## **Supporting Information**

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## **SI Materials and Methods**

**Constructs for Transfection Studies.** SND1 (At1g32770), MYB46 (At5g12870), and MYB58 (At1g16490) were cloned into the vector p2GW7 (1). The fragment (1-3414) of the vector pPGWL7 (1), which contains the firefly luciferase ORF was transferred to p2GW7 to generate a small high-copy vector with the firefly luciferase sequence. The promoters (≈1-kb regions) of F5H, C4H, COMT, and CCoAOMT were cloned into pENTR vector and then switched into the modified vector. The primers used for promoter cloning were: ProF5H-Forward 5- CACCGATTCTCACATTG-TGTTTCTATTATATATG-3, ProF5H-Reverse 5-GTTAACTTGTG-TGTTTCTATTTTTGGTTTC-3; ProC4H-Forward 5-CACCCT-CTAATTGAATAGCATGTTACCCGGTCCAACC-3, ProC4H-Reverse 5- TGTTGAGTGTGTTTTTGTGTTTTTGGGTAATG-AAA-3; ProCOMT-Forward 5-CACCGGTCACGTACATTTA-GTTGTGTGTGTGTGTGACCATATTTTC-3, ProCOMT-Reverse 5-GGT-

1. Karimi M, Inzé D, Depicker A (2002) GATEWAY vectors for Agrobacterium-mediated plant transformation. *Trends Plant Sci* 7:193–195.

TGTGATTGATGAGGTTTTTTGAGATTGTAGTGGTG-3; and ProCCoAOMT-Forward 5-CACCGCATCAAAAACATGTC-AAAACTAGCAATTCACTC-3, ProCCoAOMT-Reverse 5-TG-TTAAATTCTTGGAATGTTTTGCTTCTTTTCTG-3.

**F5H** Promoter Fragments for Competiton EMSA Assays. P1 to P6 are 50-bp fragments from -349 to -100 of the F5H promoter region, with 10-bp overlaps between each other. Their sequences are:

- P1, 5'-atcaagcatatcaaacgtgtgttaaatatctattctatcctcaaatctca-3';
- P2, 5'-ctcaaatctcaatgttacgttttttcctccaaatgctttattagttgcgt-3';
- P3, 5'-ttagttgcgtccacttgcattatatatatatctattgatacatatattttt-3';
- P4, (5'-catatatttttcttaaccaaggtttacactcttcaagcaactattatata-3';
- P5, 5'-tattatataagaccccataatatatctcacaattcagcaacccttagagt-3';
- P6, 5'-accettagagttgcaatctcacatcaactctcatcacccaacaacaaca-3').



Fig. S1. Subclade containing Arabidopsis F5H proteins. This subclade is taken from Fig. 6A and shows the evolutionary orthologs of Arabidopsis F5H proteins in other organisms.



Fig. 52. Subclade containing Arabidopsis NAC proteins. This subclade is taken from Fig. 6B and shows the evolutionary orthologs of Arabidopsis NST proteins in other organisms.



Fig. S3. Subclade containing the N-terminal domain of Arabidopsis MYB58 protein. This subclade is taken from Fig. 6C and shows the evolutionary orthologs of Arabidopsis MYB58 proteins in other organisms.



Fig. S4. Subclade containing the C-terminal domain of Arabidopsis MYB58 protein. This subclade is taken from Fig. 6C and shows the evolutionary orthologs of Arabidopsis MYB58 proteins in other organisms.



**Fig. S5.** Subclade containing the full-length region of *Arabidopsis* MYB58 protein. Proteins in Figs. S3 and S4 were collected, a multiple sequence alignment was performed, and then a new phylogeny was rebuilt using the full-length regions. This was done because the evolutionary orthologs of *Arabidopsis* MYB58 proteins in other organisms are not consistent between Figs. S3 and S4.



Fig. S6. Subclade containing the N-terminal domain of Arabidopsis MYB46 protein. This subclade is taken from Fig. 6C and shows the evolutionary orthologs of Arabidopsis MYB46 proteins in other organisms.

DNA C



Fig. 57. Subclade containing the C-terminal domain of Arabidopsis MYB46 protein. This subclade is taken from Fig. 6C and shows the evolutionary orthologs of Arabidopsis MYB46 proteins in other organisms.



Fig. S8. Model of the transcriptional network of secondary cell wall biosynthesis. Both SND1/NST1 and MYB46/MYB83 can activate the entire secondary cell wall biosynthesis program. F5H is regulated by SND1/NST1 whereas other lignin genes are regulated by MYB58/MYB63.

## Table S1. The 21 land-plant and algal species used in the phylogenetic analyses

Species	Clade	No. of UniGenes/ proteins
Cyanidioschyzon merolae 10D	Unicellular red algae	5014
Chlamydomonas reinhardtii	Chlorophytic green algae	14598
Mesostigma viride	Green algae	7371
Marchantia polymorpha	Liverwort	10959
Physcomitrella patens ssp patens	Moss	35938
Selaginella moellendorffii	Spike moss	34697
Adiantum capillus veneris	Fern	16944
Cycas rumphii	Gymnosperms	10901
Cryptomeria japonica	Gymnosperms	24299
Ginkgo biloba	Gymnosperms	10210
Picea abies	Gymnosperms	5184
Picea allspecies	Gymnosperms	72011
Picea engelmannii x Picea glauca	Gymnosperms	13880
Picea engelmannii x Picea sitchensis	Gymnosperms	13880
Picea glauca	Gymnosperms	53255
Picea sitchensis	Gymnosperms	29178
Pinus pinaster	Gymnosperms	12901
Pinus taeda	Gymnosperms	77540
Welwitschia mirabilis	Gymnosperms	6606
Arabidopsis thaliana	Dicot	33410
Medicago truncatula	Dicot	53423

Six sequenced plant genomes are underlined; the remainder are EST assemblies taken from plant GDB. Species are ordered from lower plants to higher plants.

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