

**Electronic Supporting Information:**  
**(Sustainable Energy & Fuels)**

**Laccase-Natural Mediator Systems for “Green” Synthesis of Phenolic Monomers from Alkali Lignin**

Bin Yao,<sup>a</sup> Praveen Kolla,<sup>b</sup> Ranjit Koodali,<sup>c</sup> Selvaratn Balaranjan,<sup>c</sup> Sunav Shrestha<sup>c</sup> and Alevtina Smirnova<sup>a,b,†</sup>

<sup>a</sup> Materials Engineering and Science Program, South Dakota School of Mines and Technology, Rapid City, SD, 57701.

<sup>b</sup> Chemistry and Applied Biological Sciences Department, South Dakota School of Mines and Technology, Rapid City, SD, 57701.

<sup>c</sup> Chemistry Department, University of South Dakota, Vermillion, SD, 57069.

†Corresponding authors:

Alevtina Smirnova: Tel: (860)428-0446. E-mail: [Alevtina.Smirnova@sdsmt.edu](mailto:Alevtina.Smirnova@sdsmt.edu)

The alkali lignin purchased from Sigma-Aldrich was characterized by nuclear magnetic resonance (NMR) and Fourier transform infrared spectroscopy (FTIR).

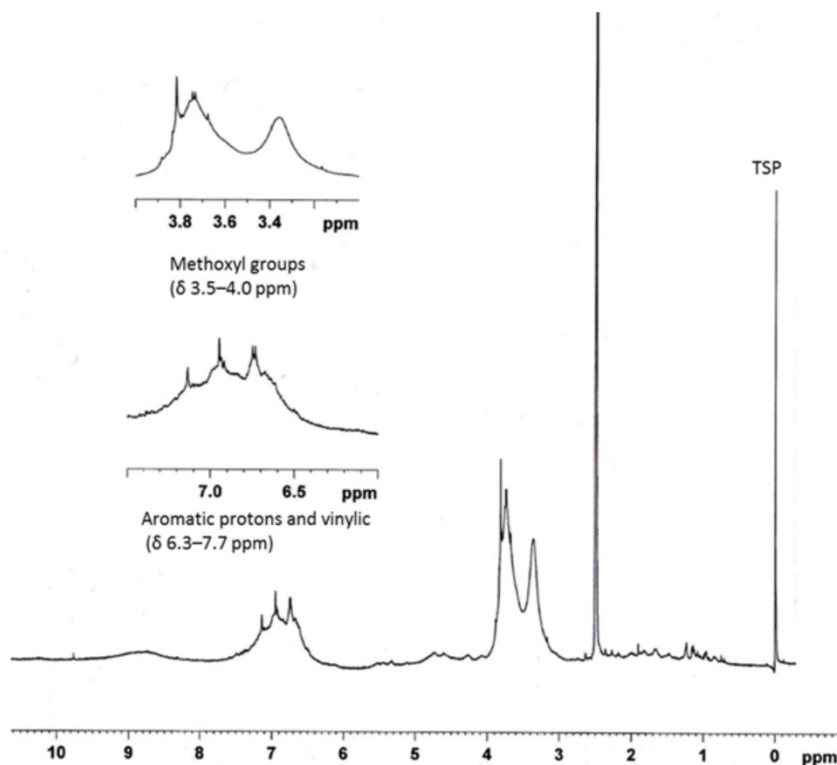


Fig. 1. The <sup>1</sup>H-NMR spectrum of alkali lignin before LMS treatment.

The alkali lignin before treatment by LMS was characterized by NMR (Bruker, 300HZ). The  $^1\text{H}$  assigned to the methyl group is at between  $\delta$  3.5 and 4.0 ppm. The protons of the aromatic and vinylic group are at between  $\delta$  6.3- 7.7 ppm.  $\text{H}_\alpha$  in  $\beta$ -O-4 structures is around 6.05 ppm.

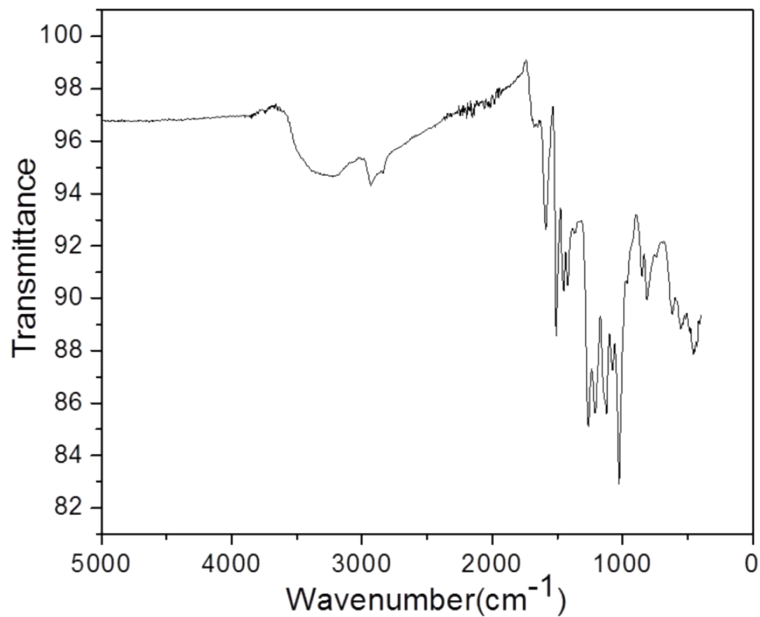


Fig. 2. The FT-IR spectrum of alkali lignin before LMS treatment.

The alkali lignin before treatment by LMS was characterized by FT-IR (Cary 660 FTIR spectrometer) from Agilent Technologies (Fig. 2). The absorption at  $3500\text{ cm}^{-1}$  is caused by the  $-\text{OH}$  stretching vibration of aliphatic and aromatic groups in the lignin. The absorption peak at  $2900\text{ cm}^{-1}$  is assigned to the stretching vibration of the  $\text{C}-\text{H}$  band of  $\text{CH}_2$ ,  $\text{CH}_3$ , and  $\text{CH}_3\text{O}$  groups in lignin. The peaks found at  $1465$  and  $1426\text{ cm}^{-1}$  are assigned to the stretching in the  $\text{Aryl}-\text{O}-\text{CH}_2$  bonds of the lignin. The peak at  $1268\text{ cm}^{-1}$  can be assigned to  $\text{C}-\text{O}$  stretching and  $\text{C}-\text{O}$ . The peak at  $1268\text{ cm}^{-1}$  belongs to  $\text{C}-\text{O}$  stretching and  $\text{C}-\text{O}$  linkage in guaiacyl aromatic methoxyl groups.

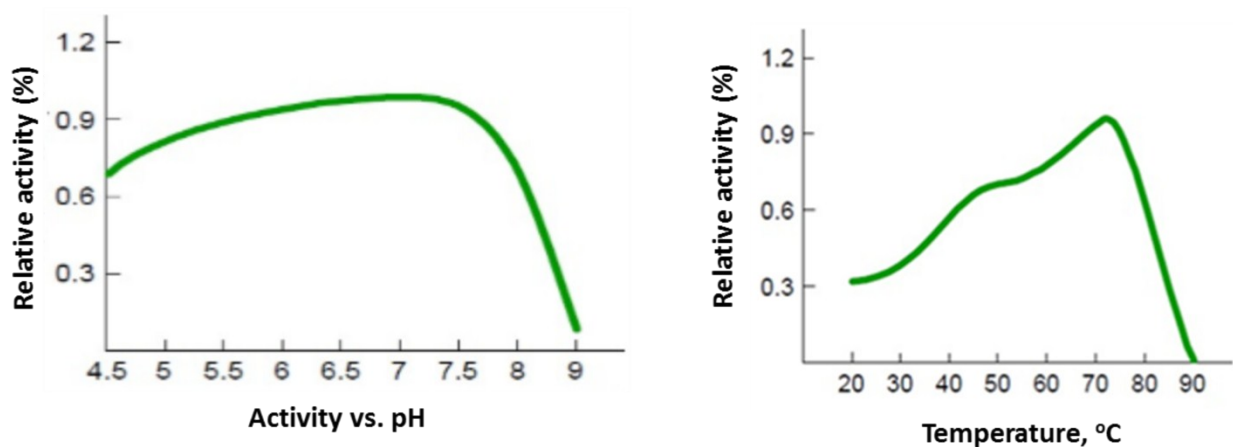


Fig. 3. Optimized pH and temperature for Laccase I (*Myceliophthora thermophila*)

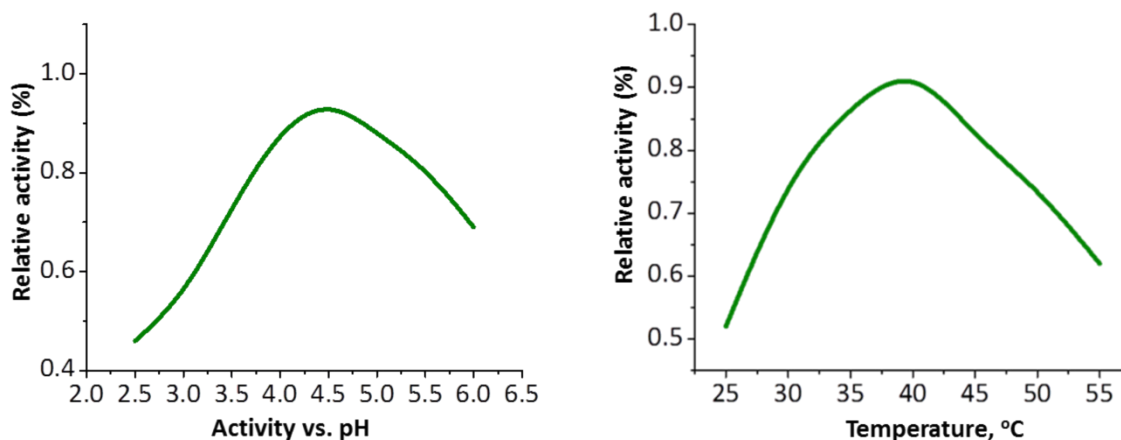


Fig. 4. Optimized pH and temperature of Laccase II (*Trametes versicolor*)

**Supplementary Table 1: Phenolic compounds identified by using pyr-GCMS in the solid-state lignin residue after treatment with LMT-MS and LTV-MS systems in comparison to the raw alkali lignin.**

Name of the compound *	Raw lignin			5 wt. % LMT-5 wt. % MS			5 wt. % LTV-5 wt. % MS		
	RT**	Area	% Area	RT**	Area	% Area	RT**	Area	% Area
Phenol, 2-methyl-	14.9	5.80.10 <sup>7</sup>	0.94	15.0	1.01.10 <sup>8</sup>	1.34	14.9	1.32.10 <sup>8</sup>	1.44
Phenol, 3-methyl-	15.8	1.29.10 <sup>8</sup>	1.47	15.9	1.00.10 <sup>8</sup>	1.32	15.9	2.01.10 <sup>8</sup>	2.05
Phenol, dimethyl-				17.4	6.34.10 <sup>7</sup>	0.55	17.8	6.34.10 <sup>7</sup>	0.98
Phenol, dimethyl-				18.0	2.10.10 <sup>8</sup>	1.88	17.9	2.38.10 <sup>8</sup>	2.77
Phenol, dimethyl-				18.1	7.89.10 <sup>7</sup>	0.66	18.2	8.21.10 <sup>7</sup>	0.48
Hydroquinone	20.3	3.87.10 <sup>8</sup>	4.43	20.4	4.88.10 <sup>8</sup>	4.47	20.4	5.56.10 <sup>8</sup>	4.78
1,2-Benzenediol, 3-methoxy-	22.5	1.98.10 <sup>8</sup>	2.01		2.12.10 <sup>8</sup>	2.34		2.52.10 <sup>8</sup>	2.88
1,2-Benzenediol, 4-methyl-								1.55.10 <sup>8</sup>	1.76
2-methoxy-4-methylphenol	24.3	2.33.10 <sup>7</sup>	0.55	24.9	0.98.10 <sup>8</sup>	1.12	25.1	1.02.10 <sup>8</sup>	1.23
Phenol, 4-ethyl-2-methoxy-				26.3	1.57.10 <sup>8</sup>	1.59	26.2	1.67.10 <sup>8</sup>	1.83
3-allyl-6-methoxyphenol				27.1	1.45.10 <sup>8</sup>	1.50	27.2	1.48.10 <sup>8</sup>	1.54
Phenol, 2-methoxy-4-(2-propenyl)-, acetate	28.8	2.76.10 <sup>8</sup>	3.12	28.8	3.34.10 <sup>8</sup>	3.87	28.9	3.12.10 <sup>8</sup>	3.33
4'-Hydroxy-3'-methoxyacetophenone				30.1	1.78.10 <sup>8</sup>	1.68	30.0	1.88.10 <sup>8</sup>	1.93
Benzoic acid, 4-(acetyloxy)-3-methoxy-, methyl ester				38.7	1.56.10 <sup>8</sup>	1.58	38.8	1.82.10 <sup>8</sup>	1.92
Benzoic acid, 3,5-dimethoxy-, methyl ester				57.9	5.56.10 <sup>7</sup>	0.43	56.7	8.76.10 <sup>7</sup>	0.99
[1,1'-Biphenyl]-4,4'-diol, 3,3'-dimethoxy-	58.9	8.18.10 <sup>7</sup>	1.11						
1,1'-Biphenyl, 6-hydroxy-2',3',4'-trimethoxy-	60.3	4.23.10 <sup>7</sup>	0.76						

\*The names of compounds are based on the identification using the MS NIST library; RT\*\* - Retention time.