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Integral process assessment of sugarcane agricultural crop residues conversion to ethanol

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ABSTRACT

This work focuses a whole process assessment on post-harvesting sugarcane residues for 2G ethanol production by different saccharification-fermentation conditions at high solids loading, performed after steam explosion, alkaline and acidic pretreatments. Carbohydrate recoveries and enzymatic digestibility results showed that alkali and steam explosion pretreatments were effective for the biomass assayed. Due to a significant improvement (60%) of the glucose released by combining hemicellulases and cellulases only after the NaOH pretreatment, the most favorable process settled comprised an alkali-based pretreatment followed by a pre-saccharification and simultaneous saccharification and fermentation (PSSF). The produced ethanol reached 4.8% (w/w) as a result of an 80% conversion of the glucose from the pretreated biomass. Finally, an ethanol concentration of 3.2% (w/w) was obtained by means of a steam explosion followed by PSSF, representing a suitable start point to further develop a low environmental impact alternative for ethanol production.

1. Introduction

Towards a progressive substitution of fossil fuels for renewable energies, the use of non-food vegetable biomass for second generation (2G) ethanol production even now lacks enough economic feasibility. Accordingly, agricultural and forest lignocellulosic residues are targeted for extensive research and viability studies, as they are considered a large source of carbohydrate for chemical fuels (Sánchez and Cardona, 2008). Among them, sugarcane post-harvesting residues comprising part of the leaves, tops and trash are described as sugarcane straw or agricultural crop residues (ACR), excluding the sugarcane bagasse. This residue is an abundant lignocellulosic source in the North West of Argentine, where the sugarcane (*Saccharum* spp.) based industry is a primary driver of the economy. This activity reached 18,436,082 t of sugarcane cultivated in 2016 (Sugar harvest in Argentine, 2016), generating large amounts of residues that are partially left in the field to sustain soil quality, prevent erosion and to improve water retention. But most of them could provide an inexpensive and readily available source of lignocellulosic biomass (Sindhu et al., 2016).

For a suitable enzymatic mediated releasement of fermentable sugars from lignocellulose, the pretreatment represents a crucial step. The use of different pretreatment methods has a remarkable impact on the global ethanol production process as they substantially affect enzymatic hydrolysis rates, enzyme loading, fermentation variables and even downstream procedures. Therefore, their conditions need to be carefully considered for each type of biomass (Tomás-Pejó et al., 2011; Bermúdez Alcántara et al., 2016). For sugarcane straw biomass, the pretreatments evaluated so far included milling (da Silva et al., 2010), diluted acid (Mesa et al., 2017), alkali (Carvalho et al., 2015), microwave (Moretti et al., 2016), un-catalyzed steam explosion (Oliveira et al., 2013), extrusion (Kuster Moro et al., 2017), and sequential pretreatment with glycerol assisted ferric chloride (Raghavi et al., 2016) and combination of wet disk milling and ozonolysis pretreatments (Barros et al., 2013). Between them, steam explosion is one of the most successful and widely used methods for fractionating and enhancing the enzymatic digestibility of lignocellulose (Duque et al., 2016). Also, the usefulness of diluted acid has been appointed because it allows a high recovery of pentoses (Alvira et al., 2010), whereas alkali-based pretreatments are advantageous given that they are carried out in relatively mild conditions producing high glucose yields, low inhibitor formation plus low capital costs (Kim et al., 2016).

Once the recalcitrant structure of lignocellulose is opened, 2G ethanol production can be accomplished by simultaneous

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saccharification and fermentation (SSF) or pre-saccharification and simultaneous saccharification and fermentation (PSSF) configurations. The SSF approach allows the sugar consumption by the yeast as it is produced by enzymatic hydrolysis, minimizing the glucose accumulation and the inhibition by cellobiose (Moreno et al., 2017). Alternatively, the utilization of a PSSF scheme involves the substrate incubation with hydrolytic enzymes during a relatively short period of time (8–24 h); then, the SSF proceeds when the microorganism is inoculated. This procedure permits to overcome enzymatic hydrolysis constraints due to the different temperature optima between the enzymes (50–55 °C) and most industrial yeasts (30 °C). Moreover, PSSF has shown to be suitable for elevated solids loads since it reduces the initial viscosity of the system thus facilitating the subsequent fermentation step (Jørgensen et al., 2007).

On post-harvesting residues from sugarcane, most of the available reports have separately focused on the individual stages of the processes, disregarding their integrated effect on the final product. Therefore, the aim of this study was to perform an overall assessment of 2G ethanol production from sugarcane agricultural crop residues (ACR). Our work encompassed the evaluation of different pretreatments on ACR by analyzing both, the chemical composition and the enzymatic digestibility of the solid fractions obtained. Then, saccharification conditions were adjusted by combining last generation cellulases and hemicellulases, and 2G ethanol production was carried out at high solids loading (20%, w/v) according to two strategies, SSF and PSSF (Fig. 1).

2. Materials and methods

2.1. Raw material and pretreatments

The agricultural crop residue (ACR) samples from sugarcane were kindly supplied by EEAOC (Estación Experimental Agroindustrial Obispo Colombres). The material was cut at particle size of 10–12 cm, water-washed, air-dried at 40 °C until moisture content near 10% and stored in dry place till used. For the biomass composition analysis, a portion of the ACR was milled at 1–2 mm particle size.

Un-catalyzed steam explosion of ACR was carried out at 204 °C and 20 min in a batch unit equipped with a 2 L reactor. The working conditions were selected according to our previous optimization assays by means of response surface methodology, aimed to maximize the overall glucose yield from this substrate. Briefly: a water impregnation was carried out by soaking 125 g (dry matter) of ACR in 1.5 L of water overnight. The liquid in excess was then removed by filtration and the resulting moisture content of the impregnate raw material was ~60%. The pressure reactor was preheated at the set pretreatment temperature with saturated steam, thus less than 60 s were needed for the material to reach the working temperature. The exploded material (slurry) was recovered into a cyclone connected to the outlet of the reactor and cooled and filtered to recover both, liquid and solid fractions. Liquid fractions were analyzed for sugar content and water-insoluble solid (WIS) fractions were washed with deionized water until pH 7 and stored at 4 °C till further processing.

Acid and alkali pretreatments were carried out in 2 L bottles with 10% (w/v) of raw material as follows: aliquots of ACR involving 100 g (dry weight basis, dwb) were treated with 2% (w/v) of NaOH or H_2SO_4 in autoclave at 121 °C during 60 min. The slurry was filtered and the liquid and WIS fractions were processed as is described above. These working conditions were selected based on bibliographic data (Li et al., 2014).

2.2. Characterization of the pretreatments

Following the pretreatments, the washed water insoluble fractions (WIS) were dried at 45 °C and milled at 1 mm particle size for composition analysis. The moisture content and chemical composition were analyzed according to the methods described below (Section 2.5). Solid recovery (SR) values were estimated as the dry weight of WIS remaining after pretreatment referred to 100 g of raw material (dwb). Cellulose, hemicellulose and lignin recovery on the WIS was calculated as follows:

$$\text{Component recovery}\,(\%) = (\text{g compound in 100 g of WIS}) \times \text{SR}/(\text{g compound in 100 g of raw material}) \times 100) \quad (1)$$

The sugars content (xylose, glucose, arabinose, mannose and galactose) of the liquid fractions was determined by HPLC before and after

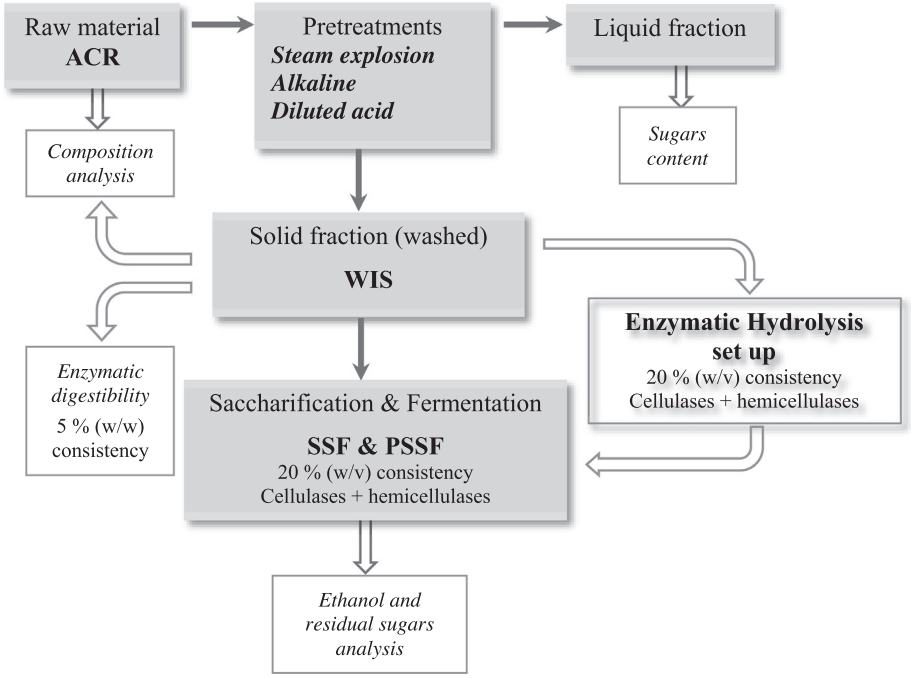

Fig. 1. Schematic flowsheet of the 2G ethanol production processes assayed and the complementary analyses performed.