Supporting information for:

Structural Motifs of Wheat Straw Lignin Differ in Susceptibility to Degradation by the White-Rot Fungus *Ceriporiopsis subvermispora*

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Supporting Information contents:

Carbohydrate content and composition. Carbohydrate content and composition was determined in duplicate as constituent monosaccharides after acid hydrolysis by a modified method reported by Englyst & Cummings.[1] Ten mg of each sample was treated with 72% (w/w) H_2SO_4 for 1 h at 30 °C followed by 1 M H_2SO_4 for 3 h at 100 °C. Subsequently, samples were centrifuged (9,000*xg*, 5 min, 20 °C) and the supernatants were diluted (20x) before analysis. Degradation of monosaccharides during hydrolysis was corrected for by including monosaccharide standard mixtures in hydrolysis. Analysis was performed on a High Performance Anion Exchange Chromatography (HPAEC) Dionex ICS-5000 system (Thermo Scientific, Synnyvale, CA, USA). The system was equipped with a CarboPac PA-1 column (250 mm x 2 mm ID) in combination with a CarboPac guard column (50 mm x 2 mm ID) with pulsed amperometric detection (PAD) (all Dionex). 10 µL of sample was injected and eluted at a flow rate of 0.4 $mL \cdot min^{-1}$ using a combination of three mobile phases: A) 0.1 M NaOH, B) 1 M NaOAc in 0.1 M NaOH and C) H_2O. The elution profile used was as follows: 0-35 min isocratic on 100% C; 35-50 min linearly from 100% A to 40% B; 50-55 min isocratic on 100% B; 55-63 min isocratic on 100% A; 63-78 min isocratic on 100% C. Post-column addition of 0.5 M NaOH at 0.1 $mL \cdot min^{-1}$ was performed between 0-35 min and 63-78 min. Data was processed by using Chromeleon 7 (Thermo Scientific). The uronic acids released after the acid hydrolysis step, were determined in duplicate as anhydrouronic acid content by an automated meta-hydroxydiphenyl assay with addition of sodium tetraborate using an auto-analyzer (Skalar Analytical BV, Breda, The Netherlands).[2] Glucuronic acid (Fluka AG, Busch, Switzerland) was used as a reference (0 – 100 µg mL^{-1}). Total carbohydrate content was calculated as the sum of neutral anhydrocarbohydrates and anhydrouronic acids.

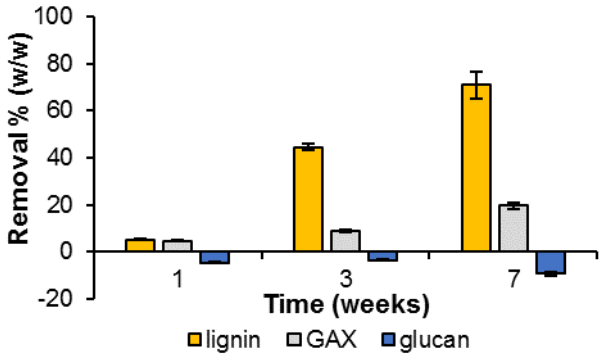
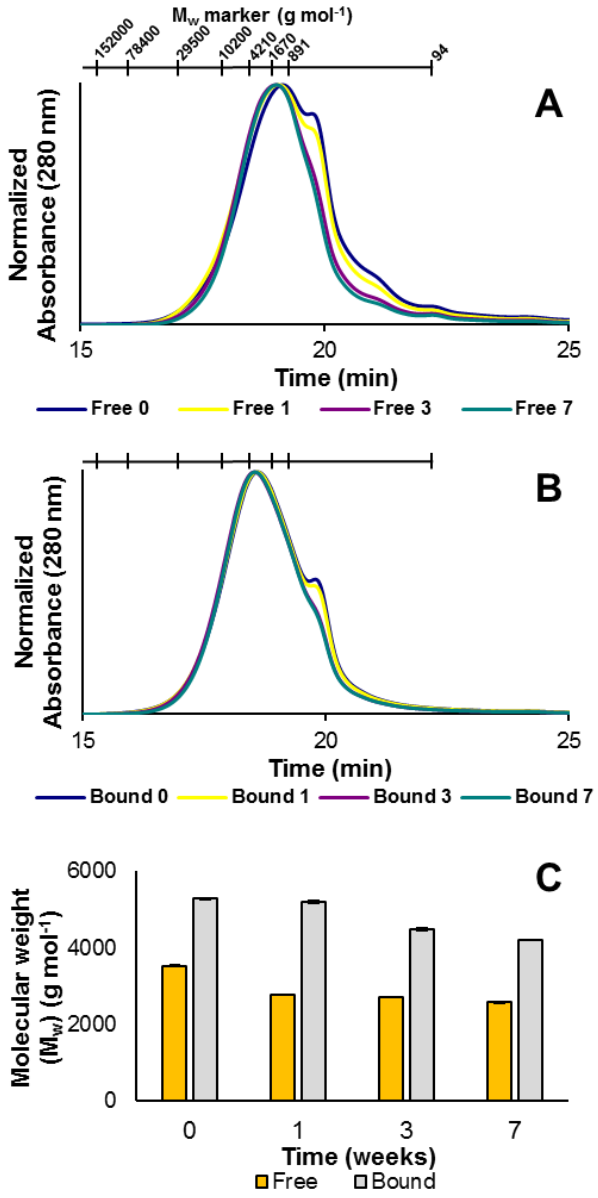


Figure S1. Lignin, glucuronoarabinoxylan (GAX) and glucan (cellulose) removal during fungal growth. Compositional analysis by using quantitative ^{13}C-IS py-GC-MS (lignin) and constituent monosaccharide analysis after H_2SO_4 hydrolysis (carbohydrates). Average and standard deviation of analytical triplicates on pooled biological triplicates. The negative values for glucan are caused by the production of fungal β-glucan, which cannot be distinguished from cellulose in constituent monosaccharide analysis. Note that the 7 weeks fungal treated sample has been previously analyzed and reported in van Erven et al. (2018).[3] This time-point was reanalyzed for this work.

Compositional analyses showed that lignin degradation by *C. subvermispora* was evident from the first week of growth on wheat straw (Figure S1). In the early stage, lignin and hemicellulose (glucuronoarabinoxylan, GAX) were removed to similar extent (~ 5% (w/w)), while cellulose appeared unaffected. The relatively unselective action in the initial stage of growth is explained by the availability of (more) easily degradable carbohydrates. As soon as these were depleted, lignin degradation became more selective. After 3 weeks of growth, up to 45% (w/w) lignin was removed at the expense of 9% (w/w) GAX. During further growth, delignification continued and reached 71% (w/w) after 7 weeks, while GAX removal increased to 19% (w/w).[3] Still, after 7 weeks of treatment more than 90% of the initial carbohydrates were retained in the residue, from which it is concluded that *C. subvermispora* delignified wheat straw in a highly effective and selective manner.



D

	M_n (g mol^{-1})	M_w (g mol^{-1})	M_w/M_n
Free 0	1015	6855	6.8
Free 1	1120	7370	6.6
Free 3	1040	6355	6.1
Free 7	965	5355	5.5
Bound 0	2265	13310	5.9
Bound 1	2550	13480	5.3
Bound 3	2520	14190	5.6
Bound 7	2400	11245	4.7

E

	M_n (g mol^{-1})	M_w (g mol^{-1})	M_w/M_n
Free 0	4215	17630	4.2
Free 1	4600	18760	4.1
Free 3	4315	16560	3.8
Free 7	4030	14350	3.6
Bound 0	8200	29920	3.6
Bound 1	8940	30340	3.4
Bound 3	9025	32050	3.5
Bound 7	8615	26580	3.1

Figure S2. Normalized alkaline SEC chromatograms of free (A) and bound (B) isolates, weight average molecular weight (M_w) of lignin isolates (C). Statistical moments of molar masses of lignin isolates by organic SEC analyses calculated based on PSS standards (D) and statistical moments of molar masses of lignin isolates by organic SEC analyses calculated with MALS-corrected standards (E). Average and standard deviation in (C) of analytical duplicates of mixed isolation duplicates. For estimation of M_w the small shoulders appearing in the alkaline SEC traces were excluded.

Table S1. ^{13}C-IS py-GC-MS relative abundance of lignin compounds in unfractionated wheat straw during growth of *C. subvermispora*. Corrected for relative response factors and relative abundance of ^{13}C analogues. Sum on the bases of structural classification according to van Erven et al.[3-4] Average and standard deviation of analytical duplicates on pooled biological triplicates. Additional data (timepoints 1 and 3 weeks) compared to van Erven et al. (2018).[3]

	Total sample (weeks)			
	0	**1**	**3**	**7**
Lignin subunits (%)				
H	10.3 ± 0.0	11.7 ± 0.3	13.1 ± 1.2	15.6 ± 0.2
G	64.0 ± 0.7	61.1 ± 1.2	59.6 ± 1.0	60.3 ± 1.3
S	25.7 ± 0.7	27.2 ± 0.9	27.3 ± 0.3	24.1 ± 1.1
S/G	0.40 ± 0.0	0.44 ± 0.0	0.46 ± 0.0	0.40 ± 0.0
Structural moieties (%)				
Unsubstituted	4.2 ± 0.1	4.9 ± 0.1	8.5 ± 0.5	10.7 ± 0.3
Methyl	2.4 ± 0.0	2.2 ± 0.1	2.8 ± 0.3	3.1 ± 0.2
Vinyl	30.9 ± 0.5	31.9 ± 0.9	34.0 ± 0.4	32.7 ± 1.3
4-VP[a]	8.7 ± 0.0	10.0 ± 0.3	10.2 ± 1.1	11.5 ± 0.0
4-VG[b]	19.7 ± 0.5	19.3 ± 1.3	20.8 ± 0.5	18.9 ± 1.3
C_α-ox	3.7 ± 0.0	3.8 ± 0.1	8.2 ± 0.2	11.4 ± 0.8
diketones	0.1 ± 0.0	0.2 ± 0.1	1.6 ± 0.1	3.1 ± 0.6
C_β-ox[c]	1.5 ± 0.0	1.5 ± 0.0	2.5 ± 0.1	2.8 ± 0.2
C_γ-ox	53.7 ± 0.6	52.5 ± 1.2	40.3 ± 0.3	35.8 ± 0.3
Miscellaneous	3.6 ± 0.1	3.3 ± 0.2	3.7 ± 0.0	3.4 ± 0.1
PhC$_\gamma$[d]	58.9 ± 0.6	57.4 ± 1.1	48.8 ± 0.2	46.1 ± 0.6
PhC$_\gamma$-diketones[e]	58.8 ± 0.6	57.3 ± 1.0	47.1 ± 0.2	43.0 ± 0.1

[a] 4-vinylphenol. [b] 4-vinylguaiacol. [c] excluding diketones. [d] phenols with intact α,β,γ carbon side chain. [e] phenols with intact α,β,γ carbon side chain, excluding diketones.

Table S2. Composition of free and bound lignin isolates. Lignin (% w/w) was determined by ^{13}C-IS py-GC-MS in duplicate on fractionation duplicates. Carbohydrates (% w/w) were determined by constituent monosaccharide analysis after H_2SO_4 hydrolysis in duplicate on pooled fractionation duplicates and are presented as anhydrosugars. Values given represent averages and standard deviation. Note that ^{13}C-IS py-GC-MS resulted in slight overestimation of lignin contents of bound lignin isolates (week 0 and 1).

	Lignin	Glucose	Xylose	Arabinose	Uronic acid	Galactose
Free lignin (weeks)						
0	75.1 ± 2.5	4.5 ± 0.2	18.1 ± 1.4	1.5 ± 0.1	2.1 ± 0.1	0.2 ± 0.0
1	74.7 ± 5.8	3.4 ± 0.2	18.6 ± 1.2	1.6 ± 0.1	2.0 ± 0.1	0.2 ± 0.0
3	54.8 ± 1.6	2.6 ± 0.1	15.8 ± 1.6	1.1 ± 0.1	2.0 ± 0.1	0.2 ± 0.0
7	45.0 ± 1.3	2.8 ± 0.2	12.7 ± 0.8	1.1 ± 0.1	1.7 ± 0.1	0.2 ± 0.0
Bound lignin (weeks)						
0	103.7 ± 5.4	1.3 ± 0.1	10.6 ± 0.6	2.0 ± 0.1	1.4 ± 0.2	0.2 ± 0.0
1	102.1 ± 2.2	1.6 ± 0.1	10.9 ± 0.8	2.1 ± 0.1	1.3 ± 0.2	0.1 ± 0.0
3	79.7 ± 3.9	1.5 ± 0.1	11.0 ± 0.7	2.0 ± 0.1	1.2 ± 0.2	0.1 ± 0.0
7	70.8 ± 1.8	1.5 ± 0.1	10.1 ± 0.8	1.8 ± 0.1	1.1 ± 0.2	0.0 ± 0.0

Table S3. ^{13}C-IS py-GC-MS relative abundance of lignin compounds in ball-milled water extracted residue R4 of wheat straw during growth of *C. subvermispora*. Corrected for relative response factors and relative abundance of ^{13}C analogues. Sum on the bases of structural classification according to van Erven et al.[3-4] Average and standard deviation of analytical duplicates on pooled biological triplicates.

	R4 (weeks)			
	0	**1**	**3**	**7**
Lignin subunits (%)				
H	11.3 ± 1.0	11.2 ± 0.4	11.7 ± 0.6	13.0 ± 0.7
G	59.4 ± 0.5	58.8 ± 0.4	60.6 ± 0.6	64.3 ± 0.4
S	29.3 ± 0.9	30.1 ± 0.1	27.7 ± 0.6	22.7 ± 0.4
S/G	0.49 ± 0.0	0.51 ± 0.0	0.46 ± 0.0	0.35 ± 0.0
Structural moieties (%)				
Unsubstituted	3.9 ± 0.3	4.4 ± 0.2	6.9 ± 0.6	7.3 ± 0.4
Methyl	2.1 ± 0.0	2.2 ± 0.2	3.4 ± 0.8	3.6 ± 0.7
Vinyl	29.1 ± 2.2	28.6 ± 0.9	32.5 ± 3.0	34.5 ± 0.7
4-VP[a]	10.1 ± 0.9	9.9 ± 0.4	9.1 ± 0.4	9.4 ± 0.7
4-VG[b]	17.0 ± 1.2	16.6 ± 0.7	20.3 ± 2.5	21.9 ± 0.8
C_α-ox	3.9 ± 0.2	4.1 ± 0.1	7.0 ± 0.3	8.2 ± 0.4
diketones	0.1 ± 0.0	0.2 ± 0.0	1.1 ± 0.1	1.6 ± 0.1
C_β-ox[c]	1.5 ± 0.1	1.5 ± 0.1	2.4 ± 0.2	2.6 ± 0.2
C_γ-ox	56.3 ± 2.9	55.6 ± 1.5	43.6 ± 5.4	39.3 ± 1.4
Miscellaneous	3.5 ± 0.3	3.6 ± 0.1	4.2 ± 0.6	4.5 ± 0.4
PhC_γ[d]	61.3 ± 2.6	61.1 ± 1.3	51.8 ± 4.7	48.6 ± 1.1
PhC_γ-diketones[e]	61.2 ± 2.7	60.9 ± 1.3	50.6 ± 4.7	47.0 ± 1.2

[a] 4-vinylphenol. [b] 4-vinylguaiacol. [c] excluding diketones. [d] phenols with intact α,β,γ carbon side chain. [e] phenols with intact α,β,γ carbon side chain, excluding diketones.

Table S4. ^{13}C-IS py-GC-MS relative abundance of lignin compounds in lignin isolates of wheat straw during growth of *C. subvermispora*. Corrected for relative response factors and relative abundance of ^{13}C analogues. Sum on the bases of structural classification according to van Erven et al.[3-4] Average and standard deviation of analytical duplicates on pooled biological triplicates.

	Free (weeks)				Bound (weeks)			
	0	**1**	**3**	**7**	**0**	**1**	**3**	**7**
Lignin subunits (%)								
H	8.6 ± 0.1	8.4 ± 0.1	8.6 ± 0.2	9.2 ± 0.1	7.9 ± 0.1	8.0 ± 0.0	7.8 ± 0.2	8.5 ± 0.1
G	62.8 ± 0.4	62.0 ± 0.4	64.7 ± 0.7	67.2 ± 0.8	57.1 ± 0.1	57.3 ± 0.4	60.6 ± 0.2	63.7 ± 0.3
S	28.6 ± 0.4	29.6 ± 0.4	26.6 ± 0.7	23.6 ± 0.6	35.0 ± 0.1	34.7 ± 0.4	31.7 ± 0.1	27.8 ± 0.5
S/G	0.45 ± 0.0	0.48 ± 0.0	0.41 ± 0.0	0.35 ± 0.0	0.61 ± 0.0	0.61 ± 0.0	0.52 ± 0.0	0.44 ± 0.0
Structural moieties (%)								
Unsubstituted	4.6 ± 0.1	4.5 ± 0.0	5.8 ± 0.0	6.8 ± 0.2	3.7 ± 0.1	3.8 ± 0.0	4.8 ± 0.1	5.0 ± 0.4
Methyl	1.5 ± 0.0	1.6 ± 0.0	1.9 ± 0.0	2.0 ± 0.1	1.5 ± 0.0	1.6 ± 0.0	1.9 ± 0.1	2.0 ± 0.1
Vinyl	18.8 ± 0.2	19.0 ± 0.0	19.0 ± 0.3	19.1 ± 0.5	19.8 ± 0.2	20.1 ± 0.1	21.5 ± 0.2	22.5 ± 0.5
4-VP[a]	6.8 ± 0.0	6.5 ± 0.2	6.2 ± 0.2	6.4 ± 0.4	6.8 ± 0.1	6.9 ± 0.1	6.3 ± 0.1	6.9 ± 0.2
4-VG[b]	10.3 ± 0.2	10.5 ± 0.2	11.0 ± 0.1	11.3 ± 0.1	10.7 ± 0.1	11.0 ± 0.1	12.9 ± 0.3	13.5 ± 0.3
C_α-ox	3.9 ± 0.1	4.2 ± 0.1	6.7 ± 0.1	9.0 ± 0.1	3.4 ± 0.0	3.6 ± 0.0	5.1 ± 0.1	5.3 ± 0.0
diketones	0.1 ± 0.0	0.2 ± 0.0	0.7 ± 0.0	1.8 ± 0.1	0.1 ± 0.0	0.1 ± 0.0	0.5 ± 0.0	0.7 ± 0.0
C_β-ox[c]	1.3 ± 0.0	1.3 ± 0.0	1.7 ± 0.0	1.8 ± 0.1	1.2 ± 0.0	1.2 ± 0.0	1.6 ± 0.0	1.6 ± 0.0
C_γ-ox	67.0 ± 0.3	66.4 ± 0.1	61.9 ± 0.4	59.3 ± 0.2	67.4 ± 0.3	66.6 ± 0.1	62.0 ± 0.3	60.6 ± 0.8
Miscellaneous	2.8 ± 0.0	2.9 ± 0.0	3.0 ± 0.0	3.1 ± 0.0	2.9 ± 0.0	3.0 ± 0.0	3.1 ± 0.0	3.1 ± 0.0
PhCγ[d]	71.8 ± 0.3	71.4 ± 0.1	68.4 ± 0.4	66.4 ± 0.2	72.0 ± 0.3	71.3 ± 0.1	67.7 ± 0.3	66.5 ± 0.7
PhC$_\gamma$-diketones[e]	71.7 ± 0.3	71.2 ± 0.1	67.7 ± 0.4	64.5 ± 0.2	71.9 ± 0.3	71.2 ± 0.1	67.2 ± 0.3	66.0 ± 0.7

[a] 4-vinylphenol. [b] 4-vinylguaiacol. [c] excluding diketones. [d] phenols with intact α,β,γ carbon side chain. [e] phenols with intact α,β,γ carbon side chain, excluding diketones.

Table S5. ^{13}C-IS py-GC-MS relative abundance of lignin compounds in unextractable residue R7 of wheat straw during growth of *C. subvermispora*. Corrected for relative response factors and relative abundance of ^{13}C analogues. Sum on the bases of structural classification according to van Erven et al.[3-4] Average and standard deviation of analytical duplicates on pooled biological triplicates.

	R7: dioxane insoluble residue (weeks)			
	0	**1**	**3**	**7**
Lignin subunits (%)				
H	12.7 ± 0.3	13.3 ± 0.6	12.4 ± 0.4	14.8 ± 0.6
G	55.3 ± 0.5	54.7 ± 0.3	56.8 ± 0.3	60.2 ± 1.0
S	32.1 ± 0.6	32.0 ± 1.0	30.7 ± 0.7	24.9 ± 0.5
S/G	0.58 ± 0.0	0.58 ± 0.0	0.54 ± 0.0	0.41 ± 0.0
Structural moieties (%)				
Unsubstituted	4.5 ± 0.1	4.9 ± 0.1	6.3 ± 0.3	7.4 ± 0.5
Methyl	2.5 ± 0.1	2.5 ± 0.1	2.8 ± 0.2	3.5 ± 0.1
Vinyl	27.1 ± 0.4	27.7 ± 1.1	27.9 ± 0.7	31.2 ± 1.3
4-VP[a]	10.8 ± 0.3	11.3 ± 0.5	9.9 ± 0.3	11.2 ± 0.7
4-VG[b]	13.5 ± 0.1	13.7 ± 0.5	15.0 ± 0.4	17.2 ± 0.5
C_α-ox	3.6 ± 0.1	3.9 ± 0.2	6.0 ± 0.1	7.0 ± 0.2
diketones	0.1 ± 0.0	0.2 ± 0.0	0.9 ± 0.1	1.2 ± 0.1
C_β-ox[c]	1.7 ± 0.1	1.7 ± 0.1	2.2 ± 0.1	2.6 ± 0.1
C_γ-ox	56.7 ± 0.5	55.3 ± 1.6	50.8 ± 1.2	44.1 ± 1.5
Miscellaneous	4.0 ± 0.0	4.0 ± 0.3	3.9 ± 0.1	4.2 ± 0.1
PhC_γ[d]	62.5 ± 0.4	61.3 ± 1.3	58.3 ± 1.0	52.6 ± 1.5
PhC_γ-diketones[e]	62.4 ± 0.4	61.1 ± 1.3	57.4 ± 1.0	51.4 ± 1.4

[a] 4-vinylphenol. [b] 4-vinylguaiacol. [c] excluding diketones. [d] phenols with intact α,β,γ carbon side chain. [e] phenols with intact α,β,γ carbon side chain, excluding diketones.

Table S6. Assignments of the lignin ^{13}C-^{1}H correlation peaks in the HSQC spectra of untreated and fungal-treated wheat straw lignin fractions.

label	δ_C/δ_H (ppm)	Assignment[a]
CHK	51.5/3.58	C-H in methoxyls of cyclohexadienone ketals (t)
B_β	53.0/3.43	C_β-H_β in phenylcoumaran substructures
C_β	53.6/3.05	C_β-H_β in resinol substructures
-OCH_3	55.6/3.72	C-H in methoxyls
A_γ	59.6/3.37, 59.8/3.59, 59.9/3.22	C_γ-H_γ in β-*O*-4' substructures
I_γ	61.4/4.09	C_γ-H_γ in cinnamyl alcohol end-groups
A'_γ	62.8/4.23, 63.9/4.18	C_γ-H_γ in γ-acylated β-*O*-4' substructures
C_γ	71.0/4.16, 71.1/3.80	C_β-H_β in resinol substructures
A_α(G)	70.9/4.71	C_α-H_α in β-*O*-4' substructures linked to G-unit
A_α(S)	71.8/4.81	C_α-H_α in β-*O*-4' substructures linked to S-unit
$A_{ox\beta}$	82.8/5.10	C_β-H_β in C_α-oxidized β-*O*-4' substructures
A_β(H)	83.1/4.49	C_β-H_β in β-*O*-4' substructures linked to H-unit
A_β(G)	83.5/4.27	C_β-H_β in β-*O*-4' substructures linked to G-unit
$A_\beta(S)_{erythro}$	85.9/4.09	C_β-H_β in β-*O*-4' substructures linked to S-unit
A_β(T)	86.2/4.36 and 86.7/4.26	C_β-H_β in β-*O*-4' substructures linked to tricin
$A_\beta(S)_{threo}$	86.9/3.97	C_β-H_β in β-*O*-4' substructures linked to S-unit
C_α	84.9/4.64	C_α-H_α in resinol substructures
B_α	86.9/5.43	C_α-H_α in phenylcoumaran substructures
T_8	94.1/6.57	C_8-H_8 in tricin
T_6	98.8/6.21	C_6-H_6 in tricin
T_{free3}	103.5/6.96	C_3-H_3 in free tricin
$S_{2,6}$	103.9/6.68	C_2-H_2 and C_6-H_6 in S-unit
$T_{2'6'}/T_{free2'6'}$	104.0/7.31	$C_{2'}$-$H_{2'}$ and $C_{6'}$-$H_{6'}$ in tricin and in free tricin
T_3	104.6/7.02	C_3-H_3 in tricin
$Sox_{2,6}$	106.4/7.30	C_2-H_2 and C_6-H_6 in C_a-oxidized (C_a=O) S-unit
$Sox_{2,6}$	106.5/7.19	C_2-H_2 and C_6-H_6 in C_a-oxidized (C_aOOH) S-unit
G_2	110.8/6.96	C_2-H_2 in G-unit
FA_2	110.9/7.34	C_2-H_2 in ferulate
$GoxI_2$	111.4/7.51	C_2-H_2 in C_a-oxidized G-unit
$GoxII_2$	112.4/7.44	C_2-H_2 in C_a-oxidized G-unit
FA_β/pCA_β	113.6/6.26	C_β-H_β in ferulate/*p*-coumarate
$H_{3,5}/FA_5$	114.5/6.69	C_3-H_3 and C_5-H_5 in H-unit, C_5-H_5 in FA
$G_5/G_6/pCA_{3,5}$	115.0/6.93 and 115.5/6.78	C_5-H_5 and C_6-H_6 in G-unit, C_3-H_3 and C_5-H_5 of pCA
G_5	119.0/6.78	C_5-H_5 in G-unit
$GoxI_6$	122.7/7.47	C_6-H_6 in C_a-oxidized G-unit
FA_6	123.1/7.12	C_6-H_6 in ferulate
$GoxII_6$	125.8/7.42	C_2-H_2 in C_a-oxidized G-unit
$H_{2,6}/PHE_{3,5}$	127.8/7.18	C_2-H_2 and C_6-H_6 in H-unit, C_3-H_3 and C_5-H_5 in phenylalanine
$PHE_{2,6}$	128.9/7.21	C_2-H_2 and C_6-H_6 in phenylalanine
$pCA_{2,6}$	130.1/7.42	C_2-H_2 and C_6-H_6 in *p*-coumarate
FA_α/pCA_α	145.1/7.57	C_α-H_α in ferulate/*p*-coumarate

[a]: assignment by comparison with literature.[4-11] (t): tentatively assigned

Table S7. Semi-quantitative HSQC NMR structural characterization of wheat straw lignin isolates in the absence and presence of chromium (III) acetylacetonate ($Cr(acac)_3$) as relaxation agent. Note that the untreated and *C. subvermispora* treated wheat straw lignin isolates shown here originate from a different batch of straw and should therefore solely be used for comparisons of $Cr(acac)_3$ effects.

	Untreated wheat straw (free + bound)		7 weeks fungal treated wheat straw (free)	
	Without $Cr(acac)_3$	With $Cr(acac)_3$	Without $Cr(acac)_3$	With $Cr(acac)_3$
Lignin subunits (%)[a]				
H	3	3	3	2
G	62	63	53	55
G_{ox}	0	0	10	11
S	35	34	24	23
S_{ox}	0	0	9	9
S/G	0.6	0.5	0.5	0.5
Hydroxycinnamates (%)[b]				
***p*-coumarate**	8	6	17	13
ferulate	5	3	6	4
Flavonolignin (%)[b]				
tricin	15	9	21	15
Lignin interunit linkages (%)[b,c]				
β-*O*-4' G+H	20 (35)	22 (37)	18 (38)	19 (38)
β-*O*-4' S	22 (38)	24 (40)	14 (29)	16 (32)
β-*O*-4' C_α-ox	1 (1)	1 (2)	0 (0)	0 (0)
β-*O*-4' tricin	8 (15)	8 (13)	11 (22)	11 (23)
total β-*O*-4' aryl ethers	51 (89)	55 (92)	43 (89)	46 (93)
β-5' phenylcoumarans	5 (8)	4 (6)	3 (7)	2 (5)
β-β' resinols	1 (2)	1 (2)	2 (4)	1 (2)
total	57 (100)	60 (100)	49 (100)	50 (100)
Side-chain γ-acylation (%)[d]	21	20	17	16
***Erythro*/*threo* of *β*-*O*-4**[e]	2.6	2.5	3.9	4.0

[a] relative distribution of lignin subunits ($H+G+G_{ox}+S+S_{ox}=100$)
[b] relative volume integral of substructure versus volume integral of total lignin subunits
[c] relative distribution of total interunit linkages in parentheses
[d] percentage of β-*O*-4' aryl ethers
[e] ratio of $A_\beta(S/G\text{-}S)_{erythro}$ and $A_\beta(S/G\text{-}S)_{threo}$, diastereomers for β-*O*-4' aryl ethers coupled to G-units are not resolved

Table S8. Hydroxyl group content of lignin isolates of wheat straw during growth of *C. subvermispora* determined by ^{31}P NMR after phosphitylation.

	Free (weeks) (mmol/g lignin)[a]				Bound (weeks) (mmol/g lignin)[a]			
	0	**1**	**3**	**7**	**0**	**1**	**3**	**7**
Aliphatic OH + carb. OH	7.49	7.77	9.40	10.15	5.81	5.86	6.73	7.28
S-OH + 5-sub. G-OH	0.56	0.62	0.95	1.23	0.39	0.43	0.63	0.71
G-OH	0.74	0.72	0.88	1.01	0.53	0.53	0.61	0.71
H-OH + *p*CA-OH	0.57	0.57	0.78	0.93	0.42	0.42	0.53	0.65
Total phenolic OH	1.87	1.91	2.61	3.17	1.34	1.38	1.76	2.07
COOH	0.28	0.36	0.79	1.33	0.14	0.17	0.34	0.43

[a]contents per g biomass corrected for lignin contents as determined by quantitative ^{13}C-IS py-GC-MS

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