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Diverse cell wall composition and varied biomass digestibility in wheat straw for bioenergy feedstock

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ABSTRACT

Wheat straw has a vast potential as feedstock for biofuel production in China. However, little information is available regarding variation in cell wall composition and enzymatic digestibility of wheat straw. This study investigated cell wall compositions and biomass digestibility of the straw of 115 wheat accessions from central China using a 1% NaOH pretreatment and mixed enzymatic hydrolysis. Significant variation in cell wall composition and sugar release was observed, with a coefficient variation (CV) ranging from 4.7% to 21.2%. Cellulose, hemicelluloses, alkali detergent hemicelluloses (ADH), and acid insoluble lignin (AIL) positively correlated with each other, and they all negatively correlated with acid soluble lignin (ASL). Hexose yields had a negative correlation with ADH and AIL, and positive correlation with ASL. No apparent undesirable correlation was found between sugar release and grain yield, thus yield and biomass convertibility can be potentially improved simultaneously. Furthermore, the features of the cell wall constitutions were compared with other plants and their implication in determining the best possible conversion strategy was discussed. This initial study is essential to understand the cell wall composition of wheat straw and to explore the potential of wheat straw as feedstock for biofuel production.

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1. Introduction

Biomass utilization is increasingly considered as a practical way of creating a sustainable energy supply while considering

a long-term protection of the environment around the world. Biofuel production from lignocellulosic feedstock, oil plants and microalgae represents a promising source of renewable energy worldwide [1-4]. Lignocellulosic biomass can be acquired from dedicated biomass crops, or from agricultural and



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forestry residuals. In China, approximately 700–900 million tons of agricultural residues are produced each year, half of which could be potential feedstock for biofuel production. Among these agricultural residues, nearly 75% of biomass resources were produced by rice, wheat and maize, three major food crops [5,6].

Because lignocellulosic biomass is made up of the complex structures of cellulose, hemicelluloses and lignin, such feedstock is highly resistant to bioconversion of its carbohydrates into ethanol, biochemical, or byproducts [7–9]. Pretreatment of lignocellulosic materials before saccharification and yeast fermentation is of great importance to break down the recalcitrance. In past years, much progress has been achieved in lignocellulose pretreatment technologies such as thermal pretreatment, acid pretreatment, lime pretreatment, oxidative pretreatment, and ammonia recycling percolation [10,11]. Despite their efficiency in unlocking cellulosic resources, most of the pretreatment technologies are energy-intensive, some of which even caused secondary environmental pollution [12,13]. Recent results have indicated that only a mild pretreatment is necessary in an industrial economically feasible system and some studies have demonstrated that dilute alkaline pretreatment is very effective for enzymatic saccharification of agricultural residues [14,15]. Much research on alkaline pretreatment focused on the optimal reaction conditions and reagent loading levels under higher temperatures or pressures for specific cellulosic biomass [16,17]. Few studies were conducted to compare the effects of the pretreatment methods on different cellulosic biomass. In essence, the lignocellulose conversion rate is determined by the wall polymer features and interaction styles such as cellulose crystallinity and lignin linking-styles [18–21]. The effect of the pretreatments largely depends on biomass composition and operating conditions [11]. The pretreatment methods should work for a wide range of lignocellulosic materials to provide a cellulosic stream that can be efficiently hydrolyzed with low concentrations of enzyme [22].

While optimizing processing techniques will initially pave the path to reduce biofuel production costs and increase ethanol yield of feedstock, further enhancement of the economics may be attained by improving feedstock quality [23]. To explore the potential of crop straw for biofuel production, it is essential to investigate the cell wall composition and capacity for producing fermentable sugars from straw of different cultivars and to estimate correlations between these traits. Recently, several studies on glucose release and stover or straw quality for cellulosic ethanol production have been published in maize and wheat [24-26]. It was found in corn stover samples that the cultivar variation of ethanol yield ranged between 45% and 73%, and it showed a strong negative correlation with lignin content [24]. Considerable differences in enzymatic digestibility were also found in wheat [25], the moderate heritability was observed for the sugar release from wheat straw that did not show an obvious adverse correlation to agronomic traits [26]. Therefore, screening wheat cultivars for less recalcitrance to enzymatic hydrolysis following pretreatment will facilitate selection of cultivars with improved biofuel feedstock of wheat straw.

China is the largest wheat producing country in the world. A total of 126.6 million tons wheat straw was harvested in 2009.

The wheat straw accounted for 15.7% of the total yield of crop residues, which would be a huge source of crop residual biomass with a vast potential as feedstock for biofuel production in China [6]. Also there is a great genetic diversity in 11,694 wheat landraces and 11,441 cultivars in China [27,28]. Although the wall polymer features of wheat straw have been recently reported as predominant factors on biomass enzymatic digestibility [29], it remains obscured about the relationships among agronomic traits, cell wall composition and biomass saccharification, due to lack of large population of wheat biomass resource. In this study, hence, we performed an initial large-scale analysis of the traits including six agronomic traits, three major wall polymers, and sugar (hexoses and pentoses) release after alkali pretreatment and enzymatic hydrolysis in 115 wheat germplasm collections from central China, and then evaluated their correlations and breeding potential.

2. Materials and methods

2.1. Plant materials

A total of 115 wheat germplasm accessions from the collection at Hubei Agricultural Science Institute in Hubei Province, China, which represent a wheat gene pool adapted to central China and Yangzi River region, were planted in the experimental farm at Huazhong Agricultural University, Wuhan, China. Field management essentially followed normal local wheat cropping practices. The lines were harvested individually at maturity to prevent seed pollution. The mature stem tissues were collected and dried at 50 °C after inactivation at 105 °C for 10 min. The dried tissues were ground and sieved to less than 40-mesh and stored in a dry container until use.

2.2. Determination of biomass digestibility

2.2.1. Alkali (NaOH) pretreatment

Alkali (NaOH) pretreatment and enzymatic hydrolysis of the residues were performed as previously described by Huang et al. [30] with minor modification.

NaOH pretreatment: The well-mixed powder of biomass sample (0.3 g) was added with 6 mL 1% (w/v) NaOH, shaken at 150 rpm for 2 h at 50 °C, and centrifuged at 3000 g for 5 min. The pellet was washed three times with distilled water, and stored at -20 °C for enzymatic hydrolysis. All supernatants were collected for determination of total sugars released, and samples with 6 mL distilled water were shaken for 2 h at 50 °C as control. All samples were carried out in biological triplicate.

2.2.2. Enzymatic hydrolysis

The pretreated biomass samples were used for enzymatic hydrolysis as described by Xu et al. [21] with minor modification. The remaining residues from various pretreatments were washed 2 times with 10 mL distilled water, and once with 10 mL mixed-cellulases reaction buffer (0.2 M acetic acid–sodium acetate, pH 4.8). The washed residues were incubated with mixed-cellulases (containing β -glucanase $\geq 2.98 \times 10^4$ U and cellulase ≥ 298 U and xylanase $\geq 4.8 \times 10^4$ U from Imperial Jade Bio-technology Co., Ltd) with a final enzyme concentration of 1.6 g/L for the biomass samples harvested from field or 0.8 g/

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