Supplementary Information (SI) for Green Chemistry.

Supporting Information

A Green Strategy for Co-production of Xylooligosaccharides and Fermentable Sugars from Birch via Tween-assisted Tartaric Acid Pretreatment

Rutong Jiang[a 1], Bin Bian[a 1], Ruoyan Li[a 1], Qijun Wu[a], Daihui Zhang[a], Caoxing Huang[a], Chenhuan Lai[a, b *], Mi Li[c*], Qiang Yong[a, b]

[a]State Key Laboratory for Development and Utilization of Forest Food Resources, Nanjing Forestry University, Nanjing 210037, China

[b]Jiangsu Co-Innovation Center of Efficient Processing and Utilization of Forest Resources, College of Chemical Engineering, Nanjing Forestry University, Nanjing 210037, China.

[c]Center for Renewable Carbon, School of Natural Resources, University of Tennessee, Knoxville, TN 37996, USA

*Corresponding author

Email: lch2014@njfu.edu.cn (C. Lai); mli47@utk.edu (M. Li)

[1]Rutong Jiang, Bin Bian and Ruoyan Li contributed equally to this work, regarding as the first author

Table S1. Chemical composition of birch and birch pretreated with tartaric acid (TA).

Temperature （℃）	Time（min）	TA concentration （%）	Contents (%)			Recovery Yield (%)		Delignification (%)
			Glucan	Xylan	Lignin	Glucan	Xylan	
	Raw birch		39.6 ± 0.5	24.3 ± 0.3	27.7 ± 0.2	/	/	/
170	30	0.0	42.2 ± 0.4	19.4 ± 0.1	23.6 ± 0.2	94.8	29.3	4.6
		0.1	42.9 ± 0.1	18.7 ± 0.1	21.1 ± 0.7	95.4	32.3	15.6
		0.2	45.7 ± 0.1	16.4 ± 0.1	24.1 ± 0.2	98.8	42.2	6.0
		0.3	46.8 ± 0.2	14.6 ± 0.1	24.5 ± 0.4	97.1	50.9	8.5
		0.4	49.2 ± 1.0	13.5 ± 0.1	24.3 ± 0.1	97.6	64.5	9.7
		0.5	50.0 ± 0.6	11.4 ± 0.1	29.1 ± 0.2	95.0	64.7	0.6
		0.6	51.2 ± 0.4	9.9 ± 0.2	30.1 ± 0.5	93.3	70.8	1.3
170	30	0.4	49.2 ± 1.0	13.5 ± 0.0	24.3 ± 0.1	97.2	56.5	13.6
	40		50.2 ± 0.2	12.3 ± 0.2	25.1 ± 0.3	98.2	68.9	6.9
	50		56.7 ± 0.2	7.8 ± 0.1	29.8 ± 0.1	98.4	77.9	6.7
	60		57.0 ± 0.1	7.7 ± 0.1	29.9 ± 0.6	97.5	78.7	7.8
	70		57.5 ± 0.5	6.0 ± 0.1	30.4 ± 0.3	97.5	83.4	7.2
150	40	0.4	40.2 ± 0.2	19.2 ± 0.3	21.9 ± 0.1	97.4	24.2	4.5
160			44.1 ± 0.3	17.9 ± 0.0	23.1 ± 0.3	95.9	36.8	9.5
170			50.2 ± 0.2	12.3 ± 0.2	25.1 ± 0.3	94.9	62.2	8.2
180			56.1 ± 0.5	6.7 ± 0.2	29.8 ± 0.2	96.8	81.2	7.4
190			59.0 ± 0.2	3.9 ± 0.0	31.7 ± 0.2	93.0	90.0	10.0

Table S2. Chemical composition and cellulose accessibility of Tween-assisted tartaric acid pretreated birch.

Biomass[a]	Glucan (%)	Xylan (%)	Lignin (%)	Solid recovery (%)	Glucan recovery (%)	Xylan removal (%)	Lignin removal (%)	Accessibility (mg/g)
TA	55.5 ± 0.3	11.2 ± 0.1	30.2 ± 0.2	66.7 ± 0.2	93.4 ± 0.4	69.0 ± 0.5	8.2 ± 0.3	266.1
0.5%TW-TA	53.4 ± 0.2	11.3 ± 0.0	29.4 ± 0.7	70.9 ± 0.1	95.6 ± 0.2	67.0 ± 0.1	5.3 ± 0.5	320.7
1%TW-TA	55.1 ± 0.1	9.8 ± 0.1	29.3 ± 0.1	70.0 ± 0.1	97.4 ± 0.3	71.5 ± 0.7	6.6 ± 0.6	325.3
2%TW-TA	55.1 ± 1.0	11.1 ± 0.0	26.7 ± 0.1	67.5 ± 0.6	93.9 ± 0.1	69.1 ± 0.1	18.1 ± 0.3	332.5

[a] TA refers to birch pretreated with 0.4% (w/v) tartaric acid at 170 °C for 40 min; 0.5%TW-TA refers to birch pretreated with 0.4% (w/v) tartaric acid and 0.5% (w/v) Tween at 170 °C for 40 min.

Table S3. Semi-quantitative analysis of inter-unit linkages and subunits in lignins from pretreated birch.

Lignin substructure	MWL[a]	MWL-0.5%TW[b]	MWL-2.0%TW[c]
β-O-4[d]	76.4(55.2)	75.4(54.0)	73.2(45.0)
β-*β*	19.3(14.0)	20.0(14.3)	21.2(13.0)
β-5	4.3(3.1)	4.6(3.3)	5.6(3.4)
Syringyl（S）	74.4	72.6	71.8
Guaiacyl（G）	25.6	27.4	28.2
S/G	2.9	2.6	2.5

[a] MWL refers to milled wood lignin isolated from tartaric acid pretreated birch;
[b] MWL-0.5%TW refers to milled wood lignin isolated from tartaric acid pretreated birch with 0.5% Tween;
C MWL-2.0%TW refers to milled wood lignin isolated from tartaric acid pretreated birch with 2.0% Tween;

[d] Linkages contents expressed per 100 Ar (and as percentages of total side chains).

Table S4. Contribution of amino acid residues, interaction forces, and binding energies in the molecular docking results between lignin model compounds and endoglucanase.

Lignin model[a]	Amino acid residue					Binding energy (kcal/mol)
	Hydrogen bonding forces	Van der Waals force	π−π	π−Alkyl	Amide−π	
SG	ASN328, SER332	GLN325, ASN328, ASN334, GLY333, ASN227, GLY230, TRP329, SER340, SER324, ALA222	/	ALA335	ASN328	-7.644
SG-TWN	SER292, TYR326, ASN227, SER318, ARG108, ASN328, SER340, GLY223	THR341, ALA222, SER340, GLN325, ALA208, GLY225, ASN227, GLY230, ALA335, GLY223, SER324, TRP329, ASN334, GLY333, SER332, GLN325, GLN178, SER221, ASP172, SER295, THR341, ILE290, THR289, PHE226, THR210, HIS212, SER144, SER318, TYR38, SER106, GLN178, TYR170	TRP320	TYR326, PRO176	/	-6.815

[a] SG refers to syringyl–guaiacyl β-O-4 lignin dimer;

[b] SG-TWN refers to Tween-modified SG molecular models.

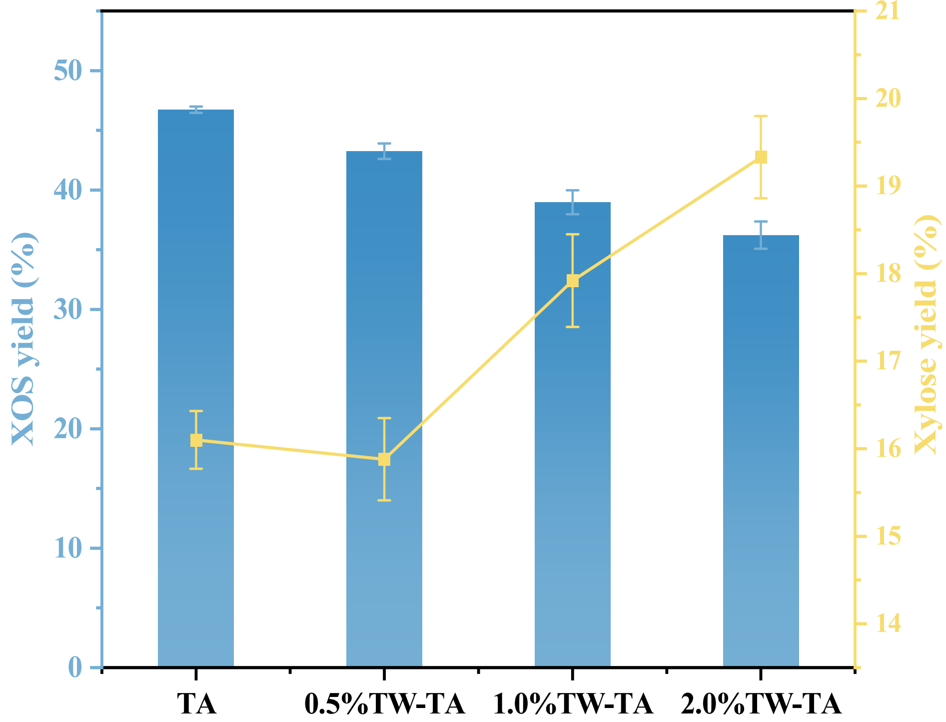

Fig. S1. Effects of Tween-assisted tartaric acid pretreatment on XOS production from birch.

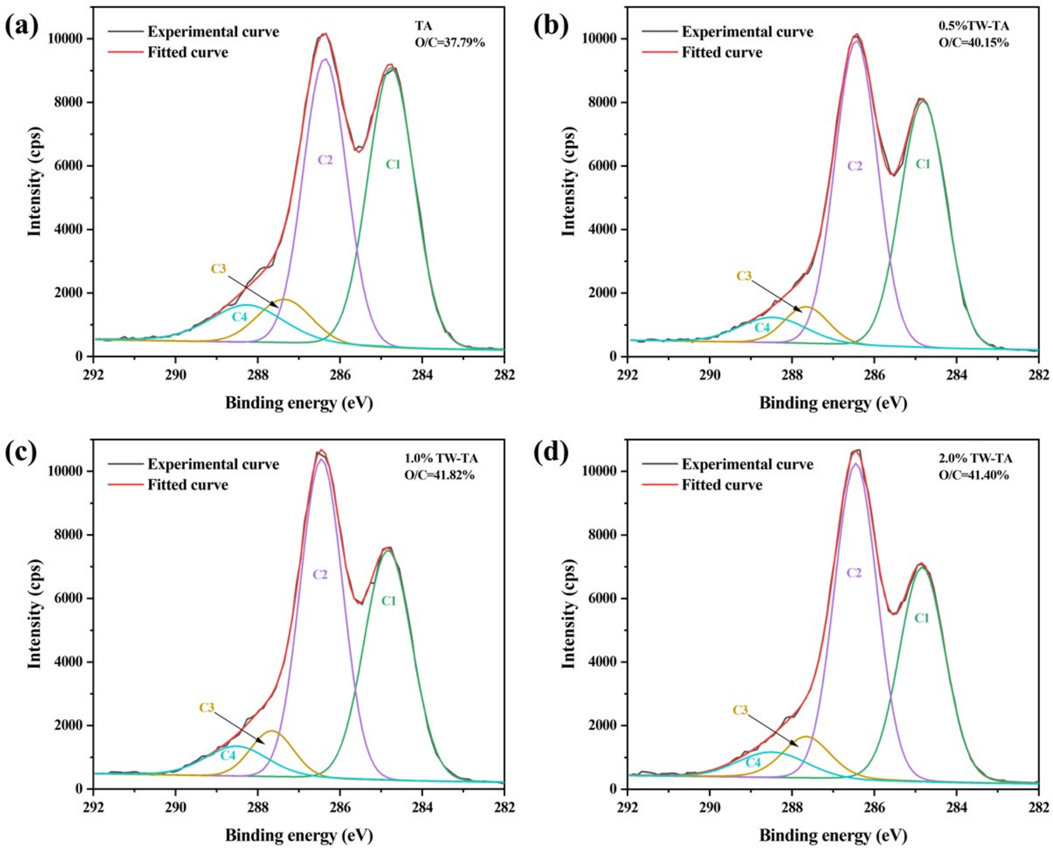

Fig. S2. Curve-fitted high-resolution XPS spectra of Tween-assisted tartaric acid pretreated birch: (a) TA (without Tween); (b) 0.5%TW-TA (with 0.5% Tween); (c) 1.0%TW-TA (with 1.0% Tween); (d) 2.0%TW-TA (with 2.0% Tween).

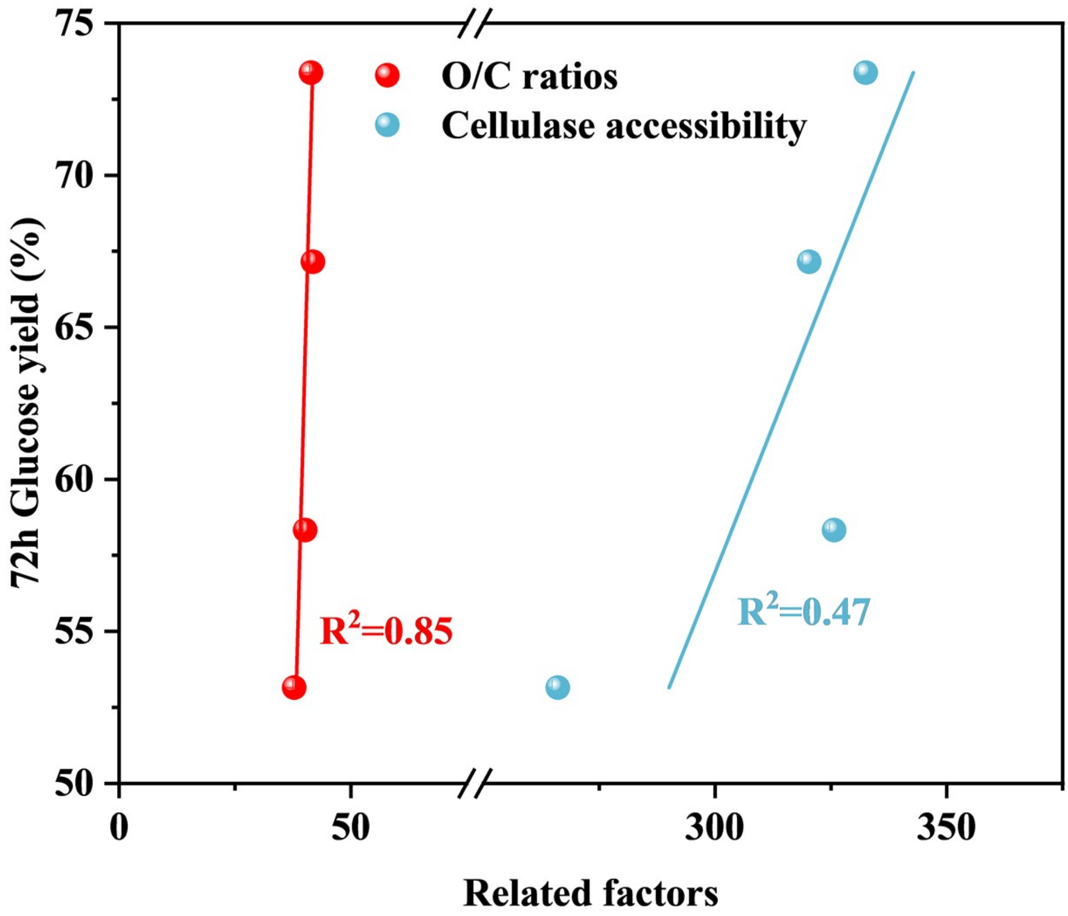

Fig. S3. Correlation analysis between O/C ratio and cellulose accessibility with 72 h glucose yield of pretreated substrates.

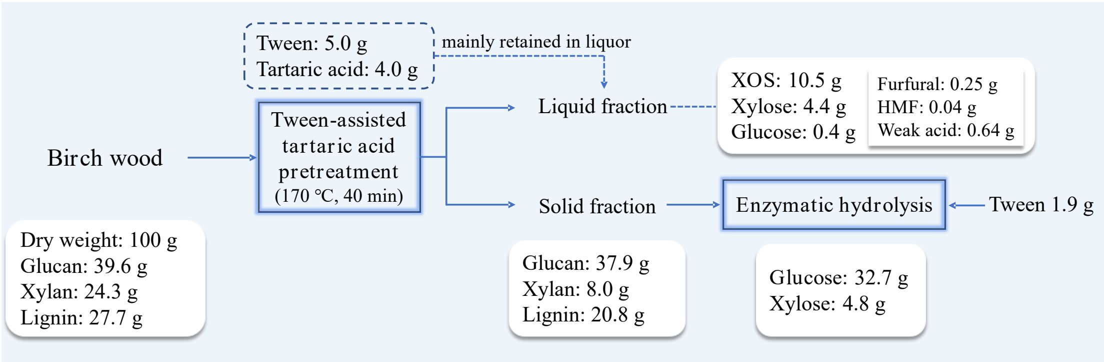

Fig. S4. Mass balance for the co-production of XOS and fermentable sugars based on Tween-assisted tartaric acid pretreatment.

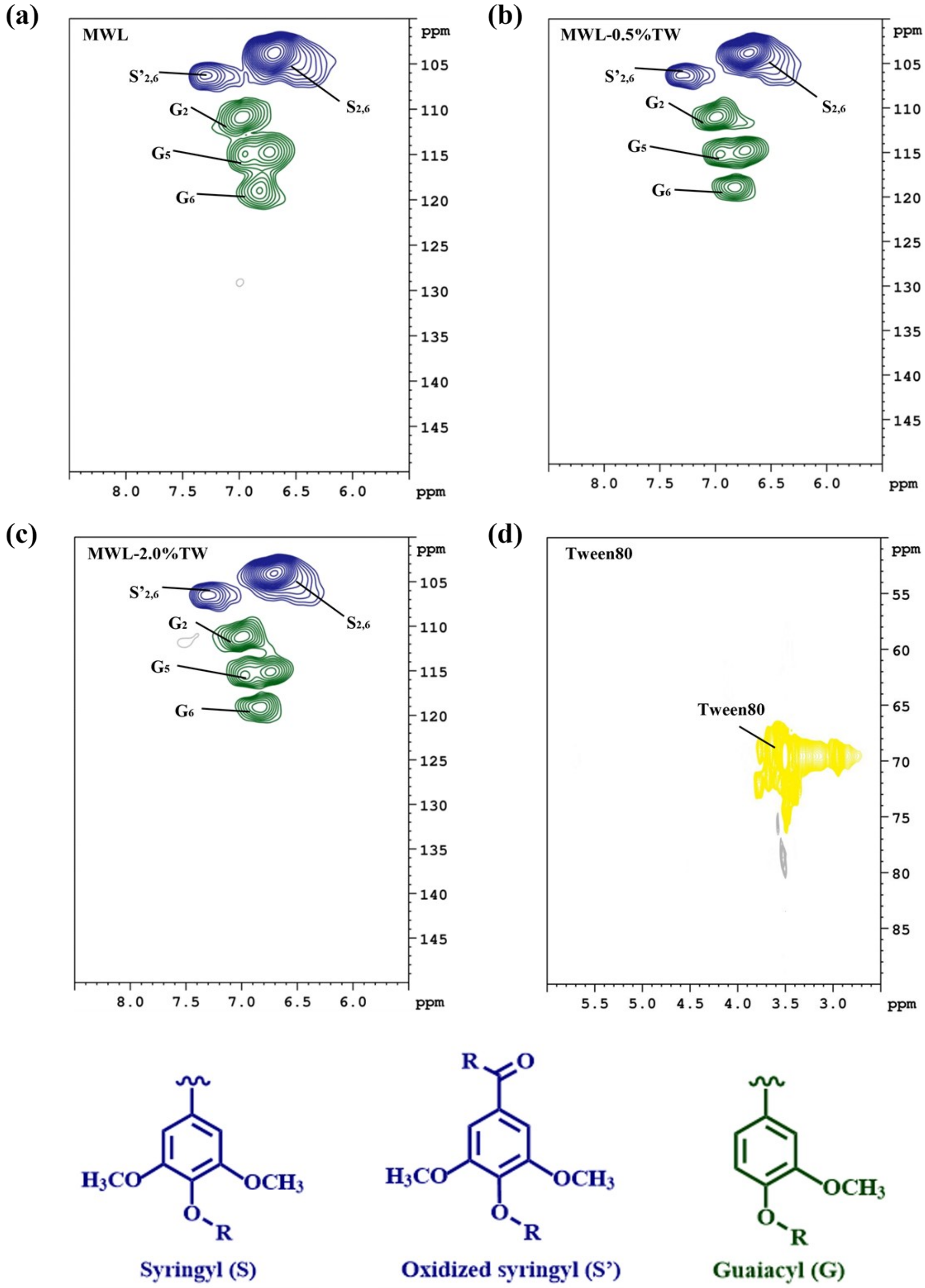

Fig. S5. Aromatic region in 2D HSQC NMR spectra of the milled wood lignins isolated from pretreated substrates: MWL (a); MWL-0.5%TW (b); MWL-2.0%TW (c); side-chain region in 2D HSQC spectra of Tween80 (d).

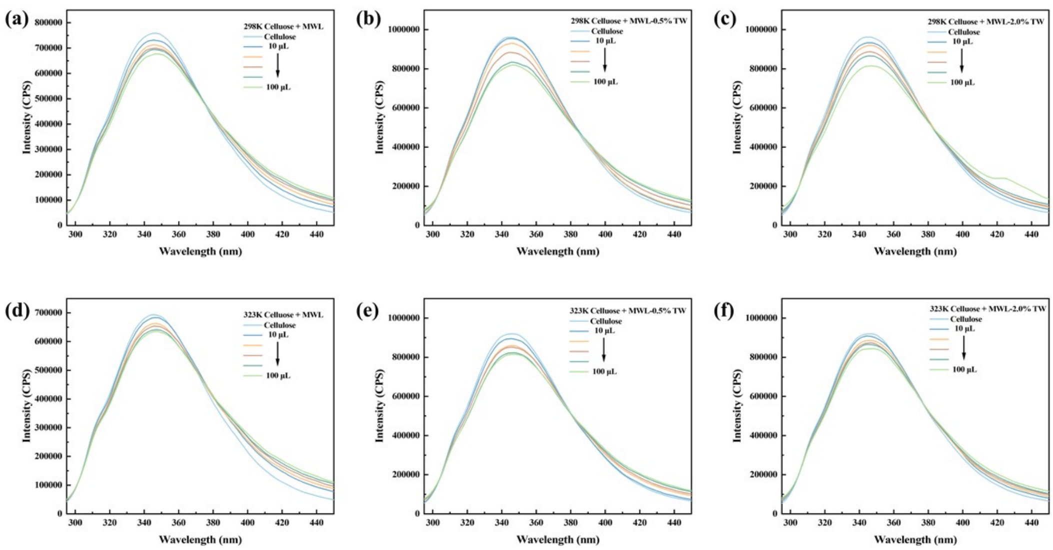

Fig. S6. Fluorescence emission spectra of cellulase titrated with different concentrations of MWL, MWL-0.5%TW, MWL-2.0%TW at 298 K (a, b, c) and 323 K (d, e, f). λex = 280 nm, λem = 295–500 nm.

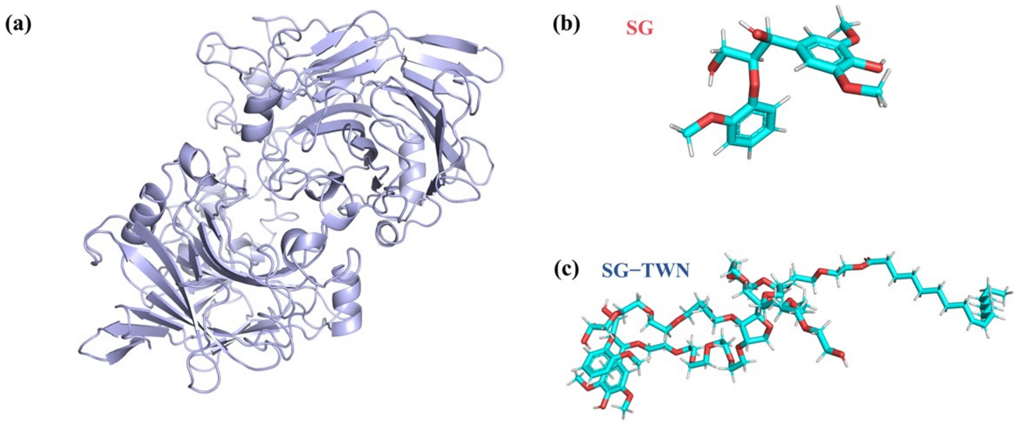

Fig. S7. Structure of endo-β-1,4-glucanase (a, PDB ID: 1EG1), SG (b), and SG−TWN (c).