Desalination and Water Treatment
www.deswater.com

doi: 10.5004/dwt.2020.25397

185 (2020) 51–61
May

Sustainability metrics on microalgae-based wastewater treatment system

Aline Meireles dos Santos, Mariany Costa Deprá, Alberto Meireles dos Santos, Alexandre José Cichoski, Leila Queiroz Zepka, Eduardo Jacob-Lopes*

Food Science and Technology Department, Federal University of Santa Maria, UFSM, Roraima Avenue 1000, 97105-900, Santa Maria, RS, Brazil, Tel. +555532208822; emails: jacoblopes@pq.cnpq.br (E. Jacob-Lopes), alinemeirelessm@hotmail.com (Aline Meireles dos Santos), marianydepra@gmail.com (M.C. Deprá), albertomsrs@gmail.com (Alberto Meireles dos Santos), ajcichoski@gmail.com (A.J. Cichoski), lqz@pq.cnpq.br (L.Q. Zepka)

Received 10 July 2019; Accepted 22 December 2019

ABSTRACT

This paper presents a life cycle assessment of a simulated large-scale process of agroindustrial wastewater treatment through microalgal heterotrophic bioreactors. The study focuses on establishing sustainability metrics for recovering energy and nutrients from wastewater to produce bulk oil and lipid extracted algae (LEA) in an integrated process. The experimental data, obtained from a bench-scale facility, were used to estimate the life cycle impacts of a wastewater treatment plant with a capacity of 16,000 m^3/d, with a production of 4.32 ton/d of biomass, fractioned in 0.84 ton/d of bulk oil and 3.45 ton/d of LEA. The sustainability metrics of the integrated process indicate a net energy ratio of 0.41, reduction of 98% of the water footprint, global warming potential of 47×10^6 $kgCO_{2eq}$/y, eutrophication potential of 5×10^4 kg eq PO_4/y, acidification potential of 7×10^4 kg SO_2-eq/y and ozone depletion potential of 3.33 kg CFC-11-eq/y. The assessment of the life cycle demonstrated that this technological route presents itself as a new sustainable approach for wastewater treatment plants and their implementation and dissemination can help to support a change towards resource recovery and a sustainable circular economy.

Keywords: Microalgae/cyanobacteria; Nutrient cycling; Biodiesel; Animal feed; Process integration; Impact categories

1. Introduction

Sustainability is one of the main concerns in many industrial areas, which is particularly true when concerning indispensable commodities such as food, feed, clean water, and energy. With the increasing populations as well as industrial activities, countries face global water stress, both in the aspect of water scarcity and deteriorated quality given the large electricity costs and greenhouse gas emissions associated with water delivery and wastewater treatment [1].

Historically, wastewater treatment plants have been designed and operated to minimize the environmental effects focusing only on treatment efficiency and final effluent quality. Economic and technical aspects are still today the major concerns of environmental projects. Though will economic criteria continue to lead the way for industrial development, this is not well enough in the near future. Sustainability metrics will be inserted in the projects and will be a determinant criterion in decision-making [2]. Thus, the full quantification of adverse environmental effects in addition to the establishment of recovery indices of energy and nutrients will be fundamental to consolidate sustainable technological routes [3,4].

Microalgae-based systems play a central role in this process by incorporating and redistributing dissolved organic matter and inorganic nutrients in the environment. These microorganisms show wide potential for use as

---

* Corresponding author.

biocatalysts in environmental biotechnology processes due to their toughness and simple nutritional requirements. Also, microalgae can manufacture a wide range of commercially attractive biobased chemicals [5]. However, sustainability metrics of microalgae-based processes need to be studied before the large-scale implementation of these strategies [6].

Given these aspects, the life cycle assessment (LCA) of processes and products is the most consistent tool to the transition of the theory to the practice in the development of technologies to sustainable development, once that assists evaluation of inputs and outputs of the system, demonstrating the environmental performances throughout the entire life cycle of the process [7–9].

Therefore, this article is a sequence of proof-of-principle experimental and theoretical studies investigating the nutrient cycling of poultry and swine slaughterhouse wastewater, through a microalgal heterotrophic bioreactor [10–12]. The results previously published showed that the microalgal heterotrophic bioreactor is capable of simultaneously converting organic matter, nitrogen, and phosphorus of the wastewater into biomass with great potential to be applied as biofuel and animal feed, being able to support the oscillations of organic matter typical of agroindustrial processes. Besides that, it presents a competitive cost. Building on these promising results, an LCA was performed to establish sustainability metrics on microalgae-based wastewater treatment systems. The study focuses on establishing energy resources, water footprint, global warming potential (GWP), eutrophication potential, acidification potential, and ozone depletion potential (ODP) for recovering energy and nutrients from wastewater to produce bulk oil and lipid extracted algae (LEA) in an integrated process.

2. Material and methods

2.1. Microorganism and culture conditions

Axenic cultures of *Phormidium autumnale* were used in the experiments. Stock cultures were propagated and maintained in solidified agar-agar (20 g/L) containing synthetic BG11 medium [13]. The incubation requirements used were 25°C, a photon flux density of 15 μmol/m^2/s, and a photoperiod of 12:12 h (light:dark).

2.2. Wastewater

The wastewater was collected from the discharge point of an equalization tank over one year in an industry located in Santa Catarina, Brazil (27°14′02″S, 52°01′40″W), whose activity covers the slaughter and processing of poultry and swine. Standard Methods for the Examination of Water and Wastewater [14] were utilized to measure pH, chemical oxygen demand (COD), total nitrogen (N–TKN), total phosphorus (P–PO_4^{-3}), total solids (TS), suspended solids (SS), volatile solids (VS), and fixed solids (FS). The average composition of the wastewater has the following composition (mg/L): pH of 5.90 ± 0.05, COD of 4,100.00 ± 874.00, N–TKN of 128.50 ± 12.10, P–PO_4^{-3} of 2.84 ± 0.02, TS of 3.80 ± 2.70, SS of 1.90 ± 0.81, VS of 2.90 ± 1.42, and FS of 0.90 ± 0.31, carbon/nitrogen (C/N) ratio of 31.90 ± 0.30 and nitrogen/phosphorous (N/P) ratio of 45.24 ± 0.54. The C/N and N/P were calculated through COD, N–TKN, and P–PO_4^{-3}.

2.3. Bioreactor configuration

Measurements were made in a bubble column bioreactor. The system was built of borosilicate glass and it had an external diameter of 12.5 cm and a height of 16 cm, resulting in a height/diameter (h/d) ratio equal to 1.28 and a nominal working volume of 2.0 L. The dispersion system of the reactor consisted of a 2.5 cm diameter air diffuser located inside the bioreactor. The airflow was monitored by a flow meter (KI-Key Instruments®, Trevose, PA, USA).

The experiments were performed in bioreactors, operating in a batch system, fed to 2.0 L of the poultry and swine slaughterhouse wastewater. The operational conditions were an initial cell concentration of 100 mg/L, constant aeration of 1.0 volume of air per volume of culture per minute (VVM), pH adjusted to 7.6, temperature 25°C, and the absence of light [10–12].

2.4. Life cycle assessment

2.4.1. Goal and scope definition

The LCA was performed according to the ISO standards [15]. The experimental data were obtained from laboratory experiments, where the elementary flows of the process were selected. Subsequently, the data were extrapolated for an industrial scale where system boundaries comprised a wastewater treatment plant with a capacity of 16,000 m^3/d, with a production of 4.32 ton/d of biomass, fractioned in 0.84 ton/d of bulk oil and 3.45 ton/d of LEA, for a period one year. The theoretical scale-up of the process was performed, as described by Santos et al. [10].

Since there is an absence of data on large-scale microalgae wastewater treatment, laboratory remarks combined with published data of known industrial procedures have been employed and extrapolated [10,16–18,]. Besides, promising technologies that can be commercialized shortly were employed as the basis for the experimental calculation to determine the sustainability metrics of the proposed process. Fig. 1 shows the scope of the proposed process.

2.4.1.1. Step 1: agroindustrial wastewater treatment

The unit operations proposed in step 1 was based on a patent application developed by Jacob-Lopes et al. [19]. A primary treatment composed by a fine screen, Parshall flume, rotary sieve, and equalization tank was used. The core of this step was the biological treatment in a bench-scale heterotrophic microalgal bioreactor, as described in item 2.3. The performance parameters are described in Table 1 [10]. After the biological treatment, the microalgal sludge was processed by a decanter and a belt filter to obtain biomass.

Furthermore, the bench-scale facility had all the necessary equipment to convert the pollutants of the agroindustrial wastewater into dried microalgal biomass and the fractionation of biomass into bioproducts.

2.4.1.2. Step 2: biodiesel production

In a bench-scale, the modified Bligh and Dyer method [20] was applied to extract the lipid content of the biomass. Based on laboratory experiments, an industrial process was

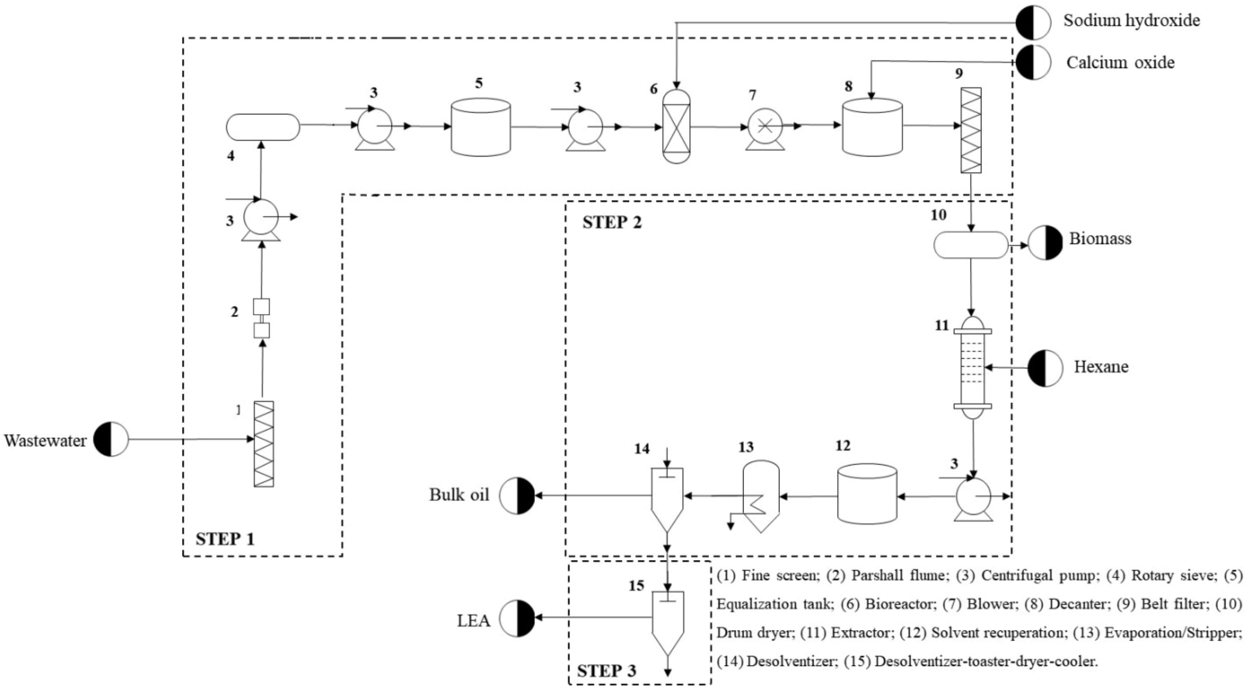

Fig. 1. Process flow diagram of the agroindustrial wastewater treatment in an integrated process. Adapted from Santos et al. [10].

Table 1
Performance parameters of the heterotrophic microalgal bioreactor. Adapted from Santos et al. [10]

Parameter	Value
$RE_{(COD)}$ (%)	97.6 ± 1.64
$RE_{(N-TKN)}$ (%)	85.5 ± 2.37
$RE_{(P-PO4^{-3})}$ (%)	92.4 ± 0.22
P_X (kg/m^3/d)	0.27 ± 0.01
HDT (d)	1.67 ± 0.00

$RE_{(COD)}$: COD removal efficiency; $RE_{(N-TKN)}$: N–TKN removal efficiency; $RE_{(P-PO_4^{-3})}$: P–PO_4^{-3} removal efficiency; P_X: average biomass productivity; HDT: hydraulic retention time.

proposed for oil extraction of the dried biomass (Fig. 1). The hexane extraction was defined as the method for large-scale applications [21].

2.4.1.3. Step 3: animal feed production

Drying in a vacuum oven at 60°C simulating an industrial process was used for hexane separation and drying of the LEA on a laboratory scale. On industrial scale, it was proposed the desolventizer-toaster-dryer-cooler (DTDC) [22].

2.4.2. Life cycle inventory (LCI)

A summary of elementary flows of the process is presented in Table 2. These data were analytically evaluated to quantify and compile all the input and output flows for each step within the process scope. The impacts of production were based on the Ecoinvent database, and are shown in Supplementary Material (Table S1).

2.4.3. Life cycle impact assessment (LCIA)

2.4.3.1. Net energy ratio

The characterization of the net energy ratio (NER) was performed as described by Jorqueira et al. [23] in Eq. (1).

$$\text{NER} = \frac{\sum E_{\text{out}}}{\sum E_{\text{in}}} \tag{1}$$

where NER is net energy ratio, E_{out} is the energy produced, and E_{in} is the total input energy required, expressed in Joule/Joule (dimensionless).

2.4.3.2. Water footprint

The water footprint is a metric that counts the direct and indirect use of water as well as pollution. The characterization of the total water footprint was performed as described by Hoekstra and Mekonnen [24] in Eq. (2).

$$\text{WF} = \sum \text{WF}_{\text{blue}} + \text{WF}_{\text{green}} + \text{WF}_{\text{grey}} \tag{2}$$

where WF is the total water footprint, WF_{blue} is the blue water footprint, WF_{green} is the green water footprint, and WF_{gray} is the gray water footprint, expressed in m^3/y.

The blue water footprint is an indicator of consumptive use of so-called water fresh or groundwater, evaporated or incorporated into the products, and it was quantified according to Eq. (3):

$$\text{WF}_{\text{blue}} = \text{BW}_e + \text{BW}_i + L_{\text{rf}} \tag{3}$$

Table 2
Elementary flows for the different steps of process scope

Steps	In/Out	Utilities/materials	Base case
Wastewater treatment	Input	Wastewater, m³/h	666.66
		Fine screen, kWh	4.80
		Parshall flume, kWh	32.05
		Centrifugal pump, kWh	617.27
		Rotary sieve, kWh	0.72
		Centrifugal pump, kWh	617.27
		Equalization tank, kWh	1,799.80
		Centrifugal pump, kWh	617.27
		Sodium hydroxide, kWh	4,000.00
		Heterotrophic microalgal bioreactor, kWh	6.45
		Blower, kWh	9.43
		Decanter, kWh	599.90
		Calcium oxide, kWh	266.66
		Belt filter, kWh	183.30
	Output	Biomass, kg/h	180.00
Biodiesel production	Input	Drum dryer, kWh	1,249.98
		Extractor, kWh	12.77
		Hexane, kg/h	1,310.00
		Centrifugal, kWh	18.80
		Solvent recuperation, kWh	22.29
		Evaporation/Stripper, kWh	4.17
		Desolventizer, kWh	7.05
	Output	Bulk oil, kg/h	36.00
Animal feed production	Input	Residual biomass, kg/h	144.00
		DTDC, kWh	19.82
	Output	LEA, kg/h	144.00

where WF_{blue} is blue water footprint, BW_e is blue water evaporation, BW_i is blue water incorporation, and L_{rf} is the lost return flow, expressed in m³/y.

The green water footprint is the consumed of water, evaporated or absorbed during the production of biomass, and it was quantified according to Eq. (4):

$$WF_{green} = GW_e + GW_i \tag{4}$$

where WF_{Green} is green water footprint, GW_e is green water evaporation, GW_i is green water incorporation, expressed in m³/y.

The grey water footprint is the volume of water needed to dilute the pollution generated in the production process to the levels established by current environmental standards [25,26] and it was quantified according to Eq. (5):

$$WF_{grey} = \frac{L}{c_{max} - c_{nat}} \tag{5}$$

where WF_{grey} is the grey water footprint, L is the pollutant load, in mass/time, c_{max} is the ambient water quality standard for that pollutant and c_{nat} is the natural concentration in the receiving water body. c_{max} and c_{nat} are based on the values established by current legislation, expressed in m³/y.

Nitrogen and phosphorus were the pollutants chosen to calculate the gray water footprint of the process, considering their concentrations in the raw wastewater, quality standards according to Brazilian and European laws (both 75% removal efficiency) and concentrations in the waterbody in their natural state, according to Brazilian law (N = 3.7 mg/L and P = 0.1 mg/L) [25,26].

2.4.3.3. Global warming potential

The GWP is the sum of direct and indirect greenhouse gas emissions. Direct emissions are greenhouse gases generated during the wastewater biological treatment process, and they were calculated by the stoichiometry of the biochemical oxidation of organic compounds. The indirect emissions are those attributable to carbon footprints associated with the operation of the proposed process due to the inputs of power and chemicals. The characterization of the GWP was performed as described by Laratte et al. [27] in Eq. (6).

$$GWP = \sum i GWP_i \times m_i \tag{6}$$

where GWP is the global warming potential, i is the time horizon of 100 years, GWP_i is the equivalence factor for a substance *i* and, m_i is the emission of substance *i*, expressed in kgCO_2eq/y.

2.4.3.4. Eutrophication potential (EP)

The characterization of the eutrophication potential was performed as described by Hauschild et al. [28] in Eq. (7), and their specific characterization factors are shown in Supplementary Material (Table S2).

$$\mathrm{EP} = \sum i\mathrm{EP}_i \times m_i \tag{7}$$

where EP is the eutrophication potential, EP_i is the equivalence factor for a substance *i* and, m_i is the emission of substance *i*, expressed in kg eq PO_4/y.

2.4.3.5. Acidification potential (AP)

The characterization of the acidification potential was performed as described by Hauschild et al. [28] in Eq. (8).

$$\mathrm{AP} = \sum i\mathrm{AP}_i \times m_i \tag{8}$$

where AP is the acidification potential, AP_i is the equivalence factor for substance *i*, and m_i is the emission of substance *i*, expressed in kg SO_2-eq.

2.4.3.6. Ozone depletion potential

The characterization of the ODP was performed as described by Hauschild and Wenzel [29] in Eq. (9).

$$\mathrm{ODP} = \sum \mathrm{ODP}_i \times m_i \tag{9}$$

where ODP is the ozone depletion potential, ODP_i is the equivalency factor for a substance *i* and, m_i is the mass of the emission substance *i*, expressed in kg CFC-11-eq.

2.4.4. Normalization

The impact categories were normalized as described by International Reference Life Cycle Data System (ILCD) [30,31] in Eq. (10). During this step, each of the environmental impact potentials was divided by reference value, corresponding to the global normalization factors from European Commission (Table S3).

$$N_k = \frac{S_k}{R_k} \tag{10}$$

where *k* is the impact category, N_k is the normalized results, S_k is the characterized impact of the impact category *k* of the system under study, and R_k is the characterized impact of the impact category *k* of the reference system.

3. Results and discussion

3.1. Resource use

3.1.1. Net energy ratio

The NER is a basic relation that helps with understanding the energy effectiveness of a particular system. The correlation between the renewable energy produced and the fossil energy required in the production process implies ideally in an NER higher than 1.0 [32]. In this sense, the energy balances of the process were examined, and the results are presented in Fig. 2.

According to the results, inputs of 127 × 10^6 MJ/y, 37 × 10^6 MJ/y and 0.5 × 10^6 MJ/y of fossil energy is required to generate 32 × 10^6 MJ/y, 11 × 10^6 MJ/y, and 23 × 10^6 MJ/y of renewable energy in the steps of wastewater treatment, biodiesel production and animal feed production resulting in NERs of 0.25, 0.31 and 42.00, respectively. In addition, the agroindustrial wastewater treatment in microalgal heterotrophic bioreactor and the production of bulk oil and LEA in an integrated process require an input of 164 × 10^6 MJ/y of fossil energy to generate 66 × 10^6 MJ/y of renewable energy, resulting in a NER of 0.41. Microalgae-based processes potential for bioproducts production such as biofuels has been proven in some studies. However, it is associated with an NER of less than one. Many authors highlight the indirect effects of fossil energy consumption in reducing the NER [33–35].

Under the aspect of fossil energy consumption, there are a few stages that make up a large proportion of primary energy consumption. Unit operations for agroindustrial wastewater treatment (step 1) consume approximately 77% of the fossil energy of the proposed process, being the primary wastewater treatment responsible for most of this consumption (3,689.16 kWh). The unit operations for biomass harvesting are responsible for 21% of fossil energy consumption in step 1 (792.62 kWh). The dilute nature of microalgal cultures (0.27 kg/m^3/d) creates a huge energetic cost during harvest, which results in a low net energy rate for microalgae-based products [36]. There is no superior method of dewatering microalgae, and this is a bottleneck of the process [37]. Unit operations for biodiesel production (step 2) consume approximately 23% of the fossil energy in the process, being the biomass drying responsible for most of this consumption (1,249.95 kWh). The drying of microalgae sludge for large-scale hexane extraction may not be economically viable in terms of absorbed energy for biofuels. However, for being a developed technology and with well-established economics for practice on a large scale for oilseeds, this unit operation is steadily used in the economic assessments of microalgal biofuel production [9,38,39]. The animal feed production (step 3) from the residual biomass consumed only 0.33% (19.79 kWh) of the total fossil energy process. This step presents a potential solution towards reducing the energy demand of this process once the renewable energy produced is greater than the required fossil energy.

In this context, wastewater treatment in microalgal heterotrophic bioreactor represents an interesting alternative for conventional activated sludge treatment plants once that they require significantly lower energy consumption (0.00968 kW/m^3) than conventional activated sludge systems (0.0132 kW/m^3) [40]. In addition, the conventional system shows a high biomass yield coefficient ($Y_{X/S}$), usually in the order of 0.3 kg $_{\mathrm{sludge}}$/kg$_{\mathrm{BOD}}$ resulting in massive production of sludge with low use potential that will require adequate treatment and disposal, further burdening the process [41].

3.1.2. Water footprint

The use of nutrient-rich wastewater in the microalgae-based processes can contribute to energy recovery and significantly reduce the water footprint [42]. In this sense, Fig. 3 presents a flow diagram of the water footprint of the integrated process.

From the data analysis, it is possible to verify that in the integrated process, the water footprint is composed only of the blue and green fractions. The blue water footprint considered in this study refers to the volume of water consumed in the lipid extraction stage of the biomass (1.11 m³/y). In addition, the green water footprint is directly related to the water volume incorporated into the biomass (1,056,000 m³/y), representing about 20% of the wastewater. The rate evaporated during the process represents about 528,000 m³/y. Thus, the total water footprint of the integrated process results in values in the order of 1,584,001.11 m³/y.

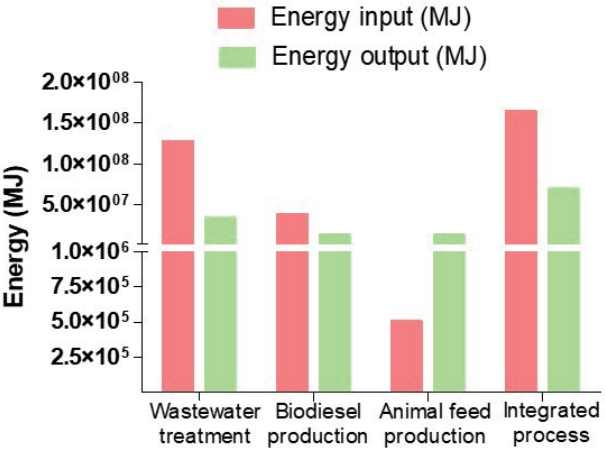

Fig. 2. Energy balances for the different steps and integrated process over one year.

The gray water footprint is generally not accounted for in the water footprint calculation of industrial processes due to the unavailability and/or poor quality of data on a global scale [43,44]. However, considering the gray footprint before the wastewater treatment, as a theoretical reference for assimilation of the body of water, this would result in values in the order of 1,07,713,204.76 m³/y.

Given these volumes, the process proposed in this study presents itself as a potential technology, since the total water footprint (1,584,001.11 m³/y) represents close to 2% when compared to the gray water footprint, which corresponds to 100%. Therefore, this technology could represent the prevention of more than 98% of the total water demand, if the gray footprint was considered usually as an alternative for nutrient assimilation.

Moreover, from the economic point of view, considering that the average water price in the world show values of approximately 1.50 USD/m³ [45], the total water footprint of the integrated treatment wastewater process costs around USD 276001.66.

3.2. Climate change

3.2.1. Global warming potential

GWP is related to emissions of greenhouse gases, including CO_2, CH_4, and N_2O. This impact category being among the aspect that has become key-factors concerning the overall performance of the wastewater treatment plants, once

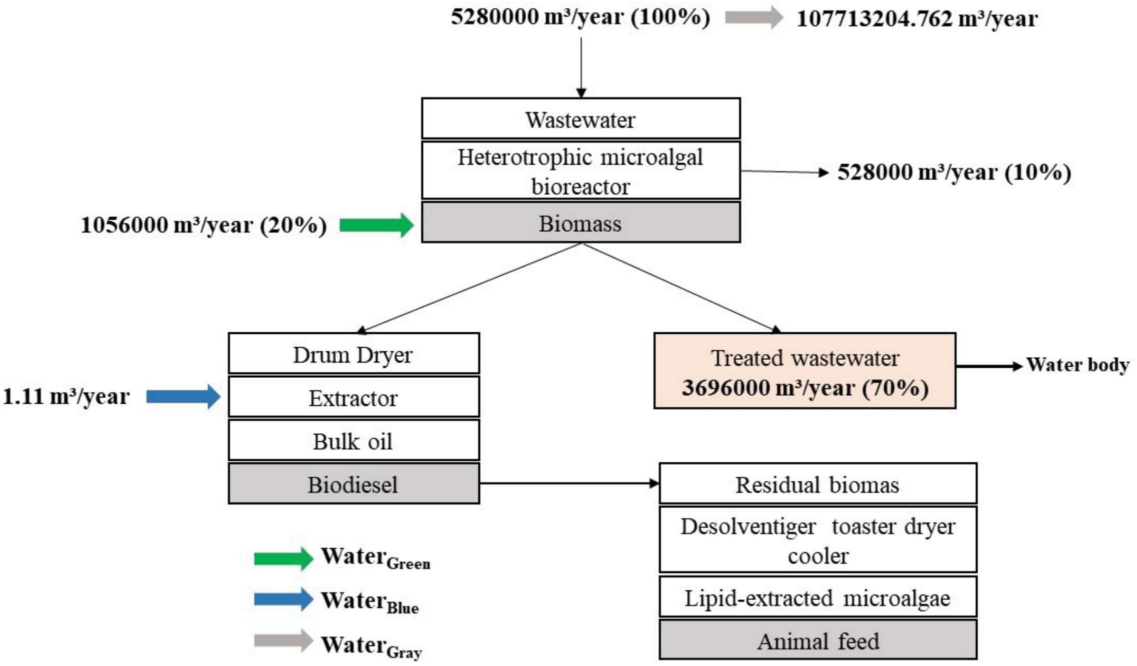

Fig. 3. Flow diagram of the water footprint of the process.

sustainability metrics are under consideration to ensure the viability of this type of process [2]. In this sense, Table 3 presents the results of GWP for the agroindustrial wastewater treatment in the integrated process.

From the data analysis, it is possible to verify that the sum of the direct and indirect emissions results in GWPs of 34 × 10^6, 12 × 10^6 and 0.08 × 10^6 kgCO_{2eq}/y in the steps of wastewater treatment, biodiesel production, and animal feed production, respectively. The agroindustrial wastewater treatment in microalgal heterotrophic bioreactor and production of bulk oil and LEA in the integrated process resulted in a GWP of 47 × 10^6 kgCO_{2eq}/y, being the biochemical oxidation, use of chemical reagents, and demand for fossil energy responsible for 1%, 46% and 53% of total emissions, respectively. Understanding the dynamics of greenhouse gas emissions in microalgae-based processes is of the utmost importance to improve strategic investments in the areas of climate change. Although these estimates are uncertain, due to the lack of large-scale applications, they sustain a case for improving mechanistic understanding of pathways and main inducements for this type of process [46].

Given the aspect of emissions generated by the employed of chemical reagents, the use of 31,680 ton/y of sodium hydroxide to control the culture pH and 2,112 ton/y of calcium oxide to separate the extracellular water from the microalgae suspension are responsible for 14 × 10^6 kgCO_{2eq}/y of the indirect emissions of greenhouse gases related to the use of chemical reagents in step 1. The use of 10,375 ton/y of hexane for biodiesel extraction is responsible for 7 × 10^6 kgCO_{2eq}/y of the indirect emissions of greenhouse gases related to the use of chemical reagents in step 2. Greenhouse gas emissions released due to the use of chemical reagents are inevitable in wastewater treatment plants. However, they can be reduced by enhancing the energy efficiency of the wastewater treatment plant and reducing the use of chemicals [2].

Additionally, considering the initial concentrations of nitrogen (678,480 kg/y) and phosphorus (14,982 kg/y) in the composition of poultry and swine slaughterhouse wastewater (Table 1) and their removal efficiencies of approximately 86% and 93% (Table 2), respectively, it is estimated a biorecovery of 583,492.8 $kg_{nitrogen}$/y and 13,933.26 $kg_{phosphorus}$/y. If these nutrients were reusing for fertilizer production could be avoid close to 3 million kgCO_{2eq} [47].

In terms of emissions generated by the demand for fossil energy, coal, and oil-fired plants represent 29% and 31% of the world total primary energy supply, being responsible for approximately 46 %and 34% of global CO_2 emissions, respectively [48]. Due to heavy carbon content per unit of energy released, approximately 0.34 and 0.26 kgCO_2 are emitted to generate 1 kWh of energy from coal and oil, respectively [49].

The GWP of the integrated process (47 × 10^6 kgCO_{2eq}/y) is 1.4 times greater than the value emitted just in the wastewater treatment step (34 × 10^6 kgCO_{2eq}/y). However, with the integration of the steps, there will be the possibility of valorization of agroindustrial wastewater from the recovery of energy and nutrients through the production of biodiesel and animal feed. The nutrient cycling from microalgae integrated to the wastewater treatment plant has long been proposed as a means to reduce the environmental impact of commodities output such as biofuels [50,51].

3.3. Ecosystem quality

3.3.1. Eutrophication potential, acidification potential, and ODP

Rupture of natural nitrogen and phosphorus cycles causes an excess of nutrients of anthropic origin on aquatic ecosystems resulting in loss of ecosystem services and species extinctions. Therefore, it is important to quantify the eutrophication potential of production processes and minimize their impacts on ecosystem quality [52]. Thus, the result obtained for the eutrophication potential of the integrated process is presented in Table 4.

The microalgal heterotrophic bioreactor is attractive for organic matter and nutrients recovery due to its great removal efficiency of the COD (97.6%), total nitrogen (85.5%) and total phosphorus (92.4%) present in agroindustrial wastewater. From these removal efficiencies, it is possible to verify a reduction of 80 × 10^4 kg eq PO_4/y for 5 × 10^4 kg eq PO_4/y on the eutrophication potential of the integrated process (Table S4). Nutrient recovery reduces the consumption of bioresources, and it saves costs related to N and P removal, thus minimizing the environmental impact of wastewater treatment [53]. Additionally, it also saves the costs of fertilizing once that the prices of ammonium nitrate and superphosphates are approximately USD/ton 550 and 300, respectively [54]. Studies conducted by Zhang and Kendall [55] report the requirements of ammonium nitrate 0.15 kg/kg of biomass and superphosphate in the order of 0.10 kg/kg of biomass. Therefore, in an estimated production of 1 ton of microalgal biomass, the required costs involving nutrient demand would be around 82.5 USD for nitrates and USD 30.00 for phosphates.

Additionally, the acidification potential and ODP are impact categories related to emissions of air pollutants that result in environmental problems such as global warming,

Table 3
Global warming potential for the individual steps and integrated process

GWP (kgCO_{2eq}/y)		Step 1 (kgCO_{2eq}/y)	Step 2 (kgCO_{2eq}/y)	Step 3 (kgCO_{2eq}/y)	Integrated process (kgCO_{2eq}/y)
Direct	Biochemical oxidation	0.05 × 10^6	NA	NA	0.05 × 10^6
Indirect	Fossil energy	19 × 10^6	5 × 10^6	0.08 × 10^6	25 × 10^6
	Chemicals	14 × 10^6	7 × 10^6	NA	22 × 10^6
Total		34 × 10^6	12 × 10^6	0.08 × 10^6	47 × 10^6

NA: not applicable.

Table 4
Impact categories that damage ecosystem quality

Impact category	Integrated process
Eutrophication potential	5×10^4 kg eq PO_4/y
Acidification potential	7×10^4 kg SO_2-eq/y
Ozone depletion potential	3.33 kg CFC-11-eq/y

acid rain, and ozone layer depletion [56,57]. Thus, sustainability metrics that predict the environmental impacts associated with emissions of air pollutants is indispensable to minimize negative environmental effects. In this sense, the results obtained for the acidification potential and ODP (Table 4) indicate values of 7×10^4 kg SO_2-eq/y and 3.33 kg CFC-11-eq/y, respectively. The results are related to the input of fossil energy resources to operate the integrated process, and agroindustrial wastewater treatment and biodiesel production steps are the main responsible for these impacts. Fossil fuels consumption to operate wastewater treatment plant is the largest contributing parameter for pollutant emissions, once only 20%–30% of the chemical energy of the fuel burned is typically transformed into useful work or heating, the rest is dissipated into the atmosphere as polluting gases [58].

The elementary input flows for sodium hydroxide, calcium oxide, and hexane were disregarded in the characterization of the acidification potential and ODP since these chemical reagents contribute lightly to these impact categories.

The comparison of the sustainability metrics obtained in this study with other technologies for wastewater treatment is limited by the fundamentals of the methodology used. The LCAs are usually performed using similar methodologies; however, there are variabilities in tool application, in terms of choice of allocation, system boundary and procedures followed to data collection. In addition, the comparative LCA applied to wastewater treatment systems is severely hampered by the choice of characterization factors, which vary in function of the database and reports considered in the life cycle impact assessment phase [59]. In this sense, considering these aspects, it's possible to reference other studies as the work of Pasqualino et al. [60] that shows sustainability metrics of 0.5214 $kgCO_{2eq}/m^3$, 3.36×10^{-4} kg eq PO_4/m^3, 4.61×10^{-3} kg SO_2-eq/m^3 and 4.2×10^{-8} CFC-11-eq/m^3 for a system of aerobic digestion applied to urban wastewater treatment. Additionally, the sustainability metrics showed by Buonocore et al. [61], Garfí et al. [62], and Li et al. [63] can be used as the comparison referential.

3.4. Normalization

Normalization is a step in LCIA that helps decision-makers interpret LCA results and determine where to prioritize efforts aimed at reducing a products environmental impact. This step serves to calculate the relevance of the results of the impact category indicators related to reference information [64]. Thus, for a better understanding between the scores and to compare the magnitude of the emissions of the integrated process, the gate-to-gate impact categories were normalized. Fig. 4 illustrates the results after normalizing the impact categories.

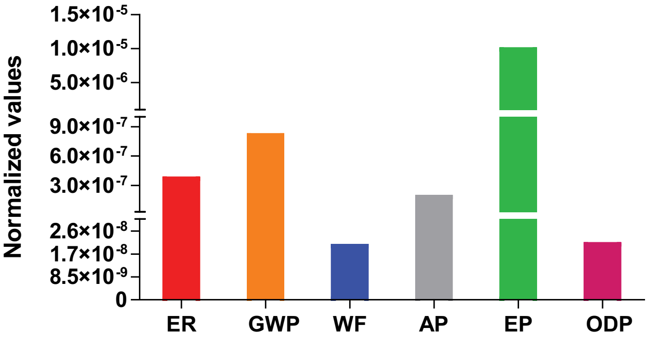

Fig. 4. Normalized values of the impact categories for the integrated process. ER: Energy resource; GWP: Global warming potential; WF: Water footprint; AP: Acidification potential; EP: Eutrophication potential; ODP: Ozone depletion potential.

As shown in Fig. 4, once normalized, the integrated process presented EP as the most significant impact (9.88×10^{-7}), followed by GWP (8.11×10^{-7}), AP (1.82×10^{-7}), ER (3.68×10^{-7}), ODP (2.06×10^{-8}) and WF (2×10^{-8}). Most of the emissions of the integrated process are related to the indirect effects of fossil energy consumption. According to Gao et al. [65], the total of pollutants emitted per unit of energy produced by fossil fuels is typically two orders of magnitude bigger than those emitted by non-fossil energy.

In this sense, the transition from fossil energy matrix to non-fossil energy has become an urgent issue for all wastewater facilities, once that these energies can supply two-thirds of the total global energy demand, and they contribute to the greenhouse gas emissions reduction. Furthermore, the development of renewable energy technologies would greatly promote the realization of a circular economy [66]. Policies and incentives of governments improve the prospects for clean energy. The total generation from renewable resources increases by 2.8% annually [48]. Although it is reasonable to expect that renewables will come to provide a growing share of the global energy supply, it should be noted that replacing fossil energy with non-fossil fuels remains challenging, especially for future global policy [67].

4. Conclusion

The LCA results demonstrate that the process integration mediated by microalgae is an appropriate and innovative approach to comply with green engineering requirements, through nutrient cycling. However, the biggest challenge will be implementing these heterotrophic systems in process chains operating across wastewater management already existing.

In this sense, new approaches to process engineering must be oriented towards promoting the exploitation and development of a commercially viable and integrated microalgae-based wastewater treatment in the future. Process integration, process intensification, and the implementation of the biorefinery concept have been considered as the main process engineering strategies that, in the medium term, will consolidate.

References

[1] A. Pyka, Transformation of Economic Systems: The Bioeconomy Case, In: Knowledge-Driven Developments in the Bioeconomy, Springer, Cham, 2017, pp. 3–16.

[2] J.L. Campos, D. Valenzuela-Heredia, A. Pedrouso, A. Val del Rio, M. Belmonte, A. Mosquera-Corral, Greenhouse gases emissions from wastewater treatment plants: minimization, treatment, and prevention, J. Chem., 2016 (2016) 12p.

[3] UNFCCC, Synthesis Report on the Aggregate Effect of the Intended Nationally Determined Contributions, Conference of the Parties, United Nations, 2015.

[4] H. Awad, M.G. Alalm, H.K. El-Etriby, Environmental and cost life cycle assessment of different alternatives for improvement of wastewater treatment plants in developing countries, Sci. Total Environ., 660 (2019) 57–68.

[5] A.M. Santos, A.M. Santos, I.A. Severo, M.I. Queiroz, L.Q. Zepka, E. Jacob-Lopes, Nutrient Cycling in Wastewater Treatment Plants by Microalgae-Based Processes, S.N. Barton, Ed., Industrial Waste: Management, Assessment, and Environmental Issues, Nova Science Publishers, Inc., New York, 2016, pp. 41–63.

[6] L.M. González-González, D.F. Correa, S. Ryan, P.D. Jensen, S. Pratt, P.M. Schenk, Integrated biodiesel and biogas production from microalgae: towards a sustainable closed-loop through nutrient recycling, Renewable Sustainable Energy Rev., 82 (2018) 1137–1148.

[7] A. Padilla-Rivera, L.P. Güereca, A proposal metric for sustainability evaluations of wastewater treatment systems (SEWATS), Ecol. Indic., 103 (2019) 22–33.

[8] B. Agarski, I. Budak, D. Vukelic, J. Hodolic, Fuzzy multi-criteria-based impact category weighting in life cycle assessment, J. Cleaner Prod., 112 (2016) 3256–3266.

[9] M.C. Deprá, E. Jacob-Lopes, L.Q. Zepka, Life Cycle Assessment of Biofuels from Microalgae, E. Jacob-Lopes, L.Q. Zepka, M.I. Queiroz, Eds., Energy from Microalgae, 1st ed., Springer, Switzerland, 2018, pp. 141–155.

[10] A.M. Santos, G.R. Roso, C.R. Menezes, M.I. Queiroz, L.Q. Zepka, E. Jacob-Lopes, The bioeconomy of microalgal heterotrophic bioreactors applied to agroindustrial wastewater treatment, Desal. Wat. Treat, 64 (2017) 12–20.

[11] A.M. Santos, A.M. Santos, R.B. Sartori, L.Z. Queiroz, J.S. Barin, E. Jacob-Lopes, Nutrient cycling in meat processing industry by microalgae-based processes, Desal. Wat. Treat., 100 (2017b) 91–99.

[12] A.M. Santos, A.M. Santos, R.B. Sartori, L.Z. Queiroz, E. Jacob-Lopes, Influence of poultry and swine blood shocks on the performance of microalgal heterotrophic bioreactor, Desal. Wat. Treat., 114 (2018) 128–134.

[13] R. Rippka, J. Deruelles, J.B. Waterbury, M. Herdman, R.Y. Stanier, Generic assignments, strain histories and properties of pure cultures of cyanobacteria, Microbiology, 111 (1979) 1–61.

[14] APHA, Standard Methods for the Examination of Water and Wastewater, American Public Health Association, Washington, D.C., 2005.

[15] ISO 14040, International Standard, Environmental Management–Life Cycle Assessment – Principles and Framework, International Organisation for Standardization, Geneva, Switzerland, 2006.

[16] L. Xu, D.W.F. Brilman, J.A.M. Withag, G. Brem, S. Kersten, Assessment of a dry and a wet route for the production of biofuels from microalgae: energy balance analysis, Bioresour. Technol., 8 (2011) 5113–5122.

[17] N. Pragya, K. Panved, Life cycle assessment of green diesel production from microalgae, Renewable Energy, 86 (2016) 623–632.

[18] V.O. Adesanya, E. Cadena, S.A. Scott, A.G. Smit, Life cycle assessment on microalgal biodiesel production using a hybrid cultivation system, Bioresour. Technol., 163 (2014) 343–355.

[19] E. Jacob-Lopes, A.M. Santos, D.B. Rodrigues, M.C.Y. Lui, C. Souza, D. Prudente, L.Q. Zepka, Bioprocess of Conversion of Hybrid Wastewaters, Heterotrophic Bioreactor, Bioproducts, and Their Uses, Brazilian Patent Application BR 10 2013 020490-0 A2, 2013.

[20] E.G. Bligh, W.J. Dyer, A rapid method of total lipid extraction and purification, Can. J. Biochem. Phys., 37 (1959) 911–917.

[21] Y. Peralta-Ruiz, A.D. González-Delgado, V. Kafarov, Evaluation of alternatives for microalgae oil extraction based on exergy analysis, Appl. Energy, 101 (2013) 226–236.

[22] G.R. Roso, A.M. Santos, M.I. Queiroz, J.S. Barin, L.Q. Zepka, E. Jacob-Lopes, The econometrics of production of bulk oil and lipid extracted algae in an agroindustrial biorefinery, Curr. Biotechnol., 4 (2015) 547–553.

[23] O. Jorqueira, A. Kiperstok, E.A. Sales, M. Embirucu, M.L. Ghirardi, Comparative energy life-cycle analyses of microalgal biomass production in open ponds and photobioreactors, Bioresour. Technol., 101 (2010) 1406–1413.

[24] A.Y. Hoekstra, M.M. Mekonnen, The water footprint of humanity, Proc. Nat. Acad. Sci., 109 (2012) 3232–3237.

[25] CONAMA, Resolução n° 357/2005, Dispõe sobre a classificação dos corpos de água e diretrizes ambientais para o seu enquadramento, bem como estabelece as condições e padrões de lançamento de efluentes, in: Conselho Nacional do Meio Ambiente, Ed., Brasília, BR, 2005.

[26] Council Directive, Council Directive 91/271/EEC of 21 May 1991 Concerning Urban Waste-water Treatment, Official Journal L 135, European Union, 1991, pp. 40–52.

[27] B. Laratte, B. Guillaume, J. Kim, B. Birregah, Modeling cumulative effects in life cycle assessment: the case of fertilizer in wheat production contributing to the global warming potential, Sci. Total Environ., 481 (2014) 588–595.

[28] M.Z. Hauschild, H. Wenzel, M. Hauschild, Environmental Assessment of Products: Scientific Background, Vol. 2, Springer Science & Business Media, London, 1997.

[29] M.Z. Hauschild, H. Wenzel, Environmental Assessment of Products, Vol. 2, Kluwer Academic Publishers, Chapman & Hall, Hingham, United Kingdom, 1998.

[30] European Commission - Joint Research Centre - Institute for Environment and Sustainability: International Reference Life Cycle Data System (ILCD) Handbook – General guide for Life Cycle Assessment - Detailed guidance, First edition March 2010, EUR 24708 EN, Publications Office of the European Union, Luxembourg, 2010.

[31] S. Sala, E. Crenna, M. Secchi, R. Pant, Global Normalisation Factors for the Environmental Footprint and Life Cycle Assessment, EUR (28984), Publications Office of the European Union, Publications Office of the European Union, Luxembourg, 2017, ISBN 978-92-79-77213-9, doi:10.2760/88930, JRC109878.

[32] M.R. Tredici, N. Bassi, M. Prussi, N. Biondi, L. Rodolfi, G.C. Zittelli, G. Sampietro, Energy balance of algal biomass production in a 1-ha "Green Wall Panel" plant: how to produce algal biomass in a closed reactor achieving a high net energy ratio, Appl. Energy, 154 (2015) 1103–1111.

[33] L. Lardon, A. Helias, B. Sialve, J.P. Steyer, O. Bernard, Life-cycle assessment of biodiesel production from microalgae, Environ. Sci. Technol., 43 (2009) 6475–6481.

[34] M.O.P. Fortier, G.W. Roberts, S.M. Stagg-Williams, B.S. Sturm, Life cycle assessment of bio-jet fuel from hydrothermal liquefaction of microalgae, Appl. Energy, 122 (2014) 73–82.

[35] M. Morales, P. Collet, L. Lardon, A. Hélias, J.-P. Steyer, O. Bernard, Chapter 20 - Life-cycle assessment of microalgal-based biofuel, In: Biofuels from Algae Sustainable Platform for Fuels, Chemicals and Remediation Biomass, Biofuels, Biochemicals, 2019, pp. 507–550.

[36] C. Zamalloa, E. Vulsteke, J. Albrecht, W. Verstraete, The techno-economic potential of renewable energy through the anaerobic digestion of microalgae, Bioresour. Technol., 102 (2011) 1149–1158.

[37] A.M. Santos, A.M. Santos, L.Q. Zepka, E. Jacob-Lopes, Downstream Recovery of Microalgal Bioproducts with Highlights on Biorefineries, In: Biorefineries: Concepts, Advancements, and Research, Nova Science Publishers, Inc., New York, 2017, pp. 1–38.

[38] P.D. Patil, V.G. Gude, A. Mannarswamy, S. Deng, P. Cooke, S. Munson-McGee, I. Rhodes, P. Lammers, N. Nirmalakhandan, Optimization of direct conversion of wet algae to biodiesel under supercritical methanol conditions, Bioresour. Technol., 102 (2011) 118–122.

[39] A. Sathish, R.C. Sims, Biodiesel from mixed culture algae via a wet lipid extraction procedure, Bioresour. Technol., 118 (2012) 643–647.

[40] A.M. Santos, M.C. Deprá, A.M. Santos, L.Q. Zepka, E. Jacob-Lopes, Aeration energy requirements in microalgal heterotrophic bioreactors applied to agroindustrial wastewater treatment, Curr. Biotechnol., 5 (2015) 249–254.

[41] M. Von Sperling, R.F. Gonçalves, Lodo de esgotos: características e produção. In: C.V. Andreoli, M. Von Sperling, F. Fernandes, Lodo de esgotos: tratamento e disposição final, Belo Horizonte: Departamento de Engenharia Sanitária e Ambiental, UFMG, Curitiba: SANEPAR, 6 (2001) 17–67.

[42] B.G. Subhadra, M. Edwards, Coproduct market analysis and water footprint of simulated commercial algal biorefineries, Appl. Energy, 88 (2011) 3515–3523.

[43] A.Y. Hoekstra, The global dimension of water governance: why the river basin approach is no longer sufficient and why cooperative action at global level is needed, Water, 3 (2011) 21–46.

[44] P.R. Van Oel, A.Y. Hoekstra, Towards quantification of the water footprint of paper: a first estimate of its consumptive component, Water Resour. Manage., 26 (2012) 733–749.

[45] J. Clere, True Cost of Water: Monetization of Water Risks, Shared Value Creation, and Local Acceptability of Extractive Projects, J. Field Actions: Field Actions Science Reports, (Special Issue 14), (2016), http://journals.openedition.org/factsreports/4069.

[46] M. Plouviez, A. Shilton, M.A. Packer, B. Guieysse, Nitrous oxide emissions from microalgae: potential pathways and significance, J. Appl. Phycol., 31 (2019) 1–8.

[47] EC, European Commission Environment, 2019. https://ec.europa.eu/ [accessed June 2019].

[48] IEA International Energy Agency, CO_2 Emissions from Fuel Combustion Highlights 2018, International Energy Agency, 2018. http://www.indiaenvironmentportal.org.in/files/file/CO2_Emissions_from_Fuel_Combustion_2018_Highlights.pdf [accessed June 2019].

[49] US Energy Information Administration, Ed., Annual Energy Outlook 2019 with Projections to 2050, Government Printing Office, 2019.

[50] C.Q. Arita, J.J. Sheehan, T.H. Bradley, Life cycle net energy and greenhouse gas emissions of photosynthetic cyanobacterial biorefineries: challenges for industrial production of biofuels, Algal Res., 26 (2017) 445–452.

[51] C.Q. Arita, C. Peebles, T.H. Bradley, Scalability of combining microalgae-based biofuels with wastewater facilities: a review, Algal Res., 9 (2015) 160–169.

[52] F. Gao, Y.-Y. Peng, C. Li, W. Cui, Z.-H. Yang, G.-M. Zeng, Coupled nutrient removal from secondary effluent and algal biomass production in membrane photobioreactor (MPBR): effect of HRT and long-term operation, Chem. Eng. J., 335 (2018) 169–175.

[53] T. Yan, Y. Ye, H. Ma, Y. Zhang, W. Guo, B. Du, Q. Wei, D. Wei, H. H. Ngo, A critical review on membrane hybrid system for nutrient recovery from wastewater, Chem. Eng. J., 348 (2018) 143–156.

[54] USDA, U.S. Department of Agriculture, 2019. https://www.usda.gov/ [accessed June 2019].

[55] Y. Zhang, A. Kendall, Effects of system design and co-product treatment strategies on the life cycle performance of biofuels from microalgae, J. Cleaner Prod., 230 (2019) 536–546.

[56] J.O. Reuss, D.W. Johnson, Acid Deposition and the Acidification of Soils and Waters, Vol. 59, Springer Science & Business Media, New York, 2012.

[57] M. Z. Hauschild, M. Goedkoop, J. Guinée, R. Heijungs, M. Huijbregts, O. Jolliet, S. Sala, Identifying best existing practice for characterization modeling in life cycle impact assessment, Int. J. Life Cycle Assess., 18 (2013) 683–697.

[58] S. Bilgen, Structure and environmental impact of global energy consumption, Renewable Sustainable Energy Rev., 38 (2014) 890–902.

[59] S. Fazio, V. Castellani, S. Sala, E.M. Schau, M. Secchi, L. Zampori, Supporting Information to the Characterisation Factors of Recommended EF Life Cycle Impact Assessment Methods, EUR 28888 EN, European Commission, Ispra, 2018.

[60] J.C. Pasqualino, M. Meneses, F. Castells, Life cycle assessment of urban wastewater reclamation and reuse alternatives, J. Ind. Ecol., 15 (2011) 49–63.

[61] E. Buonocore, S. Mellino, G. De Angelis, G. Liu, S. Ulgiati, Life cycle assessment indicators of urban wastewater and sewage sludge treatment, Ecol. Indic., 94 (2018) 13–23.

[62] M. Garfí, L. Flores, I. Ferrer, Life cycle assessment of wastewater treatment systems for small communities: activated sludge, constructed wetlands and high rate algal ponds, J. Cleaner Prod., 161 (2017) 211–219.

[63] Y. Li, X. Luo, X. Huang, D. Wang, W. Zhang, Life cycle assessment of a municipal wastewater treatment plant: a case study in Suzhou, China, J. Cleaner Prod., 57 (2013) 221–227.

[64] ISO 14044 International Standard, In: Environmental Management – Life Cycle Assessment – Requirements and Guidelines, International Organisation for Standardisation, Geneva, Switzerland, 2006.

[65] C.K. Gao, H.M. Na, K.H. Song, N. Dyer, F. Tian, Q.J. Xu, Y.H. Xing, Environmental impact analysis of power generation from biomass and wind farms in different locations, Renewable Sustainable Energy Rev., 102 (2019) 307–317.

[66] D. Gielen, F. Boshell, D. Saygin, M.D. Bazilian, N. Wagner, R. Gorini, The role of renewable energy in the global energy transformation, Energy Strategy Rev., 24 (2019) 38–50.

[67] J. Chen, Y. Wu, C. Xu, M. Song, X. Liu, Global non-fossil fuel consumption: driving factors, disparities, and trends, Manage. Decis., 57 (2019) 791–810.

Supplementary information

Table S1
Factors of characterization of environmental impacts of energy in process

Impact category	Electricity
Global warming potential (kg CO_2 eq)	5.45×10^{-1}
Acidification potential (kg SO_2 eq)	1.64×10^{-3}
Ozone depletion potential (kg CFC-11 eq)	9.39×10^{-8}

Ecoinvent 2.2 database

Table S2
Characterization factors referring the eutrophication category [47]

Nutrient	PO_4 equivalence factor
1 kg Total nitrogen (water)	0.420 kg eq PO_4
1 kg Total phosphorous (water)	3.070 kg eq PO_4
1 kg Chemical oxygen demand (COD)	0.022 kg eq PO_4

Table S3
Global normalization factors for emissions and resource extraction (Sala et al. [31])

Impact categories	Unit	Reference value
Energy resource	MJ	4.50×10^{14}
Global warming potential	kg CO_2 eq	5.79×10^{13}
Water footprint	m^3	7.91×10^{13}
Acidification potential	kg SO_2 eq	3.83×10^{11}
Eutrophication potential	kg PO_4 eq	1.22×10^{12}
Ozone depletion potential	kg CFC11 eq	1.61×10^{8}

Table S4
Eutrophication potential reduction behavior on wastewater treatment in a heterotrophic microalgal bioreactor [25,26]

Parameter	Wastewater	Treated wastewater
COD (kg PO_4 eq)	476,256	11,430
N–TKN (kg PO_4 eq)	284,961	41,319
P–PO_4^{-3} (kg PO_4 eq)	46,034	3,493
EP (kg PO_4 eq)	807,252	56,242