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Abstract: The use of diesel fuel in crop and transportation operations is responsible for one third of the carbon emissions in sugarcane biorefineries. A possible solution is to replace it with biodiesel from lipids, directly produced from sugarcane by highly productive heterotrophic microalgae. In this study a heterotrophic microalgae biodiesel plant, integrated with a typical Brazilian sugarcane bio-refinery, was designed and evaluated. Molasses, steam, and electricity from sugarcane processing were used as inputs for microalgae production. For a non-integrated plant, the production cost of the microalgae biodiesel was estimated at 2.51 and 2.27 $/liter for fed-batch and continuous processes, respectively. Equipment for cultivation and carbon sources was the highest cost affecting the financial feasibility of the proposed design. For the integrated plant, at present ethanol and biodiesel selling prices, the profitability would be lower than a first-generation sugarcane bio-refinery using fossil diesel fuel for its operations. However, the CO_2 emissions would be reduced by up to $50\,000 \times 10^3$ kg per year at a cost of \$83 10^{-3} kg^{-1} CO_2-eq. If carbon credits are considered, the process becomes economically profitable even at present fuel prices. © 2020 The Authors. *Biofuels, Bioproducts, and Biorefining* published by Society of Chemical Industry and John Wiley & Sons, Ltd

Supporting information may be found in the online version of this article.

Key words: biodiesel; bio-refinery; heterotrophic microalgae; techno-economic assessment; sugarcane

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Introduction

The continuous improvement of sugarcane biorefineries has resulted in a large reduction in greenhouse gas (GHG) emissions and fossil fuel consumption.[1] However, crop and harvesting operations still depend heavily on diesel consumption, estimated at around 200–300 L ha^{-1} $year^{-1}$.[1,2] For a sugar mill with capacity for processing 5×10^9 kg of sugarcane per year, around 20 million liters of diesel oil are consumed.[3] Considering the potential emission of 2.63 kg of CO_2eq per liter of diesel,[4] the resulting emissions may reach almost 50 million kilograms of CO_2eq, which is equivalent to one third of all the emissions produced during sugarcane processing.[5] Hence, substitution for a renewable fuel might lead to a significant reduction in GHG emissions and fossil fuel consumption, improving the renewability of sugarcane processing and reducing the vulnerability of production costs to oil price fluctuations.

Although numerous technologies have been proposed to produce drop-in fuels from renewable sources, biodiesel from vegetable oils still dominates the market worldwide,[6] corresponding to 17.9% of global biofuel production in the period of 2010–2012.[7]

Microalgae are regarded as the most productive culture for oil and biomass production, with theoretical yields of up to 40×10^3 kg of oil per hectare per year.[8] On a large scale, however, the productivity still falls short and capital and energy costs associated with cultivation and harvesting are the main cost drivers.[9] Microalgae can be cultivated under autotrophic or heterotrophic conditions. In the former, light, CO_2, and nutrients must be provided for photosynthesis to occur. This process is usually run in either open raceway ponds or closed photobioreactors,[10,11] but mutual shading of cells invariably limits the biomass concentration to about 1–5 g L^{-1}. In heterotrophic cultivation, organic carbon is instead utilized as a structural and energy source and no light is needed to support growth. Thus, heterotrophic organisms can grow in closed bioreactors, reaching biomass concentrations up to 100 g L^{-1} [12–14] and lipid productivities as high as 10–15 g $L^{-1}d^{-1}$ [15] (Table S1 - Supplementary data). Such biomass concentrations are about 100 times higher than those obtained in autotrophic cultivation, which reduces dilution and harvesting costs. Closed bioreactors also allow for steady conditions throughout the year, regardless of environmental conditions.[16]

Despite the higher productivity of heterotrophic cultivation, the need for organic carbon sources adds relevant feedstock costs. On the other hand, cultures such as sugarcane can yield up to 150×10^3 kg ha^{-1} $year^{-1}$ of plant biomass,[17] and are currently only used to produce first- and second-generation bioethanol. If these highly productive crops could be converted to oils by microorganisms, high productivity of oil per area could be obtained.

To reduce feedstock costs, inexpensive substrates have been utilized successfully by heterotrophic microalgae, such as sugar cane molasses,[18] crude glycerol from biodiesel production,[11] and rice straw hydrolysate.[19] In this work, on-site biodiesel production by heterotrophic microalgae is proposed as alternative for total substitution of diesel consumed by a sugarcane bio-refinery (see Fig. 1). The main goal of an integrated bio-refinery approach is to optimize the use of resources and profitability, while reducing waste generation.[20] We present a techno-economic assessment of the microalgae biodiesel plant based on the conversion of molasses into microalgae lipids. The lipids produced are then extracted and converted to biodiesel through transesterification.

Methods

Process simulation software and basic assumptions

Superpro Designer™ 9.0 software was used for process design and simulation. The proposed microalgae plant was designed to provide all the fuel consumed by a first-generation sugarcane bio-refinery in Brazil, with capacity for processing 5×10^9 kg of sugarcane per year. According to recent estimates, the diesel consumption during sugarcane processing is around 4 L $\times 10^{-3}$ kg^{-1} sugarcane,[3] which results in a yearly demand of 20 million liters of diesel for the bio-refinery considered in this study.

The sugarcane mill process was considered as a black box with inputs and outputs based on previous works,[21] summarized in Fig. 1A. A few modern flexible sugarcane refineries can operate in different product ratios (usually from 30:70 to 70:30 sugar:ethanol), but here a 50:50 refinery is considered. The main assumptions for the base case refinery are described in Table 1.

In the proposed integrated plant, heterotrophic microalgae use molasses produced during sugar crystallization as a substrate for growth and lipid accumulation. The lipids are extracted and converted to biodiesel for internal consumption, while the remaining cell debris is dried and sold as protein rich meal for animal feed. Other potentially high-value by-products such as carotenoids and poly-unsaturated fatty acids are not considered but could offer additional revenue potential. The microalgae biodiesel production process (Fig. 1B) was also evaluated as a stand-alone plant for comparison purposes.

For the biomass and lipid production stage, the main assumptions are taken from experimental data obtained with

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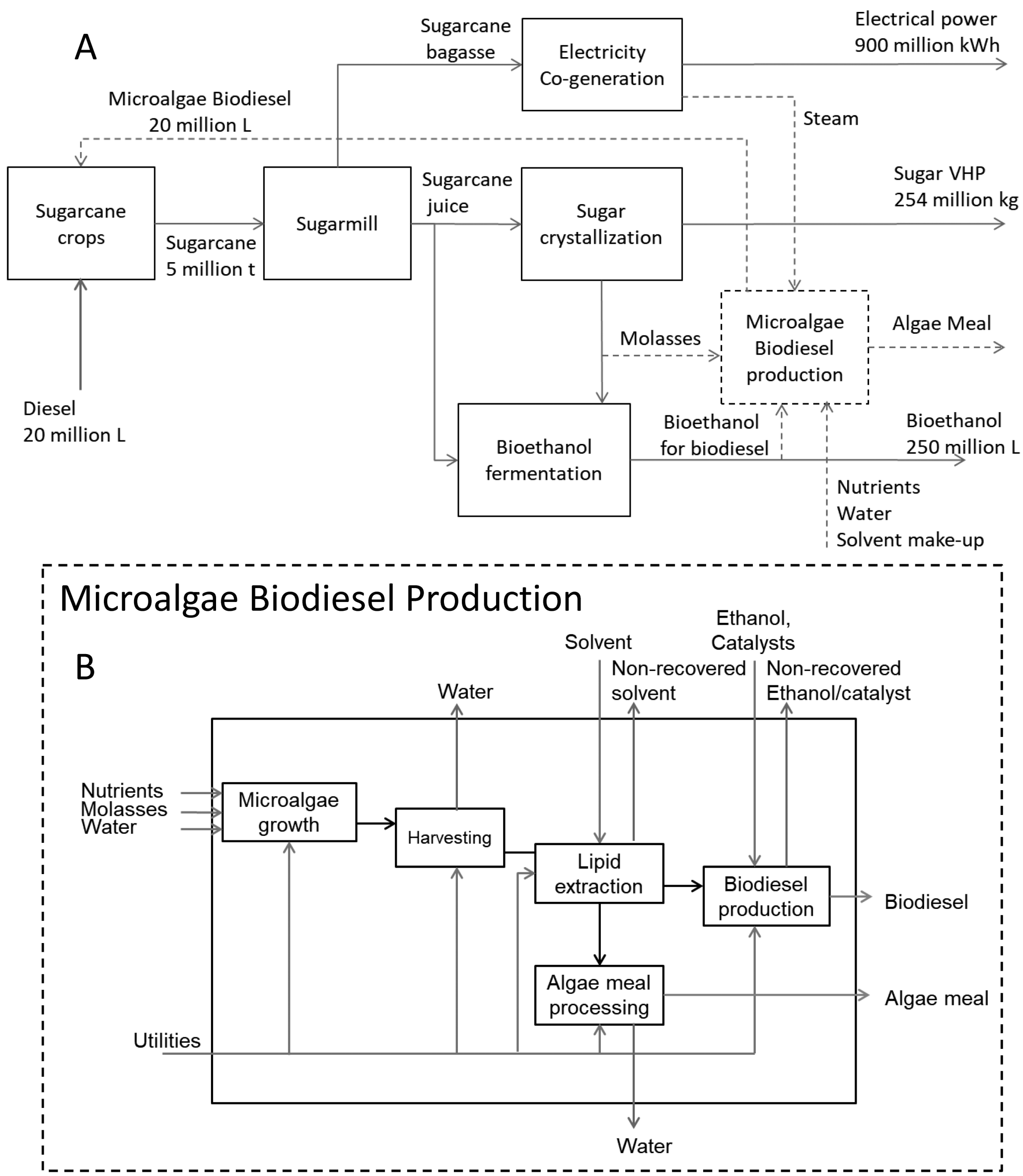

Figure 1. A: Block-flow diagram of proposed integrated sugarcane refinery. B: Block-flow diagram of microalgae production with inputs and outputs.

the microorganism *Auxenochlorella protothecoides*.[14] The following parameters were considered: specific growth rate: 0.04 h^{-1}; yield of biomass from sugars: 0.5 g g^{-1}; yield of lipids from sugars 0.25 g g^{-1}; maximum cell density: 144 g L^{-1}; final lipid content: 50%.

Microalgae biodiesel plant process description

Biomass production

Figure 2 shows the flowsheet as implemented in SuperPro Designer™. Molasses from sugar crystallization, which contains circa 40% m/m of sucrose, is hydrolyzed in a reactor with HCl at 0.25% for 1 h at 85 °C.[23] Afterwards, the mixture is neutralized by addition of lime, forming $CaSO_4$ and other salts, which are then removed in a rotary vacuum filter, while the clarified molasses is sent to sterilization. Other culture medium components are blended in a separate tank and sterilized in a continuous sterilizer before mixing with the molasses stream inside the bioreactors. The carbon-to-nitrogen (C:N) ratio is adjusted during cultivation for biomass growth (C:N 20) or lipid accumulation (C:N 40). In addition to the sugars from molasses, a stream of glycerol, a by-product from oil transesterification, is also recirculated into cultivation as a secondary carbon source.

Table 1. Main assumptions of base sugarcane refinery from literature.

Parameter	Value	Reference
Diesel consumption	4 L/ton of sugarcane	3
Total sugars in sugarcane	150 g/ton sugarcane	22
Ethanol production from sugarcane (50–50 sugar/ethanol ratio)	40.4 kg/ton sugarcane	21
Sugar production from sugarcane (50–50 sugar/ethanol ratio)	50.9 kg/ton sugarcane	21
Electricity from co-generation	182 kwh/ton sugarcane	21
Sugar selling price	$ 0.43/kg	21
Anhydrous ethanol selling price	$ 0.6/L	21
Diesel selling price	$ 1.06/L	22
Electricity selling price	$ 0.058/kWh	21
Algae meal	$ 0.45/kg	3
Sugar production cost	$ 0.25/kg	21
Ethanol production cost	$ 0.45/L	21
Electricity production cost	$ 0.052/kWh	22
Molasses sugar content	55%	21

Airlift bioreactors of 500 m^3 are used for cultivation, operating in fed-batch or continuous mode. The feasibility of operating large capacity bioreactors for heterotrophic microalgae cultivation has been previously validated by companies such as Martek Biosciences (DSM), based in Heeren, The Netherlands.[24] Airlift bioreactors are more easily scaled up to high volumes due to their simple design, and they demand less power input for agitation. The vessels are aerated at a rate of 0.5 v.v.m. with centrifugal compressors.

The fed-batch process takes 210 h with 12 h for the charging, cleaning, and sterilization procedures. The discharge of each bioreactor takes 24 h, during which all downstream operations are performed. To allow maximum capacity utilization, 20 bioreactors operate in 10 staggered racks of two bioreactors each (see Appendix S1 in the supplementary material). Downstream processing of culture broth is run continuously from the bioreactor discharge (two bioreactors a day). For the continuous process, we assumed the lipid productivity of 8.2 $g\,L^{-1}\,d^{-1}$ obtained in a previous work,[14] but running in steady state with a lower biomass concentration of 35 $g\,L^{-1}$ (dry-weight) and 50% (m/m) lipid content, as has been observed in other continuous cultivations.[11,25] This leads to a dilution rate of 0.02 h^{-1}, and a total of 15 continuous bioreactors of 500 m^3, operating with a residence time of 50 h. The feasibility of continuous cultivation of *Chlorella* has been reported in the literature, reaching lipid productivities of up to 9.76 $g\,L^{-1}\,d^{-1}$.[26]

Harvesting and oil extraction

The bioreactors operate at relatively high densities and throughputs, so decanter centrifuges were chosen for biomass harvesting. This operation concentrates the biomass stream up to a maximum solid's concentration of 30% (w/w) to ensure slurry pumpability.[27] The total throughput to the harvesting section was calculated as 35 $m^3\,h^{-1}$. The cell-limiting size for separation was 1 µm, as cell size ranges from 2 to 5 µm.[28] The centrifuge operates in continuous mode, independently of the operating mode of the bioreactors. The concentrated algal slurry is transferred to a high-pressure homogenizer for cell disruption. High-pressure homogenization is an easily scalable technology and is highly effective for processing wet biomass, eliminating the need for a preliminary drying stage.[29] It is assumed that at a pressure drop of 800 bar the resulting slurry contains 95%[30] of disrupted cell material. Lipids are subsequently extracted from the microalgal biomass through conventional liquid–liquid solvent extraction. Differently from the other downstream operations, this stage is performed in batches. The disrupted microalgae slurry is transferred to a stirred tank to which hexane at 60 °C is added and mixed for five hours to solubilize the lipids with a yield of 95%. The mixture containing lipids and hexane is then transferred to a decanter centrifuge for phase separation. The lighter phase, containing hexane and lipids, is sent to a single-stage evaporator for removal of solvent. The evaporated solvent is then recovered in a condenser and recirculated in the extraction system. The solvent-free extracted lipids are transferred to the transesterification reactor for biodiesel synthesis. The aqueous phase, containing water and cell debris, is transferred to the algae meal section (see below).

Biodiesel production

Biodiesel is usually synthesized through the transesterification of fatty acids with methanol in the

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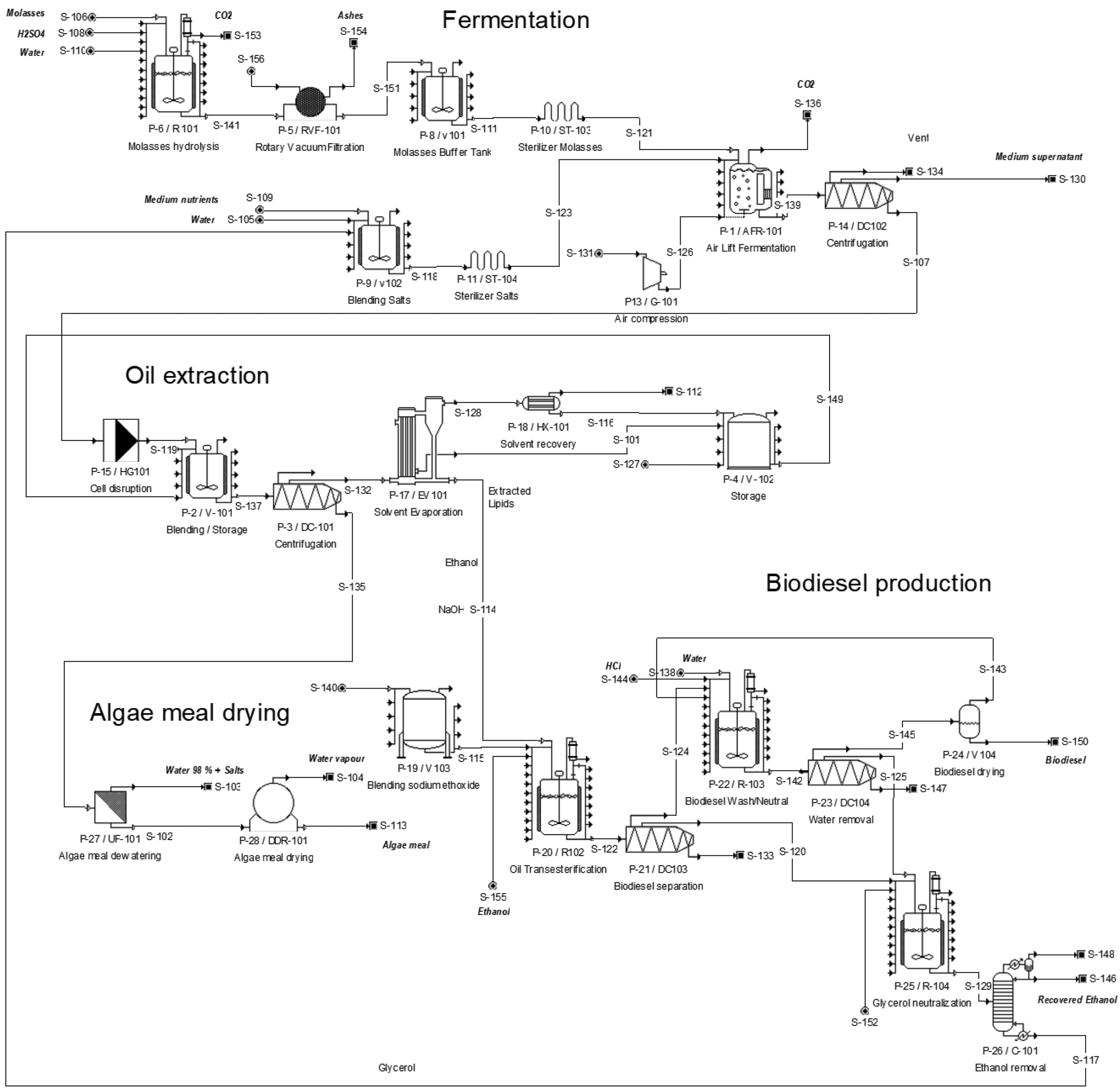

Figure 2. Process flowsheet of the proposed microalgae biodiesel plant.

presence of an acid or alkaline catalyst.[31] However, in our proposed integrated process ethanol is used instead because it is produced on site. Ethanol also offers advantages over methanol such as renewability, less toxicity, and higher cetane numbers of ethyl esters compared to methyl esters,[32] and currently there are no commercial bio-methanol plants. On the other hand, stable emulsions may occur during ethanol-based transesterification reactions, requiring phase separation by centrifugation.[33]

We considered that the extracted oil contains a maximum free fatty acid (FFA) content of 10% (w/w) and, therefore, pre-esterification of the FFA with an acid catalyst is necessary to reduce acidity before the addition of alkaline catalyst to avoid emulsion formation.[34] Hence, biodiesel synthesis is performed in a two-step reaction batch in the same reactor. In the first stage, sulfuric acid (1% of total FFA mass) and ethanol (6:1 molar ratio alcohol:FFA) are mixed with the extracted oil and reacted for 1 h at 60 °C for esterification of

FFA. The resulting mixture contains ethanol, sulfuric acid, esterified fatty acids (10% of total fatty acids), glycerol, and the remaining lipids (circa 90% of fatty acid content).

In the second stage, sodium hydroxide (1% of total lipid mass) is mixed with ethanol (6:1 molar ratio alcohol:lipid) to form sodium ethoxide, before being added to the lipid mixture. The alkaline transesterification reaction is run at 60 °C for 1 h, after which 95% of the total fatty acids are esterified. The reacted mixture is sent to a decanter centrifuge. The biodiesel output stream is sent to a second reactor for neutralization of remaining catalyst by addition of HCl and washing with water at 50 °C to remove remaining glycerol, soap, catalyst, and unreacted fatty acids. The neutralized mixture is centrifuged once more to remove most of the remaining water, glycerol, and ethanol. The oil phase output of the centrifuge is then heated to 220 °C and flash evaporated to remove the last remainders of water and ethanol.[33] After neutralization, ethanol is recovered from glycerol through distillation and reused in the transesterification reaction. Glycerol, free from ethanol, is sent to the biomass production section and used as a secondary carbon substrate.

Algae meal processing

Algae meal has been successfully tested as substitute for corn or soybean meal in animal feed.[35] In the proposed design, the remaining biomass from oil extraction is dried and sold as a protein-rich supplement for animal feed.

The bottom phase of the lipid extractor contains 17% (m/m) de-fatted biomass, 80% (m/m) water and a remaining fraction composed by ash, soluble salts, and 0.8% (m/m) non-extracted lipids. After concentration of the solid matter by ultrafiltration, drum drying is performed to reach a final water content of 5%.

Economic analysis

Direct fixed cost (DFC) estimation

Direct fixed costs include (a) purchase cost of all equipment; (b) bulk items, such as pipes, valves wiring, instruments, and others; (c) construction costs of buildings, roads, and auxiliary facilities; (d) engineering costs; and (e) contingency costs, to deal with uncertainties about the project cost estimates.[36,37] Equipment purchase costs (EPC) were estimated through built-in cost models from the Superpro Designer™ database or estimated from literature sources,[36] and adjusted through the Chemical Engineering Index. Costs and specifications of equipment for biodiesel processing and algae processing were also based on previously published research.[33,37] Auxiliary equipment that is not itemized in the equipment list, such as pumps, valves, and auxiliary tanks, was estimated as 20% of the EPC. A factorial method was used to estimate the DFCs,[36,38] applying the factors listed in Table 2.

Working capital

Working capital was estimated as the amount necessary for 30 days of raw materials, labor utilities and waste treatment costs:

$$30 \text{ days' cost} = 30 \times (\text{annual cost} / 330)$$

Table 2. Capital costs estimation parameters.

Item	Contribution	Typical ranges[35,36]
a. Equipment Installation	0.5	0.3–0.6
b. Piping	0.6	0.2–0.8
c. Instrumentation	0.15	0.2–0.3
d. Electrical	0.1	0.1–0.2
e. Buildings	0.5	0.3–0.5
f. Yard improvement	0.05	0.02–0.05
g. Auxiliary facilities construction	0	0.4
	DC = Direct capital Cost = EPC × (1 + (a + b + c + d + e + f + g))	
h. Design and Engineering	0.1	0.1–0.3
	TPC = Total plant cost = DC × (1 + h)	
i. Contractor's fee	0.05	0.02–0.06
j. Contingency	0.1	0.05–0.15
	DFC = Direct fixed Capital = TPC × (1 + h + i)	

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Operating costs (OPC)

The OPC were estimated by:

OPC = materials costs + utilities costs + waste treatment costs + labor costs + facility related costs

where utilities costs stand for electricity, steam, high-pressure steam, cooling, and chilled water costs. The OPC are described in different sections as annual total costs and cost per liter of biodiesel. The latter is calculated simply by dividing the total costs by the total production of biodiesel, ignoring other sources of revenue.

Materials costs were estimated from the ICIS pricing database,[39] previous work,[3] and the SuperPro Designer™ built-in database. Utilities costs were estimated from published studies on sugarcane biorefineries[3,21,22] and the SuperPro Designer™ built-in database. To estimate the labor costs, the number of operators was estimated for each item of equipment described in the process design, at a cost of \$11.50 per work hour.[40] Waste streams (98% water) treatment cost was estimated at $\$1.5 \times 10^{-3}\ kg^{-1}$.[36] Solid wastes (up to 50% water content) were disposed in landfills.

The facility related costs are defined as:

facility related costs = depreciation costs + maintenance costs + insurance (1% DFC) + taxes (2% of DFC) + miscellaneous factory expenses (1% DFC)

Depreciation costs were calculated based on the total EPC to be depreciated over a period of 10 years. For the estimate of the annual depreciation cost, a linear calculation was used:

depreciation = (DFC + start-up and validation costs (5% DFC)) / 10 years

Economic evaluation

Economic evaluation of the process was performed by assessing the 30 year net present value (NPV) and internal rate of return (IRR). Sensitivity analysis was used to determine the most important factors for the plant profitability and 30 year NPV assessments were applied to determine the minimum selling price of the biodiesel for profitability under different conditions.

Stand-alone microalgae plant versus integration with sugarcane bio-refinery

For the stand-alone plant, raw materials and utilities are purchased from external suppliers at market prices and biodiesel and algae meal are sold as commercial products. In the integrated model, the microalgae plant uses molasses generated during sugar crystallization as raw material and electricity and steam from bagasse burning and co-generation. Unlike the stand-alone model, the substrate is transferred directly from the crystallization process to the microalgae production plant. Apart from utilities, the equipment costs for microalgae-based biodiesel production are assumed to be identical in both set-ups.

Results and discussion

Capital costs

The equipment list and purchase costs for fed-batch and continuous production are detailed in Fig. 3. The biomass production section is by far the costliest for the capital estimation, with more than 64% and 55.5% of total EPC for the fed-batch and continuous process, respectively. The bioreactors alone are responsible for 95% of the total purchase cost of biomass production section, with a high number (20) of large-scale bioreactors.

In fed-batch mode, the need for an inoculum cultivation structure with multiple staggered seed bioreactors is a source of high capital costs, representing 15% of the EPC (see Appendix S1). Accordingly, the lower demand of inoculation operations in continuous mode results in 15% lower capital expenses. Although the lower product concentration obtained in continuous cultivation increases the size of medium preparation tanks and biomass harvesting centrifuges, the cost differences of seed and production bioreactors are not significant, as demonstrated in Fig. 3.

The DFC estimation for the continuous process (\$130 million) was circa 79% of the DFC for the fed-batch process (\$165 million), given the lower number of bioreactors needed

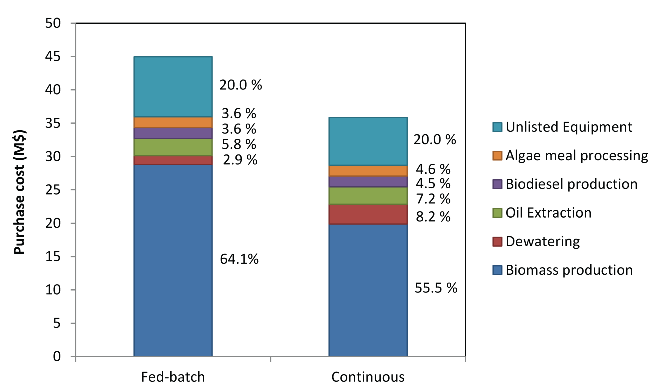

Figure 3. Equipment purchase cost by process section for fed-batch and continuous cultivation of the microalgae biodiesel plant.

for continuous cultivation. The estimation of DFC can largely vary depending on the specifications, suppliers, materials, and location factors. A brief review of similar studies and process design literature reveals a wide variability of bioreactor purchase costs, from $220 000 to $4.9 million for equivalent bioreactors (see Appendix S1). The impact of the bioreactor purchase cost and biomass productivities on the minimum biodiesel price was therefore assessed through a sensitivity analysis (Appendix S1).

Operating costs

Total OPCs were calculated for fed-batch and continuous cases and are shown in Table 3. The cost of molasses represents 56% of the total raw materials cost, followed by glycine and KH_2PO_4. Glycine is not the most economical choice of nitrogen source; however, the highest biomass productivities to date were obtained when using it.[11,14]

The utilities demand and costs were calculated for each section. Most of the electrical power is consumed in biomass production section for aeration of bioreactors. The highest demands of utilities were cooling water in the biomass production section, chilled water during oil extraction for solvent recovery through condensation, and steam in the algae meal section for drum drying. The main energy sources consumed were electricity and steam at 152 °C. The process simulation indicated an overall consumption of electricity of approximately 4.2 kWh per liter of biodiesel. The continuous and fed-batch processes had very similar overall energy demands. Although the higher product concentration in the fed-batch process resulted in less energy consumption in biomass centrifuging operation (44% lower than in continuous cultivation), the higher demand of energy for cultivation and seed bioreactors resulted in a 2% higher total consumption of electricity in fed-batch than in continuous operation. However, this translates to only $0.04 per liter of biodiesel, with $1.12 and $1.08 L^{-1} for fed-batch and continuous cultivation, respectively.

The labor demand in continuous mode was 33% lower than in fed-batch mode. The difference was due to the assumption that the sequential operations of inoculum preparation, biomass production, discharging, cleaning, and sterilization in fed-batch processing are more labor demanding than long-term continuous cultivation.

As mentioned before, the aqueous waste remaining from algae cultivation is recovered from biomass centrifugation

Table 3. Summary and breakdown of microalgae biodiesel production operating costs.

Cost source	Fed-batch Annual cost (M$)	$/L	%	Continuous Annual cost (M$)	$/L	%
Waste	0.70	0.03	1.2	1.87	0.09	3.50
Solid waste	0.43			0.43		
Liquid Waste	0.27			1.44		
Labor	3.82	0.19	6.6	2.55	0.13	4.79
Utilities	6.81	0.34	11.7	6,66	0.33	12.50
Electricity	4.4			4.3		
Steam	1.6			1.6		
Cooling	0.8			0.7		
Facility related	21.85	1.09	37.6	17.31	0.87	32.50
Maintenance (10% EPC)	4.49			3.59		
Depreciation	15.71			12.42		
Insurance (1% DFC)	1.65			1.30		
Materials	24.89	1.24	42.9	24.89	1.24	46.71
Molasses	14.04			14.04		
Glycine	5.98			5.98		
Medium salts	3.72			3.72		
Acids/bases	0.11			0.11		
Hexane	0.09			0.09		
Ethanol	0.93			0.93		
Total	58.1	2.90		53.3	2.66	
Total – algae meal revenues $		2.51			2.27	

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and algae meal ultrafiltration. The more diluted culture in continuous cultivation produces a larger volume of aqueous waste and therefore results in higher waste treatment costs. Both streams are composed of 98–99% (m/m) water, cell debris (around 0.1% m/m), remaining salts from culture medium, and ashes (around 0.8–1.0% m/m). From molasses clarification, a solid ash / gypsum-based residue with 28% water content is separated in a decanter centrifuge. As 70% of the solid residue is composed of minerals from sugarcane and gypsum formed during molasses clarification, this material could theoretically be used as fertilizer in sugarcane crops or as a supplement in animal feed. If all the solid residue generated from molasses clarification was re-applied to the soil as fertilizer, around 4250×10^3 kg year^{-1} of K_2O could be replaced, resulting in total savings of \$2 975 000. We considered, however, that the solid residue would be disposed in landfills because there is not enough data to support its direct use as fertilizer. If proven technically feasible, this option could bring significant gains.

Biodiesel cost

Revenues from algae meal were discounted from the total costs to determine the biodiesel final cost. The simulation indicated the annual production of 17.5 million kg of algae meal and the selling price of \$0.45 per kg was considered for revenue calculation. The total cost per liter of biodiesel was calculated as \$2.51 for the fed-batch process and \$2.27 for the continuous process, considering a fixed IRR of 7%.

Material costs was the most important fraction of cost composition, with 43 and 47% of the total costs for fed-batch and continuous operation, respectively. Facility related costs accounted for 38 and 32% of the total production costs for fed-batch and continuous operation, respectively, which reflects the high capital costs of the process, especially in the case of fed-batch cultivation.

Financial Analysis

Microalgae plant as independent business

The feasibility of the microalgae process as an independent business was assessed through NPV-analysis. The minimum biodiesel selling price that equals the NPV of the plant to 0 at a fixed IRR was calculated for different conditions. From the OPC, it is clear that the materials and capital expenses are the most important factors affecting the profitability of the business. Moreover, the co-product algae meal plays an important role, significantly increasing the total revenues.

The sensitivity analysis (Fig. 4) shows the effect of different values of IRR and other parameters on the biodiesel minimum selling price. An IRR of 12% is usually taken as the standard for attractive rate of return,[41] so \$2.96 and \$2.63 would be the minimum required selling prices to

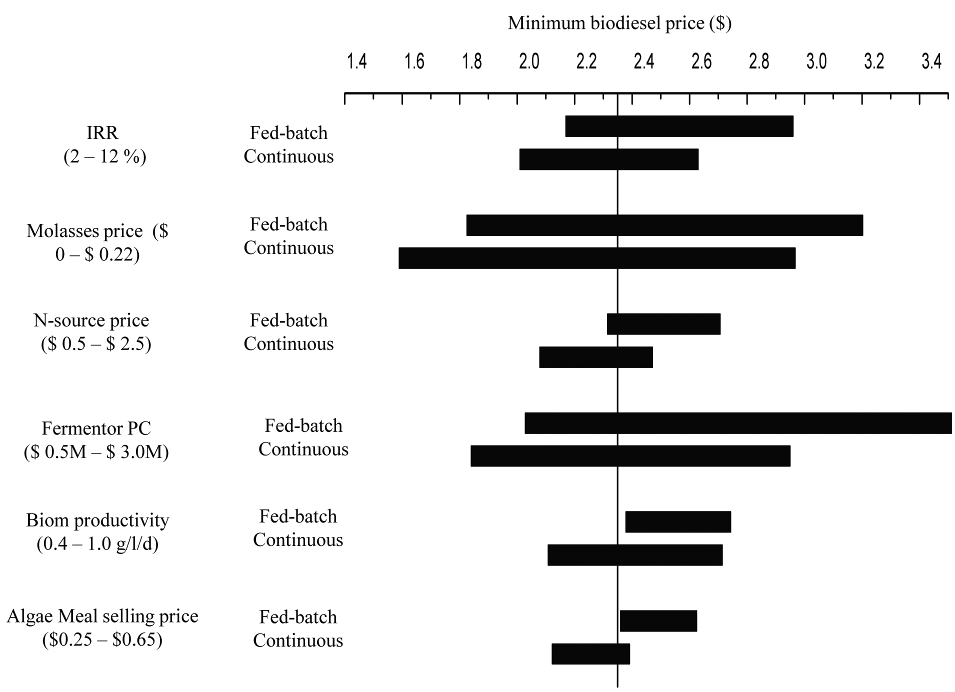

Figure 4. Sensitivity analysis of minimum biodiesel selling price for an independent (non-integrated) microalgae plant against the most important cost factors.

ensure the financial feasibility of the business. These prices are nearly three times higher than present diesel selling prices, which indicates that biodiesel and algae meal do not generate sufficient revenues to support the total costs of the plant. While several techno-economic assessments of photoautotrophic microalgae biodiesel production have been published in recent years, with a large variability in results (biofuel costs ranging from $ 0.45 to $ 8.7 per liter),[42] the results of this study are not directly comparable with those, as heterotrophic production is based on a different energy conversion concept, and the costs associated, as well as the technologies involved, are remarkably different. A more comparable process is the production of biodiesel from oleaginous yeast, which also heterotrophically converts organic carbon sources into lipids. Sae-Ngae (2020) estimated the minimum operating costs (CAPEX excluded) for producing biodiesel from lipids of oleaginous yeast would be $3.0 per kg.[43] Another recent study has estimated the minimum total cost of yeast oil (not converted to biodiesel) as $2.5 per kg.[44] These results are in relative accordance with costs estimated in this study (around $ 3.0 per kg of biodiesel), despite the differences in methodology and final products considered in these studies.

Molasses and bioreactor purchase costs had strong impact over the minimum biodiesel selling price. For molasses, a variation of $1.4 per liter between the limit values was observed. Different bioreactor purchase costs resulted in a variation of $1.5 in the minimum biodiesel prices for fed-batch mode and $1.1 for continuous mode. N-source and algae meal resulted in price variations of $0.5–2.5 and $0.25–0.65, respectively, and did not have a large impact on the biodiesel minimum price (12–15%).

Integrated bio-refinery

The main objective of the microalgae integrated system is the substitution of diesel consumed by sugarcane biorefineries. In such a configuration, utilities and molasses are supplied by the first-generation bio-refinery, which impacts bioethanol and energy production. The electricity sales revenue from cogeneration is also influenced as part of the power surplus is used in biodiesel production. A simplified analysis of gross profits was applied for feasibility assessment. The gross profit of the base sugarcane bio-refinery was calculated from literature data and compared with the estimate gross profit of the integrated microalgae-sugarcane plant.

The base sugarcane bio-refinery (Fig. 5) outputs were calculated from the data available in literature.[21] A sugar mill that processes 5×10^9 kg of sugarcane is expected to produce $254\,000 \times 10^3$ kg of sugar and 255 million liters of bioethanol. A surplus of 900 million kWh of electricity is produced through cogeneration from burning of bagasse. The gross profit is calculated as $90 350 000 million per year.

For the integrated plant, the molasses and utilities demand were calculated as described above. As these resources were now partially consumed in biodiesel production, the ethanol and electricity revenues were reduced. The new electricity revenue was calculated by:

electricity revenue (B) = electricity revenue (A) – microalgae plant demand × electricity selling cost

The reduction of ethanol production was calculated based on the conversion of the sugar content of the molasses that is used for biodiesel production, by using conversion factors from data in the literature ($Y_{\mathrm{ethanol/molasses}} = 0.45$ g ethanol / g sugar from molasses):[45]

ethanol revenue (B) = ethanol revenue (A) – molasses for biodiesel × $Y_{\mathrm{ethanol/molasses}}$

In the integrated model, purchased diesel was replaced by microalgae biodiesel. The operation costs of biodiesel and algae meal production were added to the model, as well as algae meal revenues. Molasses costs and depreciation were not considered, and electricity costs were accounted as equal to the production cost of $0.052/kWh.

The gross profit from the integrated sugarcane-microalgae bio-refinery was ca. $4.2 million lower than the base case bio-refinery. The feasibility of the integrated bio-refinery thus depends on the selling prices of ethanol and biodiesel. To investigate this, the difference in gross profit between the base and the integrated refinery were calculated for different prices of ethanol and biodiesel. The results are shown in Table 4. The integrated refinery would only be feasible with a significant increase in biodiesel selling prices and reduction in ethanol prices. At a selling price of $\$0.6\,\mathrm{L}^{-1}$ ethanol, the biodiesel price must be higher than $\$1.2\,\mathrm{L}^{-1}$.

One way to increase the profitability of the integrated plant is through the exploration of high-value products from microalgae, in a similar way to the approach taken by companies such as Martek S/A and Terravia/Corbion for carotenoids and polyunsaturated fatty acids.[46,47] Although some companies started their activities aiming at biofuel production, most of them are now focusing on specialty products, in order to finance their high production costs.[48] Some of the latest technoeconomic assessments of algae technologies also concluded that the commercial bioenergy production from microalgae can only be achieved when coupled with the production of high value-added products, such as β-carotene, polyunsaturated fatty acids (PUFAs), bioplastics and others.[20,49] Recent studies also concluded that the technology readiness level (TRL) of algal-based

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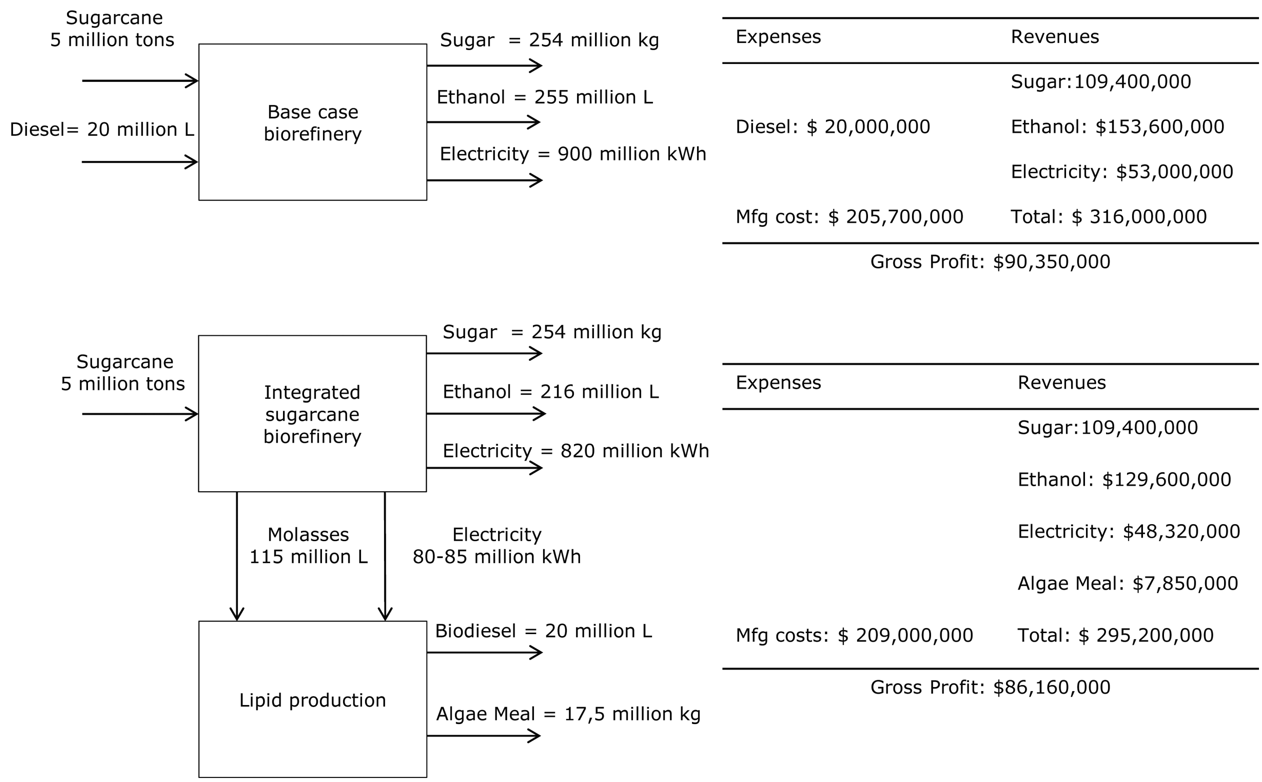

Figure 5. Inputs and outputs and profitability comparison between the base case sugarcane refinery and the integrated microalgae-sugarcane refinery.

Table 4. Calculated values in relation to ethanol and biodiesel prices: Profitability difference between base sugarcane bio-refinery and integrated microalgae-sugarcane bio-refinery / cost of reduction of CO_2 emissions in \$ ton^{-1} CO_2

		Biodiesel selling price ($/L)				
		0.6	0.8	1	1.2	1.4
Ethanol selling price ($/L)	0.9	**−22800k / 455**	**−18800k / 375**	**−14800k / 295**	**−10800k / 215**	**−6800k / 135**
	0.8	**−19200 / 385**	**−15200 / 305**	**−11200k / 225**	**−7200k / 145**	**−3200k /** 65
	0.7	**−15700k / 314**	**−11700k / 234**	**−7700k / 154**	**−3700k /** 74	0.3k / −6
	0.6	**−12160k / 243**	**−8160k / 163**	**−4160k /** 83	**−0.16k /** 3	3800k / −77
	0.5	**−8600k / 173**	**−4600k /** 93	**−600k /** 13	3370k / −67	7370k / −147
	0.4	**−5100k / 102**	**−1100k /** 22	2900k / −58	6900k / −138	10900k / −218
	0.3	**−1560k /** 31	2440k / −49	6440k / −129	10440k / −209	21500k / −430

Bold values: Higher profitability of base biorefinery compared to integrated refinery

biorefineries is still immature and further technological development is needed to reach a commercial scale.[50]

The substitution of fossil-based diesel can improve the bio-refinery overall renewability by eliminating the use of fossil fuels. Considering a total reduction of 50000 ton of CO_2-eq year^{-1}, the cost per ton of CO_2 reduction can be calculated. Carbon pricing can be considered a measure of the economic impact due to domestic and global benefits from reduced air pollution and climate change, and usually varies between \$30 and \$120 ton^{-1} CO_2-eq depending on the criteria.[51] Considering the goal carbon price of \$100 ton^{-1} CO_2-eq and the ethanol price of \$0.6 per liter, the selling price of biodiesel at \$1 is already advantageous compared to the base case, because the cost of reduction of one ton of CO_2-eq is calculated as \$83. Recent country-specific regulations can also boost renewable fuel producers' competitiveness. For example, the Renovabio program is the main instrument of the Brazilian government to increase the share of biofuels in the country's energy transport matrix. The national emission reduction targets for the fuel matrix were defined for the period from 2019 to 2029.[52] As a means of certification of biofuel production, different grades will be given to each biofuel producer and importer, in an inversely proportional value to the carbon emission intensity of the biofuel produced. The best ranked producers are then granted commercial benefits such as preferential selling orders to fuel distribution companies.

Conclusion

At present fuel prices (~\$0.6 L^{-1} ethanol and ~\$0.8 L^{-1} biodiesel), the integrated plant would be less profitable than a first-generation bio-refinery. However, inaccurate pricing estimation for equipment and materials can have a huge impact on the calculation of costs. Replacement of diesel in crop / transportation operations increases the overall renewability and reduces carbon emissions. A biodiesel derived from another sustainable feedstock may collaborate to reduce carbon emissions of sugarcane mills.

We conclude that the introduction of microalgae biodiesel in sugarcane sector depends on scale-up and learning effects; valorization of value-added and commodity by-products, e.g. fertilizers and biogas; and fiscal measures like carbon taxation. Regarding additional costs, the biodiesel-opportunity is comparable with other CO_2-emission reduction measures on a carbon price basis. Hence, innovation and political willingness are key for the success of the technology.

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References

1. Agostinho F and Siche R, Hidden costs of a typical embodied energy analysis: Brazilian sugarcane ethanol as a case study. *Biomass Bioenergy* **71**:69–83 (2014).
2. Macedo IC, Seabra JEA and Silva JEAR, Green house gases emissions in the production and use of ethanol from sugarcane in Brazil: The 2005/2006 averages and a prediction for 2020. *Biomass Bioenergy* **32**:582–595 (2008).
3. Souza SP, Gopal AR and Seabra JEA, Life cycle assessment of biofuels from an integrated Brazilian algae-sugarcane biorefinery. *Energy* **81**:373–381 (2015).
4. Edwards R, Hass H, Larivé J-F, Lonza L, Mass H, Rickeard D, et al. Well-to-Wheels analysis of future automotive fuels and powertrains in the European context WELL-TO-TANK (WTT) Report. Version 4. Jt Res Cent EU Ispra, Italy. 2013.
5. de Oliveira Bordonal R, Barretto de Figueiredo E, Aguiar DA, Adami M, Theodor Rudorff BF and La Scala N, Greenhouse gas mitigation potential from green harvested sugarcane scenarios in São Paulo State, Brazil. *Biomass and Bioenergy* **59**:195–207 (2013).
6. Atabani AE, Silitonga AS, Badruddin IA, Mahlia TMI, Masjuki HH and Mekhilef S, A comprehensive review on biodiesel as an alternative energy resource and its characteristics. *Renew Sustain Energy Rev* **16**:2070–2093 (2012).
7. Drabik D, de Gorter H and Timilsina GR, The effect of biodiesel policies on world biodiesel and oilseed prices. *Energy Econ* **44**:80–88 (2014).
8. Wijffels RH and Barbosa MJ, An outlook on microalgal biofuels. *Science* **329**:796–799 (2010).
9. Hall CAS and Benemann JR, Oil from Algae? *Bioscience* **61**:741–742 (2011).
10. Brune DE, Lundquist TJ and Benemann JR, Microalgal biomass for greenhouse gas reductions: Potential for replacement of fossil fuels and animal feeds. *J Environ Eng* **135**:1136–1144 (2009).
11. Cerón-García MC, Macías-Sánchez MD, Sánchez-Mirón A, García-Camacho F and Molina-Grima E, A process for biodiesel production involving the heterotrophic fermentation of *Chlorella protothecoides* with glycerol as the carbon source. *Appl Energy* **103**:341–349 (2013).
12. Zheng Y, Li T, Yu X, Bates PD, Dong T and Chen S, High-density fed-batch culture of a thermotolerant microalga Chlorella sorokiniana for biofuel production. *Applied Energy* **108**:281–287 (2013).
13. Doucha J and Lívanský K, Production of high-density Chlorella culture grown in fermenters. *J Appl Phycol* **24**:35–43 (2012).
14. De la Hoz Siegler H, McCaffrey WC, Burrell RE and Ben-Zvi A, Optimization of microalgal productivity using an adaptive, non-linear model based strategy. *Bioresource Technology* **104**:537–546 (2012).
15. Rismani-Yazdi H, Hampel KH, Lane CD, Kessler BA, White NM, Moats KM *et al*., High-productivity lipid production using mixed trophic state cultivation of Auxenochlorella (Chlorella) protothecoides. *Bioprocess Biosyst Eng* **38**:639–650 (2015).
16. Perez-Garcia O, Escalante FME, de- Bashan LE and Bashan Y, Heterotrophic cultures of microalgae: Metabolism and potential products. *Water Res* **45**:11–36 (2011).
17. Goes T, Marra R, De Araújo M, Alves E and Oliveira De Souza M, Sugarcane in Brazil current technologic stage and perspectives. *Rev Política Agrícola* **1**:52–65 (2011).
18. Liu J, Huang J, Jiang Y and Chen F, Molasses-based growth and production of oil and astaxanthin by Chlorella zofingiensis. *Bioresour Technol* **107**:393–398 (2012).
19. Miao X, Li P, Li R and Zhong J, In situ biodiesel production from fast-growing and high oil content *Chlorella pyrenoidosa* in rice straw hydrolysate. *J Biomed Biotechnol* **2011**:1–9 (2011).
20. Khoo CG, Dasan YK, Lam MK and Lee KT, Algae biorefinery: Review on a broad spectrum of downstream processes and products. *Bioresour Technol* **292**:121964 (2019).

21. Cavalett O, Junqueira TL, Dias MOS, Jesus CDF, Mantelatto PE, Cunha MP *et al*., Environmental and economic assessment of sugarcane first generation biorefineries in Brazil. *Clean Technol Environ Policy.* **14**:399–410 (2012).
22. Dias MOS, Junqueira TL, Cavalett O, Cunha MP, Jesus CDF, Mantelatto PE *et al*., Cogeneration in integrated first and second generation ethanol from sugarcane. *Chem Eng Res Des* **91**:1411–1417 (2013).
23. Ranken MD, Kill RC and Baker C, Sugar and chocolate confectionery, in *Food Industries Manual*. Boston, MA: Springer, pp. 406–443 (1997).
24. Behera BK, Varma A. Microbial Resources for Sustainable Energy 2016.
25. Anschau A, Xavier MCA, Hernalsteens S and Franco TT, Effect of feeding strategies on lipid production by *Lipomyces starkeyi*. *Bioresour Technol* **157**:214–222 (2014).
26. Palabhanvi B, Muthuraj M, Kumar V, Mukherjee M, Ahlawat S and Das D, Continuous cultivation of lipid rich microalga Chlorella sp. FC2 IITG for improved biodiesel productivity via control variable optimization and substrate driven pH control. *Bioresour Technol* **224**:481–489 (2017).
27. Jones S, Zhu Y, Anderson D, Hallen RT, Elliott DC. Process design and economics for the conversion of algal biomass to hydrocarbons: Whole algae hydrothermal liquefaction and upgrading. PNNL 2014.
28. Ahmad AL, Mat Yasin NH, Derek CJC and Lim JK, Crossflow microfiltration of microalgae biomass for biofuel production. *Desalination* **302**:65–70 (2012).
29. Spiden EM, Yap BHJ, Hill DRA, Kentish SE, Scales PJ and Martin GJO, Quantitative evaluation of the ease of rupture of industrially promising microalgae by high pressure homogenization. *Bioresour Technol* **140**:165–171 (2013).
30. Coons JE, Kalb DM, Dale T and Marrone BL, Getting to low-cost algal biofuels: A monograph on conventional and cutting-edge harvesting and extraction technologies. *Algal Res* **6**:250–270 (2014).
31. Rawat I, Ranjith Kumar R, Mutanda T and Bux F, Biodiesel from microalgae: A critical evaluation from laboratory to large scale production. *Appl Energy* **103**:444–467 (2013).
32. Levine RB, Pinnarat T and Savage PE, Biodiesel production from wet algal biomass through in situ lipid hydrolysis and supercritical transesterification. *Energy Fuels* **24**:5235–5243 (2010).
33. Haas MJ, McAloon AJ, Yee WC and Foglia TA, A process model to estimate biodiesel production costs. *Bioresour Technol* **97**:671–678 (2006).
34. Van Gerpen JH and He BB, Biodiesel and renewable diesel production methods, in *Advances in Biorefineries: Biomass and Waste Supply Chain Exploitation*. Cambridge, UK: Woodhead Publishing, (2014).
35. Beal CM, Gerber LN, Sills DL, Huntley ME, Machesky SC, Walsh MJ *et al*., Algal biofuel production for fuels and feed in a 100-ha facility: A comprehensive techno-economic analysis and life cycle assessment. Algal Research. **10**:266–279 (2015).
36. Towler G and Sinnott RK, *Chemical Engineering Design - Principles, Practice and Economics of Plant and Process Design*, 2nd edn. Oxford, UK: Butterworth-Heinemann, (2013).
37. Klein-Marcuschamer D, Turner C, Allen M, Gray P, Dietzgen RG, Gresshoff PM *et al*., Technoeconomic analysis of renewable aviation fuel from microalgae, Pongamia pinnata, and sugarcane. *Biofuels Bioprod Biorefin* **7**:416–428 (2013).
38. Peters MS, Timmerhaus KD and West RE, *Plant Design and Economics for Chemical Engineers*, 5th edn. Boston, US: McGraw-Hill, (2003).
39. ICIS, ICIS. Indicative Chemical Prices A-Z. *ICIS Chemical industry news & chemical market* **1**:A–Z (2015).
40. Services RE, *Richardson's International Construction Factors: Location Cost Manual*. 2007 Pahrump, NV USA: Richardson Engineering Services, Incorporated, (2007).
41. Mariano AP, Dias MOS, Junqueira TL, Cunha MP, Bonomi A and Filho RM, Butanol production in a first-generation Brazilian sugarcane biorefinery: Technical aspects and economics of greenfield projects. *Bioresour Technol* **135**:316–323 (2013).
42. Quinn JC and Davis R, The potentials and challenges of algae based biofuels: A review of the techno-economic, life cycle, and resource assessment modeling. *Bioresour Technol* **184**:444–452 (2015).
43. Sae-Ngae S, Cheirsilp B, Louhasakul Y, Suksaroj TT and Intharapat P, Techno-economic analysis and environmental impact of biovalorization of agro-industrial wastes for biodiesel feedstocks by oleaginous yeasts. *Sustain Environ Res* **30**:3627–3658 (2020).
44. Bonatsos N, Marazioti C, Moutousidi E, Anagnostou A, Koutinas A and Kookos IK, Techno-economic analysis and life cycle assessment of heterotrophic yeast-derived single cell oil production process. *Fuel* **264**:116839 (2020).
45. Basso LC, Basso TO, Rocha, ed. by Bernardes MADS, *Ethanol Production in Brazil: The Industrial Process and Its Impact on Yeast Fermentation. Biofuel Production – Recent Developments and Prospects*. IntechOpen, Rijeka, pp. 85–100 (2011).
46. Ward OP and Singh A, Omega-3/6 fatty acids: Alternative sources of production. *Process Biochem* **40**:3627–3652 (2005).
47. Ziolkowska JR and Simon L, Recent developments and prospects for algae-based fuels in the US. *Renew Sustain Energy Rev* **29**:847–853 (2014).
48. Benemann J, Woertz I and Lundquist T, Life cycle assessment for microalgae oil production. *Disrupt Sci Technol* **1**:68–78 (2012).
49. Thomassen G, Van Dael M and Van Passel S, The potential of microalgae biorefineries in Belgium and India: An environmental techno-economic assessment. *Bioresour Technol* **267**:271–280 (2018).
50. Thomassen G, Egiguren Vila U, Van Dael M, Lemmens B and Van Passel S, A techno-economic assessment of an algal-based biorefinery. *Clean Technol Environ Policy* **18**(6):1849–1862 (2016).
51. Parry IAN, Veung C and Heine D, How much carbon pricing is in countries' own interests? The critical role of co-benefits. *Clim Chang Econ* **06**:1550019 (2015).
52. CNPE. Resolution no 16, of 24th of June 2019. National Council for Energy Policy - Brazil 2019.

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