



# On-site measurement and modeling of rheological property of corn stover hydrolysate at high solids content



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## ABSTRACT

Hydrolysis of lignocellulose at high solids content is the prerequisite condition to obtain high ethanol titer broth and reduce the distillation cost. Design of large scale bioreactors requires the determination of rheological properties of the hydrolysate, but regular rheometers are no longer applicable to high solids content hydrolysate feedstocks. This study reported an on-site method using the torque meter equipped on hydrolysis reactors for its rheological property measurement. The measured torque data were transformed into the apparent viscosity and then the rheological parameters of the power law model after correlation. The determined parameters were applied to develop the computational fluid dynamics (CFD) model for simulating mixing efficiency and power consumption. This study provided a practical method for measurement of rheological parameters and design of optimal structure of large scale bioreactors of high solids content hydrolysis system.

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## 1. Introduction

Design and structure analysis of hydrolysis reactors on lignocellulose hydrolysis require characterization of its rheological properties of the hydrolysate system [1–3]. In practical applications, high feedstock solids are loaded into bioreactors for the purpose to obtain high fermentable sugar containing sugars or high ethanol titer fermentation broth [4]. Generally, rheological property of lignocellulose hydrolysate is measured using rheometer. The vane style rheometer were the most commonly used machine for measurement of rheological properties of lignocellulose hydrolysate and these high viscous slurries showed obvious shear thinning properties [5–10]. The plate style rheometer and helical ribbon agitated rheometer were also used for measurement of rheological property at high lignocellulose solids content hydrolysate and the apparent viscosity and yield stress increased with increasing solids concentration [11–13]. On the other hand, special methods such as magnetic resonance imaging (MRI) method coupled with pressure drop measurement [14] was used to measure the yield stress of pretreated corn stover. These rheometer based measurement of rheological properties are well applied to

the low solids content hydrolysis systems, but not accurate for the hydrolysate containing considerably high solid lignocellulose particles due to the irregular signal response [15] and particle sedimentation, especially in the early stage of hydrolysis at which the maximum power input and most intensive mixing are required.

In this study, an on-site method based on torque measurement using torque meter equipped on hydrolysis reactors was applied to the enzymatic hydrolysis of pretreated corn stover (CS) at the solids loading up to 30% (w/w). The determined torque values were transformed into the rheological parameters and then applied to establish the CFD model of high solids content corn stover hydrolysate for hydrolysis reactor design. This study provides a method for concentrated hydrolysis or fermentation system at high solid loading.

## 2. Materials and methods

### 2.1. Raw materials and reagents

Corn stover was harvested in fall 2011, Dancheng, Henan, China. 36.18% of cellulose, 19.83% of hemicellulose, and 4.98% of ash were contained in corn stover measured according to NREL protocols [16,17]. The pre-handling of corn stover included water-washed to remove the field dirt, stones and metals, then dried and milled using a beater pulverizer and screened through a mesh with the

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circle diameter of 10 mm. The pre-handled corn stover was stored in sealed plastic bags until use.

Cellulase enzyme Youtell #6 was provided by Hunan Youtell Biochemical Co., Yueyang, Hunan, China. Its filter paper activity of Youtell #6 was 135 FPU/g according to the NREL protocols LAP-006 [18], and the cellobiase activity was 344CBU/g using the method described by Ghose [19]. All chemicals including H<sub>2</sub>SO<sub>4</sub>, NaOH, and CaCO<sub>3</sub> were purchased from the local supplier Linfeng Chemical Reagent Co., Shanghai, China.

## 2.2. Pretreatment and Enzymatic Hydrolysis Operations

Corn stover was pretreated using dry dilute sulfuric acid pretreatment (DDAP) according to Zhang et al. [21] and He et al. [20]. Briefly, dry corn stover and 5% (w/w) sulfuric acid solution were co-currently fed into the pretreatment reactor at a solid/liquid ratio of 2:1 (w/w) under helically agitated mixing at 50 rpm, and then pretreated at 175 °C for 5 min. The solids content of the pretreated slurry was around 50% (w/w) and no wastewater was generated. The pretreated materials were cooled down and stored at ambient temperature (18–25 °C) until use.

The enzymatic hydrolysis was carried out in a 5 L bioreactor with the inner diameter of 166 mm and the height of 240 mm equipped with a helical impeller as described in our previous study [22]. Pretreated corn stover was fed into the bioreactor to reach the solids content of 30% (w/w) by adding distilled water at the cellulase dosage of 15 FPU/g DM, 50 °C for 12 h. The constant pH of 5.0 was maintained by automatic addition of 5 M NaOH solution.

## 2.3. Apparent viscosity measurement using rheometer

5 mL of hydrolysate sample was withdrawn from the bioreactor periodically and boiled for 5 min to inactive enzyme activity. Then the apparent viscosity of the sample was measured using an Anton Paar MCR501 rheometer (Ashland, VA, USA) with parallel plate geometry. Samples were pre-sheared 1 min at 100 s<sup>-1</sup> shear rate, and then determined apparent viscosity at different shear rates for fitting rheological parameters.

## 2.4. Apparent viscosity measurement using on-site method

Mixing torque of the pretreated corn stover and high solids content hydrolysate on the same 5 L bioreactor was measured using a torque meter (HX-901, Huaxin Mechanical and Electrics Co., Beijing, China) installed on the bioreactor. The torque data were recorded every 5 s and lasted for one min at different agitation rates (50–150 rpm) for pretreated corn stover and the specific agitation rate of 150 rpm for hydrolysate slurry during enzymatic hydrolysis. The zero torque value  $T_{i0}$  was defined as the torque data in the empty bioreactor without feedstock loading. The torque value with full loading of feedstock was recorded as  $T_i$ . The mixing torque value was defined as  $M = T_i - T_{i0}$ .

The calculation method of apparent viscosity  $\eta_a$  was referred to Zhang et al. [23]. Briefly, the dimensionless power number  $N_p$  is correlated to Reynolds number  $Re_m$  as  $N_p = C \times Re_m^x$ , where  $C$  is a geometry parameter of the reactor used and independent of the fluid properties,  $x$  is a dimensionless factor with the values of  $C = 147.12$  and  $x = -1$  for the reactor used [23].  $Re_m$  is calculated by  $Re_m = \frac{\rho Nd^2}{\eta_a}$  where  $\rho$  is the density of the fluid (kg/m<sup>3</sup>) which was measured by a graduated cylinder,  $N$  is the impeller rotation rate (rev/s),  $d$  is the impeller diameter (m),  $\eta_a$  is the apparent viscosity (Pa s). Then  $N_p$  can be represented as  $N_p = C \times Re_m^x = \frac{147.12\eta_a}{\rho Nd^2}$ .

On the other hand,  $N_p$  can be calculated in a stirred vessel by  $N_p = \frac{P}{\rho N^3 d^5}$  according to Metzner et al. [24], where the power consumption  $P$  (W) is represented as  $P = 2\pi NM$ . Combining the two  $N_p$

expressions together to give the working equation for calculating the apparent viscosity  $\eta_a$  using the measurable parameters  $M$ ,  $N$ , and  $d$ :

$$\eta_a = \frac{2\pi M}{CNd^3} = \frac{\pi M}{73.56Nd^3} \quad (1)$$

## 2.5. CFD modeling

A commercial grid-generation tool, ICFM CFD 14.0 (Ansys Inc., PA, USA) was used to generate the 3D grids of the reactor model in Autodesk Inventor 2014 (Autodesk Inc., CA, USA) for running Fluent 14.0 (Ansys Inc., PA, USA). The multiple reference frame approach was used to the helical ribbon agitated reactors. The upper surface of the liquid was defined as symmetry boundary, and a non-slip wall condition was used for both the walls and the surface of the impellers. The model was performed under laminar flow condition and chemical reaction was negligible. The material used was assumed to be incompressible, non-Newtonian fluid and the rheological properties were correlated by power law model. The tracer was same in physical properties as the background fluid and at the moment of injection the mass fraction of tracer in the injected area equals 1 while at all other areas it equals 0. Convergence was achieved when all normalized residuals reached values smaller than 10<sup>-4</sup>.

## 3. Result and discussion

### 3.1. Determination of rheological properties of pretreated corn stover hydrolysate

The enzymatic hydrolysis of the dilute sulfuric acid pretreated corn stover at the solids loading of 30% (w/w) was carried out for 12 h (Fig. 1). Glucose and xylose concentrations (Fig. 1a) rapidly increased, while insoluble solids content (Fig. 1b) and particle size (Fig. 1c) obviously decreased with hydrolysis processes. For rheological property measurement, two methods described in the Methods section were conducted. The regular method was conducted by taking hydrolysate samples from the hydrolysis reactor and measuring on rheometer. The samples of the pretreated corn stover materials for rheometer measurement were taken only after 30 min of hydrolysis because the initial hydrolysate samples contained too many solids and were not able to use rheometer. Instead, an on-site method using torque meter equipped on the reactor was tested by measuring mixing torque value of hydrolysis reactor and then transformed into the values of apparent viscosity according to the working equation Eq. (1) (Fig. 2). No sampling was required thus the measurement started from the very beginning to the end of the hydrolysis. The apparent viscosity values determined by the on-site method well agreed to the data measured using the rheometer in the late stage of hydrolysis when larger fiber particles hydrolyzed (Fig. 2). At the early stage of hydrolysis (0–2 h), the apparent viscosity values between the two methods showed difference due to the inaccuracy of rheometer on the early stage samples with high solids content. The results suggest that the on-site method for apparent viscosity measurement was applied to the high solids content hydrolysis system and could be used as general method in the similar system.

The apparent viscosity results determined by the two methods (Fig. 2) show that the apparent viscosity of pretreated corn stover decreased significantly at the early stage of hydrolysis (first one hour in the present hydrolysis condition), then changed slowly with the hydrolysis processes. The tendency of apparent viscosity did not exactly correspond to the changes in sugar generation (glucose and xylose) and water insoluble solids (WIS) (Figs. 1a and b), but did correspond to the particle size distribution in which the size

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