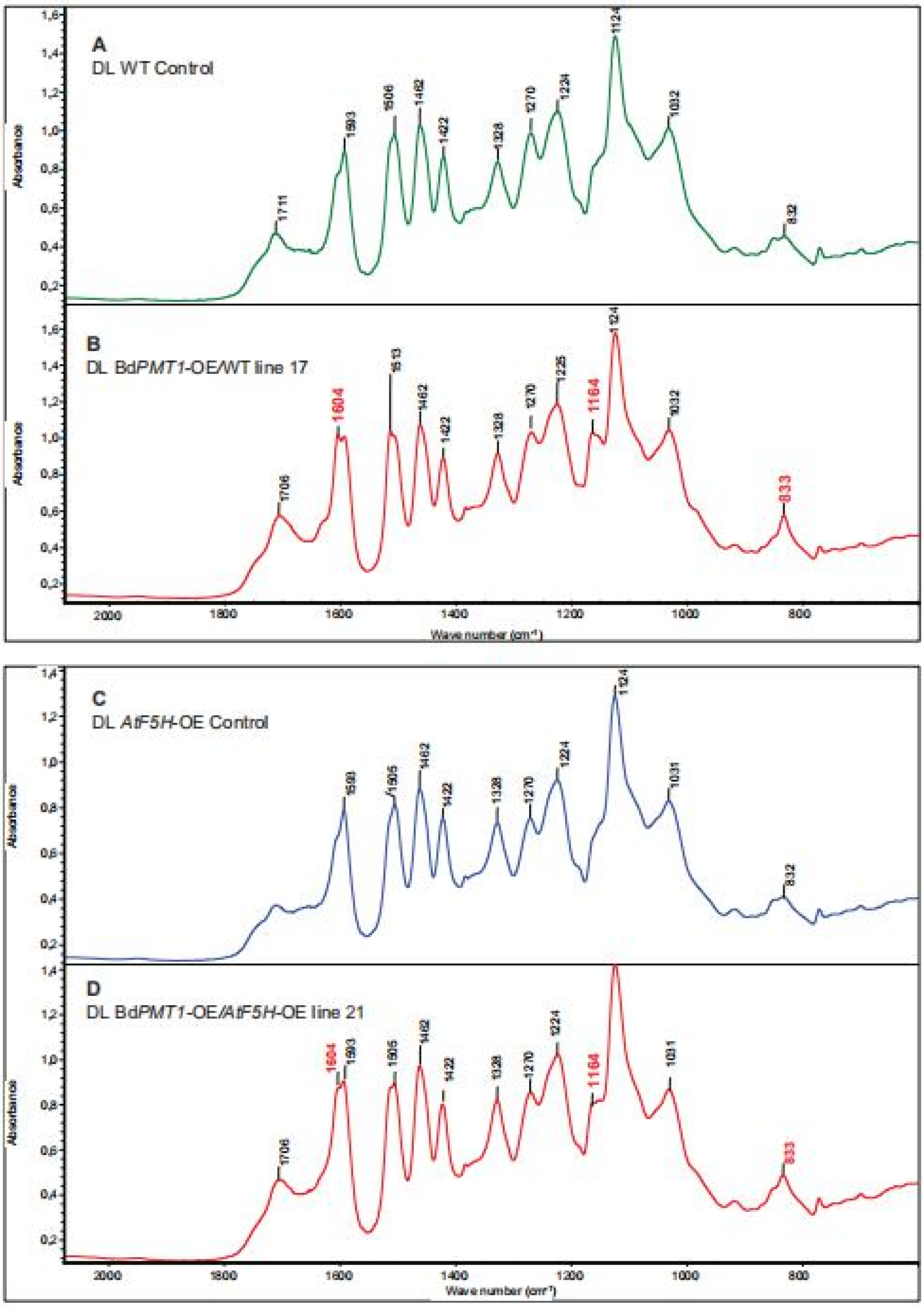
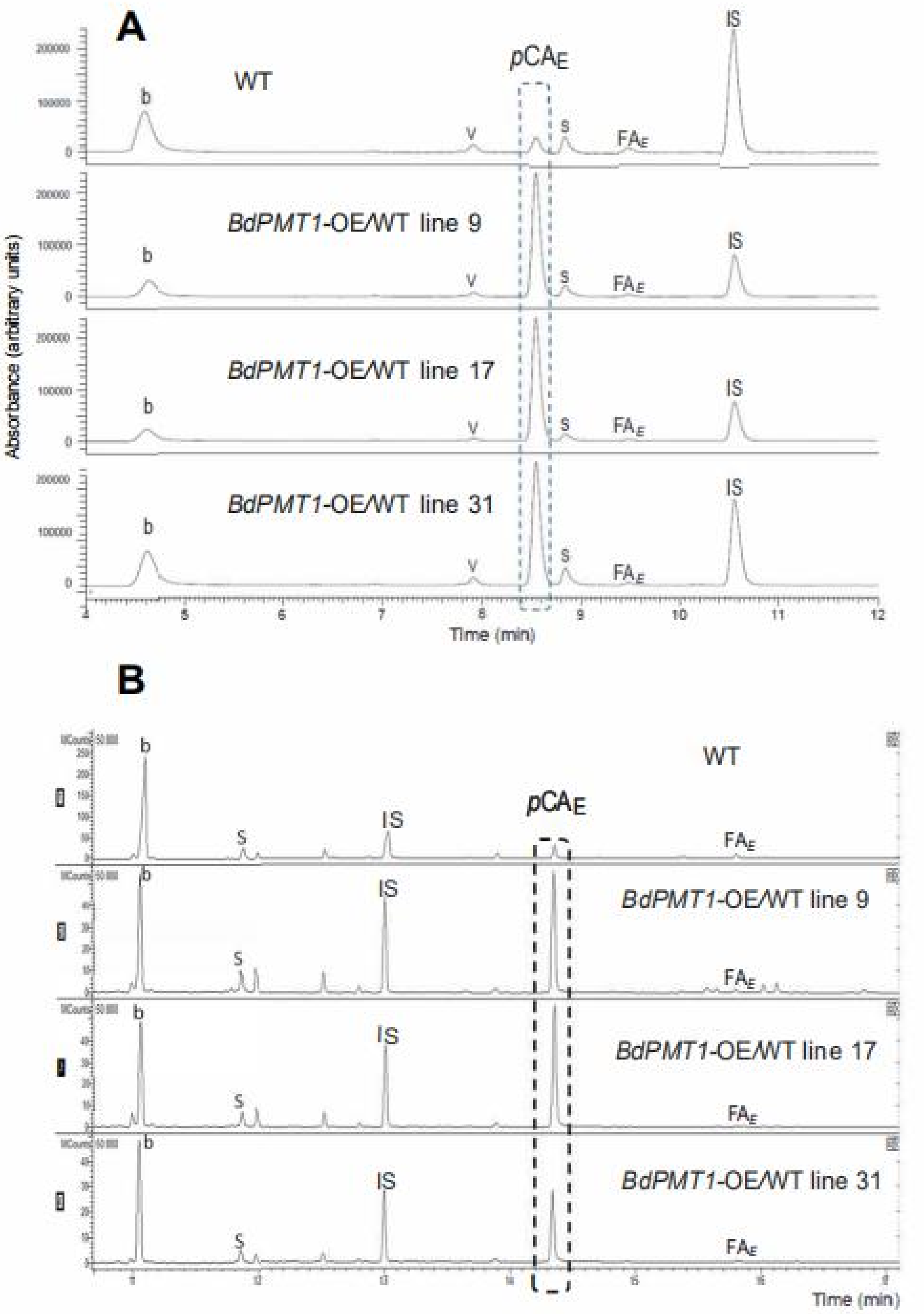
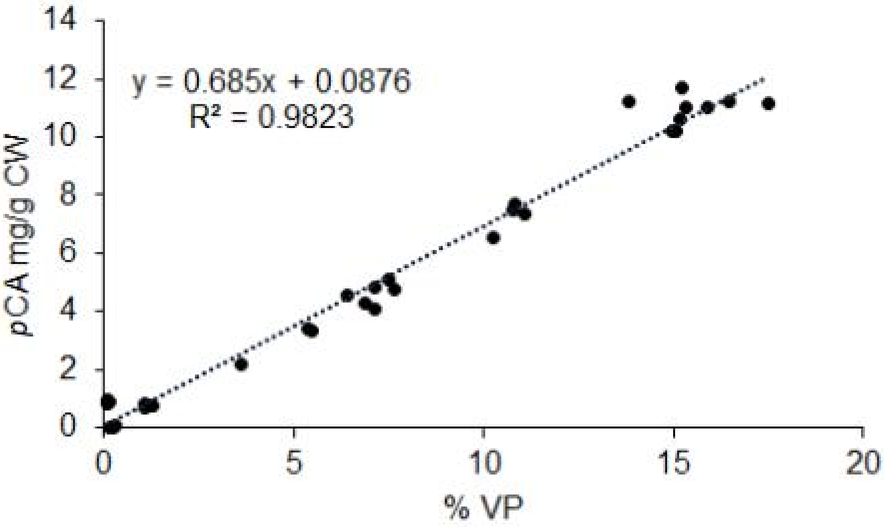
Supplemental Figure S1. Molecular analyses on greenhouse-grown plant material A. Greenhouse plants before sampling. B-C: Semi-quantitative RT-PCR using BdPMT1 and AtF5H specific primers. Reactions were carried out on three different plants of each transgenic line (only 2 plants for BdPMT1-OE/AtF5H-OE line 1).



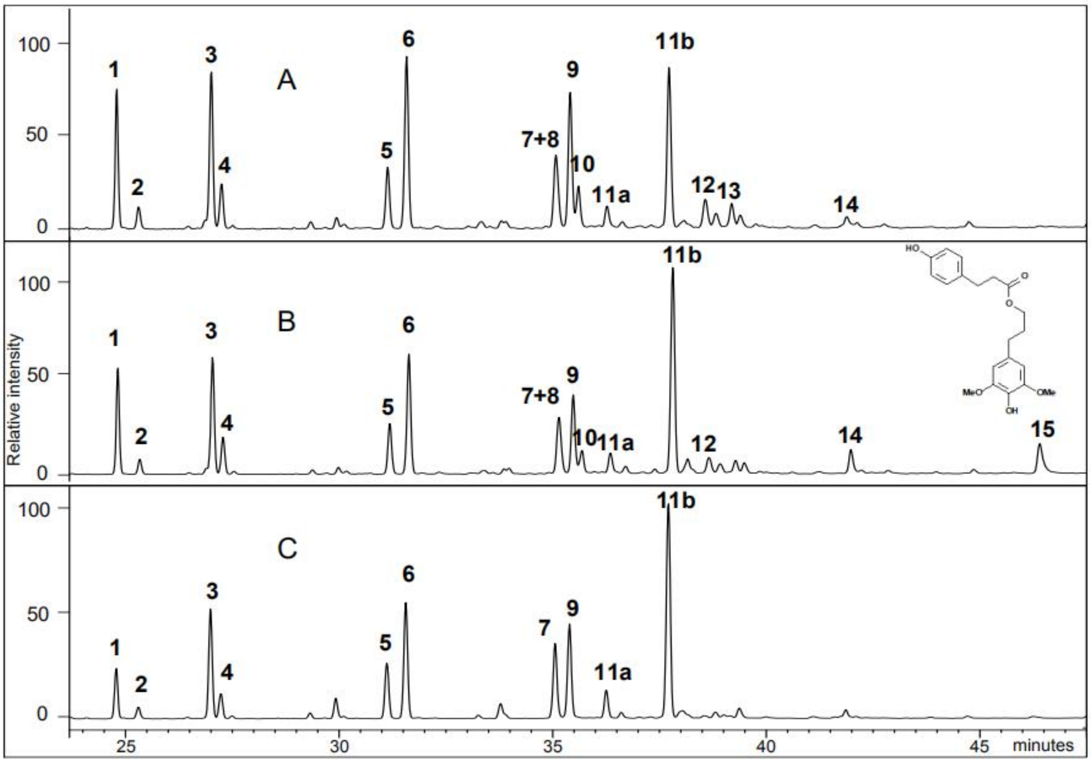
Supplemental Figure S2. IR spectra (KBr pellet) of Dioxane Lignin (DL) fractions isolated from A) WT line, B) *BdPMT1*-OE/WT line 17, C) *AtF5H*-OE and D) *BdPMT1*-OE / *AtF5H*-OE line 21. The IR wavenumbers quoted in red correspond to bands that are more pronounced in the DL samples from the transgenic lines and due to the occurrence of *p*-coumaric esters. DL extraction was performed from control samples (as biological duplicates for WT and for *AtF5H*-OE), from the *BdPMT1*-OE/WT lines 9, 17 and 31, as well as from the *BdPMT1*-OE/*AtF5H*-OE line 21. The differences outlined in this Figure (signals quoted in red) were systematically observed between any *BdPMT1*-OE line and the corresponding controls.



Supplemental Figure S3. Determination of the low-molecular weight phenolics released by alkaline hydrolysis of Dioxane Lignin (DL) fractions isolated from BdPMT1-OE/WT lines and their WT control, as measured A) by HPLC analysis with diode array detection (250-400 nm) and B) by GC/MS after trimethylsilylation. The quoted peaks correspond to : b p-hydroxybenzoic acid ; pCAE E isomer of p-coumaric acid ; FAE E isomer of ferulic acid ; IS internal standard (o-coumaric acid) ; V vanillin ; S : syringaldehyde.



Supplemental Figure S4. Relationship between the amount of p-coumaric acid *p*CA released by mild alkaline hydrolysis of control or *BdPMT1*-OE poplar cell walls and the relative percentage of 4-vinylphenol (% VP) released by analytical pyrolysis of the same poplar samples. The samples correspond to extract-free stems from 3-month-old trees (WT and *AtF5H*-OE backgrounds). The % VP is expressed as percentage of the total area of the main pyrolysis-derived phenolics.



Supplemental Figure S5. Partial GC/MS chromatograms of the main dimers (analyzed as their TMS derivatives) obtained by thioacidolysis and Raney nickel desulfuration of A) WT poplar sample with 1-hour long thioacidolysis, B) *BdPMT1*-OE/WT line 17 poplar sample with 1-hour long thioacidolysis and C) *BdPMT1*-OE/WT line 17 poplar sample with 4-hour long thioacidolysis. Peak identification is given in Supplemental Table S2. Peak 15 is observed only in B and corresponds to syringylpropanol acylated by dihydro-p-coumaric acid, as shown in the scheme.

Supplemental Table S1. Amount of p-coumaric acid (pCA) released by mild alkaline hydrolysis of grass cell walls (CW) and of the corresponding purified Dioxane Lignin (DL) fractions. The data represent means (and SD) from triplicate analyses.

Samples	*p*CA mg/g CW	*p*CA mg/g DL
Mature stems of C3 grasses		
Barley (*Hordeum vulgare*)	6.03 (0.10)	
Brachypodium distachyon Bd21-3	6.34 (0.09)	41.8 (1.6)
Oat (*Avena sativa*)	7.22 (0.05)	
Rice (*Oryza sativa*)	11.87 (0.24)	75.07 (3.77)
Wheat (*Triticum aestivum*)	4.76 (0.10)	23.8 (1.4)
Mature stems of C4 grasses		
Maize (*Zea mays*, F2 line, silage stage)	17.27 (0.71)	119.8 (2.1)
Miscanthus giganteus	17.84 (0.44)	94.7 (0.5)
Miscanthus sinensis	16.36 (0.37)	86.9 (1.8)
Sorghum (*Sorghum bicolor*)	22.43 (0.19)	121.8 (7.2)

Supplemental Table S2. Identification and relative importance (% area) of the main dimers obtained after thioacidolysis and Raney nickel desulfuration of extract free poplar stems. The peak number corresponds to the peak quotation of Supplemental Fig. S5. The main difference between the transgenic sample and its control is the higher relative importance of SS dimers 11a and 11b issued from syringaresinol structures. ND : Not detected. The relative amounts are mean values (and standard deviation) between four technical replicates of WT and line 17 samples.

Peak	Dimer type and corresponding bonding pattern[a]	M⁺ (rel intensity)	Rel. amount in WT and with 4-h long thioacidolysis	Rel. amount in *BdPMT1*-OE/WT line 17 and with 4-h long thioacidolysis [d]
1	GG Biphenyl 5-5'	474 (100)	6.86 (0.80)	5.81 (0.99)
2	GG Biphenyl ether 4-O-5'	402 (100)	1.99 (0.10)	1.78 (0.31)
3	GG Diarylpropane β-1'	418 (10)	14.46 (0.70)	13.75 (0.86)
4	SG Biphenyl ether 4-O-5'	432 (100)	4.63 (0.10)	3.72 (0.55)*
5	SG Diarylpropane β-1'	448 (25)	7.15 (0.30)	7.21 (0.51)
6	GG β-5' without CH_2OH at Cγ	460 (25)	17.76 (0.80)	16.40 (1.16)
7	SS Diarylpropane β-1'	478 (40)	7.86 (1.24)	9.00 (1.17)
8	GG β-5' with CH_2OH at Cγ	562 (25)	traces	traces
9	SG β-5' without CH_2OH at Cγ	490 (30)	18.25 (1.00)	12.69 (0.82)**
10	GG β-O-4'[b]	490 (1)	ND	ND
11a	SS β- β' from syringaresinol without CH_3 at Cγ	518 (100)	2.55 (0.18)	**3.54 (0.29)****
11b	SS β- β' from syringaresinol	532 (70)	18.50 (1.72)	**26.11 (1.92)****
12	SG β-5' with CH_2OH at Cγ	592 (30)	traces	traces
13	SG β-O-4'[b]	520 (5)	ND	ND
14	SS β-O-4'[b]	550 (2)	ND	ND
15	S-diHCA [c]	504 (100)	ND	ND

[a] G : guaiacyl unit; S : syringyl unit.
[b] Peaks 10, 13 and 14, diagnostic for residual β-O-4' dimers, are observed only with1 hour-long thioacidolysis.
[c] Peak 15 corresponds to syringylpropanol acylated by p-dihydrocoumaric acid (diHCA) and is observed only with 1-hour long thioacidolysis in BdPMT1-OE poplars.
[d] Asterisks denote significant differences (one way ANOVA) with the control (*: P <0.05, **: P < 0.01).

Supplemental Table S3. Saccharification of extract-free poplar stems corresponding to *BdPMT1*-OE lines obtained in the WT and *AtF5H*-OE backgrounds, as compared to their controls. The saccharification efficiency is evaluated both by the weight loss (%WL) and by the released glucose (Glc). The data represent mean values (and SD) from biological triplicates. A statistical test (one-way ANOVA) revealed that the transgenic samples were not significantly different as compared to the value of the corresponding control (at P < 0.05)

Extract-free poplar stem from line	% WL	Glc mg.g^{-1} sample
WT control	18.2 (0.6)	74.6 (0.4)
BdPMT1-OE/WT line 9	20.2 (3.1)	98.2 (21.2)
BdPMT1-OE/WT line 17	18.6 (0.10)	80.1 (3.4)
BdPMT1-OE/WT line 31	20.4 (2.1)	102.4 (11.5)
AtF5H-OE control	18.6 (2.4)	85.9 (20.7)
BdPMT1-OE/*AtF5H*-OE line 21	23.1 (2.2)	116.5 (16.2)