

MAURO HENRIQUE BATALHA DE SOUZA

**APLICAÇÃO DE MICROALGAS NO SOLO PARA
RECUPERAÇÃO DE NITROGÊNIO: UMA ABORDAGEM DE
CICLO DE VIDA**

Dissertação apresentada à Universidade Federal de Viçosa, como parte das exigências do Programa de Pós-Graduação em Engenharia Civil, para obtenção do título de *Magister Scientiae*.

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APROVADA: 28 de fevereiro de 2019.



Maria do Carmo Calijuri



Eduardo de Aguiar do Couto



Maria Lucia Calijuri
(Orientadora)

**Aos meus pais Mauro e Cristina;
Aos meus avós Olavo e Maria;
Ao meu irmão Gabriel;
E à minha amada, Raquel.
Dedico.**

**“Uma mente necessita de livros da mesma forma
que uma espada necessita de uma pedra de
amolar, se quisermos que se mantenha afiada”.**

(George R. Martin – Game of Thrones)

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BIOGRAFIA

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RESUMO

SOUZA, Mauro Henrique Batalha, M.Sc, Universidade Federal de Viçosa, fevereiro de 2019. **Aplicação de microalgas no solo para recuperação de nitrogênio: uma abordagem de ciclo de vida.** Orientadora: Maria Lúcia Calijuri.

A aplicação de biomassa de microalgas no solo representa uma alternativa de uso eficiente de fertilizantes. No presente estudo, os impactos ambientais gerados pela aplicação de 1 kg de nitrogênio da biomassa de algas (biofertilizante) foram mensurados por meio da análise de ciclo de vida. O nitrogênio foi recuperado de um efluente da indústria de processamento de carne em uma lagoa de alta taxa. Os impactos relacionados a toda a cadeia do biofertilizante foram significativos principalmente as mudanças climáticas (115 kgCO₂eq). Outras categorias (formação de partículas, acidificação terrestre, eutrofização de água doce e ecotoxicidade de água doce) não foram muito representativas. O cultivo da biomassa foi a etapa mais crítica em relação ao consumo de energia e tempo. Por outro lado, o uso de efluentes como meio de cultivo para o crescimento de microalgas reduziu as categorias de impacto, como a eutrofização de água doce. Os resultados mostraram que as etapas de cultivo e colheita de microalgas precisam ser tecnologicamente desenvolvidas, especialmente quando comparadas a um fertilizante convencional já estabelecido no mercado. Para tornar o biofertilizante de microalgas ambientalmente vantajoso, essas alternativas devem ser priorizadas: i) o uso de energia fotovoltaica em vez de energia hidrelétrica; ii) o uso de um efluente com maior concentração de nitrogênio; e iii) a consideração da compensação ambiental para o tratamento de efluente, desconsiderando o estágio de produção de biomassa.

ABSTRACT

SOUZA, Mauro Henrique Batalha de, M.Sc., Universidade Federal de Viçosa, February, 2019. **Soil applications of microalgae for the recovery of nitrogen: a life-cycle approach.** Adviser: Maria Lúcia Calijuri.

The application of algal biomass in the soil represents an alternative of efficient use of fertilizers. In the present study, the environmental impacts generated by the application of 1 kg of nitrogen from the algal biomass (biofertilizer) were analyzed through life cycle analysis. Nitrogen was recovered from a meat processing industry effluent in a high-rate algal pond. Impacts related to the entire biofertilizer chain were mainly impacting on climate changes (115 kgCO₂eq). Other categories (particle formation, terrestrial acidification, freshwater eutrophication and freshwater ecotoxicity) were not very representative. Biomass cultivation was the most critical step regarding energy and time consumption. On the other hand, the use of effluent as the culture medium for microalgae growth reduced impact categories, such as freshwater eutrophication. Results showed that microalgae cultivation and harvesting steps need to be technologically developed, especially when compared to a conventional fertilizer already established in the market. In order to make microalgae biofertilizer environmental advantageous, alternatives should be beforehand: i) the use of photovoltaic energy instead of hydropower energy; ii) the use of a nitrogen richer effluent; iii) and the consideration of an environmental compensation for the treatment of effluent can be accounted for, disregarding the biomass production stage.

1. APRESENTAÇÃO

Esse trabalho é parte dos estudos realizados no Núcleo de Pesquisas Ambientais Avançadas (nPA) do Departamento de Engenharia Civil da Universidade Federal de Viçosa (UFV) que englobam tratamento de efluentes e produção de biomassa algal, bem como obtenção de bioenergia e outros produtos de valor agregado derivados dessa integração. O objetivo desse estudo foi identificar os impactos ambientais em cada etapa da cadeia de produção de um biofertilizante oriundo de microalgas cultivadas em efluente industrial.

Resultados obtidos de acordo com Castro (2016) mostram que a biomassa algal pode ser utilizada para aplicação no solo, com a possibilidade de aumento de matéria orgânica e nutrientes. Além disso, se for produzida em uma propriedade onde há o exercício de atividades agrícolas, o resíduo produzido pode ser aproveitado no mesmo local gerando compensação econômica e ambiental.

A pesquisa é parte integrante do projeto “Biofilme de microalgas: tecnologia inovadora para mitigação das mudanças climáticas e desenvolvimento sustentável no setor agropecuário”, Processo nº 37/2013 – Mudanças Climáticas, aprovado em 2013, financiado pelo CNPq – Conselho Nacional de Desenvolvimento Científico e Tecnológico, Processo 404027/2013-0. O documento foi organizado em um capítulo principal, além de introdução geral, hipóteses e objetivos. O artigo aborda o impacto ambiental relacionado em todo o ciclo para utilização de microalgas como biofertilizante, desde sua produção até a disposição no solo, mostrando onde estão os gargalos de tal produção e eventuais alternativas menos impactantes do ponto de vista da avaliação de ciclo de vida.

2. INTRODUÇÃO GERAL

Muitos são os desafios que devem ser superados pela sociedade moderna: produção de alimentos e de energia, escassez quali-quantitativa de água, proteção ambiental (Batista et al., 2015), dentre outros. Segundo Oilgae (2014), a energia necessária para atividades diárias como luz e transporte dobrará de 2008 a 2035, assim como a demanda por alimentos. Além disso, a demanda por água tem estimativas de crescer 10% ao ano, na qual a agricultura corresponde a cerca de 70% do consumo total. Para resolução de tais problemas, buscam-se alternativas que venham a minimizar tanto a demanda energética quanto a demanda de água. Nesse contexto, fontes de energia renováveis, como as provenientes da biomassa algal vem sendo amplamente pesquisadas e utilizadas.

Microalgas vêm se mostrando como uma alternativa viável para tal problemática. Seu cultivo não necessita de água potável e terras aráveis (Borowitzka e Moheimani, 2013), e apresentam um percentual lipídico superior à demais culturas terrestres (Trivedi et al. 2015). Adicionalmente, podem apresentar uma gama diversa de produtos valorados energeticamente, tais como bioetanol, biodiesel, biogás e *biojets* (Sialve et al., 2009; Slade e Bauen, 2013; Castro et al., 2016).

A produção de microalgas tendo efluentes como meio de cultivo pode ser considerada um método mais econômico de produção de biomassa algal, além de ser eficiente do ponto de vista do tratamento desses efluentes (Gross et al., 2013). Águas residuárias, como o esgoto doméstico contém água e nutrientes necessários para o crescimento das microalgas, que geram biomassa capaz de ser aproveitada como matéria-prima para diversos bioprodutos, recuperando os nutrientes presentes nos efluentes (García et al, 2006; Powell et al, 2009; Park e Craggs, 2010).

Esta recuperação de nutrientes posteriormente pode ser utilizada para aplicação no solo e contribuir para redução de gases de efeito estufa (GEE) pelo solo no período de crescimento da cultura, aumento da fertilidade do solo e melhoria de suas características físico-químicas ao longo do tempo (Castro, 2016). A mesma autora adverte para a necessidade de se compreender a interação microalga-solo ao longo do tempo e verificar se outras variáveis, como a umidade do solo, influenciam nas melhorias citadas anteriormente.

Se por um lado, há o favorecimento do solo cultivado, no outro há a incerteza se a produção e o manejo das microalgas podem acarretar em impactos ambientais negativos, e ainda sobre o método de mensuração dos mesmos. Para se avaliar e contabilizar o impacto da produção de microalgas, uma importante ferramenta é a análise de ciclo de vida (ACV).

ACV é uma metodologia utilizada para avaliar aspectos ambientais associados a um produto, sendo realizada através do levantamento e quantificação da energia e dos materiais necessários a produção, entradas do sistema e dos resíduos e emissões liberados ao meio ambiente, saídas do sistema. Funciona ainda como um instrumento de gestão, auxiliando na tomada de decisões na indústria, organizações governamentais ou não governamentais, permitindo ao responsável pela decisão selecionar indicadores pertinentes ao desempenho ambiental, além do marketing, dando ao produto rotulagem ecológica ou declaração ambiental. A ACV é conhecida por ser uma metodologia capaz de compilar e avaliar entradas, saídas e potenciais impactos ambientais de um sistema ou produto ao longo do seu ciclo de vida e tem sido amplamente utilizada comparando-se a produção e utilização de subprodutos advindos de microalgas (Medeiros et al., 2015, Fortier et al., 2014, Collet et al., 2015). Atualmente, há várias iniciativas ao redor do mundo visando principalmente a padronização dos estudos e construção de banco de dados em escala local, consolidando assim a metodologia (Fortier et al., 2014; Gutierrez, 2014).

A evolução tem-se dado, principalmente, pois os principais estudos vêm discutindo os dados apenas sob um prisma energético comparando-se os combustíveis oriundos de microalgas com os convencionais (Jorquera et al., 2010; Collet et al., 2011). Percebe-se que as metodologias abordadas focam somente em GEE, embora outros impactos são tão importantes quanto, como por exemplo eutrofização e uso do solo e da água.

Além disso, há carência da base de dados regionalizada e em maiores escalas, fazendo com que os resultados sejam discrepantes. Dados de plantas comerciais colaborariam para um quadro mais claro da situação (Sing e Gu, 2010).

Esta pesquisa visa avaliar os impactos ambientais potenciais relacionados ao cultivo de microalgas em efluentes, e a utilização da biomassa produzida como biofertilizante, desde o cultivo da biomassa até sua disposição no solo.

Foram adotados princípios das normas da série ABNT NBR ISO 14.040 e 14.044 para gestão ambiental e avaliação do ciclo de vida. Foi utilizado o software SimaPro®, frequentemente empregado em pesquisas de ACV (PRÉ, 2013). A unidade funcional escolhida foi tal que se conseguisse contemplar e comparar toda a cadeia envolvida. Todas as emissões, materiais e consumo de energia estão referidas a esta unidade funcional.

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3. HIPÓTESES

Para a biomassa algal produzida em efluentes:

- A recuperação de nitrogênio a partir da biomassa para produção de biofertilizante é ambientalmente menos impactante que a produção de fertilizantes sintéticos;
- Há alternativas tecnológicas capazes de fazer com que, combinadas, o impacto de produção e recuperação da biomassa algal seja reduzido.

4. OBJETIVO

4.1 OBJETIVO GERAL

Utilizar ACV como ferramenta para propor opções, menos impactantes ambientalmente, na utilização de microalgas cultivadas em efluentes para produção de biofertilizante nitrogenado.

4.2 OBJETIVOS ESPECÍFICOS

- Identificar e descrever as rotas de recuperação energética da biomassa algal considerando as opções tecnológicas utilizadas e as várias combinações de tecnologia, além disso, quantificar os fluxos de entrada e saída dos sistemas.
- Apontar possíveis oportunidades de melhoria nos sistemas em estudo quando comparado as formas tradicionais, tendo como premissas que tais melhorias sejam de cunho ambiental a partir da ACV.

5 SOIL APPLICATIONS OF MICROALGAE FOR THE RECOVERY OF NITROGEN: A LIFE-CYCLE APPROACH

Abstract: The application of algal biomass in the soil represents an alternative of efficient use of fertilizers. In the present study, the environmental impacts generated by the application of 1 kg of nitrogen from the algal biomass (biofertilizer) were analyzed through life cycle analysis. Nitrogen was recovered from a meat processing industry effluent in a high-rate algal pond. Impacts related to the entire biofertilizer chain were mainly impacting on climate changes (115 kgCO₂eq). Other categories (particle formation, terrestrial acidification, freshwater eutrophication and freshwater ecotoxicity) were not very representative. Biomass cultivation was the most critical step regarding energy and time consumption. On the other hand, the use of effluent as the culture medium for microalgae growth reduced impact categories, such as freshwater eutrophication. Results showed that microalgae cultivation and harvesting steps need to be technologically developed, especially when compared to a conventional fertilizer already established in the market. In order to make microalgae biofertilizer environmental advantageous, alternatives should be beforehand: i) the use of photovoltaic energy instead of hydropower energy; ii) the use of a nitrogen richer effluent; iii) and the consideration of an environmental compensation for the treatment of effluent can be accounted for, disregarding the biomass production stage.

Keywords: Nutrient recovery; Biofertilizer; High rate algal ponds; Life cycle analysis; Algal biomass

5.1. INTRODUCTION

Nitrogen (N) is a resource obtained predominantly by the Haber-Bosch process. Approximately 80–90% of N synthesized by this process is used to produce fertilizers to grow more than half of the food consumed worldwide, making the process larger than the sum of all natural land processes combined (Galloway and Cowling, 2002; Galloway et al., 2008). The remaining N is used in the chemical industry for the production of nylon, plastics, explosives, and animal supplements (Galloway and Cowling, 2002; Galloway et al., 2008). The magnitude of human influence on N fluxes has substantially altered the natural N cycle, which has serious consequences on water resources (e. g. eutrophication), the lithosphere (saturation of N in the soil and impacts on biodiversity), the atmosphere (greenhouse gases - GHGs), acid rain, atmospheric pollution), and human systems (resource and economic restrictions). Although N is a renewable resource, the Haber-Bosch process is extremely energy demanding, accounting for approximately 1% of the world's total energy consumption (Matassa et al., 2015).

With regard to environmental aspects, the use of any type (chemical or biological) of N fertilizers increases N content in the soil and favors the emission of nitrous oxide (N₂O), an important GHG that affects global warming. In light of this, the Intergovernmental Panel on Climate Change has predicted that food production worldwide will suffer dramatic impacts in the coming decades due to the average increase in global temperatures (IPCC, 2007). Agriculture and livestock contribute to climate change as much as they suffer its consequences. Emissions of methane (C), carbon dioxide (CO₂), carbon monoxide (CO), N₂O, and nitrogen oxides (NO_x), which are potential gases for global warming (GWP), have been generated by different agricultural practices and livestock management methods (Lima, 2002). Therefore, better soil management practices and the look for alternatives for the recycle of N from ecosystems, or at least to reduce the demand at the source, could help to minimize undesirable effects.

Among several sources of this nutrient, we highlight the effluents. Many industrial practices result in high nutrient levels and the nutrients may accumulate in ecosystems as a consequence of lack of treatment or inadequate treatment of these effluents. An alternative to the recycle, avoiding new exploitation, of these nutrients is the cultivation of microalgae associated with the treatment of wastewater. Although, it would not prevent N accumulation in ecosystems, it could allow a better management of the nutrient cycle.

Microalgae have evolved to assimilate nitrogen and phosphorus mainly in environments where nutrients are scarce, thus achieving high maximum specific absorption rates (Lehman and Scavia, 1982) and making these microorganisms extremely effective for

use in biochemical processes of nutrient recovery. Specifically, microalgae are normally used for effluent polishing, i.e. for phosphorus, ammonia, and/or nitrate removals (Silva-Benavides and Torzillo, 2012; Shoener et al., 2014; Uggetti et al., 2014; Whitton et al., 2016). Microalgae also produce biomass that can be used for fertilizers and bioenergy feedstock, providing additional revenue and improving the financial viability of the recovery method (Pittman et al., 2011). Castro et al. (2017) performed an experiment using microalgal biofilm as a nitrogen source for the cultivation of *Pennisetum glaucum*. Authors concluded that the establishment of a microalgal biofilm in the soil favored lower nitrogen loss through ammonia volatilization and increased organic matter content and cation exchange capacity in the soil. Additionally, the shoot dry matter mass production and N content assimilated by the plant were the same as those with urea fertilizer. Results were promising, indicating the potential of the microalgae biomass produced in effluents in the recovery of nutrients as a biofertilizer. However, as a new technology, information about its environmental and economical sustainability is still required.

An important tool to evaluate and account for the impact of microalgae production and valorization is the life-cycle assessment (LCA). According to ISO 14040 (2006), LCA is a methodology used to evaluate the environmental aspects associated to a product, being carried out through the collection and quantification of energy and materials needed for production, inputs of the system and waste and emissions released to the environment. It also supports decision-makers in industry, governmental or non-governmental organizations, allowing them to select indicators relevant to environmental performance, as well as marketing, giving the product eco-labeling or environmental statement. The results are dependent on the system boundary definitions, the database used for the processes, the efficiency of the processes and the functional units (Hill et al., 2006).

A problem with these types of studies is the lack of microalgae-producing facilities that operate under actual conditions and so, in many cases, extrapolation is required to estimate the performance of facilities based on real models. Consequently, a wide range of processes for microalgae production (cultivation systems, cultivated species, processes for harvesting, value products), results in a large variability of technical data. In addition, the methodological perspectives, which address the implementation stages of the LCA, such as functional units and system boundaries, are also widely variable (Collet et al., 2015). Different methodological choices cause discrepancies in current research results even when the technical data are similar (Benemann et al., 2018).

Therefore, the objective of this study was to evaluate and account for the impacts of the production, harvesting and application steps of microalgae biomass used as N fertilizer for

the cultivation of millet (*Pennisetum glaucum*) and to compare the results to the impacts of using a traditional mineral fertilizer (urea). Experimental data obtained during the study of Castro et al. (2017) were thoroughly measured in order to supply the SIMAPRO ® software and then, get real information about the sustainability in using microalgae biofilm as a biofertilizer. The LCA with primary and real experimentation data is the novelty of the work.

5.2. MATERIALS AND METHODS

5.2.1. EXPERIMENTAL CONFIGURATION AND PRIMARY DATA OBTENTION

A high rate algal pond (HRAP) (area = 3.3 m² and volume = 1 m³) operated in batch mode (14 days of operation) was used to produced algal biomass. The effluent used as microalgae culture medium (Table 1) was a primary pretreated effluent collected after a flotation unit from the wastewater treatment plant of a meat processing industry located in the city of Viçosa, Brazil.

Table 1. Wasterwater characterization

Variable	Values
pH	5.7
VSS (mg.L ⁻¹)	571.8
N-TKN (mg.L ⁻¹)	68.4
Total phosphorus (mg.L ⁻¹)	7.4
Total chemical oxygen demand (mg.L ⁻¹)	1918.7

The HRAP had a two blades paddlewheels powered by 1HP electric motor responsible for the operation of 12 ponds. Therefore, only one-twelfth of all spent energy was computed. A CO₂ injection system was also used in order to control the pH of the pond, maintaining it between 7 and 8, thus providing greater productivity and N recovery. The addition of CO₂ was carried out in the daytime using a gas cylinder containing 99% CO₂. A 20 W aquarium pump was used to recirculate the effluent in the carbonation column. The average photosynthetically active radiation (PAR) at 12 p.m. during the cultivation period was 1445.4 ± 548 (µmol/m².s).

After the production, the biomass was harvested using sodium hydroxide (NaOH) at 50% m/v, promoting a pH increasing until 12. Paddlewheels were moved simultaneously for a short period (approximately two hours), generating a hydraulic gradient favorable for coagulation and promoting the sedimentation of the biomass, which was collected after resting the HRAP for 24 h.

Two experimental plots of 4 m² were established and contained different treatments

with different N sources for the millet (*Pennisetum glaucum*) crop. The concentrated biomass was applied manually in each plot in the soil. The treatments consisted of (i) 120 kg ha⁻¹ of N supplied by algal biomass and (ii) 120 kg ha⁻¹ of N supplied by conventional urea. The experiment was conducted over a period of 60 days in the winter. During the experiment, the closed chamber method was used to measure the emissions of CH₄, CO₂, and N₂O in each plot. Moreover, volatilization of ammonia was determined according to Araújo (2009).

More details about the experimental methodology of the biomass production and application in the soil can be found in Castro et al. (2017). As this paper does not aim to evaluate microalgae effect in soil and plant, methodological and results aspects were limited to LCA content. Table 2 presents a summary of the primary data used as entrance data in the LCA.

Table 2. Primary data used as entrance in the LCA.

	Amount per operation	Time of operation	Power (kW)	Details
CO₂	0.27 kg	20 min (4 times during 5 min each)	-	CO ₂ Flux = 7 L/min 99% purity Density = 1.98 kg/m ³
NaOH	1.1515 kg	-	-	50% m/v Density = 1.515 kg/m ³
Paddewheels motor	-	24h during 14 days	0.0613	
Carbonation column pump	-	10h during 14 days	0.02	
Pump for biomass concentration	-	13h	0.02	

In each operation, 0.85 g/L of total kjeldahl nitrogen and 55 L of harvested biomass were obtained. Therefore, 22.99 operations were needed in order to get 1 kg of nitrogen (used as the system output unit for LCA). Each operation lasted 14 days (average time to get a decrease in the algal growth, which was determined by daily monitoring of chlorophyll-a).

5.2.2 LCA USING SIMAPRO 8

This study evaluated the energy consumption and impacts of the cultivation, concentration, and application of the biomass in the soil. Fig. 1 shows the boundaries of the system used in the LCA

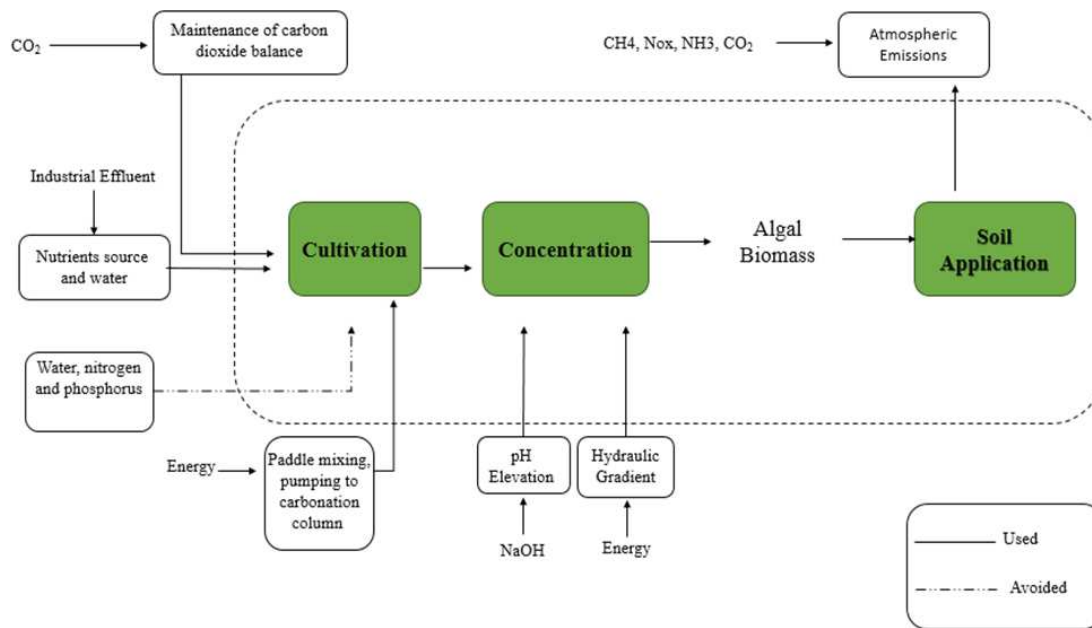


Figure 1. LCA system boundaries.

SimaPro 8.1 software was used in order to quantify the impacts of the three analyzed stages (biomass cultivation, harvesting, and soil application). Because the design of projects of this size has a useful life of approximately twenty-five years, the construction phase was not taken into account. A life-cycle impact assessment (LCIA) was conducted using the ReCiPe midpoint methodology, which is focused on environmental issues, making it, therefore, the most qualified method for this study (PRÉ, 2013). The midpoint method was used to prioritize short or medium term impacts on a constant basis. The method has 18 impact categories, although some of them, such as marine ecotoxicity and ionizing radiation, were not considered since they are not recurrent in environmental impact studies related to effluent treatment and energy recovery.

The categories used in this study were climate change, terrestrial acidification, freshwater eutrophication, human toxicity, terrestrial ecotoxicity, and particle formation. They were chosen because they suffer direct impacts during the evaluated stages.

During the production stage, the inputs were the values of electric energy (kWh) used for the paddlewheels rotation and the effluent recirculation pump into the carbonation column to add the CO_2 into the HRAP. We assumed that water and nutrients were not inputs due to the use of the effluent. In the biomass harvesting stage, the inputs were the volume of NaOH used as a coagulant and the electric energy spent for maintaining the hydraulic gradient in the pond by moving the paddlewheels for approximately two hours. It is important to note that there were two distinct scenarios for the electric energy. The first was the use of hydroelectric energy and the second was the use of photovoltaic solar energy. Finally, in the soil application stage, the inputs were the measured volumes of GHGs. Table 3 presents a summary of the

life-cycle inventories (LCI) used and their units.

Table 3. LCI Inventory.

Stage			Unit	Description
Biomass Production	Input	Industrial Wastewater	m ³	Provide nutrients and water for algal growth
		Electric Energy	kWh	Reversing and passing the effluent through the carbonation column
		CO ₂	kg	pH adjustment and CO ₂ supply for biomass
	Avoided Products	Water	m ³	Products that are no longer used once the cultivation has taken place in effluent
		Nitrogen	kg	
		Phosphorus	kg	
Biomass concentration	Input	Coagulant (NaOH)	kg	pH increasing
		Electric Energy	kWh	Promote hydraulic gradiente
Soil Application	Output	-		
		CO ₂	kg	Gases emitted during the cultivation of millet
		CH ₄	kg	
		NO _x	kg	
	NH ₃	kg		

The inputs for the conventional fertilizer (urea) consisted of the production costs of the fertilizer, which is available in the SimaPRO Software and is based on the Ecoinvent database and the maritime and terrestrial transportation costs. Maritime transport is required because the fertilizer originates in Russia and is imported to Brazil. Land transport was taken into consideration because the ports where the fertilizers arrive are relatively far from the application sites. In the soil application stage, the GHG volumes were measured in the same manner as the GHG volumes from biofertilizer. One kg of N applied to the soil was used as the system output unit.

5.2.3 POSSIBLE SCENARIOS FOR IMPACT REDUCTION

In order to minimize impacts, scenarios with different conditions to those applied in the study were proposed: i) photovoltaic energy as an alternative source, instead of using hydropower energy; ii) using other N-richer effluent; iii) environmental compensation for the

treatment of effluents, disregarding the biomass production stage.

Although considered by many as a renewable source of energy, hydroelectric power plants have a significant amount of impacts. Therefore, a scenario was proposed where all the energy used in the production and harvesting stages of the microalgae biomass (power supply for paddlewheels, effluent recirculation into the carbonation column and mixing after the use of the coagulant) was from a photovoltaic source. For this scenario, all components for the installation of a 3 kWp photovoltaic plant, energy use for the mounting, transport of materials and persons to the construction place were considered, including the disposal of components after end of life.

In the scenario of using other N-richer effluent for culture medium, a double of initial N concentration in the effluent was considered and operation time was kept the same, 14 days.

Lastly, in recent years, the pressure from environmental organizations and environmental legislation has increased and has especially targeted different industries to ensure the treatment of effluents. For this scenario, all the impacts generated for the production stage were disregarded, considering that they come from a mandatory stage in any industry - the effluent treatment process. Therefore, the recovery of the nutrients would add value for the treatment, increasing environmental and economic benefits for the industry.

5.3 RESULTS AND DISCUSSION

5.3.1 LIFE-CYCLE INVENTORY

Tables 4 and 5 show the LCI for the biofertilizer and urea, respectively. Biofertilizer results were based on experiments and, therefore, represent primary data, not literature-based data. Studies carried out in this manner are very important due to a shortage of data in these types of systems, especially in Brazil.

Table 4. Biofertilizer life-cycle inventory.

Stage			Unity	Value
Biomass Production	Input	Industrial Wastewater	m ³	22.99
		Electric Energy	kWh	531.36
		CO ₂	Kg	6.28
	Avoided Products	Water	m ³	22.99
		Nitrogen	Kg	1.07
		Phosphorus	Kg	0.019
Biomass harvesting	Input	Coagulant (NaOH)	Kg	34.83
		Electric Energy	kWh	6.12
Soil Application	Input	-		
	Output	CO ₂	Kg	6.25 x10 ⁻³
		CH ₄	Kg	-1.22x10 ⁻⁶
		NO _x	Kg	4.34 x10 ⁻⁶
		NH ₃	Kg	4.63 x10 ⁻⁵

Table 5. Urea life-cycle inventory.

Stage			Unity	Value
Fertilizer Production	Input	Ureia (46%)	Kg	2.17
Transport	Input	Maritime	Tkm*	20
		Terrestrial	Tkm*	0.8
Soil Application	Input	-		
	Output	CO ₂	kg	4.90 x10 ⁻⁴
		CH ₄	kg	-2.72 x10 ⁻⁶
		NO _x	kg	0.11 x10 ⁻⁶
		NH ₃	kg	1.89 x10 ⁻⁴

From Tables 4 and 5 it was possible to observe that values of electric energy, coagulant and CO₂ were very high for producing 1 kg of N from the microalgae biomass. The energy use can be attributed to the long operating time (14 days) of the HRAP. A slow algae growth was a consequence of the effluent characteristics, mainly the high organic load (Table 1). On the other hand, the use of a secondary treated effluent instead of a primary treated would result in lower or no N recovery.

Urea consists of 46% N (Table 5) and it requires just over two kg of urea to obtain 1 kg of N. In addition, the N from Russia has to travel 20,000 km by ship to reach Brazil and

another 800 km by truck to the site application. Although the land transport mileage is high, Brazil is a very large country; therefore, such values are acceptable for transporting goods between ports and the main agricultural regions.

The gas emission values were negative for methane due to its absorption/oxidation at the source. This phenomenon occurs due to several factors such as soil type, NH_4^+ concentration in the soil, soil moisture and temperature. Therefore, the inflow found in the field analyzes by Castro et al. (2017) was used as a compensation for GHG emissions. The high values of CO_2 emitted by the biofilm when applied to the soil were due to the development of microorganisms other than microalgae in the soil, such as bacteria and fungi. Zhang and Wang (2005) and Zhang et al. (2007) stated that CO_2 can be generated by the microbial decomposition of soil organic matter or respiration of plant roots and by microorganisms present in the soil. Similar results were observed for the N_2O emissions. Montes et al. (2014) reported that an important difference between mineral fertilizers and wastes, is that wastes contain organic carbon, which, depending on soil conditions, can affect N_2O . Marks et al. (2017) also founded that the microalgal suspension stimulated soil CO_2 production.

The lower volatilization of ammonia for the biofilm application may be related to the type of N that was applied to the soil; due to the slower degradation of the organic N, a slower nutrient release and fewer nutrient losses occurred.

5.3.2 IMPACT EVALUATIONS

5.3.2.1 IMPACTS OF THE FERTILIZERS

A comparison of the impacts of the system phases indicated that the biomass cultivation phase had the greatest impact in the climate change category of the ReCiPe method (Table 6). Pérez-López et al. (2014) found that the major contributors to the impacts on microalgae production were electricity and the use of fertilizers. Despite the substitution of fertilizers by an effluent grown biomass in the present study, the consumption of electricity was much higher in comparison to other studies (Stephenson et al., 2010; Jorquera et al., 2010; Razon and Tan, 2011). The high energy consumption was probably due to the 22.99 operations of 14 days each that were needed to produce 1 kg of nitrogen. Moreover, the 1 cv motor used to run the paddlewheels was probably super estimated.

Table 6. Impacts generated in the different phases of the process.

	Impact Category	Climate Change	Particulate Matter Formation	Terrestrial Acidification	Fresh. Eutroph.	Fresh. Ecotox.
	Unity	kgCO ₂ eq	kgPM10 eq	kgSO ₂ eq	kgP ep	kg1,4-DB eq
Biofertilizer	Production	107	0.156	0.297	0.0208	0.0338
	Concen.	8.11	0.0117	0.0229	0.00151	0.0654
	Soil Application	-3.91x10 ⁻⁶	0.0148	0.113	X	X
	Total	115.11	0.18	0.43	0.022	0.09
Urea	Production	7.38	0.02	0.06	1.01x10 ⁻³	0.09
	Land Transport	0.27	4.40x10 ⁻⁴	0.00	8.68x10 ⁻⁷	3.03x10 ⁻⁵
	Marine Transport	0.46	2.83x10 ⁻³	0.01	4.86x10 ⁻⁵	1.66x10 ⁻³
	Soil Application	-6.07x10 ⁻⁵	0.08	0.46	X	X
	Total	8.11	0.10	0.53	1.06x10 ⁻³	0.09

In addition to the high electricity consumption, the low total biomass production obtained in this study (570 mg/L) should be highlighted. The low productivity, together with the high hydraulic retention time of the HRAP (14 days) caused an increase in the inputs of the production stage. It is believed that the low growth of microalgae may be linked to the development of other microorganisms such as bacteria, fungi and protozoa. The use of the effluent resulted in competition, which hindered the growth of the microalgae.

The harvesting stage also had a large impact due to the use of NaOH as a coagulant. The coagulant NaOH was chosen because traditional coagulants such as ferric chloride and aluminum hydroxide could result in the bioaccumulation of metals in the millet. However, the quantity of the NaOH that was required to harvest the biomass was considerably larger than the amount for a traditional coagulant.

The stage of soil application had the lowest impact and zero impact occurred in half of the categories. In the climate change category, there was a negative impact, indicating that the soil application of microalgae is advantageous with regard to the effect on GHGs. According to the IPCC, methane is 25 times more polluting than CO₂, which is mainly due to the absorption/oxidation of the methane in the soil.

Table 5 also shows the impacts related to the use of 2.17 kg of urea. This value was used since N represents 46% of the total mass of urea. In most cases, the production stage had

a higher impact for urea due to high-energy system inputs (Júnior, 2011).

The transportation stages had little impact because the unit used in the software was tkm (ton-kilometer). Since only 1 kg of N was used in this study, this value is insignificant, even with high mileage. The values for the generation of gases were also low, similarly to the biofertilizer.

Arashiro et al. (2018) when comparing HRAP system for wastewater treatment where microalgal biomass was reused for nutrients recovery through biofertilizer production reported 0.2 kg CO₂eq, 0.015 kg SO₂eq, 0.0025 kgPM₁₀eq and 0.001 kPeq for the categories Climate Change, Terrestrial Acidification, Freshwater Eutrophication and Particulate Matter Formation, respectively. When comparing those impacts with a conventional technology for wastewater treatment, authors highlighted higher impacts in Terrestrial Acidification, Particulate Matter Formation, Human Toxicity and Terrestrial Ecotoxicity. NH₃ emissions to air derived from NH₄⁺ volatilization in HRAPs and heavy metals content in the biofertilizer were the main causes.

5.3.2.2 COMPARISON BETWEEN BIOFERTILIZER AND CONVENTIONAL FERTILIZER IMPACTS

Fig. 2 shows a comparison of the impacts (relative percentage) of using biofertilizer and urea. Results showed that the conventional fertilizer had much less impact than the algae biofertilizer except for terrestrial acidification and freshwater ecotoxicity. A comparison between the subcategories showed that the gases generated in the soil application stage were higher for the microalgae biofilm as mentioned earlier. In addition, the production stage was negatively highlighted, increasing the differences between the types of fertilizers mainly in the categories climate change and freshwater eutrophication.

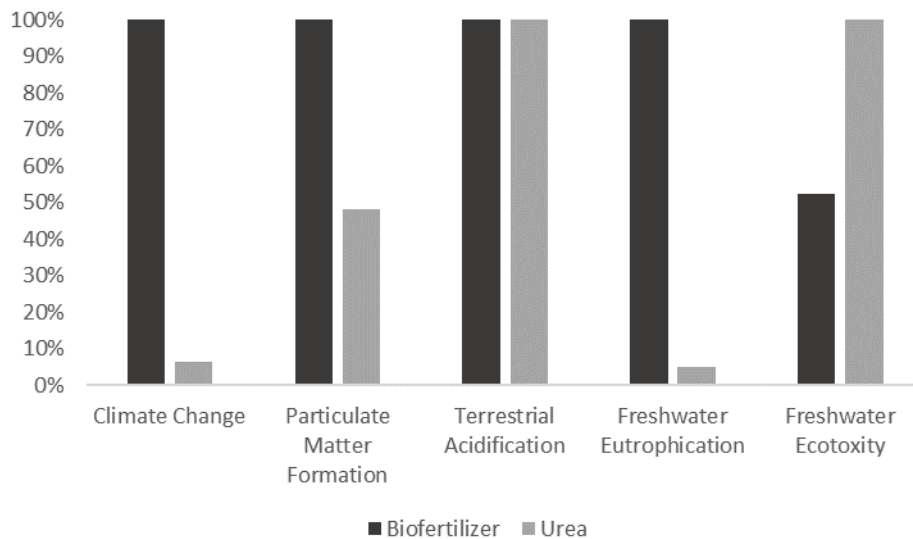


Figure 2. Percentage comparison of impacts between urea and algal biofertilizer.

The results for the types of fertilizers were similar only in the terrestrial acidification category, which was due to the high values of volatilized ammonia in the soil for the urea. According to Goedkoop et al. (2009), ammonia has a high impact in this category.

In the context of using effluent as a fertilizer, Corbala-Robles et al. (2018) through a LCA using the Recipe Midpoint methodology compared the application of pig manure effluent in the soil with and without a previous treatment. The authors concluded that the untreated effluent had the best environmental performance when compared to the treated effluent in 13 of 18 tested categories. However, authors highlighted that the comparison between the treatment scenario and the no-treatment scenario should be done with careful, as impacts at midpoint could be combined into one final score using normalisation and weighing factors depending on the questions raised.

5.3.3 POTENTIAL WAYS OF REDUCING IMPACTS

5.3.3.1 SOURCE OF ENERGY

The results and the reductions/increases in comparison with hydroelectric power plants source are shown in Table 7.

Table 7. LCI using photovoltaic energy.

Impact Category	Unity	Total	Reduction (%)
Climate Change	kgCO ₂ eq	31	73.07
Particulate Matter Formation	kgPM10 eq	0.080	56.27
Terrestrial Acidification	kgSO ₂ eq	0.301	30.47
Freshwater Eutrophication	kgP ep	0.0272	-21.92
Freshwater Ecotoxicity	kg1,4-DB eq	4.99	-4930.24

Table 7 shows that the use of photovoltaic energy generates a large reduction in the impact in most categories, mainly in the climate change category, where the largest impacts occurred.

In the categories eutrophication of water bodies and ecotoxicity of water bodies, the change in the energy source resulted in a negative impact mainly due to the presence of several carcinogenic heavy metals in the photovoltaic panels. Carcinogenicity is one of the main parameters used in the calculation of the impacts and, therefore, has considerable weight (Goedkoop et al., 2009).

5.3.3.2 EFFLUENT CHOICE

The N balance based on the Kjeldahl method is shown in Fig. 3. It is known that there are losses in all stages of the process but they were minimized in this study to obtain an efficiency greater than 66% in the recovery of all N introduced into the system. An eventual automation of some of these processes may result in even smaller losses in future studies, thus increasing the efficiency of the N recovery.



Figure 3. Nitrogen mass balance.

The losses in the production stage occurred mainly because the pH control was performed manually, which caused some failures. The loss related to the harvesting step was less than 5% and was associated with biomass lost in the clarified effluent.

It is important to note that, even if there were no losses in the system, the mass of N introduced into the system was small and several operations were required to obtain 1 kg of N. This result indicates that the meat processing industry effluent may not be the most suitable type for nutrient recovery by biomass soil application as a biofertilizer considering the entire process of batch production in the HRAPs. We suggested that a continuous operation should be performed, resulting in low energy input and increasing yields over time, moreover a nitrogen-rich effluent should be tested for biomass cultivation. Table 8 presents impacts simulation if a nitrogen-rich effluent was used.

Table 8. LCI using a nitrogen-rich effluent.

	Climate Change	Particulate Matter Formation	Terrestrial Acidification	Freshwater Eutrophication	Freshwater Ecotoxicity
Unity	kgCO ₂ eq	kgPM ₁₀ eq	kSO ₂ eq	kgP eq	kg 1,4-DB eq
Production	46.59	0.07	0.121	0.01	0.014
Reduction (%)	56	55	59	54	57
Concentration	8.61	0.0182	0.0595	0.000183	0.00657
Reduction (%)	50	50	50	50	50

Simulating twice the concentration of nitrogen in the effluent, the number of operations was halved. The avoided products – the avoided nutrient discharge in the water courses – remained the same, however, was obtained more than 50% of impact reduction in all the categories when considering the biomass production phase. Results indicated that the net gain was positive, probably due to energy lower requirement with lower operation period. For the biomass concentration phase 50% less impact was observed in all categories.

Prior to choosing the route of recovery/valorization of the biomass (biofuels, biofertilizers, among others), the effluent must be evaluated with regard to the best use of the biomass considering its characteristics. In this study, effluents with higher concentrations of N would be more suitable. However, it is important to note that a pre-treatment should be considered depending on the characteristics of the effluent and special attention should be given to the ammonia toxicity to microalgae growth.

3.3.3 ENVIRONMENTAL COMPENSATION

Fig. 4 shows the results for the case of not accounting for the production stage. In this scenario, the biofertilizer was preferred over urea in terms of environmental impacts because only an insignificant amount of electric energy was needed to harvest the biomass. The climate change was the only impact category that the biofertilizer had a higher impact, compared with

the urea.

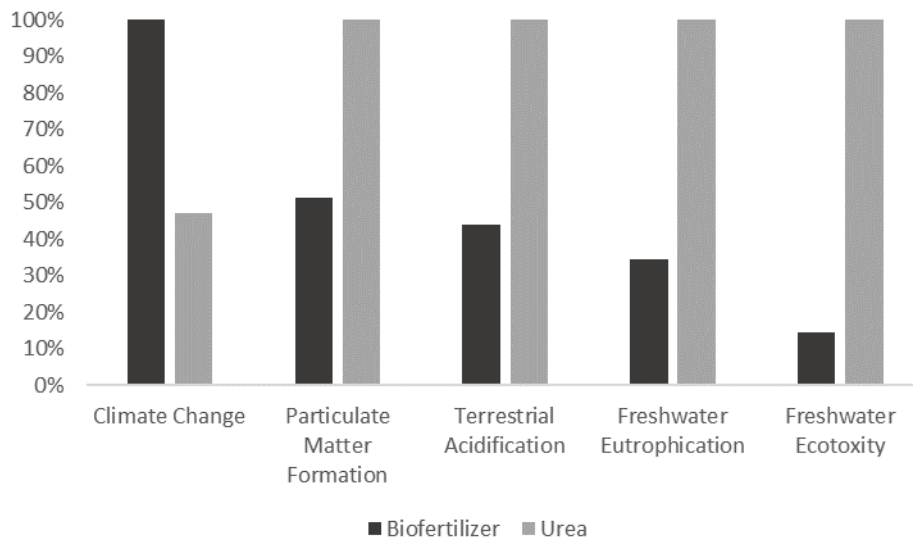


Figure 4. Percentage comparison between fertilizers excluding the biomass production stage.

5.3.4 POTENTIAL LIMITATIONS OF THE STUDY

This study compared the impacts of using a biofertilizer derived from microalgae and a conventional fertilizer (urea). Although we considered impacts inherent to fertilizers, it should be emphasized that urea is a fertilizer that is established in the market while the biofertilizer represents a relatively new technology. The scale in production step and gaps in the whole process, such as the algal biomass harvesting, resulted in greater impacts for the biofertilizer. It is important to note that many studies have shown that different types of fertilizers (urea, ammonium nitrate, ammonium sulfate, triple superphosphate, etc.) have different efficiencies and are recommended accordingly in specific situations (Júnior, 2011). For the biofertilizer used in this study, the variability is related to the composition of the biomass, which is influenced by the type of effluent, the timing in production, and other factors. Addressing these challenges will require further study.

In LCA studies, some impacts should be considered to act locally (eg, terrestrial acidification) and others globally (eg, climate change). This will affect the choice between the proposed scenarios. However, the objective of comparing scenarios is to provide a solid scientific basis for the environmental consequences of choosing either option. LCA is not sufficiently developed to present a clear conclusion of these choices (Schaubroeck et al., 2015). Schaubroeck and Rugani (2017) suggested that additional research is needed to improve LCA studies and also to integrate the social and economic impacts of the proposed

studies.

However, even if all the previous considerations were applied, at an environmental point of view, some impacts such as the climate change, have no well defined frontiers. Therefore, the discussion of local charges is made difficult.

Recently, an application of LCA tool in regions or territories was introduced (Loiseau et al., 2014) and may be of interest. While in the traditional approach, impacts mainly linked to a pre-established functional unit are calculated, the territorial approach associates the study with the area. It delivers as a result beyond the environmental impacts, functions of land use value, such as meeting the needs of a population or wealth creation. Nevertheless, the new methodology makes some LCA steps more complex, especially choosing the boundaries of the system and obtaining data that reflect regional impacts.

The evaluation of the use of microalgae as biofertilizers presents a focus on the environmental sustainability. However, other aspects can be accounted for (eg. socioeconomic), and different methods of calculating the impacts can be used, such as the Recipe Midpoint used in the present study. As for socioeconomic impacts, for example, it can be argued that, thanks to the treatment of effluents, there can be incentives and even increase the profitability of rural producers who use such practices.

5.4 CONCLUSIONS

The main contribution of this study is to evaluate the potential of microalgae biofertilizer and to use LCA tool to identify the key issues, which can serve to guide the development of new technologies and to make them more competitive when compared to traditional fertilizers.

An important result is that the more diffusion of new less environmentally impacting technologies than the use of environmental compensation can be considered to minimize the impact generated by the production of biofertilizers from algae, thus increasing their competitiveness with traditional fertilizers. The choice of the effluent based on the value of the biomass is another factor of great importance, especially considering that the biomass characteristics will vary depending on the environment in which it was cultivated. In addition, the type of harvesting system had a large impact and studies of continuous cultivation are required to determine approaches to minimize these impacts. Further research is needed to optimize the algae production chain and to determine the possibility of obtaining higher value products. At the same time, cost analyses must be performed because aside from environmental impacts, economic factors must be taken into account to choose the best

alternative.

5.5. ACKNOWLEDGEMENTS

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6. CONCLUSÕES GERAIS

A principal contribuição deste estudo foi avaliar o potencial do biofertilizante de microalgas cultivadas em efluentes e ter informações para subsidiar seu desenvolvimento, tornando-o mais competitivo ao longo do tempo. Com base nisso, foi possível concluir que:

- Os impactos ambientais do fertilizante tradicional são menores quando comparados ao biofertilizante de microalgas;
- A etapa de cultivo é a mais impactante no ciclo de vida, embora que obrigatório do ponto de vista do tratamento do efluente;
- Difusão de novas tecnologias, como por exemplo utilização de energia fotovoltaica faz com que o impacto ambiental seja reduzido;
- A utilização da compensação ambiental é uma ferramenta que aumenta a competitividade entre o fertilizante e o biofertilizante;
- A escolha do efluente com base no valor da biomassa é outro fator de grande importância, especialmente considerando que as características da biomassa irão variar dependendo do ambiente em que foi cultivada.

7. SUGESTÕES PARA PESQUISAS FUTURAS

As principais modificações e melhorias propostas para mitigação do impacto ambiental relativo ao biofertilizante foram aplicadas na etapa de produção. O mesmo não foi realizado para a etapa de colheita, na qual experimentos devem ser propostos com o intuito de diversificar as técnicas de separação e colheita, buscando sempre o menor impacto.

A otimização da cadeia de produção e obtenção de produtos de maior valor (energético e econômico) devem ser abordados, principalmente no que tange ao conceito de biorrefinaria, onde ao invés de resíduos, são gerados subprodutos diversos.

Embora o foco seja sobre ACV, análises econômicas devem ser realizadas pois são ferramentas importantes quando se pensa em aplicar tal solução no mercado. Idealmente, as duas análises devem ser levadas em conta para posterior implantação da solução desejada.