

ETHANOL PRODUCTION FROM BANANA PEEL WASTE

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ABSTRACT

The growing demand for fuels has accompanied the population increase; however, the conventional use of fossil fuels is no longer a viable alternative for future generations since these are non-renewable sources already in the process of exhaustion, and they have adverse effects. Because of the large generation of solid waste, such as fruit peels, an alternative is emerging for the production of renewable and low-pollution fuels, such as bioethanol and biogas, among others, which can be produced from the conversion of fermentable sugars into fermentable sugar products high added value aiming to make use of food waste and a sustainable production route.

Keywords: Fruit residue. Enzymatic hydrolysis. Bioethanol. Bioprocesses

1 INTRODUCTION

Fruit waste is rich in carbohydrates and minerals, has high levels of fermentable sugars, and can be used as raw material in the production of biofuels. This gives rise to a strategy for waste management and recovery of food losses unsuitable for consumption^{3,6}.

The banana peel corresponds to approximately 30% of the total weight of the fruit, which results in the annual generation of tons of waste. The peel contains a high level of fiber and free sugars, which can be used in alternative disposal strategies to valorize products with high added value⁵.

In the structure of banana peel biomass, crystalline cellulose is coated and entangled by hemicellulose structures, followed by lignin coating that reduces the surface area for the hydrolysis process, reducing the efficiency of bio-based systems such as fermentation of alcoholic beverage for ethanol production^{7,9}. In this scenario, the pretreatment step becomes a prerequisite to increase the biodegradability of lignocellulosic biomass and improve conversion to ethanol^{8,9}.

Fermenting microorganisms is a crucial part of ethanol production. On industrial scales, this process is generally carried out by yeast, with *Saccharomyces* strains traditionally used due to their high efficiency and tolerance to ethanol¹¹. In this scenario, this study's relevance stands out in developing biotechnologies with the potential to expand energy matrices, emphasizing environmentally sustainable processes with scaling feasibility. The exploration of different production routes from the banana peel in a system optimized by experimental planning for ethanol production presents the novelty elements of this study.

2 MATERIAL & METHODS

2.1 Residual biomass

Agro-industrial waste was collected at the UFFS - Campus Erechim, RS University restaurant. The banana peels were dried in an oven with air circulation (40 °C), ground in a knife mill to a particle size of 20 mesh, and stored (-20 °C) until use.

2.2 Study of different pretreatments

The study of pretreatments occurred with the application of different methodologies: acid pretreatment (H₂SO₄, 5% (v/v)), alkaline (NaOH 1% (m/v)) and in an ultrasonic bath (132W) and frequency 40kHz, and (66W) and 40kHz frequency. After pretreatments, the results were quantified in a spectrophotometer based on the release of sugars after enzymatic hydrolysis.

2.3 Enzymatic Hydrolysis

Enzymatic hydrolysis was carried out with process conditions being 1% w/v of pre-treated solids (dry mass) at pH 4.8, adjusted by the use of 0.05 mol L⁻¹ sodium citrate buffer, being hydrolyzed with 50 FPU/g of commercial enzyme cellulase (Sigma-Aldrich), and maintained at 50 °C and 150 RPM for 120 hours (GABHANE et al., 2014)⁴. Throughout the incubation period, aliquots were removed at 120 h. The samples were subjected to sugar quantification using the 3,5-dinitrosalicylic acid (DNS) method, according to MILLER¹⁰.

2.4 Ethanol Production

The yeast *Wickerhamomyces* sp. was used to produce ethanol. UFFS-CE-3.1.2. The yeast remained in a YPD culture medium with a composition of 1% yeast extract, 2% peptone, and 2% glucose, plus 2% agar until use. The cells were transferred to liquid YPD medium (10 mL) for the inoculum and incubated for 24 hours at 30 °C on an orbital shaker at 80 RPM. The fermentation stage was carried out using the most from enzymatic hydrolysis to place 90 mL of it in 250 mL Erlenmeyer flasks sterilized in an oven to avoid contamination with ethanol. The inoculum containing the liquid YPD culture medium fraction and subcultured cells was added. The Erlenmeyer flasks were placed in an orbital incubator at 30°C and 120 RPM for 48 hours. Throughout the fermentation period, aliquots were collected at times 0h, 12h, 24h, and 48h and subjected to compositional analysis on HPLC equipped with a refractive index detector (RID-10A) and Aminex® Biorad HPX-87H column, using H₂SO₄ 0.005 mol as mobile phase at a flow rate of 0.6 mL/min and a temperature of 45 °C, for quantification of sugars, ethanol, acids and other compounds^{1,2}.

3.2 Central Composite Design

The experimental design was conducted using a Central Composite Design (CCD), with variables being the mass of banana residue, the concentration of H₂SO₄ in the pretreatment stage, and the enzyme concentration in the enzymatic hydrolysis stage.

Essay	Mass (g)	H ₂ SO ₄ Concentration (%)	Enzyme concentration (FPU/g)
1	5 (-1)	10 (-1)	5 (-1)
2	15 (1)	10 (-1)	5 (-1)
3	5 (-1)	30 (1)	5 (-1)
4	15 (1)	30 (1)	5 (-1)
5	5 (-1)	10 (-1)	50 (1)
6	15 (1)	10 (-1)	50 (1)
7	5 (-1)	30 (1)	50 (1)
8	15 (1)	30 (1)	50 (1)
9	10 (0)	20 (0)	27.5 (0)
10	10 (0)	20 (0)	27.5 (0)
11	10 (0)	20 (0)	27.5 (0)

Table 1. Central Composite Design 2³ for pretreatment and enzymatic hydrolysis (actual and coded values).

3 RESULTS & DISCUSSION

3.1 Pretreatment study

The pretreatments applied demonstrated different effectiveness on biomass due to its lignocellulosic matrix. In biomasses such as banana peels, which have high levels of lignin, cellulose, and hemicellulose, chemical pretreatments tend to present better results for the release of total sugars, as in this case, 11.57g/L ± 0.03 and 7.28g/L ± 2.54 for acid and alkaline pretreatments respectively. In this case, the physical pretreatment in an ultrasonic bath did not show significant results for the release of sugars, being 1.50g/L ± 0.08 and 1.40g/L ± 0.03 for 132W and 66W, respectively.

3.2 Central Composite Design

In response, the release of total reducing sugars was analyzed. In general, the acid pretreatment combined with enzymatic hydrolysis using commercial cellulase (Sigma-Aldrich) showed good efficiency in the cleavage of glycosidic bonds, releasing fermentable sugars in significant quantities (11.88 g/l) under the best planning conditions, these being conditions, mass (15 g), H₂SO₄ (10%) and Enzyme (50 FPU/g) (Figure 1).

It is worth mentioning that, despite the significant concentration of total reducing sugars, one of the main problems in ethanol production is the generation of fermentation inhibitors along the way, such as citric acid found in high values (9.05g/L), resulting in a low ethanol yield at the end of the process (1.37 g/l).

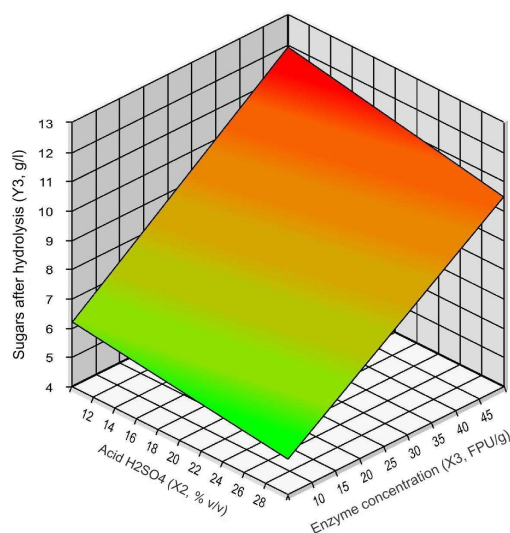


Figure 1. Mass relationship versus enzyme concentration for sugar results

4 CONCLUSION

The values of total reducing sugars confirm banana peel residues as a potential biomass for biorefinery processes due to the possibility of fermentation of these amounts of sugars; however, the high concentration of citric acid generated in the process, resulting in low ethanol yield, is a point to be addressed for better viability of the process.

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