Comparative Prospection of Bioethanol Production from *Musa* spp. Residues

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The present study explores the feasibility of bioethanol production using banana residues, focusing on its potential contribution to second-generation bioethanol. The research investigates the comparative prospecting of bioethanol production from different *Saccharomyces cerevisiae* strains, employing methods such as enzymatic hydrolysis and alcoholic fermentation. Results indicate the viability of utilizing banana residues for bioethanol production, highlighting its potential for sustainable fuel production. The findings underscore the importance of harnessing abundant agricultural byproducts for renewable energy generation, with implications for both theoretical and practical biofuel applications.
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The growing demand for renewable and sustainable energy sources has driven notable advances in second-generation ethanol production. In this context, the production of 2G ethanol contributes to reducing dependence on fossil fuels and represents a significant step towards sustainability. This approach minimizes environmental impact by using waste and promotes an economically viable, socially beneficial, and efficient solution for disposing of this waste.

Banana farming residues have aroused significant interest in being used as raw material in manufacturing bioethanol. Due to its physical-chemical characteristics, it has significant potential for fuel generation [1]. According to the Food and Agriculture Organization of the United Nations (FAO) [2], in 2011, banana production in *Musa* spp. reached more than 106.54 million tons of fruit. As a result, the fruit can be considered one of the most consumed in the world; according to Souza and colleagues (2012) [3], for each ton of banana, approximately three tons of waste are generated, including peels, leaves, pseudostems, and banana tree remains.

As mentioned by Alvarenga (2011) [4], regarding sugars during banana ripening, the amylase enzyme acts by converting 17.5 to 25.9% of the starch present in the fruit into fermentable sugars, mainly glucose and fructose, which are found in more than 20%. As presented in Table 1, the residual starch will undergo a hydrolysis process. This procedure aims to convert the starch into simpler sugars, increasing the available sugars.

To produce bioethanol with raw materials that contain starch (polysaccharide), it is necessary to carry out the hydrolysis process so that fermentable sugars (monosaccharides) are created, as described in the following reaction [5]:

$$(C_6H_{10}O_5)n + (n+m)\,H_2O \rightarrow n\,glucose + m\,maltose$$

Hydrolysis aims to convert polysaccharide-rich biomass into smaller sugars. In enzymatic hydrolysis, specific enzymes convert cellulose and hemicellulose into fermentable sugars, essential for ethanol production. This method stands out for its efficiency and lower energy demand than chemical hydrolysis [6].

Ethanol (C_2H_6O) and carbon dioxide (CO_2) can be produced through enzyme-catalyzed fermentation. This process occurs mainly in yeast to produce energy in the form of ATP. Ethyl alcohol is a byproduct of fermentation and also acts as an inhibitor of other microorganisms [7]. According to Amorim (2005) [8], strains of *Saccharomyces cerevisiae* are the most used in converting sugar into ethanol. They have an optimum temperature

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Table 1. Composition of banana sugars in the ripening phases.

Degree of Maturation	Starch (% m/m)	Total Sugars (% m/m)
Underripe	19.8	6.5
Mature	2.9	20.4
Very mature	0.63	22.7

Source: Alvarenga, 2011 [4].

of 26 to 35 °C. pH values develop in a range, with between 4 and 5 being suitable [9].

The present study aims to analyze the viability and efficiency of second-generation bioethanol production using three different *Saccharomyces cerevisiae* strains, collect data and banana residues, and evaluate residue/yeast combinations with higher alcoholic yields.

Materials and Methods

This article's methodological approach adopts a qualitative perspective, allowing a comprehensive analysis of bioethanol production from banana waste. The choice of three different commercial strains for industrial applications aims to provide comparative data for obtaining bioethanol.

Acquisition of Yeasts

The yeasts were purchased from Bahia Malt, a store selling supplies and equipment for brewing in Salvador - Bahia. The related strains of *Saccharomyces cerevisiae*, S-33, WB-06 It is K-97, were selected for the study due to their efficiency in producing beer and other alcoholic beverages.

Cultivation of Strains *Saccharomyces cerevisiae*

The cultivation of yeast strains was achieved through the use of medium Sabouraud Dextrose, in duplicate, with a composition of dextrose (20 g/L), a mixture of animal tissue peptic digestive (10 g/L) and casein pancreatic digestive (10 g/L), being autoclaved at 121°C, carrying out the inoculation of strains S-33, WB-06 It is K-97, and growth during 72h.

Preparation of Banana Waste Must and Enzymatic Hydrolysis

Hydrolysis began by treating the biomass and weighing and sanitizing the material. Approximately 782g of waste was collected and sanitized in a 10% sodium hypochlorite solution. The residue was cut, diluted in water, homogenized in a blender, and filtered. Thus, the must was prepared for enzymatic hydrolysis.

It was carried out in a 5000 mL container with a lid, in which a system was created to capture CO_2. 1,826 mL of the enzyme was inoculated into the system Attenuzyme for 4.565 kg of must was analyzed using the Dubois Sugar Determination method.

The test was monitored for 60 hours, with the analysis points removed at 0, 24, and 48 hours. The final volume of the test used for fermentation yielded 5L, and after filtration, 2.8L of the residue hydrolyzate was obtained *Musa* spp.

Alcoholic Fermentation

The alcoholic fermentation of the musts was carried out in 500 ml airtight containers, using an airlock system to monitor the fermentation process and exhaust the produced carbon dioxide. 380 ml of filtered banana hydrolysate and 20 mL of *Saccharomyces cerevisiae* inoculum was used at 23°C. The fermentation process lasted 48 hours and was halted by cooling. The alcohol content of the fermented product was measured with an alcoholmeter.

Results and Discussion

The following item provides data obtained through quantitative and qualitative analyses, such as determining Sugars by Dubois, Chemical, Enzymatic Hydrolysis, Alcoholic Fermentation, and Distillation, carried out through laboratory experiments to support the project hypotheses.

Growth of Strains *Saccharomyces cerevisiae*

The growth of the three *Saccharomyces cerevisiae* strains, which was analyzed using the Wet Weight method, showed a consonance between the growth of yeast strains in the first 24 hours, determining the minimum fermentation time.

Determination of Total Sugars in the Hydrolyzate

The analysis of total sugars using the Dubois method to attest to Attenuzyme's breakdown of complex sugars (starch) did not present significant results.

Since Table 1 indicates only 0.63% starch in the total composition of ripe bananas, it was assumed that hydrolysis did not guarantee results given the small amount of sugars to be converted into the must.

Therefore, it is suggested that the process is viable without the additional step, given that ripe bananas have a satisfactory sugar content, as described in Table 1.

Characterization of Hydrolyzed Wort

After hydrolysis and filtration, the volume obtained from the must was equivalent to 2.88 liters and presented 5°Brix, which was corrected by adding 217 g of commercial sugar, ending this process with 14.9°Brix.

Characterization of the Fermented Must of Each Strain

Based on Table 2, we assume that WB-06 is the yeast most likely to meet the parameters for the production of Hydrated Fuel Ethanol established by the National Agency of Petroleum, Natural Gas and Biofuels (ANP), as it has a higher alcohol content than the others.

Characterization of each Bioethanol Obtained

The fractional distillation process obtained ethanols. Each yeast strain provided a different volume of distillate from the same volume of wort, as shown in Table 3.

Among the three, the strain of *Saccharomyces cerevisiae* with the most significant identified productive potential is the WB-06, followed by S-33; both have notable sugar consumption and conversion capacity and ethanol production with a considerable and relevant volume of distillate. However, having observed the divergence of data obtained regarding the volume indicated by the alcohol content of the must and the distillate, an equation was developed to estimate the degree of purity of the ethanol produced:

$$P = M \times T \div D$$

Where:

P = Degree of Purity (%); M = Volume of Fermented Must (mL); T = Alcoholic Content of the Must (%); D = Distillate Volume (mL).

Table 2. Comparative analysis of fermented musts of each strain.

Yeast Strain	°Brix Post - Fermentation n	pH	Alcohol content (% v/v)
S-33	3.8	4	3
WB-06	4.5	4	4
K-97	7	4	0.5

Table 3. Volume of distillates from each strain *S. cerevisiae*.

S. cerevisiae strain	Must Volume (mL)	Distillate Volume (mL)
S-33	400	38
WB-06	400	33
K-97	400	23

Determining the degree of purity of the ethanol produced, described in Table 4, helps to compare one of the essential parameters to comply with ANP regulations. According to the requirements of this regulatory body, set out in ANP Resolution No. 907, the minimum alcohol content for Hydrated Fuel Ethanol (EHC) is 92.5%, and the minimum hydrogen potential (pH) is 6.0.

Table 4. Estimated purity content of ethanols obtained.

S. cerevisiae strain	Purity Content
S-33	31.58
WB-06	48.48
K-97	8.69

In this way, it was found that the bioethanol produced by strain WB-06 has the highest alcohol content and is close to that suitable for the Agency, followed by the ethanol from strain S-33. This information is crucial to understanding the productive potential of the analyzed strains, guiding future optimizations in the bioethanol production process from banana waste, and adapting the ethanol produced to the standards defined for Hydrated Fuel Ethanol by the ANP.

Conclusion

The present study analyzed the viability of bioethanol production from banana waste using three different strains of *Saccharomyces cerevisiae*: S-33, WB-06 It is K-97. The results demonstrated the feasibility of using banana residues for bioethanol production, confirming their potential as a sustainable source for renewable fuel generation and showing that converting them into bioethanol contributes to environmental development and provides a promising alternative to conventional biofuels. This integration involves developing advanced technologies, optimizing processing conditions, and significantly contributing to environmental sustainability with large-scale use.

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