liquid residue biodigested beef, rich in nitrate and phosphate, is an option in the cultivation of microalgae, aiming at the production of biomass for biofuel production, and thus can be an alternative for this activity environmentally economic and sustainability.

Moreover, some alternatives for biofuels have been questioned due to environmental impact in the life cycle, such as biodiesel, palm stemmed from Malaysia, according to the emissions from the transformation of the land use. In this sense, this work aims to identify preliminarily the environmental impacts of microalgae biomass produced in the laboratory. Two crop alternatives are compared: the cultivation of microalgae with synthetic nutrients and an alternative means of waste from cattle, previously biodigested in digesters.

Additionally, we will evaluate the steps that occur in greater environmental impacts using the technique of Life Cycle Assessment (LCA).

This study aims to contribute to the scientific debate on important environmental aspects and impacts on the use of manure and/or wastewater in cultures of microalgae for biomass production. This research also expand to future studies

that aim to improve the environmental performance of the product, still in the laboratory, and supports the work of research teams for the development of sustainable innovations.

2. METODOLOGY

2.1 Cultivation Conditions

The species of microalgae used for this study was the *Scenedesmus* sp. isolated in Center for Research and Development of Energy self-sustaining (NPDEAS) at the Federal University of Paraná (UFPR).

Where cultures were performed in erlenmeyer flask (borosilicate) with volume capacity of 2 L, and the useful volume used in each erlenmeyer flask was 1.8 L, as shown in figure 1. The culture media used for the growth of microalgae were half synthetic Chu (CHU, 1942) and cattle effluent from an anaerobic digester.



Figure 1. System cultivation of microalgae.

These cultures were inoculated with an initial biomass 0.1 g.l^{-1} and maintained in conical flask until the tenth day, when reached their maximum growth to be held the separation of biomass of the growth medium.

For optimum experimental condition the crops were kept in air-conditioned environment with controlled temperature to $22^{\circ}\pm 2^{\circ}$ with lighting about 111.5µmol photons. m⁻².s⁻¹ and photoperiod 12/12 obtained by means of fluorescent lamps, being constant aeration performed by air pumps, regulated to an approximate flow 0.076 l.s⁻¹ in table 1, is the quantity of nutrients used for the crops in synthetic medium.

Reagent	Fórmula	g.L ⁻¹
Sodium nitrate	NaNO ₃	25 g
Calcium chloride dihydrate	CaCl ₃ .2H ₂ O	2.5 g
Magnesium sulfate heptahydrate	MgSO ₄ .7H ₂ O	7.5 g
Potassium phosphate dibasic	K ₂ HPO ₄	7.5 g
Monobasic potassium phosphate	KH ₂ PO ₄	17.5 g
Sodium chloride	NaCl	2.5 g
Titriplex III (EDTA)	$C_{10}H_{14}N_2Na_2O_8.2H_2O$	50 g
Potassium hydroxide	КОН	31 g
Ferrous sulfate heptahydrate	FeSO ₄ .7H ₂ O	4.98 g
Boric acid	H_3BO_3	11.42 g
Zinc sulfate heptahydrate	$ZnSO_4.7H_2O$	0.00882 g
Manganese chloride tetrahydrate	MnCl ₂ .4H ₂ O	0.00144 g
Sodium molybdate	Na ₂ MoO _{4.} 2H ₂ O	0.00119 g
Copper sulphate pentahydrate	CuSO ₄ .5H ₂ O	0.00157 g
Cobalt nitrate hexahydrate	Co (NO ₃) ₂ .6H ₂ O	0.00049 g

Table 1: nutrients used in the cultivation of microalgae for the synthetic medium Chu

2.2 Life cycle assessment (LCA) Preliminary

This item presents a comparative environmental assessment was held between the process of microalgae cultivation using synthetic nutrients and the process using effluent from cattle. This study was followed by the technique of LCA, where are established four phases: LCA: scope definition, inventory analysis, impact assessment and life cycle interpretation (ABNT, 2009a and 2009b). The preliminary characteristics of the present study, however, should be considered:

- (a) the study is from cradle to gate (and therefore does not include all the stages of the product life cycle, according to requirement of NBR ISO 14 040 and NBR ISO 14 044 (ABNT, 2009a and 2009b),
- (b) the data of the processes throughout the product life cycle comes from the data base, predominantly from Europe, except the data of production of microalgae,
- (c) the environmental impact assessment method used is regionalized to European conditions.

2.2.1 Scope of Study

This work is used as the unit of reference flow analysis, by limiting itself to a study gate the gate (i.e., it is not considered the production, distribution and use of biofuel), which consists in the production of 10 g of biomass of microalgae species *Scenedesmus* sp.. Data were collected regarding theassembly processes of cropping systems, the process of acquisition of raw materials, transportation, the microalgae cultivation and microalgae biomass recovery, as represented in the product system 1 (crops with synthetic medium Chu) shown in Figure 2, and product system 2 (crops with wastewater of cattle) below in Figure 3.

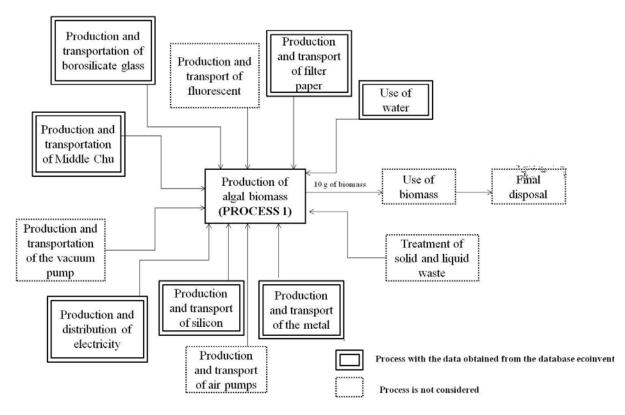


Figure 2. Border of the cultivation system in synthetic medium Chu.

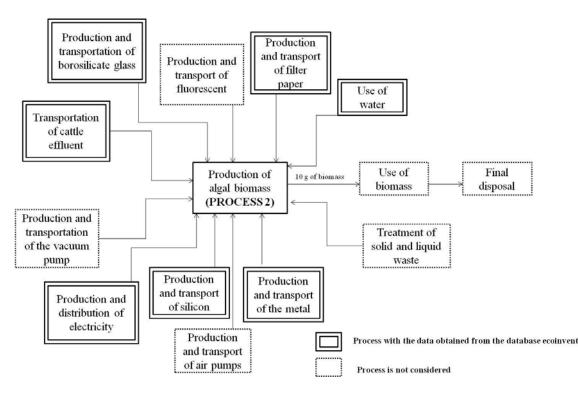


Figure 3. Border of the cultivation system with effluent from cattle.

2.2.2 Analysis of Life-Cycle Inventory and Interpretation

The data relating to the microalgae cultivation were collected in the laboratory of NPDEAS, in addition to the data concerning the Assembly of cropping systems, raw materials (synthetic nutrients), transportation, cultivation of microalgae and recovery of biomass. Electric power data were extracted from the database Center ecoinvent v.2 (Center Ecoinvent, 2010), and used the Brazilian energy matrix.

The collected data were compiled and the results obtained from the use of the software *SimaPro 7.3*, where comparative environmental assessment of the crops was chosen the CML method, because it is the most widely used in studies of LCA of microalgae. The CML method presents several environmental impact categories, converting the data flows of matter and energy in units of specific measures, according to the functional unit proposed by the study (Guinee, 2002) and for this method, five categories were selected: abiotic depletion, eutrophication, global warming potential (GWP 100), ecotoxicity freshwater and marine ecotoxicity. In this work we used the method CML 2 baseline 2000 v2.05 - World 1995.

Due to the difficulty of obtaining primary data, in this study were not included fluorescent lamps used in the artificial lighting system, compressors and centrifugal pumps, but in the case of electrical equipment, was considered only the energy consumption during the process of obtaining the reference flow of 10 g of dry biomass of microalgae.

The results obtained in steps of Inventory Analysis and impact assessment of the life cycle were shown in item 3 of this work and compared to the cropping systems.

3. RESULTS AND DISCUSSIONS

3.1 Life Cycle Inventory

In tables 2, 3 and 4 are presented the comprehensive lists of system flows for both product systems, and they refer to the reference flow proposed in item 2.2.1, respecting the boundaries of product system and including the assembly of systems to the recovery of microalgae biomass. Although this work do not reach the scale of industrial production, it presents a real data obtained *on the spot*, representing the reality of the microalgae cultivation, held at NPDEAS.

Based on life cycle inventory, it was possible to carry out the environmental impact assessment of the proposed means of cultivation, using as criteria the selected categories from CML method. The data from the tables below were obtained through enquiries during 8 months, and this time was enough for obtaining 10 g of dry biomass in laboratory scale in NPDEAS, as a study system. It was considered the necessary breaks of vitreous materials for the cultivation, as the Erlenmeyer's and beakers This way, it was possible to calculate the quantities of materials described in the tables below, for the reference unit of 10 g of dry biomass.

INPUTS	Unit	Quantity Chu	Quantity effluent	Reference
NATURAL RESOURCES				
Secondary raw material				Primary data
Glass (Erlenmeyer)	р	3	4	Primary data
Glass (pipette)	р	3	4	Primary data
Silicone (hoses)	m	1	1	Primary data
Silicone (stoppers)	р	1	1	Primary data
Metal (closet cultivation)	р	0.5	0.5	Primary data
Air pump	р	1	1	Not considered
Fluorescent lamps	р	1	1	Not considered
ENERGY RESOURCES				
Energy and transport				
Transportation (erlenmeyrs and pipette)	Tkm	2,233	2,979	Primary data
Transportation (silicone stopper - hoses)	Tkm	84.8	84.8	Primary data
Transportation (Closet cultivation)	Tkm	4,272	4,272	Primary data
Transportation (fluorescent lamps)	Tkm	-	-	Not considered
Transportation (air pump)	Tkm	-	-	Not considered
OUTPUTS				
Product				
Cropping system	р	1	1	Primary data

Table 2. Inputs and outputs of the Assembly of the systems for the cultivation of microalgae.

Table 3. Inputs and outputs of the cultivation media used.

INPUTS	Unit	Quantity Chu	Quantity effluent	Reference
NATURAL RESOURCES				
Potable water	L	20.145	19	Primary data
CO ₂	L	740.5	1089	Primary data
Nutrients				
Efluente da bovinocultura	L	-	13.5	Primary data
Sodium nitrate	g	7.65000	-	Primary data
Calcium chloride dihydrate	g	0.76500	-	Primary data
Magnesium sulfate heptahydrate	g	2.29500	-	Primary data
Potassium phosphate dibasic	g	2.29500	-	Primary data
Monobasic potassium phosphate	g	5.35500	-	Primary data
Sodium chloride	g	0.76500	-	Primary data
Titriplex III (EDTA)	g	1.53000	-	Primary data
Potassium hydroxide	g	0.94860	-	Primary data
Ferrous sulfate heptahydrate	g	0.15239	-	Primary data
Boric acid	g	0.34945	-	Primary data
Zinc sulfate heptahydrate	g	0.00027	-	Primary data
Manganese chloride tetrahydrate	g	0.00004	-	Primary data
Sodium molybdate	g	0.00004	-	Primary data
Copper sulphate pentahydrate	g	0.00005	-	Primary data
Cobalt nitrate hexahydrate	g	0.00001	-	Primary data
ENERGY RESOURCES				
Energy and transport				
Electricity (lamps)	kWh	71.8	105.6	Primary data
Electricity (air pump)	kWh	53.8	79.2	Primary data
Transportation (cattle effluent)	Tkm	-	3.105	Primary data
Transportation (nutrients Chu)	Tkm	38.6	-	Primary data
OUTPUTS				
Product				
Cultivation of microalgae	р	17	25	Primary data

INPUTS	Unit	Quantity Chu	Quantity effluent	Reference
NATURAL RESOURCES				
Secondary raw material				
Centrifugal pump	р	0.5	0.5	Not considered
Filter paper	р	374	550	Primary data
ENERGY RESOURCES				
Energy and transport				
Electricity (centrifugal pump)	kWh	4	5	Primary data
Transport centrifugal	TKm	-	-	Not considered
Transport filter paper	TKm	328.6	483.3	Primary data
OUTPUTS				
Product				
Microalgal biomass	g	10	10	Primary data
Liquid waste				
Water residuary	L	25.9	38.1	Not considered

Table 4. Inputs and outputs of microalgal biomass recovery step.

3.2. Environmental Impact Assessment

With the lifting of data and construction of the life-cycle inventory, it was possible to carry out the environmental impact assessment, using as criteria the impact categories cited in item 2.2.2. The following chart shows the results of the environmental assessment of the microalgae cultivation, using the synthetic medium Chu and with the effluent from cattle.

For the Assembly step of cropping systems, it may be noted that the Assembly for the production of biomass via effluents from cattle, in this study, exhibited higher values the cultivation system for the synthetic medium Chu. These higher values, in the five categories presented, are justified by the fact that the biomass yield per litre of cultivation with effluent from cattle (0.4 g.L^{-1}) is less than with synthetic medium cultivation Chu (0.6 g. L^{-1}), and to meet the reference of this work flow of 10 g of biomass of microalgae, a larger structure is required to provide the same amount of biomass for both systems at the same time period. This lower yield is justified by the fact that effluent from cattle has higher concentration of organic matter, making it more cloudy than synthetic medium, what hinders the passage of light, which is essential factor to the growth of algae.

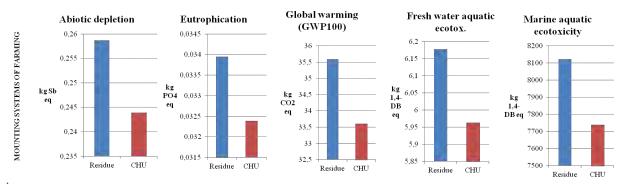
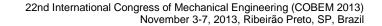


Figure 2. Step mounting systems for the cultivation of microalgae.

In step of microalgae cultivation, as shown in Figure 3, comparing the use of effluent from cattle as a cultivation means and a synthetic medium, the use of effluents presented a significant contribution in reducing environmental impacts in the categories abiotic depletion and eutrophication, and as this chart, for the category of eutrophication, the use of effluent from cattle in the cultivation of microalgae in 57.2% reduced environmental impacts in this step. For this category, the industrial processes of production of synthetic medium Chu may be directly associated to the greatest generation of environmental impacts.

For the categories of global warming, ecotoxicity and freshwater aquatic ecotoxicity marine, most of these impacts can be linked directly to greater energy consumption in cultivation with the effluent from cattle. According Galindro (2012) this large energy consumption is one of the main obstacles at the cultivation of microalgae, because the energy



generated by the burning of fuel, produced from biomass, will likely be less than the total energy used for its production.

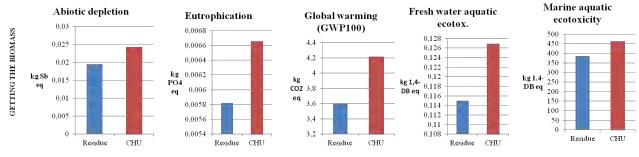


Figure 3. Cultivation of microalgae with both selected media Chu and effluent from cattle.

Therefore, to reduce the impact generated in step of cultivation of microalgae, an alternative would be to reduce the use of electric energy consumption, for example, in artificial lighting (lamps), so would imply a significant reduction of impacts, generated in this step of the product system and compared to the obtaining of biomass of microalgae, where a way to resolve that, would be the study of feasibility of the completion of cultivation in open environment, using sunlight as a source of illumination.

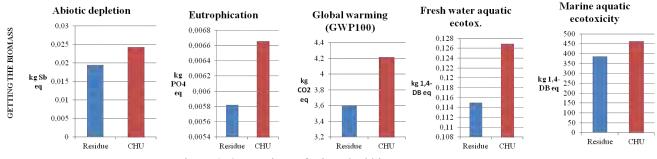


Figure 4. Comparison of microalgal biomass recovery step.

In the chart above (Figure 4), presents the comparison of biomass recovery stage for the two systems, and it should be noted that for all the selected impact categories, the use of effluent from cattle from biodigestors reduced environmental impact. These facts indicate that the use of effluent from cattle as a means of cultivation is a possible alternative to production, for generates less environmental impacts to the recovery stage of biomass of microalgae.

So, from the results presented, are demonstrated the need for adjustments in steps of Assembly of systems and cultivation for biomass production of microalgae, proposing adjustments in the production process, to achieve a more environmentally-friendly production scenario.

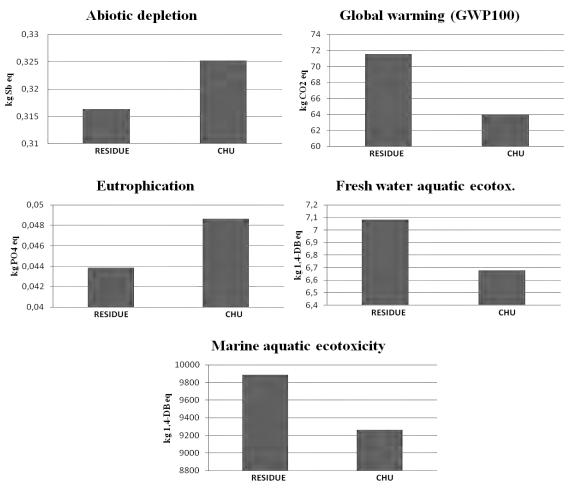


Figure 5. Graph comparing the two systems of product in three steps considered.

For a better interpretation of the results, in Figure 5, it was compared the 3 steps for obtaining the reference flow of both product systems considered in the work. This graph confirms, joining with the discussions has been made, that the microalgae cultivation reduce the categories of impact, with use of manure of cattle, and also confirms that the use of these wastes reduce nutrient impacts in the categories of eutrophication and abiotic depletion, and as has been said, LCA studies more in-depth should be made in the future.

And according to Oliveira (2013), the use of biodigested bovine effluent is an option to replace the synthetic growth medium of microalgae and it appears as an alternative to reduce the costs of its production in commercial plants, which have high nitrogen and phosphorus load, composition required for the development of microalgae, and creating an alternative to the use of synthetic nutrients. In addition of this, the microalgae is able to reduce the organic load, generating these effluents treatment, and may be disposed of an environmentally correct way.

4. CONCLUSIONS

By conducting life cycle assessment of the proposed means of cultivation, it was possible to observe that the impacts generated by the microalgae cultivation can be reduced when using the bovine effluent as growing medium, where in steps of stages of microalgae cultivation and obtaining of biomass, it was observed the reduction of environmental impacts, and according to the technique of life cycle assessment, it was possible to observe that the cropping systems analyzed, using synthetic nutrients as a means of cultivation, they present significant environmental impacts in relation to eutrophication and abiotic depletion. Before the results, the use of effluent from cattle indicates a possible alternative of production that generates less environmental impacts as it dispenses the use of synthetic nutrients as medium. Also it was possible to identify that in step cultivation systems mounting, the mounting into the middle with effluent was the main responsible for the majority of the environmental impacts in all categories, so, it's interesting to propose new alternatives for this step, aiming at the reduction of environmental impacts. Complementing this fact, it is important to emphasize the LCA studies of microalgae biomass production must include laboratory steps, therefore, from this work, it was possible to realize the relevance these processes, relative to the generation of environmental impacts. This work made it possible to assess the life cycle using effluent from cattle as an alternative source of nutrients for the production

of microalgae with better environmental performance compared to traditional synthetic medium Chu, thus completing the initial proposed objective, emphasizing the need for deeper studies.

Despite the benefits of this work, for future works we suggest a more systemic LCA, avoiding the exchange of environmental impacts.

3. ACKNOWLEDGEMENTS

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4. REFERENCES

- ABNT, 2009a. NBR ISO 14040: Gestão ambiental Avaliação do ciclo de vida Princípios e estrutura. Rio de Janeiro, p.20.
- ABNT, 2009b. NBR ISO 14044: Gestão ambiental Avaliação do ciclo de vida Requisitos e orientações. Rio de Janeiro, p.46.
- Antoni, D.; Zverlov, V. V.; Schwarz, W. H.. Biofuels from microbes. Appl. Microbiol. Biotechnol. n. 77. p.23-35, 2007.
- Clarens, A. F.; Resurreccion, E. P.; White, M. A.; Colosi, L. M. Environmental life cycle comparison of algae to other bioenergy feedstocks. Environmental science & technology, 44(5), 1813-9, 2010.
- Chehebe, J. R. B. Análise do ciclo de vida dos produtos ferramenta gerencial da ISO 14.000. Rio de Janeiro: Ed. Qualitymark, 1998.
- Chisti, Y.. Biodiesel from microalgae. Biotechnology Advances. Nº 25, p.294-306, 2007.
- Chu, S. P. The influence of the mineral composition of the medium on the growth of planktonic algae. J Ecol, v. 30, p. 284-325, 1942.
- Ecoinvent reports. Dusseldorf: Swiss Centre for Life Cycle Inventories. v.2.2, p.25, 2010.
- Galindro, B. M.. Análise Técnica e Avaliação do Ciclo de Vida de Culturas de Produção de Microalgas para Biodiesel. Dissertação (Pós-graduação em Engenharia Ambiental). Universidade Federal de Santa, Florianópolis (SC). p.92, 2012.
- Ghassan, T. A., Mohamad, I. A. L., Widyan, B., Ali, O, A.. Combustion performance and emissions of ethyl ester of a waste vegetable oil in a water-cooled furnace. Appl. Thermal Eng., v.23, p.285-293, 2003.
- Grima, E. M. *et al.*. Recovery of microalgal biomass and metabolites: process options and economics. Biotechnology Advances, v. 20, p. 491-515, 2003.
- Guinée, J. B. Handbook of Life Cycle Assessment: Operational Guide to the ISO Standards. Dordrecht: Kluwer Academic Publishers, 2002.
- Lardon, L.; Helias, A.; Sialve, B.; Steyer, J.; Bernard, O.. Life-cycle assessment of biodiesel production from microalgae. Environmental Science and Technology. n. 17, p.6475-6481, 2009.
- Lourenço, S. O.. Cultivo de microalgas marinhas princípios e aplicações. São Carlos: RiMa, p.606, 2006.
- McGINN, P. J.; Dickinson, K. E.; Bhatti, S.; Frigon, J.; Guiot, S. R.; O'Leary, S. J. B. Integration of microalgae cultivation with industrial waste remediation for biofuel and bioenergy production: opportunities and limitations. Photosynth. Res. n. 109, p.231-247, 2011.
- Oliveira, A C., Produção de biomassa de microalgas Scenedesmus sp. em efluente de bovinocultura biodigerido. 2013. 82 f. Dissertação (Mestrado) - Curso de Engenharia e Ciência dos Materiais, Universidade Federal do Paraná, Curitiba, 2013
- Rodolfi, L., Chini Zittelli, G., Bassi, N., Padovani, G., Biondi, N., Bonini, G. & Tredici, M. R. Microalgae for Oil: Strain Selection, Induction of Lipid Synthesis and Outdoor Mass Cultivation in a Low-Cost Photobioreactor. Biotechnology and Bioengineering 102: p.100-112, 2009.

5. RESPONSIBILITY NOTICE

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