Electronic Supplementary Material (ESI) for Sustainable Energy & Fuels.

E-supplementary data 2

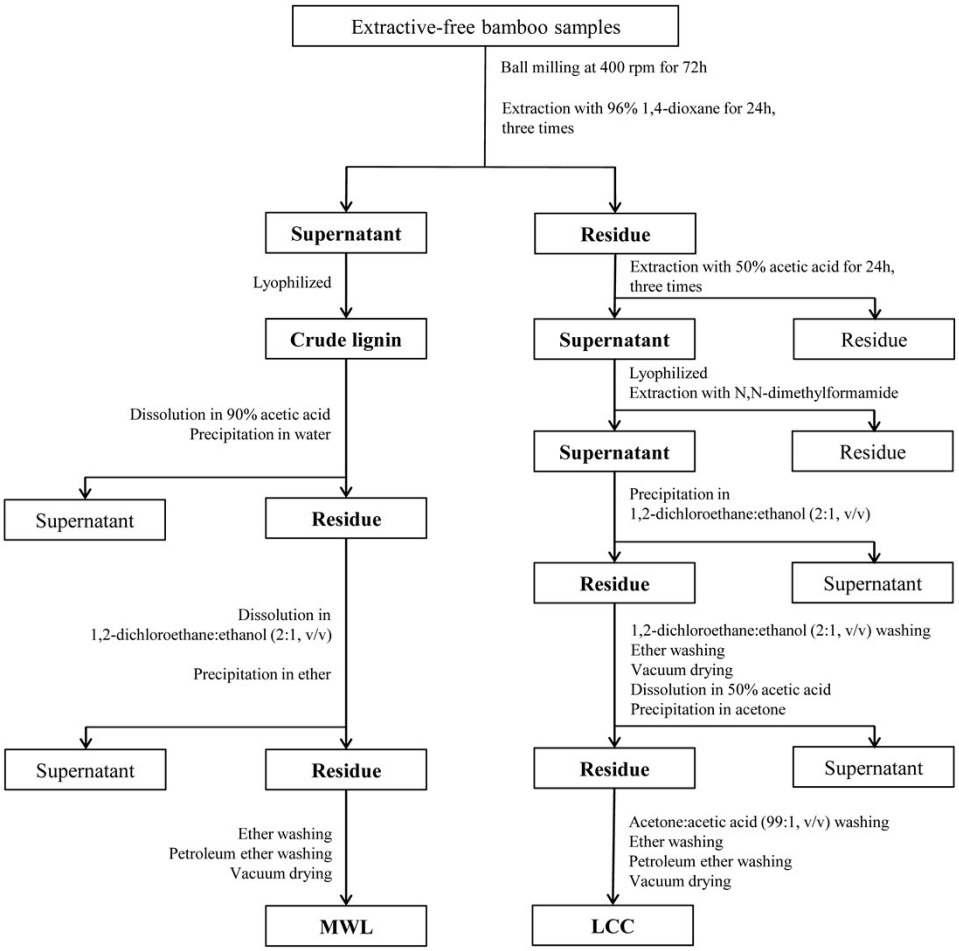

Fig. S1. The main isolation procedures of MWLs and LCCs from raw and treated bamboo.

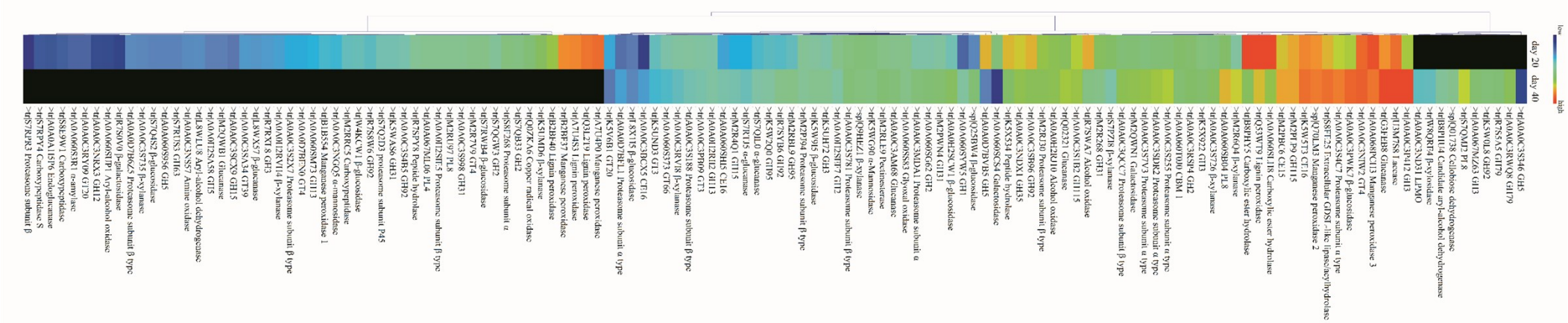

Fig. S2. Expression abundance of all secreted proteins on day 20 and day 40. Unexpressed proteins are colored black.

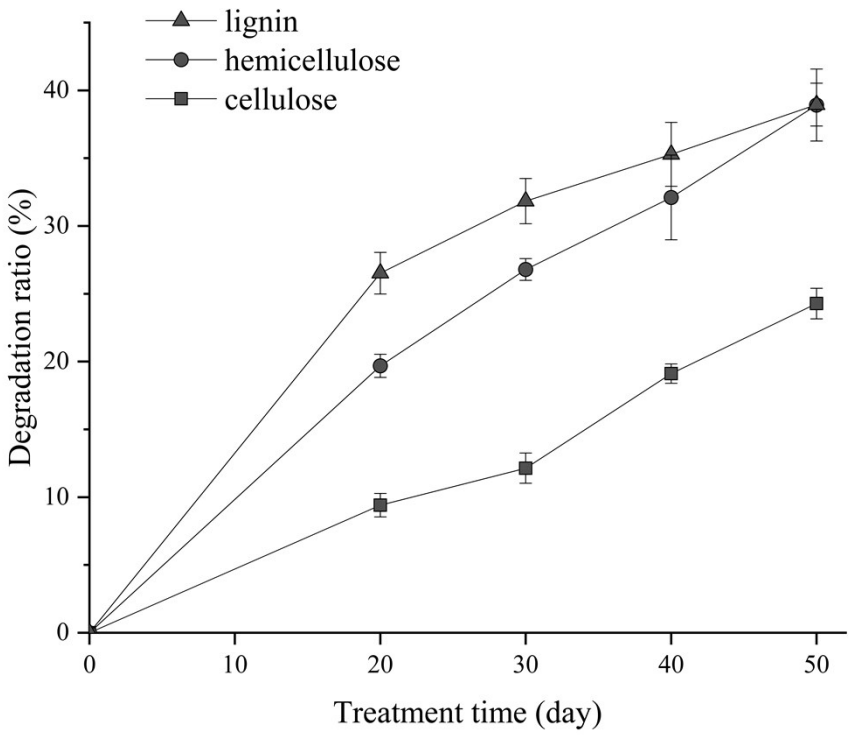

Fig. S3. The degradation ratio of lignin, cellulose and hemicellulose during fungal treatment of bamboo samples. Chemical composition including lignin, cellulose and hemicellulose composition was determined based on procedure of National Renewable Energy Laboratory (NREL, Golden, CO) [1]. The degradation ratio of the composition (%) was calculated as the formula:

$$degradation\ ratio\ of\ day\ X(\%) = \frac{content\ of\ day\ X\ (\%) \times solid\ recovery\ of\ day\ X(\%)}{content\ of\ day\ 0\ (\%)}$$

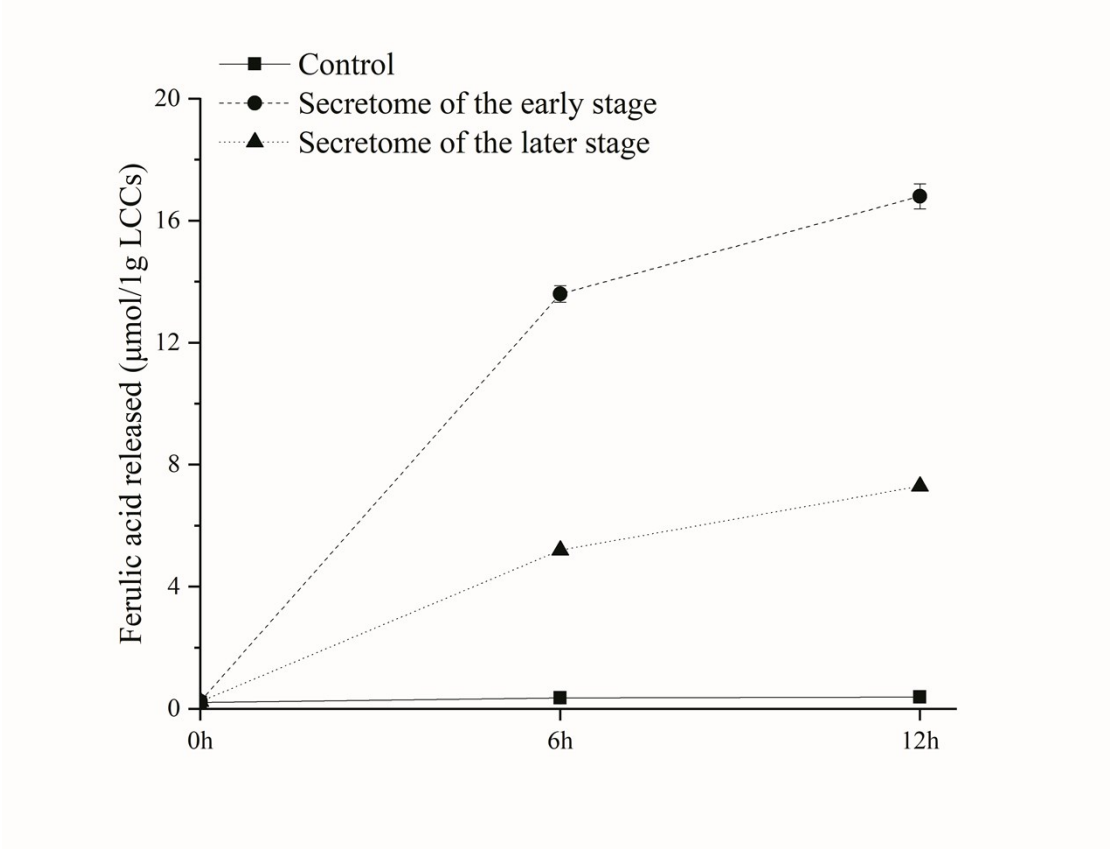

Fig. S4. FA esterase enzymatic activities of 2538 on bamboo LCCs polymers

LCCs isolated from original bamboo was used as substrate to test FA esterase enzymatic activities of extracted secretomes. 1 U of enzymatic activity was defined as 1 μmol of ferulic acid released from 1g LCCs per minute. With 13.6 μmol ferulic acid released in 6 h, 37.8 mU of FA esterase enzymatic activities was detected in fungal secretome of the early stage. At the later stage, the FA esterase enzymatic activities in fungal secretome was lower than early stage, in which only 5.2 μmol ferulic acid released in 6 h.

The reaction mixture contained 2 ml fungal secretome extract (0.05 mol/L pH 7.0 citrate-phosphate buffer as control), 1g LCCs and 0.5 ml 250 mM sodium malonate, pH 4.5. After 6 h at 25 ℃, the ferulic acid of the supernatants were determined by HPLC

using standard calibration curves. The ferulic acid of secretome was also detected and excluded in the calculation. Samples were subjected to HPLC analysis with C18 column (4.6 × 250 mm) (Waters) under the condition of 1 ml/min a flow rate with methanol-10 mM phosphoric acid as the eluent at 40 ℃. The vertical line on each point indicates the standard deviation for three replicates (SD, n = 3).

Table S2. The isolation yields of all MWL and LCC fractions.

Fraction	Fungal treated time (day)	Isolation yield (%)
MWL [a]	Untreated	15.42
	20	13.79
	30	13.31
	40	12.64
	50	9.52
LCC [b]	Untreated	8.16
	20	8.93
	30	7.75
	40	7.41
	50	6.32

[a] % of Klason lignin

[b] % of extraction-free bamboo

Table S2. The assignments of ^{13}C-^{1}H peaks in HSQC spectrum from the isolated MWLs.

Label	δ C / δ H (ppm)	Assignments
PCA_{α}	144.8/7.41	C_{α}-H_{α} in p-coumarate (**PCA**) and ferulate (**FA**)
$PCA_{2,6}$	130.1/7.45	C_2-H_2 and C_6-H_6 in p-coumarate (**PCA**)
I_{α}	128.4/6.44	C_{α}-H_{α} in cinnamyl alcohol end-groups (**I**)
I_{β}	128.4/6.23	C_{β}-H_{β} in cinnamyl alcohol end-groups(**I**)
$H_{2,6}$	127.8/7.22	$C_{2,6}$-$H_{2,6}$ in p-hydroxyphenyl units (**H**)
J_{β}	126.3/6.76	C_{β}-H_{β} in cinnamyl aldehyde end-groups(**J**)
FA_6	123.2/7.15	C_6-H_6 in ferulate (**FA**)
G'_6	123.2/7.33	C_6-H_6 in guaiacyl units (**G**)
G_6	119.0/6.78	C_6-H_6 in guaiacyl units (**G**)
G_5	115.1/6.92	C_5-H_5 in etherified guaiacyl units (**G**)
$PCA_{3,5}$	115.5/6.77	C_3-H_3 and C_5-H_5 in *p*-coumarate (**PCA**)
G_5	114.9/6.70	C_5-H_5 in guaiacyl units (**G**)
PCA_{β}	113.5/6.27	C_{β}-H_{β} in *p*-coumarate (**PCA**) and ferulate (**FA**)
FA_2	111.0/7.32	C_2-H_2 in ferulate (**FA**)
G_2	110.9/6.99	C_2-H_2 in guaiacyl units (**G**)
$S'_{2,6}$	106.3/7.32	$C_{2,6}$-$H_{2,6}$ in oxidized S units (**S ′**)
$S_{2,6}$	103.8/6.69	C_2-H_2 and C_6-H_6 in etherified syringyl units (**S**)
$T'_{2,6}$	103.9/7.34	$C'_{2,6}$- $H'_{2,6}$ in tricin (**T**)
T_6	98.9/6.23	$C_{2,6}$- $H_{2,6}$ in tricin (**T**)
T_8	94.2/6.60	C_8- H_8 in tricin (**T**)
B_{α}	86.8/5.43	C_{α}-H_{α} in phenylcoumaran substructures(**B**)
$A_{\beta}(S)$	85.9/4.10	C_{β}-H_{β} in β-O-4′substructures linked (**A**) to a S unit
D_{β}	85.3/3.85	C_{β}-H_{β} in dibenzodioxocin substructures(**D**)
F_{α}'	84.6/4.75	C_{α}'-H_{α}' in spirodienone substructures (**F**)
C_{α}	84.8/4.65	C_{α}-H_{α} in β-β′ resinol substructures (**C**)

A_β(G)	83.4/4.27	C_β-H_β in β-O-4′substructures (**A**) linked to a G unit
D_α	83.3/4.81	C_α-H_α in dibenzodioxocin substructures(**D**)
A_β(H)	82.9/4.48	C_β-H_β in β-O-4′substructures (**A**) linked to a H-unit
Aox_β	82.7/5.22	C_β-H_β in α-oxidized β-O-4′ substructures(**Aox**)
F_α	81.2/5.10	C_α-H_α in spirodienone substructures (**F**)
A'_β(G)	80.8/4.52	C_β-H_β in γ-acylated β-O-4′substructures linked to a G-unit (**A′**)
E_α	79.5/5.59	C_α-H_α in α-O-4′ substructures (**E**)
A_α(S)	71.8/4.83	C_α-H_α in β-O-4′substructures (**A**) linked to a S-unit
A_α(G)	70.9/4.71	C_α-H_α in β-O-4′ substructures (**A**) linked to a G-unit
C_γ	71.0/3.81 and 4.17	C_γ-H_γ in β-β′ resinol substructures (**C**)
A'_γ	63.5/3.83 and 4.30	C_γ-H_γ in γ-acylated β-O-4′ substructures(**A′**)
B_γ	62.6/3.67	C_γ-H_γ in phenylcoumaran substructures (**B**)
I_γ	61.3/4.08	C_γ-H_γ in cinnamyl alcohol end-groups (**I**)
F_β	59.5/2.75	C_β-H_β in spirodienone substructures (**F**)
A_γ	59.4/3.35 – 3.80	C_γ-H_γ in γ-hydroxylated β-O-4′substructures (**A**)
−OMe	55.6/3.73	C-H in methoxyls
C_β	53.5/3.05	C_β-H_β in β-β′ resinol substructures (**C**)
B_β	53.1/3.43	C_β-H_β in phenylcoumaran substructures (**B**)

Table S4. The assignments of ^{13}C-^{1}H peaks in HSQC spectrum from the isolated LCCs

Label	δ C / δ H (ppm)	Assignments
X_5	62.6/3.30 and 3.95	C_5-H_5 in β-D-xylopyranoside
X_2	72.6/3.2	C_2-H_2 in β-D-xylopyranoside
$X2_2$	73.2/4.64	C_2-H_2 in 2-O-acetyl-β-D-xylopyranoside
X_3	73.9/3.41	C_3-H_3 in β-D-xylopyranoside
$X3_3$	74.7/4.96	C_3-H_3 in 3-O-acetyl-β-D-xylopyranoside
X_4	75.4/3.68	C_4-H_4 in β-D-xylopyranoside
U_4	81.3/3.25	C_4-H_4 in 4-O-methyl-α-D-GlcUA
BE_α	81.3/4.672	C_α-H_α in benzyl ether LCC structures
αX1(R)	92.2/5.07	(1-4)-α-D-xylopyranoside (R)
βX1(R)	97.4/4.26	(1-4)-β-D-xylopyranoside (R)
$X2_1$	99.2/4.5	2-O-acetyl-β-D-xylopyranoside
$PhGlc_2$	101.2/4.65	phenyl glycoside linkages
$PhGlc_3$	101.45/4.89	phenyl glycoside linkages
$X3_1$	101.5/4.27	3-O-acetyl-β-D-xylopyranoside
X_1 /Glc_1	103.0/4.16	β-D-xylopyranoside/β-D-glucopyranoside
GlcAE	99.1/4.68	esterified 4-O-methyl-α-D-glucuronic acid units
esterified FA_β	116.5/6.33	C_β-H_β in esterified ferulate (**FA**)

Table S5. Sugar and Lignin Analysis of MWL and LCC Preparations.

Preparation	Chemical Composition [a] (% of Relative Content)		Carbohydrate Content [b] (% of Relative Molar Content)			
	Lignin[b]	Carbohydrate	Glc	Xyl	Ara	Gal
MWL-untreated	96.4	3.6	84.7	10.2	2.9	2.2
MWL-20d	95.6	4.4	79.9	12.4	4.6	3.1
MWL-30d	98.2	1.8	83.5	9.4	4.2	2.9
MWL-40d	97.1	2.9	89.2	6.7	3	1.1
MWL-50d	96.7	3.3	80.5	11.9	5.6	2
LCC-untreated	29.4	70.6	32.8	62.3	3.7	1.2
LCC-20d	26.4	73.6	27.3	68.4	2.9	1.4
LCC-30d	28.2	71.8	30.3	64.9	4.1	0.7
LCC-40d	25.7	74.3	36.1	59.7	2.5	1.7
LCC-50d	26.1	73.9	28.6	65.1	5	1.3

[a] Relative to MWL or LCC sample (%);

[b] including acid-soluble lignin and Klason lignin;

[c] Glc, glucose; Xyl, xylose Ara, arabinose; Gal, galactose.

Chemical composition was determined based on reference [2].

Reference

1. Sluiter A, Hames B, Ruiz R, Scarlata C, Sluiter J, Templeton D, Crocker D. Determination of structural carbohydrates and lignin in biomass. *Lab. Anal. Proced.* 2008.

2. Yue P-P, Hu Y-J, Fu G-Q, Sun C-X, Li M-F, Peng F, Sun R-C: Structural Differences between the Lignin-Carbohydrate Complexes (LCCs) from 2- and 24-Month-Old Bamboo (*Neosinocalamus affinis*). *Int. J. Mol. Sci*. vol. 19; 2017.