Electronic Supplementary Information

Effect of cellulase on the UCST behavior of sulfobetaine zwitterionic surfactant and cellulase recovery mechanism

Feiyun Li,^a Feiyang Qin,^a Cheng Cai,^a Yuxia Pang,^{*a} Weifeng Liu,^a Qiong Li,^a Hongming Lou,^{*ab} and Xueqing Qiu^c

^a School of Chemistry and Chemical Engineering, Guangdong Provincial Engineering Research Center for Green Fine Chemicals, South China University of Technology, Guangzhou 510641, Guangdong, P. R. China

^b State Key Laboratory of Pulp and Paper Engineering, South China University of Technology, Guangzhou 510641, Guangdong, P. R. China

^c School of Chemical Engineering and Light Industry, Guangdong University of Technology, Guangzhou 510006, Guangdong, P. R. China

*Corresponding author. Tel.: 86-20-87114722; E-mail: cehmlou@scut.edu.cn (H.M. Lou); ceyxpang@scut.edu.cn (Y.X. Pang).

1. Preparation of didecylmethyl sulfobetaine (DDSB)



Fig. S1 The scheme of the preparation of DDSB.

Didecylmethylamine (1.0 eq) was added to the dichloroethane solution of 1,3propane sultone (1.2 eq). The mixture was stirred at 85 °C for 5 h. The white solid was obtained after drying at 30 °C under vacuum. The purified product DDSB was obtained after removing remaining impurities by using diethyl ether. DDSB was characterized by maxis impact UHR-TOF (Bruker daltonics, Germany). HRMS: m/z(ESI), calculated $[M+H]^+$: 434.3590, measured: 434.3669; calculated $[2M+H]^+$: 867.718, measured: 867.7252.



Fig. S2. The HRMS spectra of DDSB.

2. UCST-responsive performance of sulfobetaines in acetate buffer solution



Fig. S3 The turbidity curve of SB3-16 and DDSB.

3. Peak thickness of SB3-18 in acetate buffer solution



Fig. S4 Peak thickness of sulfobetaines in acetate buffer solution at different temperatures (calculated according to the backscattering at 880 nm, relative threshold was 0.5).

4. Effect of sulfobetaines on the enzymatic hydrolysis of lignocellulose and Avicel

Avicel (PH101) with a mean particle size of 50 µm was purchased from U.S. Sigma-Aldrich Company (USA). CCR was provided by Shengquan Corp. Ltd. (Jinan,

China) and was the enzymatic residue from the production of functional sugars such as xylose and L-arabinose. CCR was treated at 120 °C for 1 h by auto-clave prior to use. The glucan and acid-insoluble lignin contents of pretreated CCR reached 87.5% and 7.6%, respectively. The condition of enzymatic hydrolysis was as follows. 0.60 g avicel or CCR, certain amounts of sulfobetaines and cellulase were added to 30 mL of the slurry of buffer solution. The enzyme load for avicel and CCR were 5 FPU/g and 10 FPU/g glucan, respectively. The enzymatic hydrolysis of the substrate was carried out at 50 °C at 150 rpm for 72 h in a shaker (DDHZ-300, Jiangsu Taicang Equipment Factory, China). The glucose concentration was monitored by SBA-40E (Institute of Biology, Shandong Academy of Sciences, China). The enzymatic hydrolysis efficiency was determined by glucose yield (SED@72h). The blank experiment was without SB3-18 or SB3-16. All experimental data was the average of three parallel experiments, the data deviations were shown in the figures.



Fig. S5 Effect of sulfobetaines on the enzymatic hydrolysis of Avicel and CCR (solid concentration, 2%, pH 5.0, ionic strength, 50 mM, cellulase loading, 5 FPU/g glucan for Avicel, 10 FPU/g glucan for CCR).

5. Effect of ionic strength and temperature on the stability of SB3-18 film



Fig. S6 (a) The effect of ionic strength on the adsorption of buffer on SB3-18 film (pH 5, 50-200 mM, 25 $^{\circ}$ C). (b) The effect of temperature on the adsorption of buffer SB3-18 film (pH 5, 50 mM, 25-45 $^{\circ}$ C).

6. Effect of ionic strength and temperature on the adsorption of cellulase on SB3-18 film

As shown **Fig. S7**, the adsorption capacities at 50 and 200 mM were 375.1 and 413 ng/cm², respectively, respectively. (In the **Fig. S7**, the adsorption capacity of



Fig. S7 The effect of ionic strength on the adsorption of cellulase on SB3-18 film (pH 5, 50-200 mM, 25 $^{\circ}$ C).

cellulase includes that of buffer, when ionic strength was 200 mM.). A part of the charges of SB3-18 and CTec2 was shielded by ionic, but the adsorption capacity of cellulase increased with an increasing ionic strength. The result indicated that the electrostatic interaction was not the main driving force for cellulase adsorption on the film. Combined the urea test, the hydrogen bond interaction was not the main driving force. In summary, we speculated that the hydrophobic interaction was the main



Fig. S8 The effect of temperature on the adsorption of cellulase on SB3-18 film (pH 5, 50 mM, 25-45 $\,^{\circ}\mathrm{C}$).

driving power of the adsorption of cellulase on the film. The adsorption capacity of cellulase on film increased, this result indicated hydrophobicity of cellulase was increased at higher ionic strength.

And besides, as shown in **Fig. S8**, the adsorption capacities of cellulase on the film increased as temperature increased. The adsorption capacities of cellulase on film at 25, 35 and 45 $^{\circ}$ C (pH 5, 50 mM) were 375.1, 578.2 and 660.8 ng/cm2, respectively. (In the **Fig. S8**, the adsorption capacities of cellulase includes that of buffer, when the temperature was 35 and 45 $^{\circ}$ C, respectively.) The result indicated the hydrophobicity of cellulase increased at higher temperatures. And besides, to a certain extent, the increased capacity may be related with the increased electrostatic interaction at higher temperature.