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Effect of the organizational difference of corn stalk on hemicellulose extraction and enzymatic hydrolysis

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ABSTRACT

The composition units and structure of hemicellulose are more complicated and relatively more variable compared to cellulose. In addition, hemicellulose also has the possibility of being converted to high-value products. However, a variety of factors affect the extraction and transformation of hemicellulose. To study the effect of the organizational difference of corn stalk (CS) on hemicellulose extraction and biotransformation efficiency, CS was divided into three parts: leaf, bark and pith. After water and ethanol extraction, xylans were obtained by alkaline solution, which were then enzymatically hydrolyzed to produce xylo-oligosaccharides (XOS). The results showed that the highest purity of xylan extracted from pith reached 84.89%, while the lowest color value was 1.43×10^5 and the highest hemicellulose recovery ratio was 91.03%. The biotransformation results revealed that the highest enzymatic hydrolysis ratio at 40.09%, was obtained from pith, followed by leaf at 30.57% and bark with the worst ratio at 20%. By thoroughly analyzing all relevant aspects, it was determined that the corn pith was more conducive to the subsequent hemicellulose extraction and enzymatic hydrolysis to produce XOS.

1. Introduction

Corn stalk (CS) is the most representative of the lignocellulosic materials in China, with an annual output of up to 350 million tons, which accounts for about 60% of the total straw output (Liu et al., 2016). Hemicellulose is a major component of CS. Based on its complex and diverse structural properties and its physiological functions, hemicellulose has some research and application value (Carvalheiro et al., 2008), and the development of technologies for its processing and utilization has a far-reaching economic and environmental impact (Egüés et al., 2012). The extraction of hemicellulose is a prerequisite for its efficient use. Unfortunately, in the cell wall of CS, hemicellulose is covalently linked to lignin (Buranov and Mazza, 2008; Farhat et al., 2017), and tightly connected to cellulose with hydrogen bonds and van der Waals forces (Henriksson and Gatenholm, 2001), making the efficient extraction of hemicellulose more difficult (Hamzeh et al., 2013). In many hemicellulose extraction methods, the alkaline treatment saponifies the ester bond between lignin and hemicellulose, swells the cellulose at the same time, and dissolves the hemicellulose without reducing its relative molecular weight (Jin et al., 2009; Persson et al., 2009). It is established that less saccharide component degradation is more conducive to the subsequent modification of hemicellulose (Sun et al., 2000). In addition, the alkaline pretreatment showed the best performance for the xylan-exposure effect, making it suitable for the endo-xylanase reaction, thereby increasing the yield of xylo-oligosaccharides (XOS) (AkpiNar et al., 2010).

The structural differences among the various organizations of CS are very significant. CS can be divided into leaf, bark and pith according to the organization structure (Zeng et al., 2012). They are clearly different from the aspects of apparent morphology, chemical composition and density. These factors inevitably affect the extraction process (dissolution and recovery ratio of hemicellulose), which in turn affects the quality (purity, color value and branching degree) of the extracted hemicellulose. Currently, the XOS is mainly produced by enzymatic hydrolysis of xylan in lignocellulosic materials. Accordingly, the enzyme system characteristics, the xylan structural characteristics and the effect of the substrate on the enzyme activity affect the rate of hydrolysis and the composition of the hydrolyzate (Yang and Wyman, 2008; Zeng et al., 2012). For convenience of operation, the CS is generally crushed and used as raw materials for the biorefinery, ignoring the structural differences in CS, which inevitably reduces the conversion efficiency (Chen et al., 2011). It has been reported that these structural differences have significant impact in the biological or chemical transformation of different parts of CS (Huang et al., 2015).

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Recently, a research group from China divided the CS into five parts, and studied the effects of the different parts on ethanol fermentation (Li et al., 2016), acetone-butanol-ethanol fermentation (Di et al., 2016) and corn fiber composites (Luo et al., 2017), respectively. However, the effect of different parts of CS on the alkaline extraction of hemicellulose and its enzymatic hydrolysis to produce XOS have not yet investigated by any researcher. As a functional sugar, XOS show wide applications in food, health care products, chemical, animal husbandry, pharmaceutical industries and other areas (Wang et al., 2017). Nowadays, as the most representative of lignocellulosic materials, CS will be a great candidate to produce XOS (Zhang et al., 2017). So it is a worthwhile thing to investigate effect of the organizational difference of CS on hemicellulose extraction and enzymatic hydrolysis to produce XOS. Indeed, from the perspective of industrial applications, the physicochemical properties of hemicellulose, such as purity, color value, molecular weight, is a key performance indicator determining their application value. It is of great significance to extract natural hemicellulose from lignocellulosic materials. Extraction is a prerequisite for efficient utilization, extracting hemicellulose directly affects the quality of the resulting XOS. There are obvious differences between the three parts of CS, such as apparent morphology, chemical composition and density, these factors inevitably affect the extraction process (dissolution and recovery ratio of hemicellulose), which in turn affects the quality (purity, color value and branching degree) of the extracted hemicellulose. Needless to say, the quality of XOS obtained by enzymatic hydrolysis of the extracted hemicellulose is affected. Furthermore, the structural characteristics of hemicellulose can have a certain impact on enzyme activity, thereby affecting the rate of hydrolysis and the composition of the hydrolyzate (Zeng et al., 2012). Therefore, it has greatly potential value to investigate the effect of the organizational difference of CS on hemicellulose extraction and enzymatic hydrolysis. Based on the above analyses, this study evaluated the performance of the different CS parts in the processes of hemicellulose extraction and enzymatic hydrolysis, in order to determine the factors that affect the biotransformation of CS and improve the conversion efficiency of CS.

2. Materials and methods

2.1. Materials

CS (corn variety: Beijing Agricultural Science 728, Beijing Academy of Agriculture and Forestry Sciences) was collected in Fangshan, a Beijing suburb in 2016. In order to ensure the consistency of sampling, CS was from the same field, at the same time, the CS was fully mixed after harvesting and then used for subsequent experiments. First, CS was divided into three parts: leaf, bark and pith. Then, they were air dried, milled to ≤2 mm, and stored in sealed plastic bags.

2.2. Methods

Fig. 1 was the process to investigate the effect of the organizational difference of CS on hemicellulose extraction and enzymatic hydrolysis. First of all, CS was divided into three parts: leaf, bark and pith; then, removal of the non-structural components by water and ethanol, respectively; followed by extraction of xylan by 10% NaOH; finally, xylanase was used to produce XOS by enzymatic hydrolysis.

2.2.1. Removal of non-structural components of CS

CS powders and deionized water with a pH of 7 were mixed at a solid-to-liquid ratio of 1:8 (g/mL), immersed at 80 °C for 1 h. Leaf, bark and pith were treated separately. After solid-liquid separation, the pH of the filtrate was 6.91. Then, the same amount of deionized water with a pH of 7 was added, and the mixture was kept immersed at 80 °C for 1 h to dissolve the hydrophilic substance. After the secondary treatment, the pH of the filtrate was 6.95. The pretreated samples were dried in an oven at 65 °C to obtain a water-extracted CS sample.

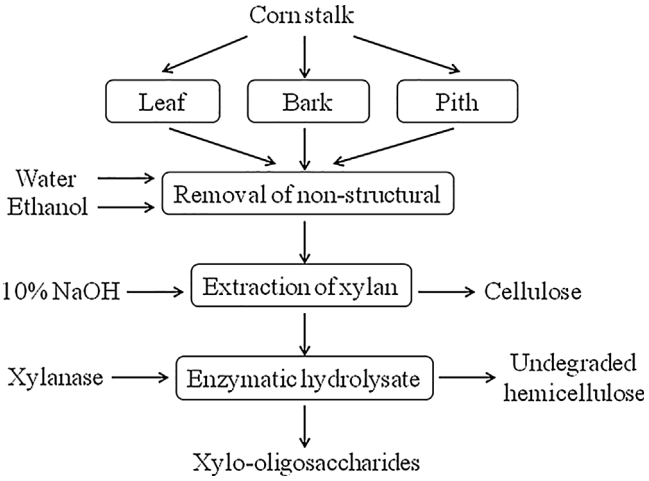

Fig. 1. The process to investigate the effect of the organizational difference of corn stalk on hemicellulose extraction and enzymatic hydrolysis.

To further remove the fat-soluble material, the water-extracted CS samples, namely leaf, bark and pith, were suspended separately in 95% ethanol (v/v), at a solid-liquid ratio of 1:5 (g/mL), and immersed at 60 °C for 1 h. After repeated treatment, the samples were placed in an oven at 65 °C to obtain the water/ethanol-extracted CS samples.

Specific methods can be found in our previously reported work (Liu et al., 2016).

2.2.2. Extraction of xylan

Xylan was extracted separately from leaf, bark and pith water/ethanol-extracted samples by the method of alkaline extraction and alcohol precipitation. Specific methods can be found in our previously reported work (Liu et al., 2016).

2.2.3. Enzymatic hydrolysis of xylan

The xylan sample (1 g) was dissolved in deionized water (50 mL), the pH of the xylan solution was adjusted to 8.0, NaCl was added to a concentration of 10 mg/mL and 1 mL 1% (w/v) xylanase solution (AU-PE89, Shandong Sukehan Bio-Technology Co., Ltd., Weifang, China) was added. Then, the enzymatic hydrolysis of xylan was carried out in a shaker at 200 rpm and 50 °C. Samples were collected at 0, 1, 2, 3, 4, 6, 8, 10, 12 and 24 h and centrifuged at 10,000 rpm for 5 min, followed by filtration through 0.45 μm polyether sulfone membrane. The monosaccharides and oligosaccharides were determined by high-performance liquid chromatography (HPLC) (Shen et al., 2016).

In this study, xylanase activity was 3.40×10^4 IU/g in accordance with the method of Bailey et al. (Bailey et al., 1992).

2.3. Analytical methods

2.3.1. Analysis of the components

The structural components of cellulose, hemicellulose and Klason lignin were determined by two-step acid hydrolysis (Li and Xu, 2013).

2.3.2. Determination of monosaccharides and oligosaccharides

XOS solution was filtered through a 0.45-μm polyether sulfone membrane (Jinteng, Tianjin, China), the contents of XOS and monosaccharides were determined using a high-performance liquid chromatography system (Agilent 1260, Agilent Technologies, Santa Clara, CA, USA) with a PL Hi-Plex-H column (300 × 7.7 mm, Agilent Technologies) at 65 °C and a refractive index detector (RID). A 5 mM H_2SO_4 solution was used as eluent with a flow rate of 0.6 mL/min. The concentrations of monosaccharides and XOS in the sugar solution were calculated by plotting the standard curves of the concentration and