BIOETHANOL PRODUCTION FROM FRUIT WASTE

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ABSTRACT

Considerable amounts of fruit waste are generated annually, and its low use represents a potential environmental problem. This study aims to use the sugars available in fruit peel mix residue (orange, banana, mango, pineapple, and lemon) for bioethanol production. For this, sugars were obtained by enzymatic hydrolysis of biomass using commercial cellulase and optimized with Central Composite Rotational Design (CCRD), evaluating the variables of the mass of the fruit peel mix and enzyme concentration. The bioethanol production step was processed by fermentation with the yeast *Wickerhamomyces* sp. UFFS-CE-3.1.2. The results demonstrated that with the optimization of enzymatic hydrolysis, it was possible to achieve satisfactory sugar yields and substantial bioethanol production from fermentation.

Keywords: Fruit waste. Sugars. Enzymatic hydrolysis. CCRD. Bioethanol.

1 INTRODUCTION

According to the Food and Agriculture Organization of the United Nations (FAO), in 2017, global fruit production exceeded one billion tonnes, resulting in an estimated amount of waste between 25 and 75 million tonnes[1]. The underutilization of these fruit residues represents an environmental problem, as well as the loss of a potential energy source and high-added-value biocompounds since the organic composition of these biomasses offers significant potential for conversion into bioproducts and energy.

Using this food waste in biorefineries is an effective strategy for valuing the production chain of foods no longer suitable for consumption[2,3]. Increasingly, new process configurations are proposed in the production of bioenergy and bioproducts from abundant biomass without the need for pretreatment with chemical additives through clean and sustainable techniques, aiming for maximum efficiency in the recovery of high-added value bioproducts and enabling the use of predominantly biological systems.

Therefore, this study uses the biorefinery concept to obtain bioethanol and explore the potential of waste from various fruits (orange, banana, mango, pineapple, and lemon). This approach aims to mitigate environmental problems and open new research perspectives in the emerging circular economy and biorefineries field, promoting integrated systems of high efficiency and low environmental impact.

2 MATERIAL & METHODS

Orange, banana, mango, pineapple, and lemon peels were collected in the university restaurant of the Federal University of Fronteira Sul, dried in an oven with air circulation at 40 ºC for 48 hours and comminuted in a knife mill (20 mesh)[4]. The biomass was washed in a Dubnoff bath to remove soluble sugars, using 10 g of dry mix mass and 100 mL of distilled water under mechanical stirring for 5 minutes at 30 °C[5]. The samples were filtered and separated into liquid and solid fractions. The solid fraction was dried in an oven with air circulation at 40°C and stored for use in enzymatic hydrolysis.

Aiming to produce sugars, the enzymatic hydrolysis was optimized using Central Composite Rotational Design (CCRD) using the mass of the fruit peel mix and the enzymatic concentration of commercial cellulase as variables (Table 1). The response was the concentration of total reducing sugars quantified by colorimetry using the 3,5-dinitrosalicylic acid (DNS) methodology[6].

The enzymatic hydrolysis assays were carried out using the mass of fruit peel mix and enzyme concentration (according to Table 1), 100 mL of 0.05 M sodium citrate buffer and pH 4.8, at 50 °C and 150 rpm for 120 h on an orbital shaker[7]. At the end of hydrolysis, samples were collected to quantify the total sugars released during the procedure.

Table 1 Central Composite Rotatable Design of enzymatic hydrolysis (actual and coded values).

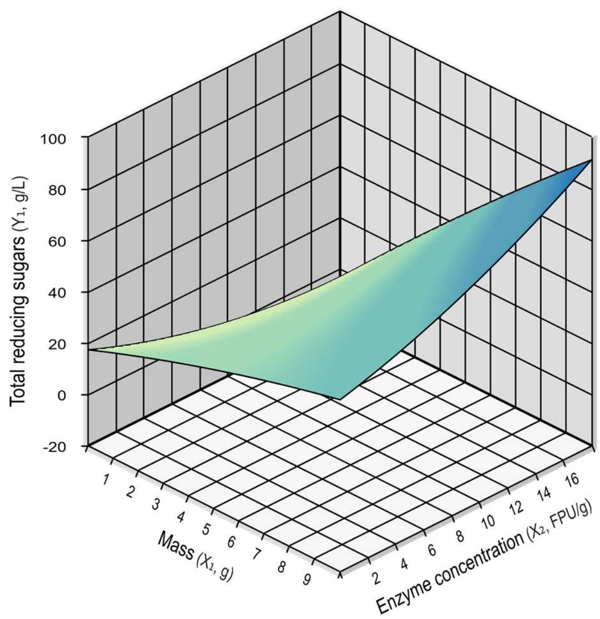
Level	Mass of the fruit peel mix (g)	Enzyme concentration (FPU/g)
1	8	15
-1	2	3
0	5	9
1.41	9.24	17.49
-1.41	0.76	0.51

Bioethanol was produced using the sugar-rich hydrolyzate generated in the hydrolysis stage and the yeast *Wickerhamomyces* sp. UFFS-CE-3.1.2. After inoculation, the fermentation process was conducted at 30°C, 120 rpm, and 48 h in an orbital shaker[5]. The responses were the sugars, acids, and bioethanol concentrations determined by HPLC-RID. Samples were collected at 0, 12, 24, and 48 h to monitor sugar consumption and bioethanol production. CCRD was carried out using the Protimiza Experimental Design software, with a confidence level of 95% ($p < 0.05$). The results of alcoholic fermentation procedures are expressed as mean ± standard deviation.

RESULTS & DISCUSSION

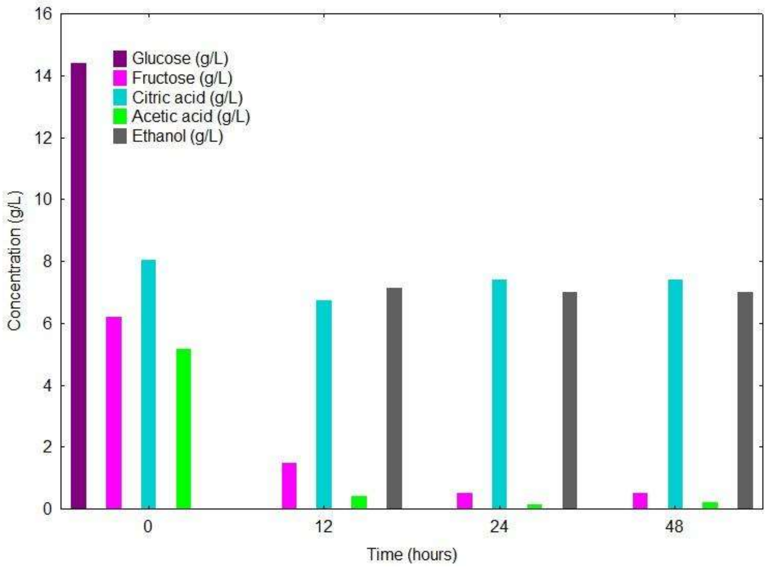
Experiment 6 of the CCRD, using the highest amount of biomass (9.24 g) and central enzyme concentration (9 FPU/g), was the one that generated a significant amount of sugars (63.90 g/L) in 120 h. According to Figure 1, it is possible to observe that the greater the mass of biomass used, the greater the production of sugars, indicating that the amount of mass of the fruit peel mix had a considerably positive influence on the yield of these compounds. Furthermore, for the enzyme concentration, little effect was observed on production, with the best result obtained with the central value of the CCRD. It is worth highlighting that experiment 6 used a lower enzyme concentration (9 FPU/g) than experiment 4 (15 FPU/g), presenting similar sugar values (63.9 and 65 g/L, respectively). This factor impacts the reduction of experimental costs due to the costs of the commercial enzyme. Mendoza et al. (2024) used a cocktail of enzymes to hydrolyze mango peel and, in 36h, obtained 81% yield, while Rodrigues et al. (2020) carried out enzymatic hydrolysis using sugarcane bagasse as a substrate and obtained 44.84 g/L of total reducing sugars with similar processes. This demonstrates that experiments like these are considerably efficient in the biotechnological field as long as the methodology is respected.

Figure 1. Variable effects, optimized with CCRD, on the enzymatic hydrolysis of fruit peel mix to produce sugars expressed by response surface.



The production of bioethanol from sugars using *Wickerhamomyces* sp. UFFS-CE-3.1.2 showed a maximum yield of 7.1 g/L in 12 hours of fermentation (Figure 2). The sugars most consumed were glucose and fructose, with the glucose concentration reaching minimum levels in the first 12 hours. Furthermore, the decrease in acetic acid occurred due to its use by yeast as a carbon source[10], while citric acid may have acted as an inhibitor of this yeast used during fermentation.

Figure 2. The concentration of compounds was monitored in fermentation with *Wickerhamomyces* sp. for 48 hours, using sugar-rich hydrolysis.



3 CONCLUSION

The results demonstrated the potential for using fruit waste to produce second-generation bioethanol. The production of sugars by optimized enzymatic hydrolysis is promising within biorefinery processes because it reduces the costs of chemical pretreatments, promoting a significant impact on large-scale production. Furthermore, the feasibility of using these fruit residues in producing bioethanol and other value-added products cooperates with the circular economy and makes it possible to mitigate environmental problems.

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