

Article

# Ethanol Production from Waste of Cassava Processing

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**Abstract:** Cassava processing produces by-products such as brown bark, between bark, disposal, bran, fiber and bagasse. Cassava bagasse is characterized as a source of starch that can be converted into sugars to obtain biofuels. The objective of this work was to produce ethanol from this cassava processing residue and to evaluate its contribution potential in the Brazilian energy matrix. Cassava processing residues were obtained from four different starch manufacturers in Brazil. Analysis of the chemical compositions of these samples provided the content of starch, sugar, crude grease, moisture, ash and also their pH values. For the ethanol process, the samples were submitted to enzymatic hydrolysis using the alpha-amylase and amyloglucosidases enzymes, followed by fermentation and distillation. The samples showed high starch indices, approximately 64% on average. The average yield of ethanol obtained was 30% after treatment of the sample like this. Considering the estimated volume of cassava bagasse in Brazil, it is possible to produce an average of 789 million cubic meters per bagasse, replacing about 24% of the first generation ethanol. Cassava bagasse can be considered an interesting biomass for the production of biofuels, contributing to the expansion of the energy matrix.

**Keywords:** biofuel; second generation ethanol; cassava bagasse

## 1. Introduction

Cassava bagasse is an inexpensive and broadly available waste by-product from cassava starch production. It contains roughly 50% cassava starch along with mostly fiber and could be a valuable feedstock for various bioproducts [1]. Cassava pulp or bagasse is a byproduct of starch production.

For each 250–300 tons of cassava root processed, about 280 tons of bagasse with 85% moisture are produced [2]. Currently, the majority of the cassava bagasse is discarded as waste; however, this residue can be considered as an excellent source of fermentation for higher—value biochemicals [3], as is the case with second generation ethanol.

Second generation ethanol production is a strategic alternative to increasing ethanol production with environmental and economic advantages. Because of this, there is currently great interest in developing technologies for obtaining cellulosic ethanol (second generation) from the conversion of cellulose and hemicellulose of the bagasse [4] Great effort has been employed on the study of the enzymatic hydrolysis of sugarcane bagasse and many different pre-treatment methods were studied to enhance its biodegradability by enzymes [5].

The use of lignocellulosic materials as feedstock for second generation bioethanol production has not yet become an industrial reality due to the lack of efficient and low cost technologies, which results in prohibitive investment and production costs as well as poor returns on investment [6].

Increased production and use of ethanol with emphasis on advanced biofuels participation (such as cellulosic ethanol, also known as second-generation or 2G) is a commitment recognized by many

countries in the world, including the Brazilian Government in its Intended Nationally Determined Contribution (INDC) to United Nations Conference on Climate Change—COP 21 [7].

Therefore, potential environmental benefits of different alternatives for 2G ethanol production should be properly quantified for supporting public policy formulation that will affect the future of climate and other environmental aspects of our planet [8].

The results obtained from the test were applied to verify the production potential in Brazil. According to [9] each ton of manioc processed about 930 kg of manioc bagasse was produced, so it was possible to obtain a survey of the Brazilian Ministry of Agriculture and Supply related to cassava production between 2017 and 2018 to estimate the second-generation ethanol production potential from cassava bagasse. Subsequently, this value was compared to the data expressed by EPE—Energy Research Company (2018), where they present the volume of ethanol produced in 2017.

In this context, the objective of this study was to analyze ethanol production from cassava processing residues and to evaluate its impact on the Brazilian energy matrix, replacing first generation biofuels.

## 2. Materials and Methods

### 2.1. Materials and Pre-Treatment

This study was conducted at different stages and laboratories, the first stage (preparation of the samples, hydrolysis and fermentation) took place in the laboratory of biotechnology and physics of soil of the State University of Western Paraná, Campus Cascavel, the second and final stage (Distillation), was held in the Analytical Chemistry laboratory of the State University of Western Paraná, Campus Toledo.

The raw material used is characterized as cassava processing waste, also known as bagasse or cassava bran. The material was given by different cassava starch manufacturers, namely: Fecularia LAR, in the municipality of Missal/Parana, Industry Amidos Mundo Novo, located in the city of Novo Mundo/Mato Grosso do Sul, MCR Amidos located in Mercedes/PR and Horizonte Amidos located in Marechal Candido Rondon/PR.

The moisture content of the bagasse was determined using an oven at 110 °C for 24 h. For the determination of ash (%), grease matter (%), and fiber (%), AOAC—Official Methods of Analysis (1995) were used. The protein content was determined by converting the total N content in protein by using the factor 6.25. The starch content was calculated by the difference of the other components of the sample (moisture, ash, protein, raw fat, fiber, and total soluble sugar) under the rules of the Adolfo Lutz Institute (2005).

### 2.2. Enzymatic Hydrolysis

The enzymes used are produced, purified and standardized by the manufacturer for industrial application of food grade. The enzymes used were provided by the companies Science Pro (Araucaria, Brazil) and LNF-Latin American obtained from Novozymes<sup>®</sup> Latin America manufacturer (Araucária, Brazil). Enzymes used were: Hydrolytic (Alpha-amylase) and Saccharifying (Amyloglucosidases). The strain used was *Saccharomyces cerevisiae* from the LNF.

As the material has a wet and disintegrated structure, it was necessary to perform moisture control factor using greenhouse and heating it to 110 °C for 24 h. After checking the moisture content of each sample, dilution was performed in distilled water. Six erlemeyers were used with 20 g of dry bagasse for each treatment, a total of 24 flasks.

To carry out hydrolysis, the solid material was suspended in distilled water. For measurement of pH, a microprocessor phmetrer bench was used, and the pH was adjusted with 0.06 mol sodium hydroxide solution. The samples were heated in a water bath to 45 °C, and Liquozyme<sup>®</sup> (Araucaria, Brazil) enzyme was added in cassava mass. Subsequently, the temperature was raised to 90 °C. The hydrolysis was carried out for 60 min. under mechanical agitation of 200 rpm and a constant temperature of 60 °C.

After dextrinization, the samples were subjected to a bath in order to reduce the temperature at 36 °C, and the pH of the dispersion was adjusted to 5.1 to 5.5 with 0.06 mol sodium hydroxide

and heated again to 60 °C. After the temperature reached 60 °C, a suspension of amyloglucosidase enzyme was added and subjected again by mechanical agitation for a predetermined time of one hour at 100 rpm. Filtering of the hydrolyzate material was performed for removing impurities (fiber) using a vacuum pump, Buchner funnel and kitassato.

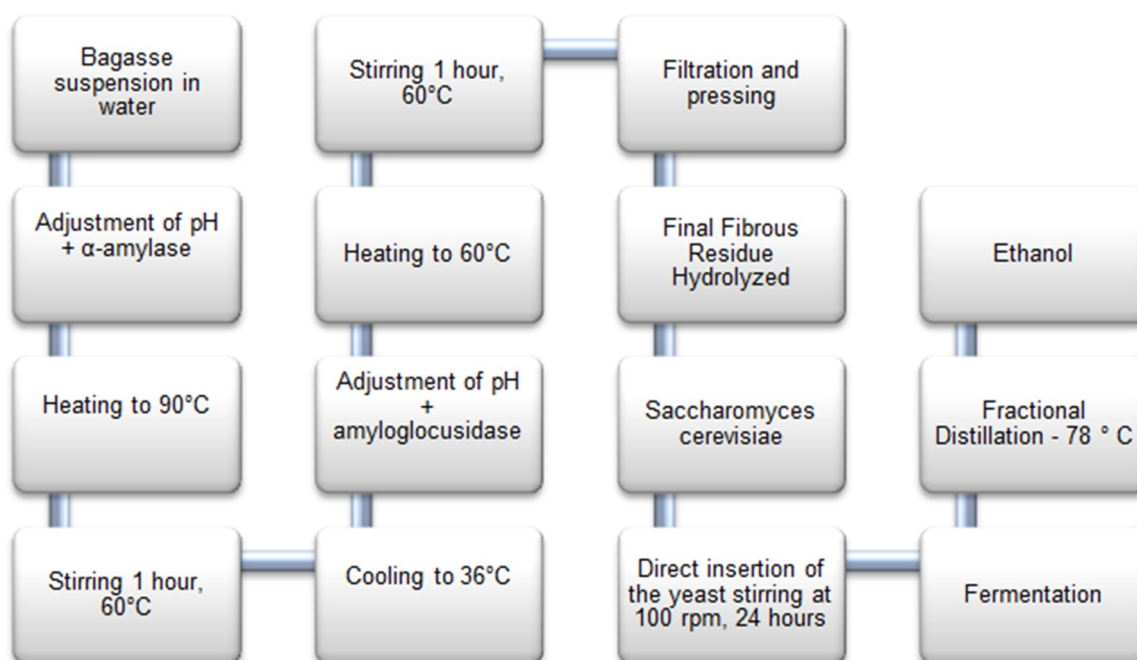
### 2.3. Fermentation and Distillation

Each assay was performed in sequence, and the hydrolysates were frozen for later fermentation and distillation. For the fermentation of the yeast strain, *Saccharomyces cerevisiae* was used. The quantity used was 100 g/L of must. In addition, 10% fermentation volume was used in water at 35 °C for diluting the yeast for 30 min. before adding samples for fermentation. The samples were subjected to constant agitation of 100 rpm and 30 °C for 24 h. Then, we realized the wine filtration process.

### 2.4. Analysis Methods

Infrared spectrometric analysis was performed prior to analysis of 24 samples, making it possible to obtain the spectra and highlighting the best samples for each treatment with the samples that have been selected with major peaks for the distillation procedure. The step of obtaining the wine was carried out by simple distillation at a concentration of 10%. The monitoring of temperature using a digital thermometer was performed.

For analysis of distilled portions, infrared spectrometric analysis was used to verify the presence of ethanol by medium infrared, with a wavelength of 1600–1800 nm. Figure 1 represents systematically the process as a whole, for obtaining ethanol.



**Figure 1.** This represents systematically the process as a whole, for obtaining ethanol.

## 3. Results and Discussion

### 3.1. Potential for Ethanol Production

Before and after drying treatment, samples A3 and A4 visually presented darker staining when compared to others revealing the differences in industrial processes. In Table 1, the values obtained from the compositional analysis of cassava bagasse are displayed.

**Table 1.** Physical and chemical characterization of samples before hydrolysis.

Samples	Starch (%)	Raw Fiber (g/100 g)	Protein (g/100 g)	Sugars (g/100 g)	Grease Matter (g/100 g)	pH	Humidity (%)	Ash (g/100 g)
A1	64.80	3.42	0.49	8.30	0.21	4.31	87.14	0.23
A2	59.90	3.85	0.53	6.61	0.20	4.45	89.29	0.29
A3	64.10	6.07	0.90	8.04	0.20	4.50	88.70	0.12
A4	68.30	3.94	0.52	8.24	0.01	4.91	86.75	0.23
Mean	64.27	4.32	0.61	7.79	0.15	4.54	87.97	0.21

The bagasse used had approximately 88% average moisture content. [10] Reported that starch-fibrous material has a large water retention capacity, and often the moisture content of the pulp leaving the process is higher than 80%. In accordance with [11], each ton of processed root yields approximately 930 kg of bagasse with 85% moisture content.

These values are specific for many bagasse cassava, as there are root variations having different chemical compositions [12]. It was observed that the residual starch concentrations are similar (Table 1). The soluble sugars were also similar, except for the sample from A2, which showed a lower value compared with other samples. The similarity between the other samples is probably due to the action of similar degrading microorganisms, indicating the possible need to use additives for fermentation.

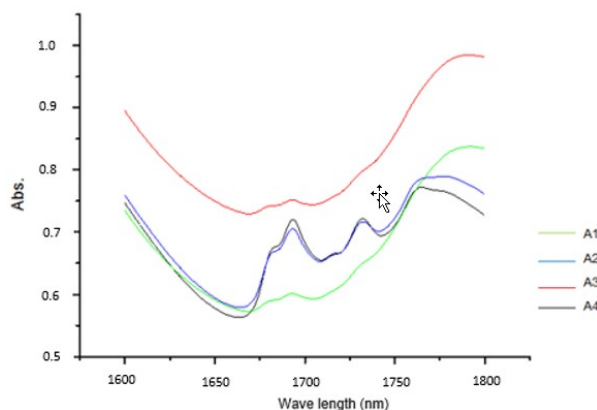
The major component, starch, comprised approximately 64% on average and was highest in A4, with a value of 68.30%. The content of starch in the bagasse is within the average value obtained by different authors, ranging from 47.10% [13] to 65.37% [14].

According to Table 1, the levels of various chemical components found may be comparable with the results obtained by [9,15], where the values found were as follows: 65.16% moisture in the humid matter, 3.0% protein, 0.30% grease matter, 2.6% fiber, 2.4% ash, and 8.2% sugar. The presence of low concentrations of protein and fatty matter found favors the fermentation process considerably by reducing foaming. Table 2 shows the values obtained after the hydrolysis process.

**Table 2.** Physical and chemical characterization of samples after hydrolysis.

Sample	Humidity (%)	% Total Solids	g/Total Solids	Initial pH	Final pH	Mass (%)	Volume before Hydrolysis (mL)	Volume after Hydrolysis (mL)
A1	64.19	35.81	7.16	6.15	5.14	27.13	150	130.67
A2	68.88	31.12	6.22	5.98	5.23	29.52	150	124.00
A3	68.59	31.41	6.28	5.94	5.37	29.24	150	132.67
A4	75.90	24.10	4.82	6.22	5.39	32.52	150	124.17

Temperature adopted 60 °C, density 1.13 g/mL, residence time 1 h, and a volume used of 20 g of substrate. Through infrared spectrometric analysis, it was found that various samples showed similarities in the spectra. As described above, the samples were distilled at 10% concentration utilizing only 5 mL from 50 mL; therefore, treatments that showed the highest amount of ethanol in distilled material were A2 and A4, respectively having 2.4 mL and 2.65 mL of ethanol from 5 mL of solution. A1 had a value of 0.63 mL ethanol from distillation. Figure 2 indicates the curves obtained in the analysis of infrared spectrometry.



**Figure 2.** Chart showing absorption spectra of the solutions A, B, C, and D to the solutions distilled to 10% by volume.

The solutions of A1, A2, and A4 have baselines close to the standard solutions and can be analyzed using the adjusted straight-line method, providing the concentrations of A1 as 12.6%, A2 as 48.5%, and A4 as 53.3%. The A3 solution had a very different baseline from the standard solution; therefore, its analysis was difficult. However, by spectral comparison between the solutions of A3 and A1, the concentrations are expected to be close to each other, obtaining a value of approximately 12% for A3.

The ethanol values obtained were found to be directly related to the amount of starch present in the sample, as can be seen in Table 3. It is noteworthy that A2 presented greater efficiency in ethanol production compared with A1 and A3, considering the starch percentage.

**Table 3.** Values after distillation.

Treatment	Starch (%)	Ethanol (%)	Ethanol (mL/kg)	Ethanol Yield (%)	Fibrous Residue (%)	Reducing Sugars (g/100 g)	Reducing Sugars (%)
A1	64.80	12.60	103.48	12.06	27.13	2.43	0.02
A2	59.90	48.50	368.21	48.50	29.52	2.27	0.02
A3	64.10	12.00	97.49	12.00	29.24	2.35	0.02
A4	68.30	53.30	461.39	53.00	32.52	3.65	0.04
Mean	64.28	31.60	251.45	31.00	29.60	2.68	0.03

The A4 had higher conversion of ethanol at 53%, slightly below the values reported by other authors, such as [16]. wherein the ethanol production obtained from cassava pulp was 78.6%. This oscillation in values in relation to ethanol production is directly related to the different process conditions that the samples have been subjected to, from the efficiency of the centrifuges to the storage model. In Table 4, we can observe the comparison between ethanol production potential from different substrates.

**Table 4.** Potential of ethanol production from different substrates.

Substrates	Starch Concentration (%)	Ethanol Production (g/g)	Authors
Cassava bagasse	64	0.21	-
Corn	69 [17]	0.32 [18]	[17,18]
Wheat	66 [17]	0.31 [18]	[17,18]
Cane bagasse	15 [19]	0.11 [12]	[12,19]
Corn stover	64	0.26 [20]	[20]

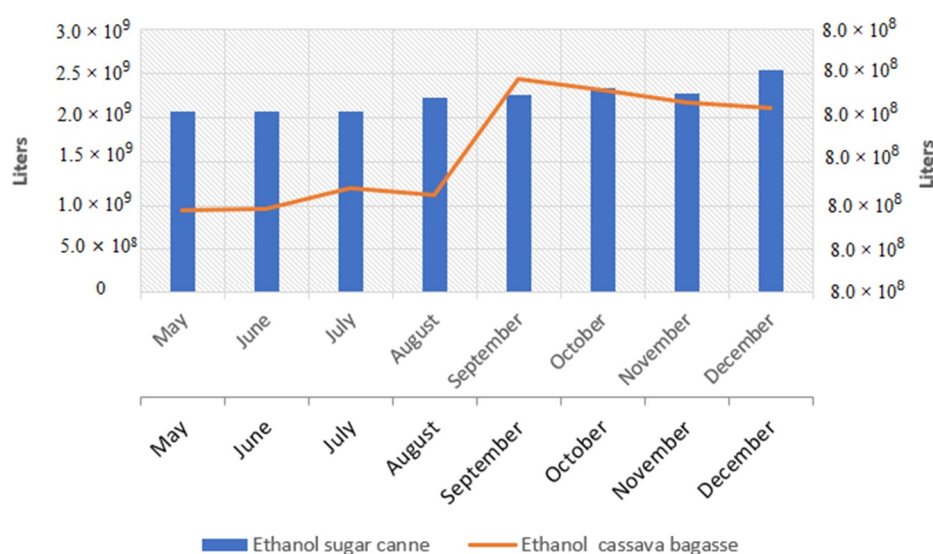
These variables make it possible to miss a part of starch, as the samples showed acidity, concluding therefore that part of the starch was converted to organic acids.

From the values described in Table 4, about 251 mL/kg, that is, 0.25 m<sup>3</sup> of ethanol per ton of cassava bagasse, were obtained. The concentration of ethanol was 31% in average. [21] obtained

by distilling 16 mL of cassava residue, about 32.4% ethanol at a temperature of 32 °C. The ethanol yield obtained was 31% on average, according to [22] who carried out research using two strains of *Saccharomyces cerevisiae* of non-manioc.

### 3.2. Second Generation Ethanol from Brazil

According to the Energy Research Company—EPE [23] by 2030, Brazil will produce approximately 2 billion liters of second generation ethanol, considering a projection for the only two plants installed in Brazil. The Union of Producers of Sugar Cane—UNICA releases monthly the production of anhydrous and hydrated ethanol. In Figure 3, the comparison between the potential of ethanol production from cassava bagasse (considering the volumes of bagasse generated) and the production of first generation ethanol produced in the months of May to December 2017 can be observed.



**Figure 3.** Comparison of the production of first and second generation ethanol; source: UNICA—Union of Producers of Sugar Cane, 2018.

With the volume generated from the cassava meal, it would be possible to produce about 9 million m<sup>3</sup> of second generation ethanol. This value inserted in the Brazilian energy matrix could replace about 24% of the first generation ethanol produced from sugarcane sugar. The decision regarding second generation ethanol production should be made considering the opportunity prices for the different biomass derived products (ethanol and other biofuels, bioelectricity and sugar, among others) [3].

Therefore, the total use of this biomass received by the industry, including generated residues, is important for the industrial economy balance. In addition, the use of this agro-industrial waste as a carbon source for another process helps to solve the pollution problems. Each ton of cassava contains 211.2 kg of native starch and, after industrial processing, 115.5–132.0 kg (Chavalparit and Ongwande, 2009) are generated as solid waste.

According to [24] if the cassava bagasse generated in Brazil in 2014 (about 3.5 million tons, dry basis) is considered, it would have the potential to produce 866.6 million liters of additional ethanol, corresponding to an increase of 3.04% of total ethanol production in Brazil during the same period, which was 28.48 billion liters, according to UNICA (2014).

Second generation ethanol on an industrial scale depends primarily on political factors and, in a second stage, on technical achievements. In fact, although the technique makes the industrial scale feasible, and constitutes a major challenge, the evolution of second generation ethanol in the industry as a whole depends crucially on policies and strategic decisions, which include the promotion of technology and policies to create artificial markets [25].

#### 4. Conclusions

- Within the experimental conditions, the results showed that cassava bagasse showed starch content with an average of 64%, making it as an efficient raw material for second-generation ethanol production process.
- There was an average ethanol production potential of 0.25 m<sup>3</sup>/ton of cassava bagasse.
- This material has potential features that make it an interesting biomass for energy matrix enlargement.
- It is important to note that using cassava bagasse to produce second generation ethanol can prevent future competition for land use between ethanol production and food production, thereby enabling an increase in ethanol production without causing an increase in cassava.

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**Conflicts of Interest:** The authors declare no conflict of interest.

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