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Prefeasibility analysis of low-scale biorefineries: annatto and açai case

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Abstract

The industrial applicability of exotic and wild fruits has been investigated as stand-alone processes framed to obtain a single main product. However, their combination and the valorization of the processing residues in a biorefinery scheme have not been explored. Therefore, this work aims to assess the technical and economic feasibility of low-scale biorefineries based on annatto and açai as raw materials. Therefore, the actual processing of both fruits was carried out experimentally. For annatto, the extraction of dye paste was performed, obtaining mass yields of up to 58.4% by working with ground seed. Regarding the açai, pulp was obtained with a polyphenol content of 23.56 mg of gallic acid equivalent per gram of sample. The residues generated in both processes were used as substrates for biogas production, achieving biomethane yields between 186–533 mL per gram of volatile solids. These results were the basis for the design and simulation of process scale-up in low-scale biorefinery contexts. Five scenarios were evaluated, involving the production of annatto dye paste, açai pulp, biogas from exhausted annatto seeds, slurry and seeds of açai, and feedstock integration. As the main energy result, it was found that the individual production of açai pulp can fully supply the required energy of the process through the self-generation index. From the economic perspective, it was found that there is economic feasibility working with flows below the regional and national production in Colombia for all scenarios.

1. Introduction

Nowadays, the worldwide concern for health welfare has allowed the consumption of fruits and natural additives to be in constant growth due to their high content of minerals, vitamins, and antioxidant compounds. The relevance of agricultural production for economic development can be seen through employment generation, economic incentives from governmental entities towards crop development, and its contribution to the gross domestic product (GDP). Most developing countries rely heavily on agriculture as a source of employment and economic income, whether for local consumption or export. For example, the EU accounts for about 50% of the world's tropical fruit exports (FruitLogistica, 2021). The most well-known and consumed fruits (major fruits) are cultivated on a large scale and industrialized. Meanwhile, wild and exotic fruits are not as widely cultivated and are consumed regionally or according to cultural practices. The trade of these fruits is seasonal, and their export is limited. The biogeographical region of Chocó (Colombia) is characterized by an extensive cultivated area of rainforest, including a great variety of exotic and wild fruits. Therefore, many efforts have been made to valorize these crops for the small-scale production chains of the local communities.

The annatto (*Bixa orellana* L.) is an exotic reddish-yellow fruit used to extract natural colorants (carotenoids) from the outer coating of the seeds. Interest in natural colorants is growing due to their minimal or null toxicity. In contrast, the World Health Organization (WHO) regulates the consumption of synthetic colorants due to health problems such as hyperthyroidism, kidney disease, insomnia, and anemia (L.C. Albuquerque & A.A. Meireles, 2012). The main pigment present in annatto is bixin, and it comprises more than 80% of the total carotenoid content in their seeds (Rodrigues et al., 2014). Annatto seeds can be processed in two main pathways to produce the dye extract. (i) Mechanical abrasion of the exocarp using a suspending agent (i.e., water, alkali, vegetable oil) followed by seed separation. Despite the high extraction yields with alkaline agents, removing these components requires several downstream unit operations with high energy requirements, increasing overall process cost. (ii) Extraction with organic; however, the dye will be subject to strong technical restrictions on the number of toxic solvent residues (Taham et al., 2015). Therefore, extraction by mechanical abrasion can be attractive for the extraction of bioactive substances used in non-technified or low-technological processing. It would help reduce processing time, decrease or eliminate the organic solvent use, and improve the quality of the extracts. Moreover, this methodology is applied in the Choco region, where ancestral processing has predominated.

Açai (*Euterpe oleracea*) is a wild fruit that can be black or purple, and white or green, which will produce a pulp according to its color (Noblick, 2019). Açai pulp is the most economically viable product used for juice production, sold unprocessed (pasteurized pulp) or in powders. The nutritional power of açai has been recognized due to more than 90 bioactive substances with antioxidative capacity, highlighting flavonoids, lignoids, phenolic compounds, proanthocyanidins, and anthocyanins (Matta et al., 2020). Furthermore, açai juice is significantly caloric (as an energy drink) given its levels of fiber, protein, carbohydrates, and lipids, where the latter has a quantity of unsaturated fatty acids greater than 65% and with a profile similar to avocado and olive oils (M. do S. P. de Oliveira & Schwartz, 2018). Therefore, açai has been considered a functional food in the food industry.

Throughout the processing of annatto and açai, considerable residues are generated. About 95% of the annatto seed is destined as waste due to the low bixin extraction yields (Taham et al., 2015). Moreover, an açai processing company produces a large amount of waste per day since 5–15% of the fruit is edible, and 80% corresponds to the seed weight (Buratto et al., 2021). Therefore, the valorization of these residual fractions is important for sustainability through the integral use of the raw material. In the literature, studies concerning annatto are focused on bixin extraction for stand-alone processes or coupled with pretreatment steps and do not contemplate the generation and valorization of residues (Albarelli et al., 2016). In contrast, for açai, some works have focused on the seed valorization towards energy products. the technical and economic feasibility of power generation through different combustion schemes (Teixeira et al., 2013), the use of downdraft gasifiers (Itai et al., 2014), and the formation of biochar through slow pyrolysis (Sato et al., 2019) have been evaluated. Based on those mentioned above, coupling the up-to-date processing of annatto and açaí fruits to an additional processing step to valorize the residues may be promising. This step should be simple, requiring a low technology readiness level, low capital and energy requirements, and that can be easily implemented due to the accessibility of exotic and wild crops. Thus, anaerobic digestion can be a waste processing alternative since it can encompass different substrates to mitigate the organic environmental load, low capital cost, and supply the energy demand of the process or surrounding urban locations. Despite several processes to produce energy from waste, anaerobic digestion can be a suitable technology to manage and dispose of food industry waste. To date, there are no studies that demonstrate the coupling of anaerobic digestion as an alternative to the integral processing of annatto and açai residues.

Biorefineries are integrated systems that can propose processing schemes to transform biomass into value-added products. It is common to find integration concepts in mass, energy, or technology (Moncada B et al., 2016). The challenge is to evaluate the process by finding a balance and analyzing the feasibility based on scale and context. Mass integration is one of the alternatives with high potential to be applied in biorefineries. This integration allows for increased yields, productivity, and overall sustainability of the process (Piedrahita-Rodríguez & Cardona Alzate, 2021). However, several types of research on this topic have covered the definition of mass integration from different points of view. For example, some authors consider the valorization of residual streams to obtain value-added products or integration by process flexibility (Moncada et al., 2014), use raw material at different seasons (Pyrgakis & Kokossis, 2018), or use more than one feedstock at a time (Zhang et al., 2018). This last consideration is one of the most interesting and offers multiple benefits to improve the viability of biorefineries. However, the studies that have been carried out so far have shown some progress, but it is not a completely standardized methodology. In this way, it would be possible to determine the main restrictions of integration from the conceptual design and how this can be improved.

This work aims to evaluate the current regional use of exotic and wild fruits such as annatto and açai for the production of dye and fruit pulp in stand-alone processes. In addition, the influence of anaerobic digestion on the residue treatment from fruit processing was analyzed. Different feedstock feed ratios were also studied in the anaerobic digestion stage in a low-scale biorefinery scheme to integrate regional raw materials and improve the technical and economic aspects of the process. For this purpose, six scenarios were evaluated for experimental and simulation purposes: (Sc1) annatto dye production; (Sc2) açai pulp production; (Sc3) production of annatto dye and biogas; (Sc4) production of açai pulp and biogas; and (Sc5) production of dye, pulp, and biogas through a feedstock integration (biorefinery scheme).

2. Methodology

2.1. Experimental procedure

2.1.1. Raw materials

The açai (*Euterpe oleracea* Mart.) was obtained from a local non-commercial crop in the tropical forest city of Quibdó, Colombia (5°41'28.9"N 76°39'57.9"W) and red annatto seeds (*Bixa Orellana* L.) from a small seed-threshing of the Unión Panamericana, Colombia (5°14'38.4"N 76°39'33.3"W).

2.1.2. Raw material characterization

The physicochemical characterization of the raw material was performed based on international standards and in triplicate. Soxhlet system was used for extract content with water and ethanol (Pagès et al., 2016) and with hexane for fats (M. D. L. De Castro & Priego-Capote, 2010). Holocellulose was estimated by the chlorination with acetic acid, while cellulose after different dosages of NaOH (Machrafı, 2012). The insoluble lignin was estimated as Klason lignin (Sluiter et al., 2012). Protein was calculated by the Kjeldahl method using a factor conversion of 6.25 (Hames et al., 2008). Total pectin was extracted with concentrated sulfuric acid at continuous stirring (Yu et al., 1996). For pectin quantification, photometric galacturonic acid measurement was implemented by the carbazole method at 240 nm (Abbaszadeh, 2008). Fats were determined after Soxhlet extraction with hexane. Fiber was calculated using the gravimetric method (S. Lee et al., 1992) and carbohydrates as nitrogen-free extract (Hall, 2003). Finally, ash was determined after heating at 500°C (Feng et al., 2005).

2.1.3. Mass integration

The feedstock integration ratios under the biorefinery concept were determined according to the methodology reported by Piedrahita-Rodriguez (2020). The raw materials considered for integration were: exhausted annatto seed (after the dye extraction process), açai seed, and açai slurry (residue left after obtaining açai pulp). Several criteria were used to find the integration rate. These criteria involve the composition of the raw material, the supply chain, its phase and particle size, pretreatment stage, market and final prices, environmental impact on its agronomic component, and availability and accessibility. The algorithm depicted in Fig. 1 considers each of these criteria. Besides, the algorithm must initially have fixed information, such as a base raw material, a clear integration objective, defined technologies, and defined products.

The first step is to define the base raw material and apply the algorithm that includes each of the criteria mentioned above. In this sense, the base raw material was selected as presenting a smaller production scale or having important restrictions relating to its availability. On the other hand, it can also be considered if the base raw material involves an urgent need. The exhausted annatto seed was selected as the base raw material for this case due to its production scale (smaller than açai) and the lack of research related to integrating processes to complement the annatto value chain. The integration objective defined for this work is to generate a sustainable process under the biorefinery concept by using two raw materials present in the region of Chocó, Colombia. Thus, the biorefinery products are annatto dye, açai pulp, and biogas through anaerobic digestion based on their residues.

Figure 1 Block-diagram algorithm for the design of biorefineries integrating raw materials.

2.1.4. Açaí pulp production

The açai fruit was processed without the agricultural residues (i.e., leaves and branches) of the crop. The pulp production was performed manually based on the expertise of a local facility (Refrescos del Litoral, Chocó, Colombia). The process started with a fruit washing (tap water) ratio of 0.75 L kg^{-1}. Thereafter, the açai was disinfected with a sodium hypochlorite solution (100 ppm) in a solid-to-liquid ratio of 4:3. Then, the fruit was blanched by heating in boiling water for 3 min, followed by cooling. Finally, the açai was manually pulped, generating three raw material fractions (i) pulp (10–13%

wt.); (ii) slurry, equivalent to retained fibers and peel (15–20% wt.); and (iii) seed (65–75% wt.). The seeds were dried at 30°C, ground, and sieved to a size of 0.44 mm. The pulp and the slurry were stored in plastic bags and frozen at -30°C for further use.

To determine total phenolic content, the pulp sample (0.150 g) was mixed with 1 mL of ethanol:HCl:water solution (93:1:6 vol.) in conical flasks and immersed for two hours in an ultrasound bath operated at 37 kHz and 22°C. Then, the extract was centrifuged (13,500 rpm, 20 min), and the supernatant was collected and stored in the dark in an amber glass bottle at 4°C prior to analysis. The total phenolic content of the extract was measured by using the Folin–Ciocalteu colorimetric method according to Singleton et al. (1999) with some modifications, 150 µL of the ethanol/HCl extract was mixed with 2.4 mL distilled water, 150 µL of Folin–Ciocalteu solution (1N) and 300 µL of sodium carbonate (20% w/v). After 120 min in the dark, the absorbance was measured at 765 nm in a UV/Visible spectrophotometer. The total phenolic content was expressed in mg of gallic acid (GA) equivalents $100g^{-1}$ of the sample.

A DPPH (2,2'-diphenyl-1-picrylhydrazyl) determination was made using a modification of the method described by Marinova and Batchvarov (2011). A sample of 2.5 mg of the radical was weighed and dissolved in 100 mL of ethanol (96%). For all samples, 3 mL of this solution and 150 mL of the extract were dissolved in ethanol in different tubes at five concentrations from 20 to 80%. The tubes were stored in the dark for 60 min and their absorbances were measured at 517 nm. The results were expressed as IC50 (amount of the extract needed to scavenge 50% of the DPPH). The anthocyanin content was determined by the differential pH method (J. Lee et al., 2001). Anthocyanin content is expressed in equivalent mg of cyanidin-3-glucoside per liter of solution or milligram of dry matter.

2.1.5. Dye production

For dye production, two types of seed size were studied: ground and natural. The ground seeds were obtained after a seed milling (blade mill) until 0.44 mm particle size. At first, 100 g of both samples were mixed with 300 mL of tap water and left to stand for 24 h. Then, each sample was manually washed several times with tap water through abrasion until a dye-depleted seed (removal of the colorant layer by abrasion) was obtained. Finally, the extract obtained (water with dye) was evaporated with magnetic stirrer assistance. The collected colorant was stored in amber flasks at -30°C.

Bixin and norbixin were quantified in the produced extract by high-performance liquid chromatography (HPLC) according to the methodology reported by Noppe et al. (2009). The separation was performed with a Kromasil C_{18} column (150 mm x 4.6 mm x 5 µm) in a linear gradient method of two mobile phases of (A) acetonitrile and (B) formic acid solution (0.1% vol.) at 0.3 mL min^{-1} and room temperature. The method begins with 80% A and 20% B, increasing to 95% A and 5% B during 10 min; then, 100% A is reached over 5 min and must be held for 5 min. Finally, the initial conditions (80% A and 20% B) are set and held for 10 min. The stock standard solutions for bixin and norbixin were prepared in ethanol. The samples and the calibration curves were dissolved in 0.1% acetic acid in acetonitrile/water (80:20 vol.) and were filtered on a 0.22 µm nylon filter.

2.1.6. Biogas production

2.1.6.1. Biochemical methane potential

The residue fractions from pulping and colorant extraction (exhausted seed) were used as the substrate for biogas production through anaerobic digestion, defined as EAS-N (natural exhausted annatto seed); EAS-G (ground exhausted annatto seed); AS (açai seed); ASL (açai slurry); EAS-N:AS:ASL (mixture of natural annatto seed, seed and slurry of açai); and EAS-G:AS:ASL (mixture of ground annatto seed, seed and slurry of açai). For this purpose, the residues were characterized by total solids (TS) and volatile solids (VS) content. Sludge from an anaerobic water treatment of coffee was used. The sludge was filtered with a 0.1 mm pore-sized nylon filter to remove the excess water, reaching a final concentration of 7.4% and 11.1% for VS and TS, respectively. The "concentrated" sludge was adapted by a degassing process at 37°C in an inert medium to avoid disturbances in biogas production measurements (VDI-Handbuch Technik Biomasse/Boden, 2016).

The biochemical methane potential (BMP) methodology was used to evaluate the degradation of organic substrates. The protocol of the international standard VDI 4630 (VDI-Handbuch Technik Biomasse/Boden, 2016) was followed. A 100 mL molded glass vial was used as the digester (90 mL of digestion volume). Two feed ratios were used: (i) 0.015 gVS of feedstock per mL of digestate and (ii) a VS ratio between feedstock and inoculum of 0.4. Subsequently, the digestion volume was supplemented with tap water and added macro and micronutrients (Angelidaki et al., 2009). Finally, the pH was adjusted to 7 with HCl or NaOH (1 N) solution. After the digestion medium was set, nitrogen was bubbled for 5 min to each sample and left in incubation at 37°C. Measurement and recording of the biogas produced (gas concentration) were carried out using a portable gas analyzer (Portable Infrared Biogas Analyzer Gasboard – 3200L) every 48 h.

2.1.6.2. Kinetic model

The experimental results of the anaerobic digestion were fitted to the Gompertz kinetic model (**Eq. (1)**). MATLAB R2018a software was used to minimize the error between the model and the experimental values. The square correlation factor evaluated the degree of determination. In **Eq. (1)**, y represents the gas accumulation yield (mL gVS^{-1}) at the time t (day) of digestion. A is the gas production potential (mL g^{-1}), μ_m is the maximum gas production rate (mL $g^{-1}day^{-1}$), and λ is the adaptation phase (day).

$$y = A\exp\left\{ - \exp\left[\frac{\mu_m e}{A} (\lambda - t) + 1 \right] \right\} \quad \text{Eq. (1)}$$

2.2. Simulation procedure

2.2.1. Simulation desccription

Each experimental process was simulated with Aspen Plus software v9.0 (Aspen Technologies, Inc., USA), using the experimental data as input parameters. Five scenarios were contemplated: the stand-alone processing of annatto seeds for dye production (Sc1) and açai for pulp production (Sc2). The valorization of annatto and açai residues to produce biogas coupled with dye production (Sc3) and pulp production (Sc4). The integration of both fruits and the valorization of their residues in a biorefinery scheme to produce annatto dye, açai pulp, and biogas (Sc5). All the simulations consider a raw material processing of ten tons per day (10 ton d^{-1}). The objective is to scale up the processes and define the industrial equipment to obtain the desired products. Therefore, the technical and economic feasibility of regional application (Chocó, Colombia) was analyzed through controlled processes of cultural or artisanal production practices. A detailed description of each scenario can be found in **Online Resource 1**.

2.2.2. Techno-economic assessment

The processes were analyzed considering mass and energy requirements and simulation results. Some mass and energy indicators were calculated using the equations shown in **Online Resource 2**. The indicators allow determining the potential of raw materials to generate several products in stand-alone or biorefinery schemes. They also allow giving comparable values with other similar processes or under different contexts that enhance the results discussion and the perspectives that may arise (Alonso-Gómez et al., 2020).

Based on the previous technical results, all the simulated scenarios were analyzed in economic terms. The costs of the equipment involved in the processes were calculated using the commercial tool Aspen Process Economic Analyzer v9.0 (Aspen Technologies, Inc., USA). Likewise, the capital (CapEx), operating (OpEx), and production costs were calculated based on the methodology presented by Peters et al. (2003). A continuous operation process with a working time of 8760 hours per year, an interest rate, and a corporate tax rate of 9.85% and 31% were considered. The economic assessment involves the US dollars as unit cost, and the operator pay rate was 1.32 USD h^{-1}. Table 1 summarizes the input and utility costs and product selling prices. Finally, all scenarios assumed the straight-line depreciation method for the plant life of 20 years.

Table 1
Cost and prices for the economic assessment

Item	Value	Units
Annatto seeds*	2.90	USD kg^{-1}
Açai fruit*	0.25	USD kg^{-1}
Sodium hypochlorite (70%)**	1.15	USD kg^{-1}
Process water***	2.95	USD m^{-3}
Cooling water***	4.95	USD m^{-3}
Low-pressure steam****	26.0	USD ton^{-1}
Electricity****	0.1	USD kWh^{-1}
Annatto dye paste*	20.28	USD kg^{-1}
Açai pulp*	5.32	USD kg^{-1}
Biogas****	0.35	USD m^{-3}
*Values from the regional market.		
**Cots assumed as an average of www.alibaba.com.		
***Values calculated using a CEPCI of 701.4 from 2021.		
****Taken from previous works (Poveda-Giraldo & Cardona Alzate, 2021).		

Table 1 Cost and prices for the economic assessment

3. Results And Discussion

3.1. Raw material composition

Table 2 shows the compositional analysis of the raw materials, as well as the polyphenolic and anthocyanin content, and the antioxidant activity. The results showed that the composition of annatto and açai are similar except for the hemicellulose content. However, the lignocellulosic composition was in agreement with other studies reported in the literature. Buratto et al. (2021) calculated an açai seeds content of, respectively, 8.5%, 48.1%, and 11.6 % for cellulose, heicellulose, and lignin. In contrast, 14.4% cellulose, 34.0% hemicellulose, and 10.8% lignin have been reported for annatto seeds (Kumar et al., 2007). The high lignocellulosic composition allows them to be a potential source of biofuel, with calorific values of 16.98 MJ kg^{-1} for açai seeds (Alves et al., 2021) and 16.0 MJ kg^{-1} for annatto seeds (Atienza et al., 2021). These values have been equal to or even higher than wood chips or pine and with a comparable açai energy density of 10.01 GJ m^{-3} with wood pellets (11.1 GJ m^{-3}) (Alves et al., 2021). The high calorific values are also explained by the high extract content found in both fruits since extractives promote biomass ignition at the initial combustion stage. The extract content is greater in this work than another açai (Souza De Oliveira et al., 2020) and annatto (Okorie et al., 2020) studies. Concerning the ash composition, values were slightly higher for acai seed than those reported by Sato et al. (2019). Meanwhile, the values for annatto seed were lower than those reported in the literature (C. Alcázar-Alay et al., 2015). These results can be explained by species and crop factors, such as the ripening stage and harvest season.

Table 2
Physicochemical characterization of annatto seeds and açai fractions

Parameter	Mass composition (%)		
	Annatto seed	Açai seed	Açai pulp
Initial moisture	40.01 (0.05)	31.26 (0.02)	89.63 (0.32)
Total extractives*	28.46 (1.91)	22.31 (0.51)	-
Fiber*	-	-	11.78 (1.36)
Total carbohydrates*	-	-	29.91 (1.20)
Cellulose*	18.81 (0.73)	13.05 (1.46)	-
Hemicellulose*	11.34 (1.20)	42.67 (1.81)	-
Lignin*	13.92 (0.35)	15.91 (6.71)	-
Fats*	2.76 (0.06)	2.98 (0.10)	51.86 (1.45)
Pectin*	16.00 (0.04)	-	-
Protein*	8.71 (0.39)	-	4.21 (0.70)
Ash*	5.39 (0.07)	7.54 (0.11)	2.24 (0.30)
Polyphenols content**	5.45 (0.26)	-	23.56 (0.72)
Antioxidant activity IC50***	443.65 (5.37)	-	285.84 (3.54)
Anthocyanins content****	-	-	22.58 (1.41)
Values in brackets refer to standard deviation.			
*Composition on a dry basis.			
**Results in mg of GA equivalents g^{-1} of the sample.			
***Results in µmol L^{-1}.			
****Results in equivalent mg of cyanidin-3-glucoside equivalent $100mg^{-1}$ of sample.			

Table 2 Physicochemical characterization of annatto seeds and açai fractions

The açai pulps are highly perishable at room temperature by the rapid action of its peroxidases and polyphenol oxidases. These enzymes cause browning, anthocyanin degradation, production of peroxide radicals, and lipid oxidation. Therefore, it is recommended a fast fruit processing maximum of 48 hours after harvest) and the application of methods such as pasteurization or novel technologies such as isostatic high-pressure processing, high-pressure processing pulsed light, ultrasound, and microwave (Dantas et al., 2021). Fats extraction from the açai pulp must be carried out to avoid deterioration. This oil is used as a supplement for animal feed, especially in lactation sheep, and for cosmetics (da S. dos Santos et al., 2019). A lipids content close to 50% (on a dry basis) allows the pulp to be an energy source for food products. The total dietary fiber content was low than other fruits, such as passion fruit (53.5%), citrus lemon (87.1%), and coconut (69.9%) (Garcia-Amezquita et al., 2018) since açai pulp was filtered and a certain fraction of the fiber was possibly discarded in this process. Meanwhile, the carbohydrate content is in agreement with the typical range of berries, as blueberries (Lucas et al., 2018) and açai (A. R. Oliveira et al., 2020).

The açai presented high values in polyphenols content than other fruits that are considered sources of these nutrients, such as European cranberry (161 mg GA Eq. $100g^{-1}$), honeyberry (311 mg GA Eq. $100g^{-1}$), black cowberry (565 mg GA Eq. $100g^{-1}$) (N. L. Oliveira et al., 2021), and blueberries (378.2 mg

GA Eq. $100g^{-1}$) (Li et al., 2017). Some biocompounds of açai, such as anthocyanin and carotenoids, have shown therapeutic, anti-inflammatory, and anticancer properties (Romualdo et al., 2015). Some studies exhibited the anthocyanin as the main compound of polyphenols in açai, cyanidin-3-glucoside, and cyanidin-3-rutinoside as the predominant forms and are responsible for the purple color of the fruit (Yamaguchi et al., 2015). Regarding the phenolic content of annatto seeds, it showed better results when there are extracted with KOH (4.47 mg GA g^{-1}) (Raddatz-Mota et al., 2016) or methanol (1.5 mg GA g^{-1}) (Chisté et al., 2011).

3.2. Dye extraction

As the main results, mass yields of dye extraction of 15.2% and 58.4% were achieved for natural and ground seeds, respectively. Furthermore, exhausted seed yields of 80.3% for the natural seed and 42.1% for the ground seed were achieved. Therefore, annatto processing faces the challenge of waste treatment due to the high generation of exhausted seed. The bottleneck in the production of this dye is the high-water demand, both for the softening and rinsing stages of the fruit, achieving a total water usage of 13.5 L kg^{-1} and 10.5 L kg^{-1} for the natural and ground seed, respectively. The extraction of bixin and norbixin was quantified in each rinse (see Fig. 2), and it is possible to analyze different issues. (i) For the ground seed, three rinses were performed, while for the natural seed, there were four. This can be explained by the decrease in particle size increases the surface area, allowing greater contact between the seed and the friction media, increasing the extraction performance. (ii) Total depletion of the dyes was not achieved (null extraction) since the seed was rinsed until it was observed that the water did not turn reddish. (iii) A total yield (dye mass per seed mass) of bixin of 44.84 and 36.66 mg g^{-1} was achieved for ground and natural seeds, respectively (productivity increase of 22.3% for ground seed). (iv) A total norbixin yield of 2.02 mg g^{-1} for ground seed and 2.79 mg g^{-1} for whole seed was achieved. No significant difference was observed in the extraction of this dye. (v) As bixin and norbixin were detected in the extraction of the dye paste, mechanical friction is not selective to only one dye. Nevertheless, the small amount of norbixin (compared to bixin) makes the separation process technically unfeasible. (vi) A potential for bixin production was found since it has been established that contents higher than 2.7% in seeds are suitable for commercialization in international markets (Giridhar & Parimalan, 2010). Based on these results, dye extraction was better for the ground seed due to its high extraction yield and high bixin and norbixin content.

Figure 2 Bixin and norbixin extraction yields of the dye throughout seed rinsing. Error bars represent the standard deviation

Although dye extraction through mechanical friction is a clean and chemical-free methodology, it is not widely studied in the literature. Therefore, the results will be compared with other methodologies reported in the literature. The present work results were in agreement with the data reported by several authors. Barrozo et al. (2013) studied the mechanical extraction of bixin powder from annatto seeds using a spouted bed. The authors concluded that the best result was 23 g $kg^{-1}h^{-1}$ of powder with a bixin content of 44% (bixin yield of 10.12 mg g^{-1} seed). Shuhama et al. (2003) evaluated a new method for preparing annatto powders based on alkaline seed extraction and subsequent drying in a spouted bed with inert particle bodies, obtaining 3 mL min^{-1} of an extract with a bixin concentration close to 13% (1.3 mg g^{-1}). Other studies have emphasized the extraction of bixin and norbixin through solvents or alkaline agents, showing lower extraction yields than the present work. Pigment extraction yields (g pigment per g seed) of 2.55–4.11% using water, 3.02–4.83% using ethanol, and 4.22–6.04% using KOH (2%) as solvents have been reported, with norbixin contents of 0.06–0.15 mg g^{-1} and bixin contents of 6.5–31.5 mg g^{-1} of seed (Raddatz-Mota et al., 2016). Taham et al. (2015) evaluated bixin extraction through a fixed-bed extractor using supercritical CO_2, ethanol, and an ethanol-water mixture as solvents, finding that the best yields were, respectively, 0.9, 8.4, and 5.5 mg g^{-1} of seed.

3.3. Integration rate definition

Table 3 shows the algorithm criteria evaluated for the base raw material and for three proposed integration ratios (1:1:1, 1:0.5:0.5, 1:2:2, EAS:AS:ASL). These integration rates are based on information about raw materials in Chocó. In this sense, for the case of the 1:2:2 EAS:AS:ASL integration (which is the highest), it would consider the projected growth of raw materials in Chocó. Besides, it is expected that the raw material processing will be more standardized to manage better the supply of residues and their application in the production of value-added products. Therefore, Table 3 is made by inquiring about the criteria that will determine the decision-making in the algorithm. At the same time, it is necessary to define a base case (EAS for this study), to subsequently evaluate whether or not the proposed integrations improve the analyzed criteria.

Table 3
Criteria information for the integration proposed. Exhausted annatto seed is the base raw material

Criteria	**Base raw material (EAS)**		**1:1:1 (EAS:AS:ASL)**		**1:0.5:0.5 (EAS:AS:ASL)**		**1:2:2 (EAS:AS:ASL)**	
	Information	**Restrictions**	**Information**	**Restrictions**	**Information**	**Restrictions**	**Information**	**Restrictions**
Scale	Pilot plant scale	Artisanal process already established in the region	More than base scale	Açai: There is no standardized cultivation, so in this region it is considered a weed	Less than integration 1:1:1, but more than base scale	Açai: There is no standardized cultivation, so in this region it is considered a weed	More than integration 1:1:1 and base scale	Açai: There is no standardized cultivation, so in this region it is considered a weed
Composition	The composition of EAS is rich in cellulose which makes it suitable for use	-	The mixing of these residues generates a considerable increase in cellulose and lignin content	Lignin content for anaerobic digestion process	The mixing generates a low increase in cellulose and lignin content	Lignin content for anaerobic digestion process	The mixing generates a high increase in cellulose and lignin content	Lignin content for anaerobic digestion process
Phase and particle size	Solid-phase, reduction of the size required	0.5-4 mm	Solid-phase, reduction of the size required for the raw materials (homogenizer)	0.5-4 mm	Solid-phase, reduction of the size required for the raw materials (homogenizer)	0.5-4 mm	Solid-phase, reduction of the size required for the raw materials (homogenizer)	0.5-4 mm
Pretreatment stage	Dryer and miller	Energy needs	Dryer and miller	More energy needs than the base case	Dryer and miller	Less energy needs than integration 1:1:1	Dryer and miller	More energy needs than the base case and integration 1:1:1
Prices of raw materials and production of value-added products	Production of bioenergy products		More proportion in bioenergy products production than the base case, and more raw material cost		More proportion in bioenergy products production than a base case but less than integration 1:1:1, and more raw material cost		More proportion in bioenergy products production than the base case and integration 1:1:1, and more raw material cost	
Environmental impact	There is no use of other crop residues, but there is use of one of the processing residues (EAS)		Use of processing residues (EAS, AS, and ASL)		Use of processing residues (EAS, AS, and ASL)		Use of processing residues (EAS, AS, and ASL)	
Availability and fluctuation in price	There are no problems with the availability or accessibility of annatto. There are no seasons of significant price variations due to suitable production conditions		There are no problems of availability or accessibility of each raw material. There are no seasons of significant price variations due to suitable production conditions		Similar to integration 1:1:1. Less amount of açai generates less cost associated with its achievement		Similar to integration 1:1:1. More amount of açai generates an increase in cost associated with its achievement	

The first criterion refers to the scale. For both raw materials, the production correspond to low scale, even for the inegration of annatto (Ávila Ávila et al., 2017) and for açai (Isaza Aranguren et al., 2014). In the case of EAS, the restrictions to applying the process are related to the management of these residues since they come from an artisanal process of obtaining annatto dye. However, if this stage can be logistically and technically improved, the scale could be much more stable. Therefore, the proposed integrations are expected to improve the viability of the process and obtain value-added products. The restriction of the integration of açai residues is focused on the problems related to establishing crops (S. Y. Castro et al., 2015). This raw material in the study region has not been established as a producer. It is considered a weed, a native plant, and there are no specific delimitations for its crop.

The second criterion is related to composition. In this criterion, it is needed to know the composition of the raw materials individually. For this purpose, the results of the characterization are analyzed. This information shows whether the integration improves according to the products and platforms defined for the process. Finally, when the best integration ratio is selected, the composition of the mixture is experimentally confirmed.

The phase and particle size (third criterion) of the raw materials determine the pre-processing conditioning requirements. In this case, the raw materials are in a solid phase and are in a small particle size, which indicates that there is no need for considerable conditioning (Centro Nacional de Tecnología Agropecuaria y Forestal, 2010). However, homogenization at the time of blending must be adequately ensured. The fourth criterion for defining the best integration ratio is directly related to the previous one. If a pretreatment step is present, the energy requirements are increased, harming the process (Amin et al., 2017). However, in this case, a dryer and a mill are necessary to homogenize the integrations. Because the raw materials are intended to be integrated during processing, the pretreatment steps are not as significant since the raw materials are already in conditions that are close or at least easy to reach.

Table 3 Criteria information for the integration proposed. Exhausted annatto seed is the base raw material

The prices of raw materials and products (fifth criterion) are evaluated individually. Then it is shown whether integration improves or not, balancing with the value of the value-added products to be obtained. In this study, açai and annatto in Chocó have a price of 0.25 USD kg^{-1} and 2.9 USD kg^{-1}, respectively. The integrations will inevitably increase this value concerning the base raw material. However, biogas as a biorefinery product provides additional income to the stand-alone annatto (or açai) process. In this sense, the information placed in Table 3 for this criterion corresponds to the comparison of bioenergy products between the proposed integrations. The environmental impact (sixth criterion) is related to analyzing whether the raw material within its agronomic or processing component mitigates negative effects on the environment. The residues are considered only in the processing stage (colorant and pulp) for this study. In this sense, residues and their impact are only reflected in this stage. The agronomic part (residues such as peels, leaves, and branches) was not considered raw materials for the integrations.

Finally, the last criterion corresponds to raw material price fluctuation and its availability. In this regard, the acquisition of the residues proposed for the integrations depends on the processing of raw materials. This criterion is related to the cultivation and harvesting of açai and annatto. In the case of açai, it has been reported that it is possible to obtain this fruit throughout the year, but the representative harvest is mainly carried out twice a year. On the other hand, in the case of annatto, there is no problem in obtaining it since in the Chocó region, it is grown practically all year round (except in February and March, when it is in short supply). Therefore, price fluctuations for both raw materials are not very high. This information was evaluated for each proposed integration.

Considering the above analysis and the score defined for each criterion in the three proposed integration ratios, the integration ratio to be worked at the experimental level will be the 1:1:1 EAS:AS:ASL ratio. However, the experimental results using this integration ratio should be analyzed to identify or make adjustments. If this happens, a new integration ratio will be defined to be used in the simulations.

3.4. Anaerobic digestion

In anaerobic digestions, VS and TS content are important variables for calculating substrate and sludge loading and estimating biogas productivity. Table 4 shows the substrate characterization in terms of solids content. A high VS content implies higher biogas productivity. However, the chemical structure and the variability of the organic composition in the samples unbalance the quantification. It should also be noted that according to the initial conditions of the sample for analysis, the results can be discrepant due to the moisture composition. For example, it can be noted that the açai seed has a higher VS content compared to the other samples. Therefore, analyzing only the VS is not completely accurate for estimating maximum biogas productivities. The biodegradability index (BI) is a more reliable parameter for substrate consumption in anaerobic digestions. This index corresponds to the ratio of volatile solids to total solids (VS/TS). Thus, all the samples have similar values that induce good performances in the biogas tests since there are no significant differences between them.

Table 4
Solid composition of the anaerobic digestion substrates

Sample	VS (% wt.)	TS (% wt.)	Biodegradability index (%)
EAS-N	24.3 (0.53)	25.4 (0.37)	95.7
EAS-G	20.2 (0.25)	20.6 (0.27)	98.1
AS	90.3 (0.11)	94.5 (0.14)	95.5
ASL	29.1 (0.89)	29.4 (0.82)	98.9
EAS-G:AS:ASL	65.7 (0.59)	66.7 (0.65)	98.5
EAS-N:AS:ASL	63.9 (0.52)	66.2 (0.63)	96.5
EAS-N: natural annatto; AEAS-G: ground annatto; AS: açai seed; ASL: açai slurry; EAS-G:AS:ASL: mixture with ground annatto; EAS-N:AS:ASL: mixture with natural annatto.			
Values in brackets refer to standard deviation.			

Table 4 Solid composition of the anaerobic digestion substrates

Figure 3a shows the results of daily productivity in terms of biogas yields. It is possible to note biogas production from the first day, indicating no microorganism adaptation to the substrate (hydrolysis phase). It was also evidenced an increase in production rates for all samples during the first three days of digestion, being the annatto the most representative, with average values of 45.1 and 34.3 mL $gVS^{-1}d^{-1}$ for ground and natural seeds, respectively. However, after the third day, a decrease in the productivity rate was observed, which can be explained by the depletion of soluble and low molecular weight compounds (such as free sugars) that are easily assimilated by the microorganism. Moreover, from the fifth day onwards, the productivities are relatively constant (between 5–35 mL of biogas per day) throughout the digestion time for the açai and mixed residue samples. This may indicate a constant consumption of more complex molecules or biopolymers such as cellulose and hemicellulose. In contrast, an increase in productivity is shown from the ninth day for the annatto samples. This variation can be explained by consuming high molecular weight carbohydrates and other major compounds in the annatto seed, such as lipids, pectin, and protein.

Figure 3 Biogas productivity performance. (a) Biogas yield; (b) methane composition; and (c) H_2S concentration. Error bars represent the standard deviation. The legend is the same for all Figures

As shown in Fig. 3a, there is variability in biogas production. Different factors can explain these differences. (i) No pretreatment of the raw material was performed to improve accessibility to biodegradation. Pretreatments decrease the crystallinity and recalcitrance and increase the surface area and porosity of the sample, leading to a higher biodegradability rate (Angelidaki & Ahring, 2000). The only treatment performed on some samples was particle size reduction. However, this treatment does not influence biogas production since annatto seeds have similar productivities (difference of 9.46%), as shown in Table 5. (ii) Partial inhibitions in the açai samples may prevent effective digestion performance. During the initial stage of anaerobic digestion, the accumulation of inhibitory volatile fatty acids (VFA), such as propionic, acetic, and butyric acids (produced during hydrolysis and acidogenesis), is promoted, leading to a decrease in the pH of the system and hindering the further growth of methanogenic bacteria (Wainaina et al., 2019). Simple and soluble sugars, hemicellulose oligosaccharides, and pectin are rapidly hydrolyzed and converted into methanogenic intermediates (mainly VFA) that partially inhibit biogas productivity (Chanakya et al., 1995). (iii) Ammonia can also affect the inhibition of anaerobic digestions if it exceeds a specific concentration of 0.1-2 g-N/L (Parawira et al., 2004). It has been reported that at concentrations above the threshold level of 1.7 g-N L^{-1}, ammonia nitrogen has a stronger effect on acetoclastic methanogenic bacteria than on hydrogenic methanogenic bacteria (Koster & Lettinga, 1984). During anaerobic digestion, the balance between the production of acetoclastic methanogenic bacteria and hydrogenic bacteria is crucial. The latter help to preserve a low hydrogen concentration to ensure a favorable thermodynamic conversion of propionic acid into hydrogen and acetic acid (methanogenic bacteria substrates) (Fujishima et al., 2000).

Table 5
Summary of anaerobic digestion results of annatto, açai, and the substrates integration

Sample	Biogas yield (mL gVS^{-1})	Biomethane yield (mL gVS^{-1})	Maximum production rate (mL d^{-1})	Average concentration	
				CH_4 (% vol.)	H_2S (ppm)
EAS-N	934.7	533.2	22.0	53.7	291.1
EAS-G	838.8	466.3	24.7	51.5	187.9
AS	219.3	129.8	8.0	59.7	44.9
ASL	213.6	144.2	6.3	66.7	43.1
EAS-N:AS:ASL	272.9	171.7	8.7	63.1	55.1
EAS-G:AS:ASL	307.6	186.8	8.8	60.6	46.9

Table 5 Summary of anaerobic digestion results of annatto, açai, and the substrates integration

Table 5 shows the results of final cumulative yields and average concentrations of biogas. It was found that the exhausted annatto seeds show the highest production yields for both biogas and biomethane. Likewise, the annatto has the largest average H_2S concentrations, decreasing its quality as a biofuel. H_2S is a pollutant gas that harms pipes and processing equipment since it promotes fouling formation, reducing their diameter and finally clogging them after a certain operating period. Furthermore, in combustion processes, the formation of H_2SO_4 that corrodes accessories and components of the biogas internal combustion engine has been reported (Mamun & Torii, 2015). Nevertheless, all samples (Fig. 3c) have concentrations below the limit (< 1000 ppm) for thermochemical biogas application technologies such as Gas Heating Boilers and Combined Heat and Power (Choudhury et al., 2019). In contrast to the annatto, the ASL has the highest average CH_4 concentration, reaching the maximum concentration of 76.5% (vol.) on day 19 (Fig. 3b). Regarding the AS sample, a cumulative biogas and biomethane yield of 219.3 and 129.8 mL gVS^{-1}, respectively, and a maximum CH_4 concentration of 66.4% (day 26) were obtained. These results are in agreement with some studies reported in the literature. Sganzerla et al. (2021) evaluated the biogas production from açai seed (derived from a food industry) in a 6.8 L biodigester at mesophilic conditions. The authors obtained a final methane yield of 158.8 mL gVS^{-1} at an average composition of 56.5% (vol.) during the first half of the digestion. In addition, Maciel-Silva et al. (2019) studied mesophilic biogas production from seeds and wastewater from açai processing following pretreatment with subcritical water. The authors obtained a biogas yield of 791.8 mL gVS^{-1} when the pretreatment was performed at 200°C and 7.8 mL gVS^{-1} in the absence of pretreatment (control assay) with a fed with 25% açai seed and 45% wastewater. It is noteworthy that the pretreatment with subcritical hydrolysis is efficient since it increases the yield by more than 100 times.

3.4.1. Substrate integration

The results of the proposed integrations showed a maximum production rate of 8.7 mL d^{-1} for EAS-N:AS:ASL and 8.8 mL d^{-1} for EAS-G:AS:AS:ASL. In this sense, both performances were lower than the exhausted annatto seed-only assays. It demonstrates that feedstock integration does not always guarantee improvements in biogas yields. It is explained by the chemical composition of the mixture, which is more complex for the metabolism of anaerobic microorganisms. Therefore, a pretreatment stage should be considered to facilitate access to platform molecules such as sugars. Another possible explanation for this decrease in productivity may be the variation of the C/N ratio. The indicator of the carbon and nitrogen content of the substrate can interact with the medium, which leads to different ammonia concentrations and inhibitory effects (Wang et al., 2014). It has been demonstrated that the mixing of substrates through co-digestion favors the C/N ratio (Shahbaz et al., 2020). However, fruit mixtures are disadvantaged due to the high protein content (Jesus et al., 2018), especially when dealing with açai slurry with considerable pulp, which finally alters the optimum C/N ratio (25–30). Analyzing these results and as mentioned in S**ection 3.2**, adjusting the initial definition of the integration rate is necessary. In global terms, it is evident that the best performance in biogas production was annatto residues. Even though the biogas quality is the best for the evaluated cases. Therefore, it would be expected that the new integration ratio should have a majority mass representation of annatto compared to açai residues. Hence, the 1:0.5:0.5 EAS-N:AS:ASL is proposed as the new integration ratio used in simulation scenarios.

3.4.2. Kinetic model

Online Resource 3 shows the mathematical fit of the Gompertz model for cumulative methane productivity of annatto samples. it was possible to observe a model deviation for the first 15 days of digestion, even with adaptation time results, as shown in Table 6. On the other hand, differences between the experimental and predicted values of 4.8% and 3.4% were obtained for the EAS-N and EAS-G, respectively, for methane production potential (A). Moreover, a difference of less than 5% was obtained between the maximum production rates of the model and the experimental results for EAS-N (24.1 mL $gVS^{-1}d^{-1}$) and EAS-G (23.1 mL gVS^{-1}d-1). Concerning the açai residues (**Online Resource 4**), both samples showed productivity from the beginning of the digestion; however, the AS was the sample with the most moderate adjustment (R^2 of 0.978). In addition, there was a difference (between experimental and model results) in the methane production potential of 7.9% for the AS and 0.2% for the ASL, respectively. For the maximum methane productivities, similarities were obtained for the AS substrate and less than 8% errors for the ASL. With these results, the model shows that there can be small variations regarding the experimental results, being the initial digestion stage the most relevant to variation.

Table 6
Parameter results of Gompertz model for anaerobic digestion

Parameter	Sample			
	EAS-N	EAS-G	AS	ASL
R^2	0.988	0.989	0.978	0.997
A (mL gVS^{-1})	626.35	520.62	334.25	154.63
μ_{max} (mL $gVS^{-1}d^{-1}$)	25.19	23.41	5.62	6.65
λ (day)	4.00	3.80	8.09	1.43

Table 6 Parameter results of Gompertz model for anaerobic digestion

3.5. Simulations analysis

3.5.1. Techno-energetic results

The techno-energetic results were determined based on mass and energy balances. Table 7 shows the results of the indicators for the proposed scenarios. In terms of Y_P index, it is possible to evidence that the yields in Sc1, Sc2, Sc3, and Sc4 are maintained for annatto seed colorant and açai pulp under the same processing scale. However, if this index is analyzed globally, the values indicate that Sc5 presents the highest raw materials yield per kg. It means that with the integration of annatto and açai, more fractions of the raw materials are used to obtain value-added products. Sc5 also presented a higher value than the other scenarios for the PMI and MLI indicators since the product determines both flows obtained in the process. Finally, in terms of the RM_I mass indicator, Sc1 presents the highest value for this indicator, while Sc5 presents the lowest value. It is explained by the amount of inputs required in the process. Sc1 requires only one input (water), while Sc5 requires three (water, NaClO, and sludge).

Table 7
Mass and energy indicators for scenarios proposed

Mass indicators				
Scenario	**Product yield (Y_P)**	**Process mass intensity index (PMI)**	**Mass loss index (MLI)**	**Renewability material index (RM_I)**
Sc1	43.48 kg dye 100kg^{-1} annatto	19.44 kg raw materials kg^{-1} dye	18.44 kg waste streams kg^{-1} dye	0.118 kg renewable feedstock kg^{-1} raw materials
Sc2	52.99 kg pulp 100kg^{-1} açai	5.86 kg raw materials kg^{-1} açai	4.86 kg waste streams kg^{-1} pulp	0.322 kg renewable feedstock kg^{-1} raw materials
Sc3	43.48 kg dye 100kg^{-1} annatto	57.20 kg raw materials kg^{-1} products	56.41 kg waste streams kg^{-1} dye	0.032 kg renewable feedstock kg^{-1} raw materials
	11.61 kg biogas 100kg^{-1} annatto		56.99 kg waste streams kg^{-1} biogas	
Sc4	52.99 kg pulp 100kg^{-1} açai	41.03 kg raw materials kg^{-1} products	40.34 kg waste streams kg^{-1} pulp	0.032 kg renewable feedstock kg^{-1} raw materials
	22.97 kg biogas 100kg^{-1} açai		40.73 kg waste streams kg^{-1} biogas	
Sc5	37.70 kg dye 100kg^{-1} raw materials	64.43 kg raw materials kg^{-1} products	63.82 kg waste streams kg^{-1} dye	0.026 kg renewable feedstock kg^{-1} raw materials
	9.48 kg pulp 100kg^{-1} raw materials		64.27 kg waste streams kg^{-1} pulp	
	13.64 kg biogas 100kg^{-1} raw materials		64.20 kg waste streams kg^{-1} biogas	
Energy indicators				
Scenario	**Overall energy efficiency (η)**	**Specific energy consumption (S_{EC})**	**Resource energy efficiency (η_E)**	**Self-generation index (SGI)**
Sc1	0.36	1.11 MJ kg^{-1}	0.43	0.67
Sc2	0.46	0.95 MJ kg^{-1}	0.53	1.06
Sc3	0.73	1.69 MJ kg^{-1}	0.47	0.95
Sc4	0.69	6.78 MJ kg^{-1}	0.58	0.40
Sc5	0.60	3.82 MJ kg^{-1}	0.97	0.47

Table 7 Mass and energy indicators for scenarios proposed

On the other hand, energy indicators make it possible to evaluate the energy efficiency of the process and energy consumption and sufficiency. Sc3 presented the highest η indicator compared to the other scenarios. It indicates that it has a much higher overall efficiency, mainly due to the equipment used in the process. Compared to Sc1, this indicator also shows that adding a processing line (biogas production for Sc3) can energetically benefit the performance of the process. Regarding the SEC indicator, Sc4 shows 6.78 MJ kg^{-1}, the highest value among all the proposed scenarios. Furthermore, the integration of raw materials (Sc5) shows by employing the η_E indicator that the transformation of raw materials into products maintains 97% of the energy. Finally, the SGI indicator evaluates the capacity of the process to supply partially or totally the energy of the process. In the case of Sc2 (with a value greater than 1.0), the process energy is not only fully supplied, but there is a surplus that can be used in part of the value chain of the products. However, Sc4 and Sc5 supply less than 50% of the process energy.

3.5.2. Economic assessment

The economic analysis was performed considering parameters such as capital expenditure (CapEx), operational expenditure (OpEx), incomes, net present value (NPV), among others, as shown in Table 8. Concerning the CapEx, the raw material integration scenario (Sc5) has the highest cost, which was expected due to both processing units of açai and annatto. This capital investment involved the cost of equipment (144.2 k-USD), instrumentation (198.5 k-USD), installation civil work (38.5 k-USD), piping (22.9 k-USD), and electrical (5.3 k-USD). On the other hand, it can be observed that although the CapEx of the stand-alone pulp production (Sc2) is slightly higher than the annatto dye paste (Sc1), the addition of an anaerobic digestion stage leads to a higher CapEx for the dye production with biogas (Sc3) compared to the processing of açai with biogas (Sc4). This can be explained by the fact that the digester has a larger processing volume leading to an increase in the direct equipment, accessories, and installation cost. Therefore, from the CapEx comparison, Sc1 is the most economical process. Regarding the revenue indicator, the scenarios of annatto dye paste production (Sc1 and Sc3) are the most relevant due to the high selling price in the market (> 20 USD kg^{-1}). Indeed, it can be observed in Table 8 that the biogas sale does not influence the total revenue, contributing 148.4 k-USD $year^{-1}$ (equivalent to 16.9 USD h^{-1}). Likewise, higher gross income is observed for the annatto scenarios. Although the açai processing (Sc2 and Sc4) have the lowest gross income, they were the scenarios that best buffered the production costs (see Table 9), reducing revenues (comparison between revenues and gross incomes) by 14–17% compared to Sc1, Sc2, and Sc5, which were 33–39%.

Table 8
Economic results summary of the proposed scenarios

Parameter	Scenario				
	Sc1	Sc2	Sc3	Sc4	Sc5
CapEx (k-USD)	358.1	366.9	415.4	408.2	559.1
OpEx (M-USD $year^{-1}$)	11.7	1.3	11.9	1.7	9.9
Raw material (M-USD $year^{-1}$)	10.6	0.9	10.7	1.0	8.8
Utilities (M-USD $year^{-1}$)	1.8	0.1	0.9	0.5	0.9
Depreciation (M-USD $year^{-1}$)	45.8	46.9	53.1	52.2	71.5
Revenue (M-USD $year^{-1}$)	32.2	10.3	32.4	10.6	26.4
Gross income (M-USD $year^{-1}$)	19.6	8.9	20.4	8.8	17.7
NPV in 20 years (M-USD $year^{-1}$)*	164.3	75.5	171.5	74.2	137.9
MPSEF* (kg d^{-1})**	73	148	84.2	161.9	88.8

*Net present value at the base flow rate of 10 ton d^{-1}.

**Minimum processing scale for economic feasibility.

Table 9
Production cost of dye paste, pulp, and biogas for the different scenarios

Scenario	Production cost (USD kg^{-1})			Profit margin (%)
	Dye paste	Pulp	Biogas*	
Sc1	7.95	-	-	60.8
Sc2	-	0.68	-	87.3
Sc3	7.47	-	0.13 (0.16)	63.2
Sc4	-	0.90	0.06 (0.07)	83.2
Sc5	7.68	2.02	0.13 (0.17)	62.1

*Values in brackets refer to the production cost in USD m^{-3}

Table 8 Economic results summary of the proposed scenarios

Table 9 Production cost of dye paste, pulp, and biogas for the different scenarios

Concerning the OpEx, Table 9 shows that the annatto processing scenarios are the most expensive, almost ten times more expensive than the processing of açai. This difference in variable costs is due to the high cost of raw materials (see Fig. 4), being 10.6, 10.7, and 8.8 M-USD $year^{-1}$ for Sc1, Sc3, and Sc5, respectively. For these scenarios, the annatto cost had the greatest influence on total raw material, representing more than 98%. The annatto seed cost was calculated based on the production cost per kg of seed in the agronomic crop stage in Chocó (Colombia), considering variables such as fertilizers, substrates, nursery construction, machinery & accessories for germination, land adaptation and harvesting, labor, transport, among others. This cost was in agreement with the annatto seeds sold by agronomists in the region. Furthermore, the raw material cost had the greatest impact on the OpEx in the açai processing scenarios, being 1.3 M-USD $year^{-1}$ for Sc2 and 1.7 M-USD $year^{-1}$ for Sc4. In terms of utilities, it was observed that they had a minor impact on the variable costs being within 7–10% of the OpEx for Sc1, Sc2, Sc3, and Sc5. In contrast, Sc4 utilities contributed 29.1%, where the low-pressure steam, cooling water, and electricity were, respectively, 60.8, 55.4, and 402.7 k-USD $year^{-1}$. The remaining OpEx (e.g., maintenance, administrative, plant overhead, laboratory charges, among others) had a low influence on total costs, ranging between 2% and 18%. Therefore, from an OpEx perspective, the açai processing scenarios are the least expensive.

Figure 4 Total distribution cost of (a) Sc1, (b) Sc2, (c) Sc3, (d) Sc4, and (e) Sc5. The legend is the same for all Figures

Table 9 shows that positive profits (> 60%) were obtained for all the proposed scenarios since the production cost is lower than the selling cost. Likewise, a positive cash flow balance was obtained over 20 years of economic analysis (see Table 8), showing that the accumulated net present value (NPV) was between USD 74 and USD 171 million per year. Therefore, economic viability is demonstrated for all the proposed scenarios. Based on the annatto processing perspective, the cumulative NPV for 20 years would be 193.7 and 500.9 M-USD $year^{-1}$ assuming the Chocó regional production (11.3 ton d^{-1}), and 202.9 and 524.5 M-USD $year^{-1}$ when working with the Colombian national production (11.8 ton d^{-1}) for Sc1 and Sc3, respectively. Meanwhile, working with 66.4-ton d^{-1} of the national production of açai would provide a NPV of 514.1 M-USD $year^{-1}$ for Sc2 and 516.3 M-USD $year^{-1}$ for Sc4. Therefore, it was proposed to perform a sensitivity analysis based on the raw material scale to find the minimum point for economic viability, a term known as minimum processing scale for economic viability (MPSEF), as shown in Fig. 5. It was found that the annatto scenarios (Sc1 and Sc3) and integration (Sc5) demonstrate the lowest processing flows (see table). In contrast, obtaining açai pulp coupled with biogas production (Sc4) requires the largest input flow (161.9 kg d^{-1}). Nevertheless, all MPSEFs are below the regional and national production of both annatto and açai.

Figure 5 NPV at the minimum processing scale for economic feasibility

Conclusions

The technical and economic feasibility of small-scale biorefineries was demonstrated through experimental and simulation data. The combination of current annatto and açai processing with anaerobic digestion schemes shows to be a promising alternative for exotic and wild crop regions. The high extraction yields of annatto paste with a rich bixin content promote the profitability of the processes due to its high selling price. A high integration ratio of the residues does not always improve the yield or the quality of the biogas. Therefore, it is necessary to analyze beyond the composition and operating factors such as the C/N ratio. However, it was possible to obtain biogas with a high biomethane content and low H_2S concentrations for future use in household or industrial networks. From the simulation perspective, the feedstock integration scheme (Sc5) showed that the energy contained in the feedstock was almost fully used since 3% was not retained in the products, unlike the other scenarios. This suggests that biorefinery integration schemes are more energy efficient. Regarding the economic analysis, although the production of dye paste with biogas showed the best economic profitability, all scenarios are economically favorable. Even smaller processing scales than the national production were achieved for economic profitability.

Declarations

Authors contribution

Jhonny Alejandro Poveda-Giraldo: Conceptualization, Methodology, Software, Validation, Investigation, Data Curation, Writing - Original Draft. **Sara Piedrahita-Rodriguez:** Conceptualization, Methodology, Software, Validation, Investigation, Data Curation, Writing - Original Draft. **Natalia Salgado Aristizabal:** Conceptualization, Methodology, Visualization. **Haminton Salas Moreno:** Resources, Visualization. **Carlos Ariel Cardona Alzate:** Conceptualization, Formal Analysis, Resources, Project administration, Funding acquisition, Writing - Review & Editing.

Data Availability Statement

Data supporting the results of this study are available upon request from the corresponding author.

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Declaration of interest

The authors declare that they do not have a conflict of interest.

Financial interests

The authors declare they have no financial interests

References

1. Abbaszadeh, A. H. (2008). *Pectin and galacturonic acid from citrus wastes* (Issue 7). Master Thesis. University of Borås.
2. Albarelli, J. Q., Santos, D. T., Cocero, M. J., & Meireles, M. A. A. (2016). Economic analysis of an integrated annatto seeds-sugarcane biorefinery using supercritical CO2 extraction as a first step. Materials, *9*(6). https://doi.org/10.3390/ma9060494
3. Alonso-Gómez, L. A., Solarte-Toro, J. C., Bello-Pérez, L. A., & Cardona-Alzate, C. A. (2020). Performance evaluation and economic analysis of the bioethanol and flour production using rejected unripe plantain fruits (Musa paradisiaca L.) as raw material. Food and Bioproducts Processing, *121*, 29–42. https://doi.org/10.1016/J.FBP.2020.01.005
4. Alves, J. L. F., da Silva, J. C. G., di Domenico, M., Galdino, W. V. D. A., Andersen, S. L. F., Alves, R. F., & de Sena, R. F. (2021). Exploring Açaí Seed (Euterpe oleracea) Pyrolysis Using Multi-component Kinetics and Thermodynamics Assessment Towards Its Bioenergy Potential. Bioenergy

Research, *14*(1), 209–225. https://doi.org/10.1007/s12155-020-10175-y

5. Amin, F. R., Khalid, H., Zhang, H., Rahman, S., Zhang, R., Liu, G., & Chen, C. (2017). Pretreatment methods of lignocellulosic biomass for anaerobic digestion. AMB Express, *7*(1). https://doi.org/10.1186/s13568-017-0375-4
6. Angelidaki, I., & Ahring, B. K. (2000). Methods for increasing the biogas potential from the recalcitrant organic matter contained in manure. Water Science and Technology, *41*(3), 189–194. https://doi.org/10.2166/wst.2000.0071
7. Atienza, A. H., Cataluña, L., Combis, A., & Cosino, V. A. (2021). Solar dryer with reheating system for Annatto fruit pericarp. *IOP Conference Series: Earth and Environmental Science*, *765*(1). https://doi.org/10.1088/1755-1315/765/1/012066
8. Ávila Ávila, M. I., Oñarte Arboleda, J., & Valencia Rentería, C. N. (2017). *Estudio de Factibilidad de creación de una Empresa Productora y Comercializadora de Tintes Orgánicos a base de Achiote*. Universidad Católica de Colombia.
9. Barrozo, M. A. S., Santos, K. G., & Cunha, F. G. (2013). Mechanical extraction of natural dye extract from Bixa orellana seeds in spouted bed. Industrial Crops and Products, *45*, 279–282. https://doi.org/10.1016/j.indcrop.2012.12.052
10. Buratto, R. T., Cocero, M. J., & Martín, Á. (2021). Characterization of industrial açaí pulp residues and valorization by microwave-assisted extraction. *Chemical Engineering and Processing - Process Intensification*, *160*. https://doi.org/10.1016/j.cep.2020.108269
11. C. Alcázar-Alay, S., P. Cardenas-Toro, F., T. Santos, D., & A. Meireles, M. A. (2015). Study of an Extraction Process as the Pretreatment Step for Sugar Production from Acid Hydrolysis. Food and Public Health, *5*(2), 47–55. https://doi.org/10.5923/j.fph.20150502.03
12. Castro, S. Y., Barrera, J. A., Carrillo Bautista, M., & Hernández Gómez, M. S. (2015). *Asaí (Euterpe precatoria) Cadena de valor en el sur de la región amazónica* (First Ed.). Instituto Amazónico de Investigaciones Científicas- Sinchi.
13. Centro Nacional de Tecnología Agropecuaria y Forestal. (2010). *El cultivo del achiote, Bixa orellana L. Manual técnico* (pp. 1–88).
14. Chanakya, H. N., Ganguli, N. K., Anand, V., & Jagadish, K. S. (1995). Performance characteristics of a solid-phase biogas fermentor. Energy for Sustainable Development, *1*(6), 43–46. https://doi.org/10.1016/S0973-0826(08)60100-3
15. Chisté, R. C., Yamashita, F., Gozzo, F. C., & Mercadante, A. Z. (2011). Simultaneous extraction and analysis by high performance liquid chromatography coupled to diode array and mass spectrometric detectors of bixin and phenolic compounds from annatto seeds. Journal of Chromatography A, *1218*(1), 57–63. https://doi.org/10.1016/j.chroma.2010.10.094
16. Choudhury, A., Shelford, T., Felton, G., Gooch, C., & Lansing, S. (2019). Evaluation of hydrogen sulfide scrubbing systems for anaerobic digesters on two U.S. Dairy farms. Energies, *12*(24). https://doi.org/10.3390/en12244605
17. da S. dos Santos, D., Klauck, V., Campigotto, G., Alba, D. F., dos Reis, J. H., Gebert, R. R., Souza, C. F., Baldissera, M. D., Schogor, A. L. B., Santos, I. D., Wagner, R., Vedovatto, M., & da Silva, A. S. (2019). Benefits of the inclusion of açai oil in the diet of dairy sheep in heat stress on health and milk production and quality. Journal of Thermal Biology, *84*, 250–258. https://doi.org/10.1016/j.jtherbio.2019.07.007
18. Dantas, A. M., Batista, J. D. F., dos Santos Lima, M., Fernandes, F. A. N., Rodrigues, S., Magnani, M., & Borges, G. da S. C. (2021). Effect of cold plasma on açai pulp: Enzymatic activity, color and bioaccessibility of phenolic compounds. *LWT*, *149*. https://doi.org/10.1016/j.lwt.2021.111883
19. De Castro, M. D. L., & Priego-Capote, F. (2010). Soxhlet extraction: Past and present panacea. Journal of Chromatography A, *1217*(16), 2383–2389.
20. de Oliveira, M. do S. P., & Schwartz, G. (2018). Açaí— Euterpe oleracea. In *Exotic Fruits* (pp. 1–5). Elsevier. https://doi.org/10.1016/b978-0-12-803138-4.00002-2
21. Feng, F., Liu, W. Y., Chen, Y. S., Guo, Q. L., & You, Q. D. (2005). Study on derivatives of gambogic acid. Journal of China Pharmaceutical University, *36*(4), 302–305.
22. FruitLogistica. (2021). *European statistics handbook - FruitLogistica 2021*. www.AMI-informiert.de
23. Fujishima, S., Miyahara, T., & Noike, T. (2000). Effect of moisture content on anaerobic digestion of dewatered sludge: Ammonia inhibition to carbohydrate removal and methane production. Water Science and Technology, *41*(3), 119–127. https://doi.org/10.2166/wst.2000.0063
24. Garcia-Amezquita, L. E., Tejada-Ortigoza, V., Serna-Saldivar, S. O., & Welti-Chanes, J. (2018). Dietary fiber concentrates from fruit and vegetable by-products: Processing, modification, and application as functional ingredients. Food and Bioprocess Technology, *11*(8), 1439–1463. https://doi.org/10.1007/s11947-018-2117-2
25. Giridhar, P., & Parimalan, R. (2010). A biotechnological perspective towards improvement of annatto color production for value addition – the influence of biotic elicitors. Asia-Pacific Journal of Molecular Biology and Biotechnology, *18*(1), 77–79. https://www.researchgate.net/publication/268393606
26. Hall, M. B. (2003). Challenges with nonfiber carbohydrate methods. Journal of Animal Science, *81*(12), 3226–3232. https://doi.org/10.2527/2003.81123226x
27. Hames, B., Scarlata, C., & Sluiter, A. (2008). Determination of Protein Content in Biomass - NREL/TP-510-42625. In *National Renewable Energy Laboratory*.
28. Isaza Aranguren, C., Galeano, G., & Bernal, R. (2014). Management of asaí (Euterpe precatoria Mart.) for fruit production in southern Colombian Amazonia. Colombia Forestal, *17*(1), 77–99.
29. Itai, Y., Santos, R., Branquinho, M., Malico, I., Ghesti, G. F., & Brasil, A. M. (2014). Numerical and experimental assessment of a downdraft gasifier for electric power in Amazon using açaí seed (Euterpe oleracea Mart.) as a fuel. Renewable Energy, *66*, 662–669. https://doi.org/10.1016/j.renene.2014.01.007

30. Jesus, A. L. T. de, Leite, T. S., & Cristianini, M. (2018). High isostatic pressure and thermal processing of açaí fruit (Euterpe oleracea Martius): Effect on pulp color and inactivation of peroxidase and polyphenol oxidase. Food Research International, *105*, 853–862. https://doi.org/10.1016/j.foodres.2017.12.013
31. Koster, I. W., & Lettinga, G. (1984). The influence of ammonium-nitrogen on the specific activity of pelletized methanogenic sludge. Agricultural Wastes, *9*(3), 205–216. https://doi.org/10.1016/0141-4607(84)90080-5
32. Kumar, P. S., Reddy, Y. R., Ramesh, S., Gobinath, S., & Ramana, D. B. V. (2007). Evaluation of Pigment Extracted Annatto Seed (Bixa Orellana) By Chemical, in-Vitro and in-Sacco Techniques in Buffaloes. Buffalo Bulletin, *26*(1), 5–9.
33. L.C. Albuquerque, C., & A.A. Meireles, M. (2012). Trends in Annatto Agroindustry: Bixin Processing Technologies and Market. Recent Patents on Engineering, *5*(2), 94–102. https://doi.org/10.2174/187221211796320738
34. Lee, J., Durst, R. W., & Wrolstad, R. E. (2001). Determination of Total Monomeric Anthocyanin Pigment Content of Fruit Juices, Beverages, Natural Colorants, and Wines by the pH Differential Method: Collaborative Study. Journal of OAC International, *88*(5), 1269–1278. https://doi.org/10.1093/jaoac/88.5.1269
35. Lee, S., Prosky, L., & de Vries, J. (1992). Determination of Total, Soluble, and Insoluble Dietary Fiber in Foods-Enzymatic-Gravimetric Method, MES-TRIS Buffer: Collaborative Study. Journal of AOAC Iinternational, *75*(3), 395–416. https://doi.org/10.1093/jaoac/75.3.395
36. Li, D., Li, B., Ma, Y., Sun, X., Lin, Y., & Meng, X. (2017). Polyphenols, anthocyanins, and flavonoids contents and the antioxidant capacity of various cultivars of highbush and half-high blueberries. Journal of Food Composition and Analysis, *62*, 84–93. https://doi.org/10.1016/J.JFCA.2017.03.006
37. Lucas, B. F., Zambiazi, R. C., & Costa, J. A. V. (2018). Biocompounds and physical properties of açaí pulp dried by different methods. LWT, *98*, 335–340. https://doi.org/10.1016/j.lwt.2018.08.058
38. Machrafi, H. (2012). Green Energy and Technology. In *Green Energy and Technology*. https://doi.org/10.2174/97816080528511120101
39. Maciel-Silva, F. W., Mussatto, S. I., & Forster-Carneiro, T. (2019). Integration of subcritical water pretreatment and anaerobic digestion technologies for valorization of açai processing industries residues. Journal of Cleaner Production, *228*, 1131–1142. https://doi.org/10.1016/j.jclepro.2019.04.362
40. Mamun, M. R. al, & Torii, S. (2015). Removal of Hydrogen Sulfide (H2S) from Biogas Using Zero-Valent Iron. Journal of Clean Energy Technologies, *3*(6), 428–432. https://doi.org/10.7763/jocet.2015.v3.236
41. Marinova, G., & Batchvarov, V. (2011). Evaluation of the methods for determination of the free radical scavenging activity by DPPH. Bulgarian J Agric Sci, *17*(1), 11–24.
42. Matta, F. v., Xiong, J., Lila, M. A., Ward, N. I., Felipe-Sotelo, M., & Esposito, D. (2020). Chemical composition and bioactive properties of commercial and non-commercial purple and white açaí berries. Foods, *9*(10). https://doi.org/10.3390/foods9101481
43. Moncada B, J., Aristizábal M, V., & Cardona A, C. A. (2016). Design strategies for sustainable biorefineries. Biochemical Engineering Journal, *116*, 122–134. https://doi.org/10.1016/j.bej.2016.06.009
44. Moncada, J., Tamayo, J. A., & Cardona, C. A. (2014). Integrating first, second, and third generation biorefineries: Incorporating microalgae into the sugarcane biorefinery. Chemical Engineering Science, *118*, 126–140. https://doi.org/10.1016/j.ces.2014.07.035
45. Noblick, L. (2019). *Guide to the Palms of Northeastern Brazil* (First ed., Vol. 1). UEFS Editora. https://www.researchgate.net/publication/336364606
46. Noppe, H., Abuín Martinez, S., Verheyden, K., Van Loco, J., Companyó Beltran, R., & De Brabander, H. F. (2009). Determination of bixin and norbixin in meat using liquid chromatography and photodiode array detection. *Food Additives and Contaminants - Part A Chemistry, Analysis, Control*, Exposure and Risk Assessment, *26*(1), 17–24. https://doi.org/10.1080/02652030802322564
47. Okorie, N., Fasogbon, S., Mohammed, U., Abubakar, U., Avwioro, O., Tsamiya, R., Mohammed, M., & Tangaza, A. (2020). Study of phytochemical compositions and separation of colours using thin layer column chromatography for Bixa orellana (annatto) seed extracts. Yenagoa Medi Cal Journal, *2*(1), 43–49. https://www.researchgate.net/publication/338224504
48. Oliveira, A. R., Ribeiro, A. E. C., Oliveira, É. R., Garcia, M. C., Soares Júnior, M. S., & Caliari, M. (2020). Structural and physicochemical properties of freeze-dried açaí pulp (Euterpe oleracea mart.). Food Science and Technology (Brazil), *40*(2), 282–289. https://doi.org/10.1590/fst.34818
49. Oliveira, N. L., Silva, S. H., Figueiredo, J. de A., Norcino, L. B., & Resende, J. V. de. (2021). Infrared-assisted freeze-drying (IRFD) of açai puree: Effects on the drying kinetics, microstructure and bioactive compounds. Innovative Food Science & Emerging Technologies, *74*, 102843. https://doi.org/10.1016/J.IFSET.2021.102843
50. Pagès, P. B., Le Pimpec-Barthes, F., & Bernard, A. (2016). Chirurgie des métastases pulmonaires des cancers colorectaux: facteurs prédictifs de survie. Revue Des Maladies Respiratoires, *33*(10), 838–852. https://doi.org/10.1016/j.rmr.2016.02.006
51. Parawira, W., Murto, M., Zvauya, R., & Mattiasson, B. (2004). Anaerobic batch digestion of solid potato waste alone and in combination with sugar beet leaves. Renewable Energy, *29*(11), 1811–1823. https://doi.org/10.1016/j.renene.2004.02.005
52. Peters, M., Timmerhaus, K., & West, R. (2003). *Plant Design and Economics for Chemical Engineers* (Fifth ed.).
53. Piedrahita-Rodriguez, S. (2020). *Design of biorefineries with multiple raw materials for the use of agroindustrial waste in post-conflict zones in Colombia - Master thesis*. Universidad Nacional de Colombia sede Manizales.
54. Piedrahita-Rodríguez, S., & Cardona Alzate, C. A. (2021). Small-scale biorefineries based on plantain and avocado residues. In *Waste Biorefinery* (pp. 349–374). Elsevier. https://doi.org/10.1016/B978-0-12-821879-2.00012-0
55. Poveda-Giraldo, J. A., & Cardona Alzate, C. A. (2021). A biorefinery for the valorization of marigold (Calendula officinalis) residues to produce biogas and phenolic compounds. Food and Bioproducts Processing, *125*, 91–104. https://doi.org/10.1016/j.fbp.2020.10.015

56. Pyrgakis, K. A., & Kokossis, A. C. (2018). An Engineering Tool to Screen and Integrate Biomass Valorization Paths in Multiple-Feedstock Biorefineries. In *Computer Aided Chemical Engineering* (Vol. 43, pp. 573–578). Elsevier Masson SAS. https://doi.org/10.1016/B978-0-444-64235-6.50103-0
57. Raddatz-Mota, D., Pérez-Flores, L. J., Carrari, F., Insani, M., Asis, R., Mendoza-Espinoza, J. A., de León-Sánchez, F. D., & Rivera-Cabrera, F. (2016). Chemical characterization and quantification of the pigment extraction yield of seven Mexican accessions of Bixa orellana. Revista Mexicana de Ingeniera Quimica, *15*(3), 727–740.
58. Rodrigues, L. M., Alcázar-Alay, S. C., Petenate, A. J., & Meireles, M. A. A. (2014). Bixin extraction from defatted annatto seeds. Comptes Rendus Chimie, *17*(3), 268–283. https://doi.org/10.1016/j.crci.2013.10.010
59. Romualdo, G. R., Fragoso, M. F., Borguini, R. G., de Araújo Santiago, M. C. P., Fernandes, A. A. H., & Barbisan, L. F. (2015). Protective effects of spray-dried açaí (Euterpe oleracea Mart) fruit pulp against initiation step of colon carcinogenesis. Food Research International, *77*, 432–440.
60. Sato, M. K., de Lima, H. V., Costa, A. N., Rodrigues, S., Pedroso, A. J. S., & de Freitas Maia, C. M. B. (2019). Biochar from Acai agroindustry waste: Study of pyrolysis conditions. Waste Management, *96*, 158–167. https://doi.org/10.1016/j.wasman.2019.07.022
61. Sganzerla, W. G., Ampese, L. C., Parisoto, T. A. C., & Forster-Carneiro, T. (2021). Process intensification for the recovery of methane-rich biogas from dry anaerobic digestion of açaí seeds. Biomass Conversion and Biorefinery. https://doi.org/10.1007/s13399-021-01698-1
62. Shahbaz, M., Ammar, M., Korai, R. M., Ahmad, N., Ali, A., Khalid, M. S., Zou, D., & Li, X. J. (2020). Impact of C/N ratios and organic loading rates of paper, cardboard and tissue wastes in batch and CSTR anaerobic digestion with food waste on their biogas production and digester stability. SN Applied Sciences, *2*(8). https://doi.org/10.1007/s42452-020-03232-w
63. Shuhama, I. K., Aguiar, M. L., Oliveira, W. P., & Freitas, L. A. P. (2003). Experimental production of annatto powders in spouted bed dryer. Journal of Food Engineering, *59*(1), 93–97. https://doi.org/10.1016/S0260-8774(02)00433-8
64. Singleton, V. L., Orthofer, R., & Lamuela-Ravent6s, R. M. (1999). Analysis of Total Phenols and Other Oxidation Substrates and Antioxidants by Means of Folin-Ciocalteu Reagent. *Methods in Enzymology*, *299*, 152–178. https://doi.org/10.1016/s0076-6879(99)99017-1
65. Sluiter, A., Hames, B., Ruiz, R., Scarlata, C., Sluiter, J., Templeton, D., & Crocker, D. (2012). Determination of structural carbohydrates and lignin in Biomass - NREL/TP-510-42618. *Laboratory Analytical Procedure (LAP)*, *April 2008*, 17.
66. Souza De Oliveira, P. R., Fernando Trugilho, P., & Pires De Oliveira, T. J. (2020). Briquettes of acai seeds: characterization of the biomass and influence of the parameters of production temperature and pressure in the physical-mechanical and energy quality. Environmental Science and Pollution Research, *29*, 8549–8558. https://doi.org/10.1007/s11356-021-15847-6/Published
67. Taham, T., Cabral, F. A., & Barrozo, M. A. S. (2015). Extraction of bixin from annatto seeds using combined technologies. Journal of Supercritical Fluids, *100*, 175–183. https://doi.org/10.1016/j.supflu.2015.02.006
68. Teixeira, M. A., Escobar Palacio, J. C., Sotomonte, C. R., Silva Lora, E. E., Venturini, O. J., & Aßmann, D. (2013). Assaí - An energy view on an Amazon residue. Biomass and Bioenergy, *58*, 76–86. https://doi.org/10.1016/j.biombioe.2013.08.007
69. VDI-Handbuch Technik Biomasse/Boden. (2016). *VDI 4630 - Fermentation of organic materials – Characterization of the substrate, sampling, collection of material data and fermentation test*. http://www.vdi.eu/uploads/tx_vdirili/pdf/2385990.pdf
70. Wang, X., Lu, X., Li, F., & Yang, G. (2014). Effects of temperature and Carbon-Nitrogen (C/N) ratio on the performance of anaerobic co-digestion of dairy manure, chicken manure and rice straw: Focusing on ammonia inhibition. PLoS ONE, *9*(5). https://doi.org/10.1371/JOURNAL.PONE.0097265
71. Yamaguchi, K. K. D. L., Pereira, L. F. R., Lamarão, C. V., Lima, E. S., & da Veiga-Junior, V. F. (2015). Amazon acai: Chemistry and biological activities: A review. In *Food Chemistry* (Vol. 179, pp. 137–151). Elsevier Ltd. https://doi.org/10.1016/j.foodchem.2015.01.055
72. Yu, L., Reitmeier, C. A., & Love, M. H. (1996). Strawberry texture and pectin content as affected by electron beam irradiation. Journal of Food Science, *61*(4), 844–846. https://doi.org/10.1111/j.1365-2621.1996.tb12216.x
73. Zhang, H., Lopez, P. C., Holland, C., Lunde, A., Ambye-Jensen, M., Felby, C., & Thomsen, S. T. (2018). The multi-feedstock biorefinery – Assessing the compatibility of alternative feedstocks in a 2G wheat straw biorefinery process. GCB Bioenergy, *10*(12), 946–959. https://doi.org/10.1111/gcbb.12557

Figures

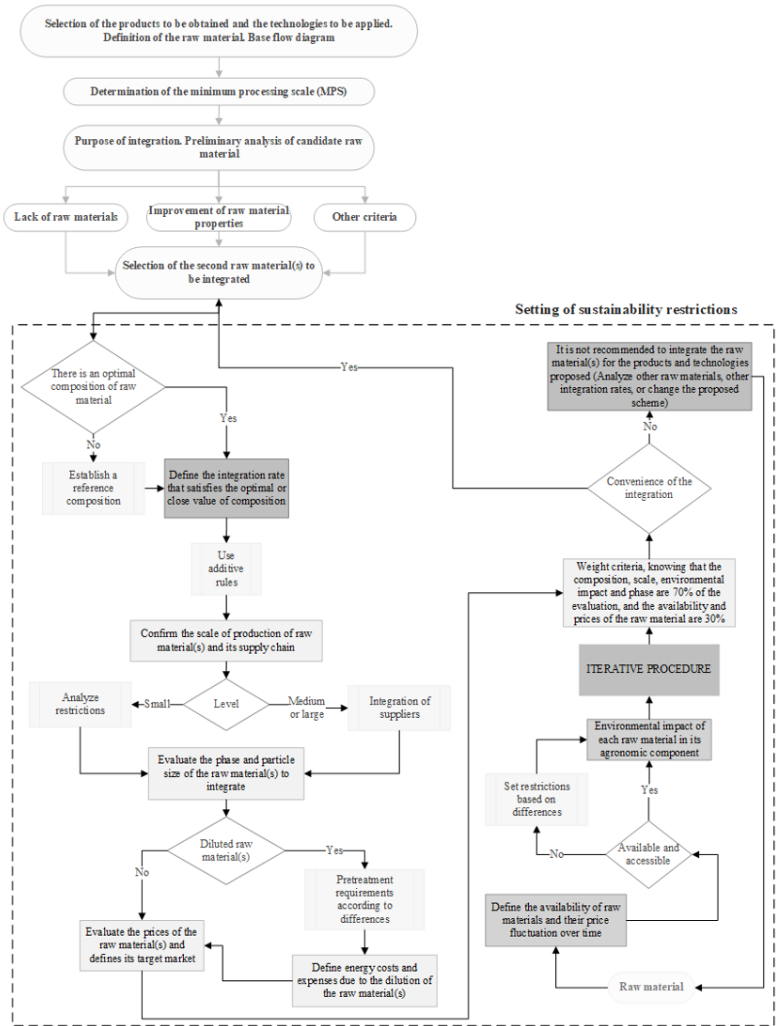

Figure 1

Block-diagram algorithm for the design of biorefineries integrating raw materials

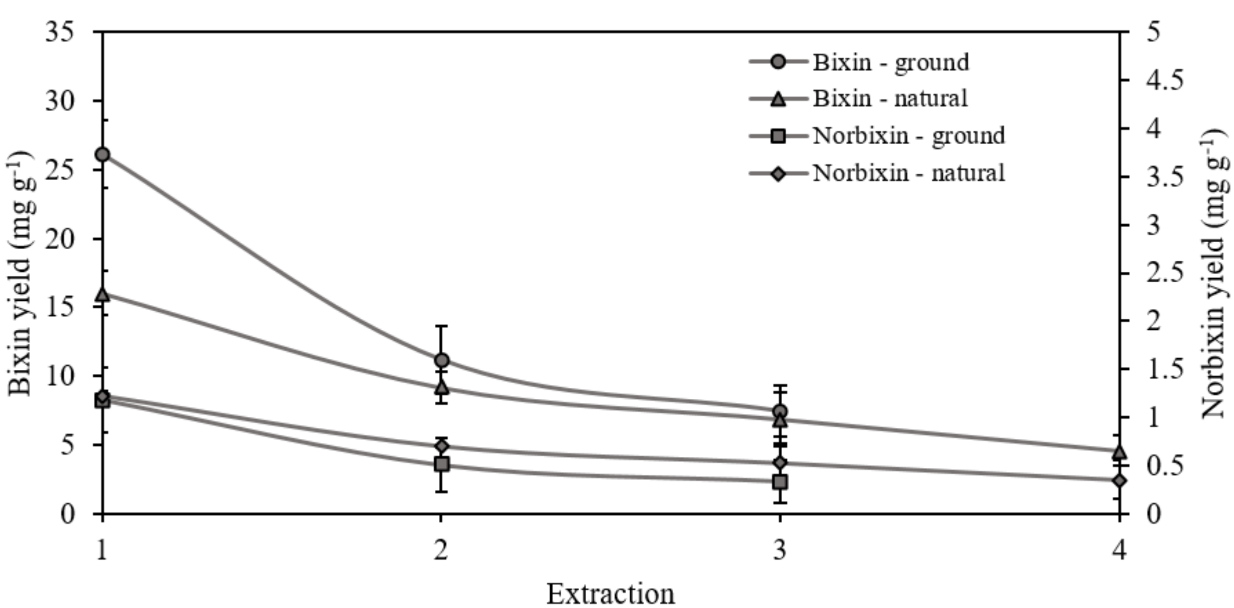

Figure 2

Bixin and norbixin extraction yields of the dye throughout seed rinsing. Error bars represent the standard deviation

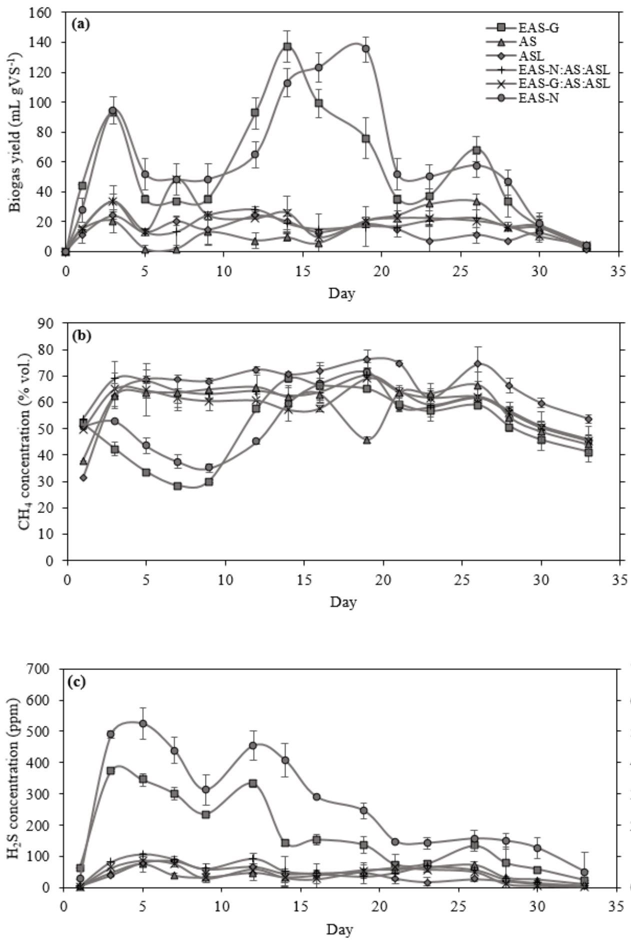

Figure 3

Biogas productivity performance. (A) Biogas yield; (B) methane composition; and (C) H_2S concentration. Error bars represent the standard deviation. The legend is the same for all Figures

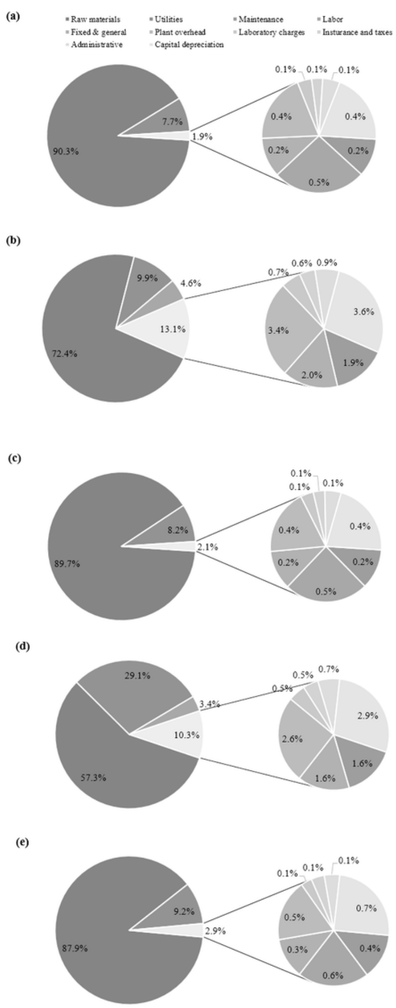

Figure 4

Total distribution cost of (A) Sc1, (B) Sc2, (C) Sc3, (D) Sc4, and (E) Sc5. The legend is the same for all Figures

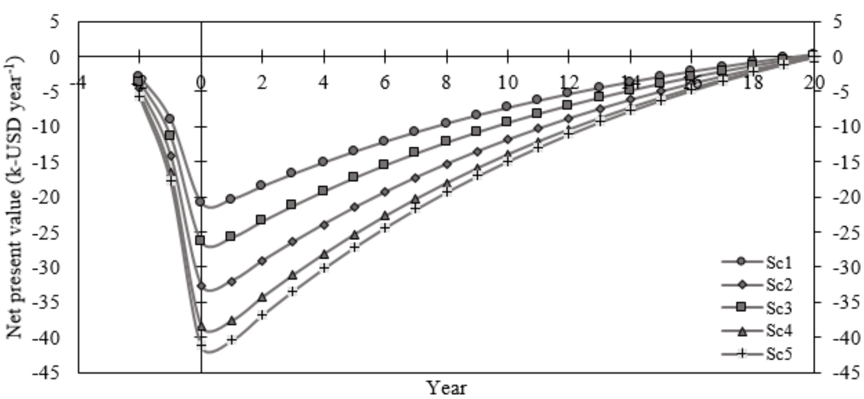

Figure 5

NPV at the minimum processing scale for economic feasibility

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- OnlineResource1.docx
- OnlineResource2.docx
- OnlineResource3.docx
- OnlineResource4.docx