Supporting Information for

Integrating Enzymatic and Acid Catalysis to Convert Glucose into 5-Hydroxymethylfurfural

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1. Experimental Section

1.1. Conversion of glucose into 5-hydroxymethylfurfural (HMF)

In a typical experiment, an aqueous solution (approximately pH 7.7, 20 mL) containing glucose (1.00 g), sodium tetraborate (for example, 1.06 g) and $MgSO_4 \cdot 7H_2O$ (50 mg, used to activate the glucose isomerase) was prepared in an Erlenmeyer flask. The resulting mixture was placed in a shaking water bath at 150 rpm and 70 ^oC, and then immobilized glucose isomerase (Sweetzyme IT, 0.50 g) was added to catalyze the isomerization reaction. After the reaction, glucose isomerase was recycled by vacuum filtration. The supernatant was collected and mixed with hydrochloric acid (37 wt%, 0.60 mL), NaCl (2.00 g, used to improve the partitioning of HMF into the organic phase) and 1-butanol (30 mL) in a reaction vessel (100 mL). The two-phase reactor was transferred into an oil bath at 190 ^oC (placed on a magnetic stirrer) to perform the dehydration reaction. The reaction was then stopped by cooling the reactor in a refrigerator (-20 ^oC). The HMF product after phase separation was analyzed and quantified by HPLC.

1.2. Analytical methods

Sugars disappearance and the reaction products were analyzed and quantified by HPLC using Bio-Rad Aminex HPX-87H column (5 mM H_2SO_4, 0.6 mL/min, 65 °C). Further analysis was carried out by on-line liquid chromatography–tandem mass spectrometry (LC–MS/MS) system equipped Surveyor PDA detector and a LCQ Advantage MAX ion trap mass spectrometer (Thermo Finnigan, San Jose, CA). Samples were loaded onto Agilent Zorbax SB-C18 reverse phase column (methanol/water gradient elution, 0.8 mL/min) for separation. Full scans were performed between m/z 100 and 800. A full-scan MS spectrum was acquired followed by tandem mass spectra using collision-induced dissociation (CID) of the three most intense precursor ions present in the MS scan.

^{11}B NMR spectra were recorded at 96.28 MHz with boric acid (50 mg/mL) as the external reference on Varian Mercury-300BB NMR spectrometer. ^{13}C NMR measurements were performed at 75.45 MHz using a Varian Mercury-300BB NMR spectrometer. In each case, the temperature was about 25 ^{o}C. The samples containing borate were prepared by dissolution of sugar (100 mg) and sodium tetraborate (105.8 mg) in D_2O (2.00 mL).

2. Supporting Figures

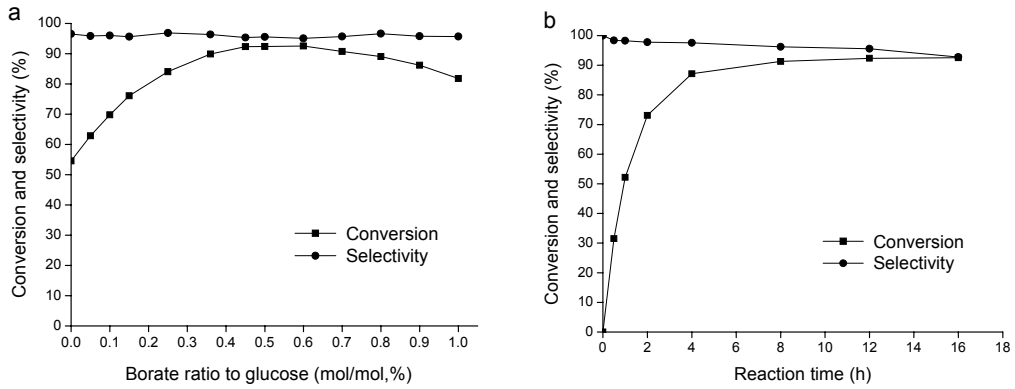


Fig. S1. Glucose conversion and fructose selectivity in the isomerization reaction. (Conversion = weight of glucose reacted / weight of glucose initial; selectivity = weight of fructose produced / weight of glucose reacted)

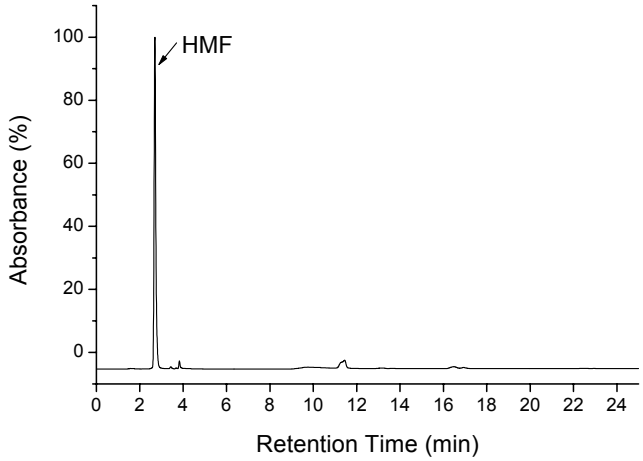


Fig. S2. HPLC analysis spectrum of liquid products. Operating conditions: Zorbax SB-C18 reverse phase column, methanol/water gradient elution (0~6 min: 7:3 water/methanol; 7~25min: 3:7 water/methanol), 0.8 mL/min, 285 nm.

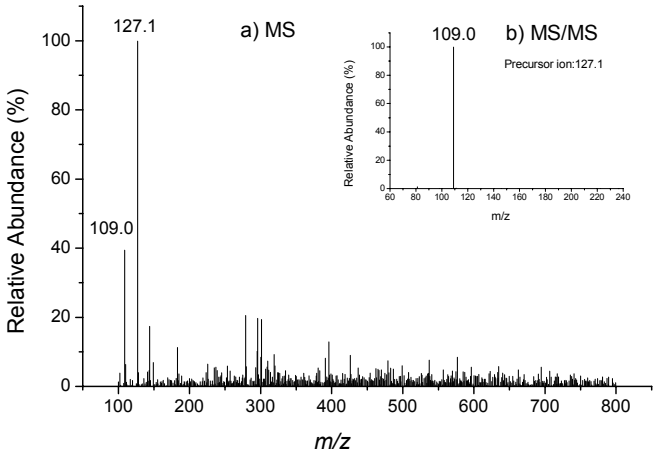


Fig. S3. a) Mass spectrum of the selected peak (assigned to HMF) in Fig.S1; b) Tandem mass spectrum of singly charged ion *m/z* 127.1. (HMF showing an intense $[M-H]^+$ signal at m/z 127.1; Loss of 18 amu [OH-H] from the molecular ion gives the peak at m/z=109.0.)

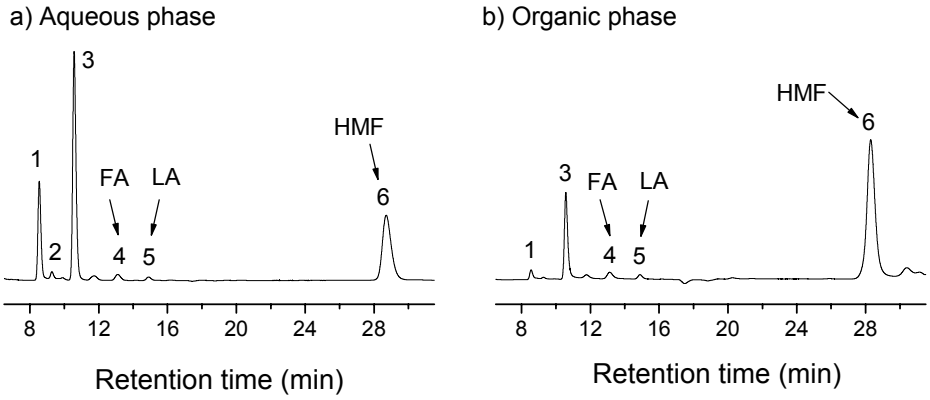


Fig. S4. HPLC analysis spectra of liquid products. Operating conditions: Bio-Rad Aminex HPX-87H column, 5 mM H_2SO_4, 0.6 mL/min, 65 °C. Peaks assignment: (1) glucose; (2) fructose; (3) sodium tetraborate; (4) formic acid (FA); (5) levulinic acid (LA); (6) 5-hydroxymethylfurfural.

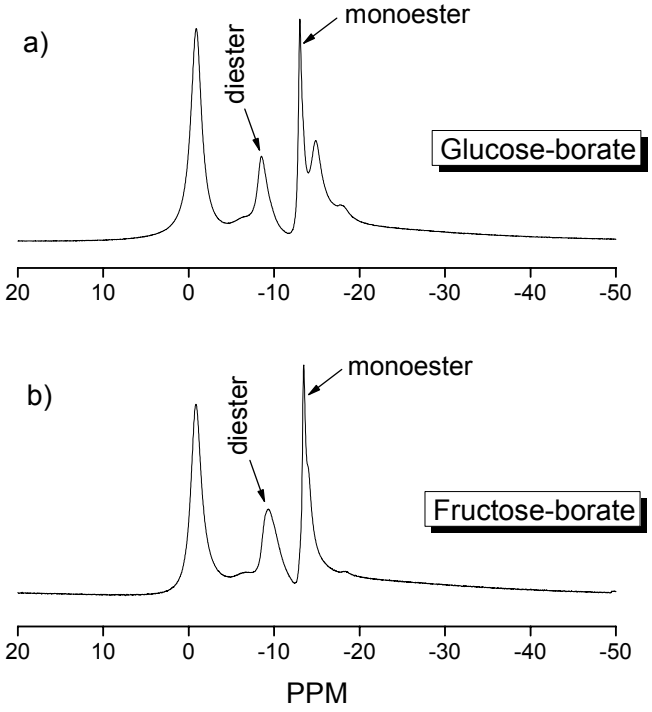
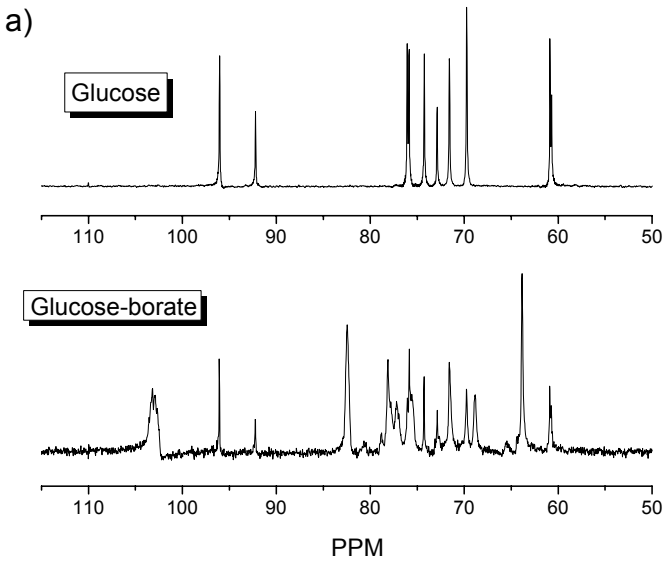


Fig. S5. ^{11}B NMR spectra of sugar–borate complexes. The spectra were recorded at 25 ^{o}C with boric acid as the external reference. Peaks assignment: a) glucose-borate (monoester, 12.98 ppm; diester, 8.45 ppm); b) fructose-borate (monoester, 13.44 ppm; diester, 9.21 ppm).



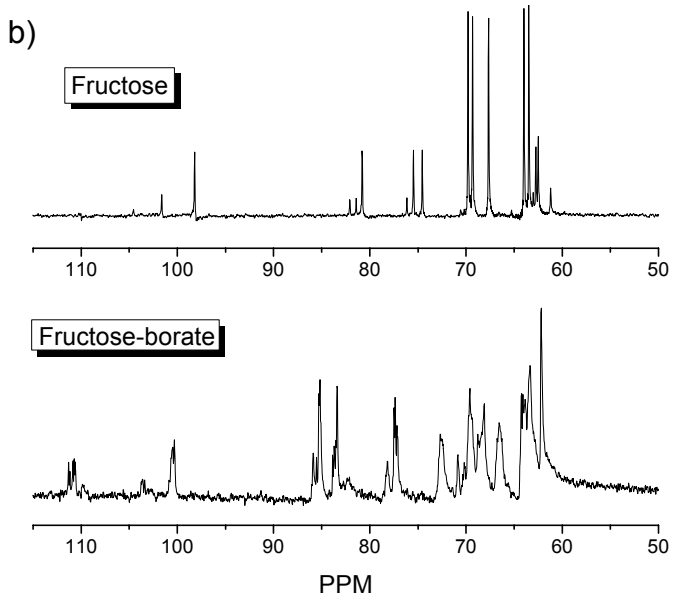


Fig. S6. ^{13}C NMR spectra of sugar–borate complexes. a) glucose and its complexes; b) fructose and its complexes. The peaks assignment was summarized in Table S1.

3. Supporting Tables

Table S1 ^{13}C Chemical shift of sugars and of their B^-L and B^-L_2 borate esters at pH 7.7 [a]

Ligand – L			^{13}C Chemical shift (ppm)					
			C-1	C-2	C-3	C-4	C-5	C-6
D-glucose	α-glucopyranose	L	92.2	71.6	72.9	69.8	71.6	60.7
		$B^-L+B^-L_2$ [b]	102.9	78.1	76.9	68.9	75.6	63.8
	β-glucopyranose	L	96.0	74.3	75.9	69.8	76.1	60.9
		$B^-L+B^-L_2$	103.2	82.5	77.2	68.9	77.8	63.8
D-fructose	β-fructofuranose	L	62.8	101.6	75.5	74.6	80.8	62.5
		$B^-L+B^-L_2$	64.2	111.3	83.4	77.4	85.2	62.2
	β-fructopyranose	L	64.0	98.2	67.7	69.8	69.3	63.5
		$B^-L+B^-L_2$	66.5	103.6	68.8	69.6	70.9	63.4

[a] Assignment of the free sugars (L) and borate eaters (B^-L and B^-L_2) according to refs.1~2.

[b] B^-L and B^-L_2 represent the 1:1 (1 molecule boron : 1 molecule sugar) and 1:2 species of borate esters, respectively.

References

[1] R. Van den Berg, J. A. Peters, H. Van Bekkum, *Carbohydr. Res.* **1994**, *253*, 1.

[2] S. Chapelle, J. F. Verchere, *Tetrahedron* **1988**, *44*, 4469.