



## Short Communication

# Modeling laccase-induced lignin removal in prehydrolysis liquor from kraft-based dissolving pulp production



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## HIGHLIGHTS

- Modeling of lignin removal during laccase treatment of PHL was developed.
- Lignin removal can be divided into first fast phase and second slow phase.
- Dissolved organic materials were negligibly affected by laccase treatment.

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## ABSTRACT

Laccase treatment is a promising approach to remove lignin from prehydrolysis liquor (PHL) for value added utilization of hemicellulose rich waste streams. Modeling the lignin removal process is of practical interest for prediction and control of laccase treatment of PHL. The present study focused on the lignin removal through variation of laccase charge and treatment time. Results showed that the lignin removal may be divided into two phases, i.e. a fast initial phase followed by a second slow phase. A kinetic model based on the experimental results was developed, which can be used to predict the lignin removal of PHL during the laccase treatment.

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## 1. Introduction

Historic utilization of renewable materials, lignocellulosics in particular, for the production of bio-materials and bio-energy is an effective way to address the depletion of fossil sources (Ragauskas et al., 2006). The integrated forest biorefinery concept is an ideal model that agrees with this strategy (van Heiningen, 2006). One of its examples can be found in kraft-based dissolving pulp production process, a commercial practice that fractionates lignocellulosic chemical components into different streams (Saadatmand et al., 2013).

Generally, the kraft-based dissolving pulp production process includes the prehydrolysis stage, cooking stage, and bleaching stage to produce desired product, i.e., high alpha cellulose content pulp (92–98%) (Martin-Sampedro et al., 2014; Miao et al., 2014). Hemicellulose, a class of “impurity” in dissolving pulp, is extracted substantially into PHL during water/steam prehydrolysis (Sixta

et al., 2013). Many valued components, such as monomeric and oligomeric sugars, acetic acid and furfural, can be found in the PHL (Wang et al., 2014). To produce value-added product from PHL, numerous technologies have been exploited (Yang et al., 2013; Liu et al., 2013; Ahsan et al., 2014). For example, solvent exchange has been used to recover acetic acid, nano-filtration was used to concentrate the sugars, both monophase and biophase systems have been used to produce furfural. However, the dissolved lignin in the PHL hampers those processes by fouling the membrane and impairing the product purity.

Several approaches have been adapted to remove the lignin from PHL. Shi et al. (2012) reported that the maximum lignin removal of 46% can be achieved when using polyethylene oxide to treatment of PHL. Yasarla and Ramarao (2012) reported that low molecular weight high charge density polymers, such as PEI and CPAM can be used to remove lignin from a wood extract of sugar maple. Shen et al. (2013) used activated carbon to adsorb lignin from PHL and found that about 90% of lignin can be removed. Subsequently, Fatehi et al. (2013) further studied the adsorption behavior of various components onto activated carbon by prepared model prehydrolysis liquor. Recently, laccase treatment has attracted

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interest to remove lignin through phenolic unit polymerization in an environmentally friendly manner (Wang et al., 2014). Similarly, Ludwig et al. (2013) reported that about 82% of phenolic compounds have been reduced using immobilized laccase treatment of the xylan rich fraction of the ethanol-based organosolv process.

In driving toward an industrial application, modeling is essential for the design of efficient reactors and optimization and control. The objective of this work was therefore to develop a model for laccase treatment of PHL in terms of lignin removal mainly focused on laccase charge and treatment time.

## 2. Experimental

### 2.1. Raw materials and laccase

Prehydrolysis liquor (PHL) was collected from a dissolving pulp mill in Eastern Canada based on the kraft technology using mixture wood of maple, poplar and birch.

The laccase (produced by *Trametes versicolor*) was purchased from Sigma–Aldrich. The activity was 12.0 U/mg as determined by UV at 420 nm ( $\epsilon = 36,000 \text{ M}^{-1} \text{ cm}^{-1}$ ) with ABTS (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) 0.5 mM as the substrate, as described in our previous study (Wang et al., 2014).

### 2.2. Laccase treatments

30 mL PHL was added into 250 mL Erlenmeyer flask. Then a required amount of  $30 \text{ U mL}^{-1}$  laccase solution was added into the flask, the reaction was carried out in shaker in a water bath. The treatment conditions were: laccase concentration of 1, 2, 3 and  $4 \text{ U mL}^{-1}$ , time of 0–300 min, pH of 3.6, and temperature of  $36^\circ\text{C}$ . Once the reaction was completed, PHL was centrifuged at 4000 rpm for 15 min.

### 2.3. Analytical methods

Lignin content of the PHL was determined based on a UV spectrometric method at 205 nm by following a known procedure (Schoening and Johansson, 1965). The lignin removal was calculated according to Eq. (1):

$$R = \frac{[l_0] - [l]}{[l_0]} \times 100\% \quad (1)$$

where  $R$  is the lignin removal (%),  $[l_0]$  is the concentration of lignin in the original PHL (g/L),  $[l]$  is the concentration of lignin in the laccase treated PHL (g/L).

The concentration of hemicelluloses in the samples was determined using an ion chromatography (IC) unit equipped with a CarboPacTMPA1 column (Dionex-300, Dionex Corporation, Canada) and a pulsed amperometric detector (PAD). Acid hydrolysis of samples was carried out with sulfuric acid at  $121^\circ\text{C}$  in an oil bath for 1 h (Neslab Instruments, Inc., Portsmouth, NH, USA). The sugar content before acid hydrolysis represented the mono-sugars, and the oligo-sugars were calculated from the difference with and without acid hydrolysis.

A Varian 300 NMR-spectrometer was employed for acetic acid and furfural concentrations determination as described previously (Yang et al., 2013), with a  $\text{D}_2\text{O}$  to water ratio of 1:4.

## 3. Results and discussion

### 3.1. Lignin removal by laccase treatment

Fig. 1 showed lignin removal under various laccase charges and treatment times. As can be seen, two distinct phases can be identified, i.e., a fast initial phase, followed by a second slow phase.

The majority of lignin removal occurred over the first 120 min for all laccase charges, and then the lignin removal was kept constant at a high level until 300 min elapsed. On the other hand, a higher laccase charge further improved lignin removal. It is known that the laccase induces radical cation formation, which subsequently deprotonates the hydroxyl group to give a radical. The radical formed tends to polymerize to high molecular mass products (Mikolasch and Schauer, 2009). During this process, a high laccase charge leads to increases in radical propagators, hence enhancing the probability of interaction. Similarly, Ludwig et al. (2013) reported that the content of phenolic compound decreased gradually with increases in laccase charge from  $0.32$  to  $0.8 \text{ U mL}^{-1}$  when using immobilized laccase detoxification of ethanol organosolv pretreated wheat straw.

### 3.2. Kinetic model development

As discussed above, the laccase treatment induced the lignin radical propagators, followed by polymerization. A proper generic chemical reaction equation for polymerization of lignin may be considered in Eq. (2):

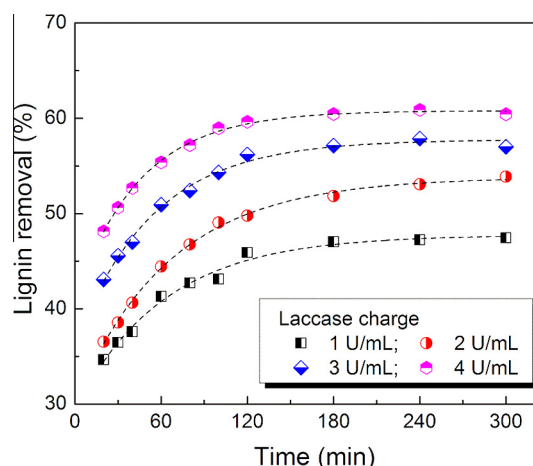
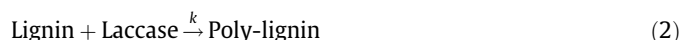


Fig. 1. Lignin removal as a function of laccase charge and treatment time.

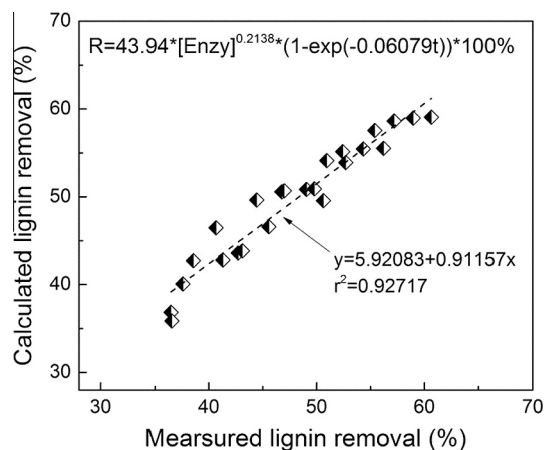


Fig. 2. Linear relationship between the calculated and the measured lignin removal of laccase treatment of PHL (Treatment condition: laccase concentrate 1, 2, 3 and  $4 \text{ U mL}^{-1}$ , treatment time of 60, 80, 100, 120, 180, 240 min).

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