

Hydrolysis of *Typha domingensis* Cellulose: Use of a New Amperometric Glucose Biosensor Based on Layer-by-Layer Films

Flávio Rodrigues¹ , Juliana Santos Graça¹ , Natalia Reigota César¹ ,
Aparecido Junior de Menezes¹ , Marystela Ferreira¹ , Vagner R. Botaro^{1,*} 

¹ Universidade Federal de São Carlos-UFSCar, Rodovia João Leme dos Santos, Km 110, Sorocaba, CEP 18052-780, São Paulo, Brazil

* Correspondence: vagner@ufscar.br; vagnerbotaro@gmail.com; Scopus ID: 6507485048

Abstract: Search for materials and fuels from renewable sources is a widespread alternative that has been intensified over the past few years. Lignocellulosic biomass consists mainly of cellulose, hemicelluloses, and lignin, cellulose being an alternative among the current integrated biorefinery processes. The central study is based on the hydrolysis of the most important polymer produced by nature, cellulose. The controlled acid hydrolysis of cellulose results in two important components: glucose as a soluble portion obtained mainly from the amorphous regions of cellulose and nanocrystals from crystalline and organized regions of the polymer. In this context, the hydrolytic or enzymatic cellulose degradation for glucose production is a promising and current study. This work's main objective is to analyze the chemical composition of a plant studied by our research group, *Typha domingensis* (Taboa), to quantify the presence of glucose after the controlled acid hydrolysis process. Two methods of quantifying glucose in solution were employed. One important method is innovative and uses biosensors in the detection process. Among the main results, glucose in solution was properly determined by the two methods that showed a minimal percentage of errors between both. The best conditions were statistically proven, and in the best of cases, the glucose concentrations obtained were approximately 0.5 g L⁻¹.

Keywords: *Typha domingensis*; acid hydrolysis; glucose.

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1. Introduction

One of the economic segments that most generate biomass as waste is biofuels production, such as ethanol. Used to minimize dependence on petroleum-based fuels, alcohol production from sugar cane has been seen as a plausible solution. The high availability of raw material, added to the need for a renewable source to obtain fuels, opens the

opportunity for technological advances that add value to the agribusiness products and, at the same time, assist in the fixation of carbon in nature [1].

Due to the enormous variety in biodiversity, fibers have considerable dimensional and mechanical variations. It is possible to find fibers that fit the needs of desired properties. In general,

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they are biodegradable materials and have low cost [2, 3]. A large number of fibers have been used basically in two ways: "*in natura*" or after some specific modification process aiming at the physical-chemical transformation that can generate co-products of commercial interest [4, 5].

Biomass is composed of lignocellulosic materials formed basically by three groups of macromolecules, cellulose, polyoses, and lignin. These materials are available from a wide variety of sources, from the plant population, agricultural inputs, food, and as a result of other processes. So, it is a source of renewable raw material, abundant, sustainable, and easily accessible. [6]

Hydrolysis of cellulose to glucose [7] can be a key point in the process for using fibers from biomass, as this polysaccharide is easily converted into several other by-products such as biofuels [8], food, and medicines [9, 10]. Cellulose may be degraded by acid hydrolysis either heterogeneously or homogeneously. In this case, homogeneity implies that the entire structure is susceptible to degradation. Heterogeneous acid hydrolysis of cellulose is performed at a low concentration of acids. It leaves a residue of cellulose not susceptible to degradation. Some diluted acid processes use high temperatures and pressures, with reaction times from seconds to a few minutes, which

facilitates the use of continuous processes [11]. Concentrated acid processes, on the other hand, are conducted in milder conditions, but with typically longer reaction times [11]. Finally, products from the hydrolysis process and sub-processes can be valuable sources of inputs that are still little explored today, such as sugars that can be recovered or treated to obtain biofuels.

In this context, this work presents the possibility of using of hydrolyzing the cellulose contained in a fast-growing plant that is very common in Brazil, *Typha Domingenes* (Taboa) which is a material normally used in decoration pieces. Additionally, it is a continuation of a previous recently published [12]. It was possible to isolate nanocrystals (NCs) from the raw material via the hydrolysis process. Nano reinforcements were used in nanocomposite materials [12]. After obtaining the nanocrystals, the soluble portion contains glucose, and this solution was treated, and glucose was isolated and quantified. Besides, this work also allowed testing a traditional methodology based on colorimetry to quantify glucose in solution and another innovative one based on LbL biosensor, which was recently developed and published in previous works [13]. The results presented by both techniques were very close in general, and the glucose isolation was possible.

2. Materials and Methods

2.1 Materials.

Acetone, ethanol, methanol, 95% sulfuric acid, sodium hydroxide, acetic acid, ethyl ether, sodium chlorite, ethyl alcohol (Synth), and (Sigma-Aldrich) were used as received. The micropoxidase-11 (MP-11) and glucose oxidase (GOx) from *Aspergillus niger* were purchased from Sigma Aldrich and Fluka, respectively. Poly(ethylene imine) (PEI) and the phospholipids 1-palmitoyl-2-oleoyl-sn-glycero-3-phosphoglycerol (POPG) and 1,2-dipalmitoyl-phosphatidylglycerol (DPPG) were purchased from Sigma-Aldrich and Avanti Polar Lipids, respectively. Glucose (Sigma-Aldrich) solutions used to determine the biosensor response were allowed to mutarotate overnight.

2.2 Methods.

The basis of the process for the isolation of glucose is the acid/hydrolytic attack on accessible cellulose chains [12]. Part of the process leads to the preparation of NCs, and a solution is obtained as a by-product. This solution, after neutralized,

contains glucose, among other possible units of sugar from the polysaccharide. Glucose was quantified and isolated, according to Fig. 1. Details of the acid hydrolysis process are described below. A complete approach concerning hydrolysis and interesting characterization of Taboa, including studies of FTIR, SEM, and X-ray diffraction, can be found in the previous work published by our research group [12].

2.2.1. *Typha domingensis* Macrophyte Leaves.

Initially, young and senescent leaves of the macrophyte *Typha domingensis* were collected in Sorocaba – SP-Brazil. These leaves were at different stages of development and growth. The young and senescent leaves were subdivided into three regions: the upper part, the central part, and the lower part, in which experiments were carried out with the central parts of the plant.

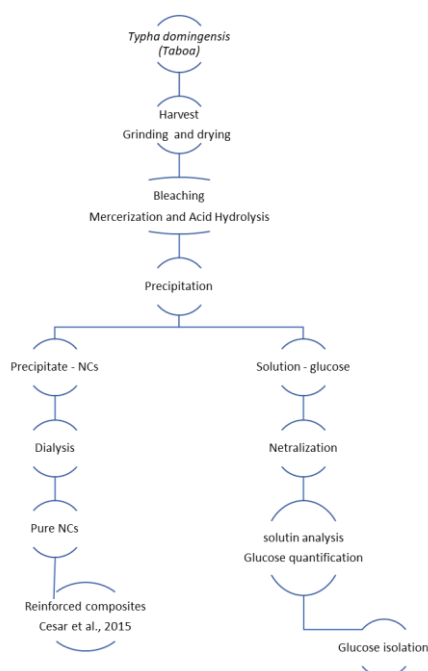


Figure 1. Illustrative flowchart representing the extraction of the cellulose NCs and the glucose-soluble solution.

2.2.2. Characterization of *Taboa*.

Typha's characterizations were previously published [12]. The contents of ash, lignin, extracts, alpha-cellulose, holocellulose, and hemicellulose were duly carried out based on known norms and established literature.

Scanning Electron Microscopy (SEM).

Fiber morphology analyzes were performed using SEM. The equipment used was the LEO 440 ZEISS / LEICA. The samples were placed in an aluminum sample holder, covered with a thin layer of conductive material (gold, Coating System MED 020 BAL-TEC).

Fourier Transform Infrared Spectroscopy (FTIR).

Analyzes were performed to identify the characteristic bands present in the samples. Tablets with KBr and fiber were prepared in the proportion of 1: 100, respectively. The analyzes were performed on a Nicolet IR 200 equipment (Thermo Scientific, USA), with a resolution of 4 cm⁻¹ and 64 scans. The spectra were obtained using the Ominic software (Thermo Scientific, USA) and were treated in Origin Pro 8.0.

Thermogravimetry (TG).

The samples' thermal stability was obtained using the TA-51 device (Shimadzu Scientific Instruments, Japan). Samples of approximately 20 mg were weighed. The measurements were carried out with a platinum sample holder, heating rate 10 °

C.min⁻¹ under nitrogen atmosphere flow 50 mL.min⁻¹ in a range of 25 ° to 1000 ° C. The initial degradation temperature (Ti) and the final degradation temperature (Tf) were determined using the DTG curves, the recorded values correspond to the temperatures at the point where the mass levels ceased to be constant (Ti) and returned to constant for Tf.

X-ray Diffraction Analysis (XRD).

X-ray diffraction analyzes were performed to evaluate the crystallinity index of the samples. The Carl-Zeis-Jena – UDR6 diffractometer was used at a speed of 1.2 ° / min, in the range of 5 ° to 70 ° (2θ). The crystallinity index (Ic) of these samples was calculated according to the method proposed by Segal and collaborators (1959) [14], according to the equation.

$$I_c = (I_{002} - I_{am} / I_{002}) \times 100 \text{ (eq 1)}$$

Where: I₀₀₂ = maximum diffraction intensity (amorphous and crystalline phase); I_{am} = diffraction intensity at 18 ° (2θ) (amorphous phase).

2.2.3. Acid Hydrolysis - Extraction of CNCs and obtaining the glucose solution.

After the bleaching step, followed by alkaline pre-treatment [12], the samples were subjected to acid hydrolysis. The methodology adopted is based on methodologies describe elsewhere: 1. CNCs from eucalyptus kraft pulp: H₂SO₄ 60 %, at a temperature of 45 °C for 30 min [15]; 2. cotton: H₂SO₄ 60 %, at a temperature of 45 °C for 75min [16]; 3. ramie: H₂SO₄ 65 %, the temperature of 55 °C for 30 min [17] and 4. blackberry: H₂SO₄ 64 %, the temperature of 60 °C for 30 min [18]. Tests for *taboa* showed that this material is less densified than the other species mentioned above. From this form, the initial studies involved concentrations of 50, 35, and 17.5% w/V sulfuric acid for degradation of cellulose chains. The previous results published by César et al. [19] have shown that the concentration of 35% is ideal since it leads to an interesting degradation of the amorphous cellulose without causing intense degradation crystalline part of the material. The mass percentage of bleached cellulose used by César and collaborators in 2018 was 17.5% in relation to the acid solution under constant agitation at 80 ° for 17 minutes. After the reaction period, the solution was centrifuged at 4000 rpm for 15 minutes, and the supernatant was stored. The samples were analyzed visually and stored in an environment with constant refrigeration from 2 ° C to 8 ° C until their eventual use. A factor analysis

was used for the statistical study of hydrolysis conditions (Table 1). Factor analysis includes the analysis of major components and common factors. It is applied when there are a large number of variables and correlated with each other, to identify a smaller number of new alternative variables, not correlated and that, in some way, summarize the main information of the original variables by finding the latent factors or variables. Thus, effects combined with this type of approach can be observed [20].

2.2.4. Treatment of supernatants obtained from hydrolysis processes.

The samples in a solution with an acid characteristic with an approximate concentration of 35% (m / v) of H_2SO_4 were subjected to neutralization with sodium hydroxide 35% (m / v) until neutral pH. The treated samples were used to determine the amount of glucose by the spectrophotometry method in the visible region using the Enzymatic-Colorimetric test with glucose oxidase, detailed in the item below.

2.2.5. Glucose determination method using the Enzyme-Colorimetric test.

Ready-made kits from BioTécnica ref 10.008.00 were used to determine the amount of glucose in the samples, which provide the reagents already buffered and with the necessary conditions for the test.

2.2.6. Glucose calibration curve using an Enzyme-Colorimetric test.

Since the colorimetric reagent's sensitivity is 0.003 g of glucose per liter of solution, and its coefficient of variation is between 1.5% and 3%, it was possible to work with minimum concentration values. These solutions were prepared from solid glucose, weighing in an analytical balance the mass required for one liter in a volumetric flask up to a concentration of 1 g.L⁻¹. From this point, all subsequent concentrations were obtained by diluting the solution to 1 g.L⁻¹.

3. Results and Discussion

3.1. Characterization of Taboa.

Fig. 3 shows Taboas's planting that gave rise to the raw material used in the treatments and obtaining glucose.

Chemical characterizations of Taboa were previously published by Cesar et al., [12], and include, among others: the contents of Ash, Moisture, Water-soluble extracts, Organic solvent-

2.2.7. Preparation of the LbL film of (PEI/MP-11)₂/(PEI/GOx + POPG+DPPG)₁.

Another method for the quantification of glucose is using LbL film as the modified electrode. The LbL films were assembled onto ITO-coated glass substrates (indium-tin-oxide). The liposome-encapsulated GOx solutions were prepared as described elsewhere [13]. Here, GOx was encapsulated into a mixture of DPPG + POPG (1:4) (w/w) liposome. The final architecture of the biosensor (PEI/MP-11)₂/PEI/(GOx + POPG + DPPG)₁ was used in the determination of glucose. More details of the LbL film is described by Graça et al. [13]. Briefly, Fig. 2 shows the idealized structure of the glucose biosensor.

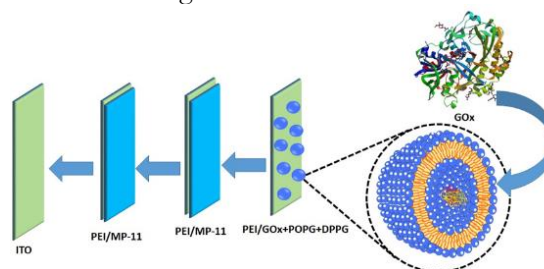


Figure 2. Idealized structure of the glucose LbL ITO/(PEI/MP-11)₂/PEI/(GOx + POPG + DPPG)₁ biosensor.

2.2.8 Procedure used to detect glucose using LbL film.

Amperometric measurements were carried out in a 3-electrode electrochemical cell (10 mL), at room temperature, using an Autolab PGSTAT 30 (Echochemie). A platinum sheet (1.0 cm²) was used as the counter electrode, and the LbL-modified ITO substrates were used as the working electrode. All potentials were referred to as a saturated calomel electrode (SCE). For this, 7mL of sodium phosphate buffer 10 mmol L⁻¹, pH 6.3 was used as an electrolyte solution. The potential was fixed at 50 mV versus SCE. After stabilizing the current, 1000 μ L aliquots of sample 1 were added under stirring [13].

soluble extracts that were approximately 8.4; 8.3; 25.2 and 11.5% respectively. Percentages of insoluble and soluble Klason lignin, alpha-cellulose, and polysols that presented values of approximately 21.1; 50.0 and 11.5% respectively [12].



Figure 3. Taboas's planting at Sorocaba – Brazil.

The calculated levels of α – cellulose, hemicellulose, and Holocellulose are $50.16\% \pm 1.09$, $11.48\% \pm 0.44$ and $6.64\% \pm 0.65$, respectively, which are by the values obtained by Khider et al. [21]. The differences in the literature between the levels obtained depend on factors such as differences in climatic and geographic parameters, temperature, altitude, wind direction, rainfall, and type of soil. Many of these factors can impose limitations on plant growth due to the ability to supply nutrients, water, and oxygen needs. Mechanical damage and pest attacks influence the concentration of nutrients in vegetables [22-24].

SEM analysis.

The micrographs of the fibers have content of roughness and superficially aggregated solids (Fig. 4). Into fiber, there are present polysaccharides of low molar mass, inorganic impurities (minerals), proteins, alkaloids in addition to lignin, hemicellulose, and cellulose [3, 12].

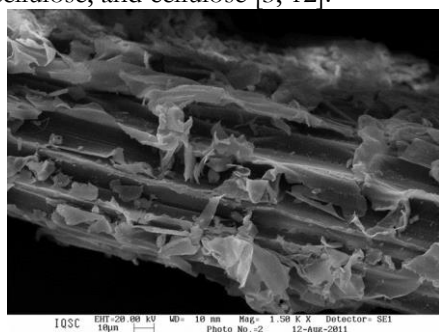


Figure 4. SEM micrographs of the fibers of Taboa with a magnification of 1500x.

FTIR analysis.

Taboa leaves were analyzed by FTIR (Fig. 5). The main groups present in the macromolecular structures that make up the vegetative tissues were observed.

Among the countless bands, some deserve attention: γ OH intra and intermolecular - cellulose and hemicelluloses 3433; γ CH - cellulose, triglycerides, esters, fatty acids, resinous acids, and sterols 2927; γ asymmetric CH₂ - hemicelluloses,

triglycerides, fatty acids, resin acids and sterols 2850; vibrations of C = O of acetyl groups and COOH groups. C = O γ in non-conjugated carbonyl ketones and other ester groups (often originated from carbohydrates) conjugated aldehydes, carboxylic acids, and triglycerides. 1731 1730; water adsorption 1637 1639 and vibration of the aromatic ring present in lignin 1511 cm⁻¹.

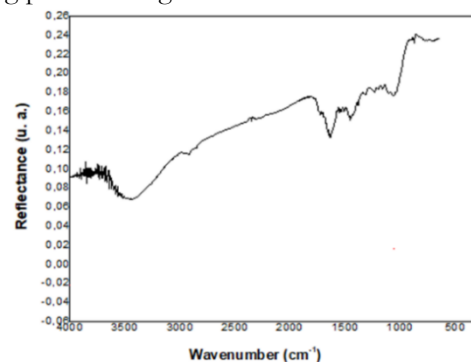


Figure 5. FTIR of Taboa leaves.

An even more complete FTIR analysis of Taboa was pointed out by César et al. [12].

X-ray diffraction.

The x-ray diffraction technique made it possible to determine the crystallinity indexes of the cellulose present in Taboa's leaves. The diffractograms are shown in Fig. 6.

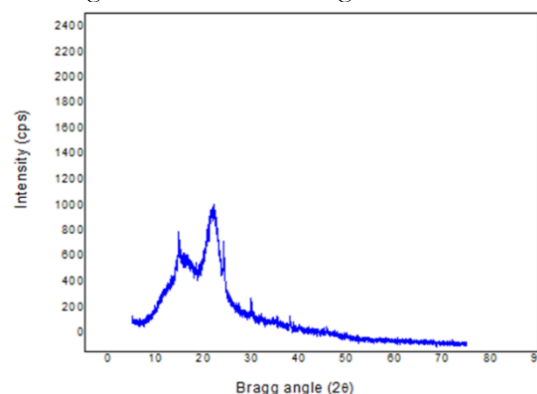


Figure 6. Diffractograms of Taboa.

The crystallinity index provides information on the proportion of crystalline regions and amorphous regions in the cellulose structure [12]. The 2 θ peaks of approximately 15°, 22°, and 34.5° indicate the presence of type I cellulose and are attributed to the planes (101), (002), and (040), respectively [16]. After obtaining the X-ray diffractogram, the crystallinity index was calculated and presented a value of 51.9%. This result is in agreement with others published in the literature [12]. The crystallinity index value around 50% is interesting because it shows that part of the cellulose is interesting for preparing cellulose nano

reinforced, as shown by César et al. [12]. The other, non-crystalline part was successfully used to obtain of glucose.

3.2. Glucose detection and determination

3.2.1. Colorimetric Method.

Standard solutions were prepared by diluting a 1 g L^{-1} concentration solution. The diluted standard glucose solutions' absorbance measurements gave rise to the calibration curve (Fig. 7). The absorbance results obtained resulted in a curve determined by the equation $y = 3.231x + 0.0004$ and with an excellent correlation factor R^2 of 0.9967, which attests to the method's reliability.

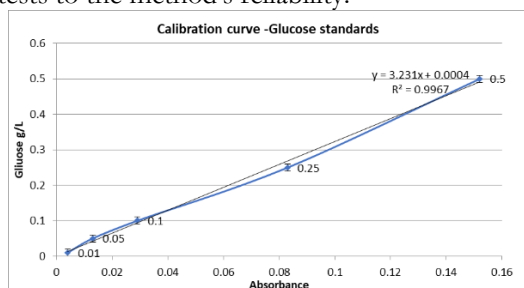


Figure 7. Calibration curve and equation obtained from colorimetric measurements for glucose quantification.

Detection limit $83.8 \pm 0.016 \mu\text{mol L}^{-1}$.

3.2.2. Calibration curve for glucose detection using LbL film.

A calibration curve was also constructed to perform the glucose concentration calculations using the biosensor method (Fig. 8). In the same way, as for the calibration curve obtained by the colorimetric method, the correlation factor obtained was by the biosensor method and showed the reliability of the method.

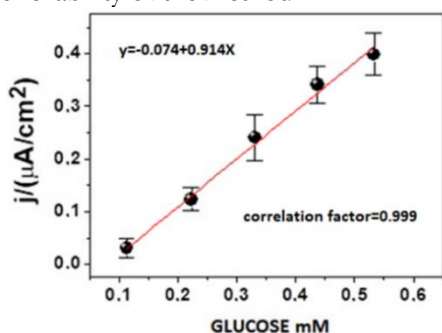


Figure 8. Glucose concentration calibration curve x

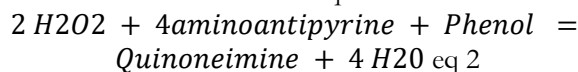
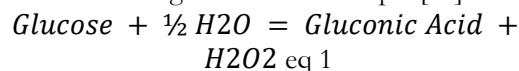
Current density used to quantify glucose using the biosensor method. Detection limit $8.6 \pm 1.1 \mu\text{mol L}^{-1}$.

3.2.3. Determination of glucose concentrations in solution. Values obtained by the colorimetric method.

Samples with a pH close to 7 were analyzed to aid the calibration curve constructed for this method (Fig. 7). Typical absorbances at a

wavelength of 505 nm were determined for each sample after the necessary dilutions.

Glucose oxidase catalyzes the oxidation of glucose to gluconic acid and hydrogen peroxide. Through an oxidative coupling reaction catalyzed by peroxidase, the hydrogen peroxide formed reacts with 4-aminoantipyrine and phenol, forming a red complex (quinoneimine), whose absorbance measured in 505 nm is directly proportional to the concentration of glucose in the sample [25].



Having chosen the colorimetric method for sugar quantification, the absorbance x concentration curve was made for samples of known values, to obtain the equation that determines the generated curve and thus the concentration of the unknown samples.

Table 1 shows the experimental conditions and the concentrations obtained for the glucose solutions. The values for glucose concentration presented are the result of an average in triplicate for each condition evaluated. Also, 2 experiments were performed in triplicate for each one (EXP A and EXP B, Table 1) to increase data reliability.

An analysis of Table 1 shows that some conditions are extremely favorable for the extraction of glucose. The conditions that used the highest acid concentration led to more hydrolyzed samples, emphasizing conditions 8A and 8B and showed the highest glucose yields. On the other hand, conditions such as 5A and 5B show no glucose sample was detected in the solutions. The conditions of extreme minimums with the lowest temperatures, times, and concentrations of reagents explain the failure to obtain glucose for experiments 5A and 5B. The use of lower temperatures and acid concentrations and shorter times did not lead to the formation of measurable glucose in the samples. According to the literature [26,27], low concentrations and insufficient hydrolysis times do not even favor the degradation of the amorphous part of cellulose. Factorial planning is fundamental for understanding the influence of variables on the hydrolysis process of cellulose chains present in the structures of Taboa.

3.2.4. Factorial Planning.

K factors were considered, all of them with two levels. Levels can be quantitative or qualitative.



A complete repetition has 2k experimental units. As these experiments only have two levels of each factor, they provide the least number of treatments; thus, they are widely used to select important factors and which may be used in a future large-scale experiment, for example. For this planning, the main parameter to be understood is the final yield of a chemical reaction varying the temperature, time, and / or varying the reagent concentration.

Statistical treatment applied was based on the dependence of the concentration of glucose in solution with three primary factors: temperature, time, and concentration of the reagent. With that, the complete factorial planning started to require the performance of $2^3 = 8$ tests. Levels 40°C and 80°C were adopted for temperature and reagent concentration of 17.5% and 35%. The list of these combinations, called the planning matrix, is presented in Table 2, together with the results obtained in the experiments carried out in duplicate. The planning matrix lists the tests in standard order with a total of k factors [20].

T-test was used to show significantly different from zero. At the 95% confidence level, the t-value corresponding to 8 degrees of freedom is 2.306. In our analysis, this means that we will only consider an effect whose absolute value exceeds $(0 + 2.306 \times 1.1) = 2.37$ to be statistically significant. Then, by analyzing the statistical significance of the data in Table 2, it appears that all the main effects are significant, and the effects of interaction between temperature, time, and concentration are also significant. The existence of a significant interaction effect indicates that the main effects must be interpreted together (Fig. 9).

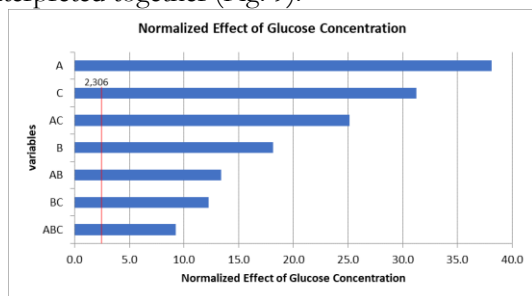


Figure 9. Pareto graph for determining glucose concentrations using colorimetric methods. (A) Temperature, (B) Time and (C) Concentration

It can be verified by the analysis of Figure 9 that the three variables used experimentally in the hydrolysis processes, (A) -Temperature, (B) -Time

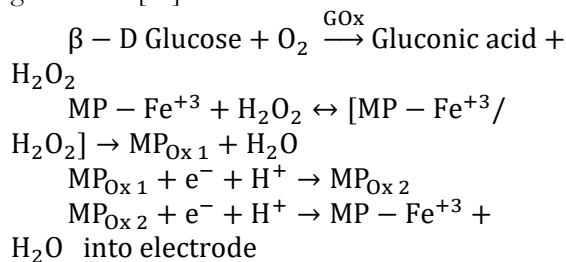
and (C) -Concentration, positively influenced the obtaining of glucose in general. The factors temperature and concentration of sulfuric acid used in hydrolysis have a high influence on the yield obtained in the processes of obtaining glucose. These results presented in Figure 5 are important and decisive for future projections on the possibility of obtaining glucose via hydrolysis at scales higher than those used in this study.

3.2.5. Glucose concentration using LbL film.

Glucose was detected using amperometric measurements, and the electrode ITO modified LbL film. After stabilizing the current, 1000 μL aliquots of sample 2A from the experiment presented in Table 1 were added under stirring.

Reaction mechanism and function of the glucose biosensor.

The mechanism of glucose detection by the biosensor consists of two stages: i) catalysis of β -D-glucose by GOx producing gluconic acid and H_2O_2 , ii) catalysis of H_2O_2 by MP-11e electrical signal generation [13].



Where: $\text{MP}_{\text{Ox}1}$ and $\text{MP}_{\text{Ox}2}$ are the two intermediate forms of MP-11.

Sample 2A was analyzed three times, with three biosensors made by the same process. Fig. 10 shows only an example of a typical curve obtained for the chosen sample. The current densities used in this case are specified in the figure legend.

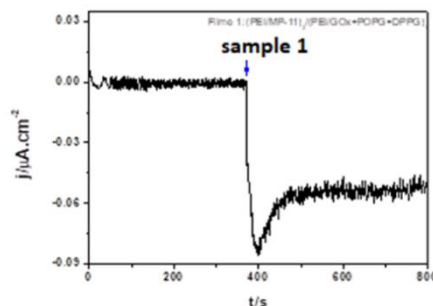


Figure 10. Example of Film analysis showing variation in current x time density ($0.0559 \mu\text{A} \cdot \text{cm}^{-2}$).

The average of the results obtained was $0.0546 \mu\text{A} \cdot \text{cm}^{-2}$, which according to the equation provided by the calibration curve $y = -0.0739 + (0.9136x)$

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(Figure 8), provides the concentration of 0.1406 mM glucose. Considering dilution factors and molar mass of glucose that is 180.16 g mol⁻¹, glucose concentration in the initial sample is 0.4053 g L⁻¹.

It is the possible notice that concentrations obtained by both methods, the colorimetric obtained by an average between experiments A and B in Table 1 and by the biosensor were 0.4208 and 0.4053 g L⁻¹ of glucose, respectively. These results are promising and very close, showing a difference of less than four percent between the two methodologies. Thus, the new methodology based on the use of a biosensor was evidenced since the result obtained is very close to the result originated by a traditional method and widely disseminated and accepted in the literature and based on the

colorimetric determination. Even being used in a specific environment for which it was developed, the latter presented a precise result. The colorimetric methodology was created to be used in blood serology. However, it did not present deviations in the tests reproduced in the laboratory when applied in a vehicle other than the blood serum. The colorimetric methodology was adopted because it is easy to access, low cost, and very accurate and can be used in the most varied installations. In contrast, the methodology through the biosensor requires a more sophisticated installation. However, with a more accurate detection limit and an entirely new methodology, it can also be applied in countless possibilities.

Table 1. Conditions for obtaining glucose and concentration in the samples for the two experiments performed.

Sample EXP A	Temperature (°C)	Time (min)	Concentration H ₂ SO ₄ (%)	Absorbance	Glucose concentration g L ⁻¹
1A	80	20	17.5	0.015	0.0977
2A	80	20	35	0.06	0.3885
3A	80	40	17.5	0.03	0.1947
4A	80	40	35	0.145	0.9378
5A	40	20	17.5	0	0
6A	40	20	35	0.005	0.0331
7A	40	40	17.5	0.003	0.0202
8A	40	40	35	0.017	0.1106
EXP B					
1B	80	20	17.5	0.011	0.0719
2B	80	20	35	0.07	0.4531
3B	80	40	17.5	0.026	0.1688
4B	80	40	35	0.143	0.9249
5B	40	20	17.5	0	0
6B	40	20	35	0.004	0.0266
7B	40	40	17.5	0.002	0.0137
8B	40	40	35	0.015	0.0977

Table 2. Planning matrix and the yields for experiments.

Factors		Lower limit (-)		Upper limit (+)		
A	T(°C)	40		80		
B	Time (min)	20		40		
C	C (g L ⁻¹)	17.5		35		
Test	A	B	C	Yield		Average
1	-	-	-	0	0	0
2	+	-	-	0.0977	0.0719	0.0848
3	-	+	-	0.0202	0.0137	0.0170
4	+	+	-	0.1947	0.1688	0.1818
5	-	-	+	0.0331	0.0266	0.0299
6	+	-	+	0.3885	0.4531	0.4208
7	-	+	+	0.1106	0.0977	0.1042
8	+	+	+	0.9378	0.9249	0.9314

4. Conclusions

Glucose was obtained using the methodology employed. With the use of factor analysis, it was possible to determine the best experimental

conditions for the hydrolysis of Taboa. Controlled hydrolysis was conducted based on the mentioned data, which resulted in important information about



its operating conditions and performance. An innovative and unprecedented technique that uses biosensors has been developed specifically for glucose analysis. The other colorimetric technique widely disseminated by biochemical laboratories in the world was used comparatively, and the results showed reliability due to the reproducibility of

results obtained by both methods. We can conclude that there is a portion of usable sugars being discarded as waste in the process of obtaining whiskers, which can be used as raw material so that it becomes a closed process where losses and rejects are the minima possible.

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Conflicts of Interest

The authors declare no conflict of interest.

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