

Figure S2

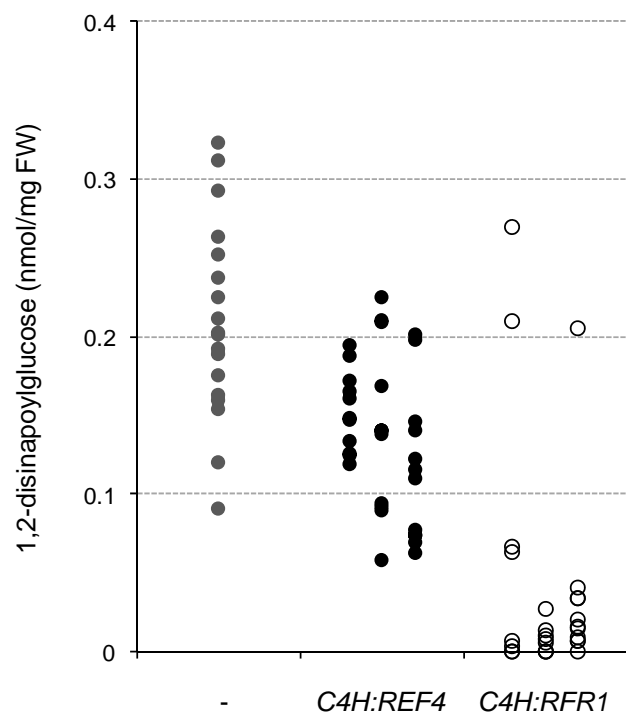
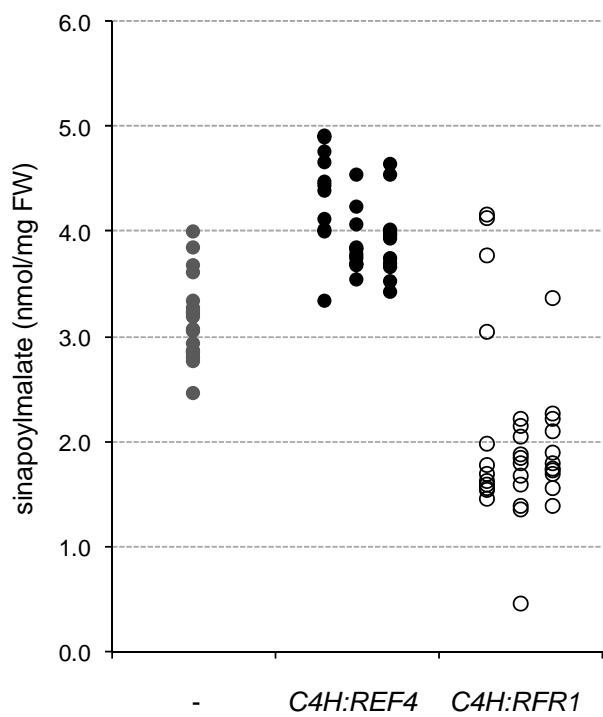
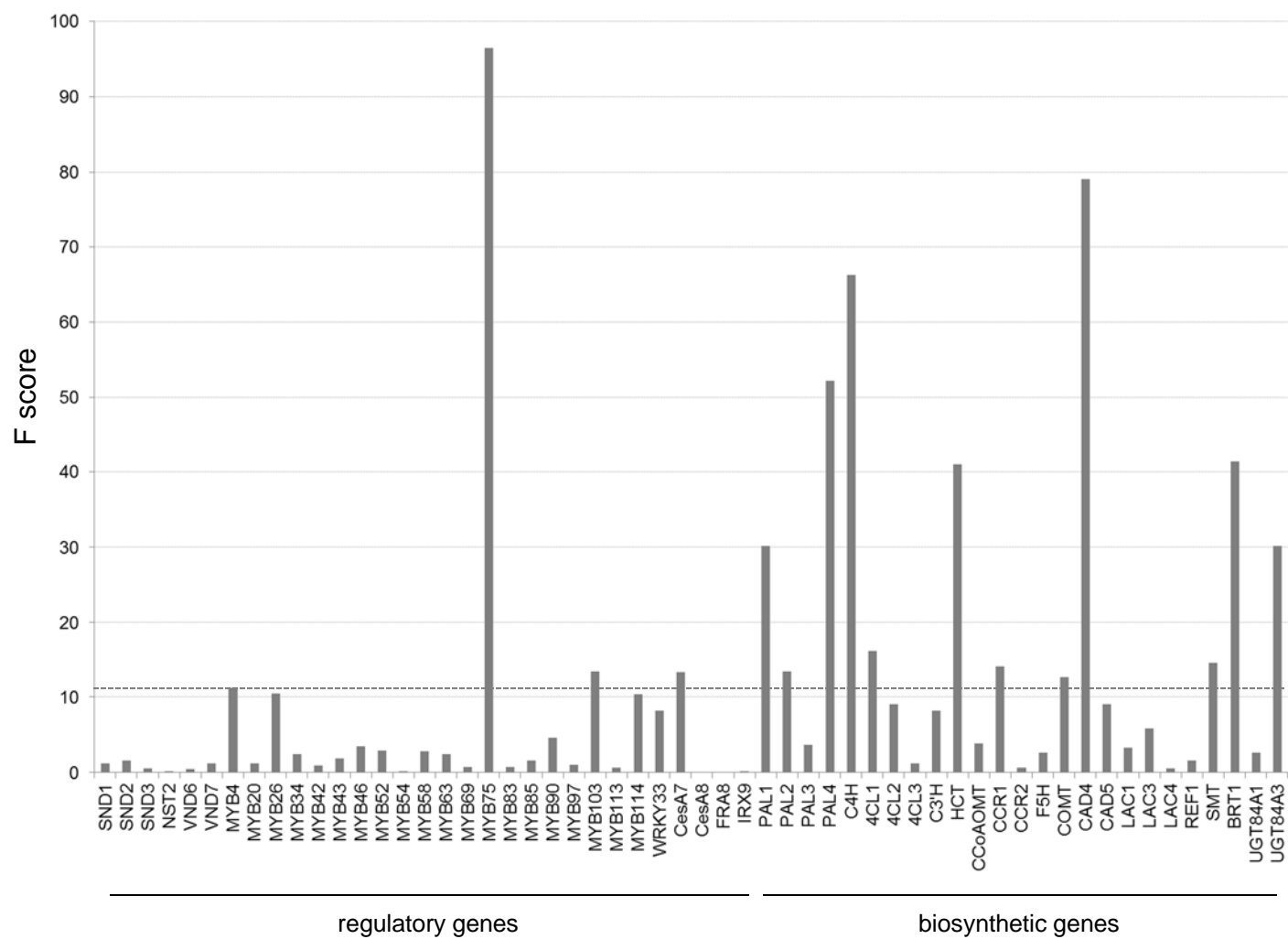


Figure S3







Supplemental Table SI. REF4/RFR1/RFR2 family members used for phylogenetic analysis

Gene	Accession number	Source
Ppatens REF4B	Pp1s106_135V6	Phytozome
Ppatens REF4A	Pp1s23_340V6.1, Pp1s23_340V6.2	Phytozome
Smoellendorffii REF4	168663	JGI
Vvinifera RFR2	XP_002264843	NCBI
Mtruncatula RFR2	Medtr7g080810	Phytozome
Ppersica RFR2	ppa017529m.g	Phytozome
Cpapaya RFR2	evm.TU.supercontig_60.37	Phytozome
Ptrichocarpa RFR2	XP_002323105	NCBI
Rcommunis RFR2	EEF42295	NCBI
Mesculenta RFR2	cassava4.1_000362m.g	Phytozome
Osativa RFR2B	EEC78956	NCBI
Sbicolor RFR2B	4894134	JGI
Zmays RFR2B	GRMZM2G358491	Phytozome
Osativa RFR2A	EEC82696	NCBI
Bdistachyon RFR2A	Bradi1g17590	Phytozome
Zmays RFR2A1	GRMZM2G151983	Phytozome
Sbicolor RFR2A	5052260	JGI
Zmays RFR2A2	GRMZM2G106790	Phytozome
Bdistachyon REF4	Bradi1g54150	Phytozome
Osativa REF4	EAZ03202	NCBI
Sbicolor REF4	4746974	JGI
Zmays REF4	GRMZM2G150754	Phytozome
Vvinifera REF4	XP_002271735	NCBI
Mguttatus REF4	mgv1a000266m.g	Phytozome
Ptrichocarpa REF4A	XP_002302829	NCBI
Ptrichocarpa REF4B	XP_002320389	NCBI
Rcommunis REF4	EEF27765	NCBI
Mesculenta REF4A	cassava4.1_000297m.g	Phytozome
Mesculenta REF4B	cassava4.1_000404m.g	Phytozome
Athaliana REF4	At3g23590	TAIR
Ppersica REF4	ppa000301m.g	Phytozome
GmaxREF4A	Glyma03g30760	Phytozome
GmaxREF4B	Glyma05g09220	Phytozome
GmaxREF4C	Glyma19g00780	Phytozome
Mguttatus RFR1	mgv1a00297m.g	Phytozome
Ppersica RFR1B	ppa017778m.g	Phytozome
Vvinifera RFR1	XP_002277484	NCBI
Csativus RFR1	Cucsa.260950	Phytozome
Ppersica RFR1A	ppa000298m.g	Phytozome
Gmax RFR1	Glyma15g10970	Phytozome
Mtruncatula RFR1	Medtr2g026510, Medtr2g06520	Phytozome
Athaliana RFR1	At2g48110	TAIR
Cpapaya RFR1	evm.TU.supercontig_16.34	Phytozome
Ptrichocarpa RFR1	XP_002311827	NCBI
Rcommunis RFR1	EEF45403	NCBI
Mesculenta RFR1A	cassava4.1_000332m.g	Phytozome
Mesculenta RFR1B	cassava4.1_028600m.g	Phytozome

Supplemental Table SII. Primers used in this study

CC1446 ACGGGGCATTGATAGAAAAA
CC1554 AGGAGGGAATCGACAATGTG
CC1662 GCGTGGACCGCTTGCTGCAACT
CC1776 TTGGCATTAGTGAGCAAGCA
CC1791 CGTCACGTTCGACAGCTTAGAGGAGGAACTGAGA
CC1792 GAGCGAATTCTATAGTTTGTGTATCCGCAATG
CC1834 TTTTATGGGCCTCTTTCGTG
CC1871 GATAAGCTTGGATCCCAGTCACGACGTT
CC1872 TATCTAGAGTTTTCCCAGTCACGACGTT
CC2206 GGGGACAAGTTTGTACAAAAAAGCAGGCTCGGTCGTCCAACAAATGG
CC2207 GGGGACCACTTTGTACAAGAAAGCTGGGTCAAGCTAATGTTGATGGCAAG
CC2204 GGGGACAAGTTTGTACAAAAAAGCAGGCTTCAAATGGTGGTTCCTGGAC
CC2205 GGGGACCACTTTGTACAAGAAAGCTGGGTGCTATGAACGCAAATGAATG
CC2946 GGGGACAAGTTTGTACAAAAAAGCAGGCTTAGGGAATATGCGTTTAGGTTTC
CC2947 GGGGACCACTTTGTACAAGAAAGCTGGGTAGCTGTTGTTTCCTCTCAAGC
CC3280 GGAAGCTTTCAAATTAGCAGGAAT
CC3281 GACTAGCGCGAGACCTTCCTTA
CC3282 ACCGGCCGTCCTAATTCC
CC3283 GCTTCTTTCGCGGTTAGCGATTC
CC3256 GCCACTAAGCCTTGCCTAATCA
CC3257 CGTGGACGTCGGAGTAAGTGT
CC3258 TCTCCTCGTGCCTCACATGA
CC3259 TGCTTTCTGCTGGGATATCGTA
CC2558 TAACGTGGCCAAAATGATGC
CC2559 GTTCTCCACAACCGCTTGGT

Supplemental Figure legends

Supplemental Figure S1. *ref4 rfr1*-deficient plants hyperaccumulate phenylpropanoids in stems and roots.

(B) HPLC profiles of methanol soluble, UV absorbing metabolites from inflorescence stems and (C) soil-grown roots of wild-type and *ref4 rfr1* null plants.

Supplemental Figure S2. RFR1 complements the metabolic phenotype of *ref4 rfr1*-deficient plants.

(A) Sinapoylmalate concentration in individual T2 *ref4 rfr1* plants transformed with a C4H-driven *RFR1* or *REF4* constructs. (B) 1,2-disinapoylglucose concentration of the plants in (A).

Supplemental Figure S3. Genetic manipulation of *REF4* and *RFR1* leads to dysregulation of phenylpropanoid biosynthetic genes, but not regulatory proteins.

Expression of the indicated genes in wild-type, *ref4-3*, and *ref4 rfr1* null plants was measured by microarray (N=3 for each) and subjected to ANOVA. The y-axis shows the resulting F score for each gene, with an F score greater than 10.9 (dashed line) coinciding with a p-value less than 0.01 for an effect of *REF4/RFR1* on the expression of a given gene. The expression of phenylpropanoid regulatory genes was relatively unaffected by *REF4/RFR1*, with the exception of *MYB75 (PAP1)*, which was upregulated in the *ref4-3* mutant. Phenylpropanoid regulatory shown along the x-axis are *SND1* (At1g32770), *SND2* (At4g28500), *SND3* (At1g28470), *NST2* (At3g61910), *VND6* (At5g62380), *VND7* (At1g71930), *MYB4* (At4g38620), *MYB20* (At1g66230), *MYB26* (At3g13890), *MYB34* (At5g60890), *MYB42* (At4g12350), *MYB43* (At5g16600), *MYB46* (At5g12870), *MYB52* (At1g17950), *MYB54* (At1g73410), *MYB58* (At1g16490), *MYB63* (At1g79180), *MYB69* (At4g33450), *MYB75* (At1g56650), *MYB83* (At3g08500), *MYB85* (At4g22680), *MYB90* (At166390), *MYB97* (At4g26930), *MYB103* (At1g63910), *MYB113* (At1g66370), *MYB114* (At1g66380), *WRKY33* (At2g38470), *CesA7* (At5g17420), *CesA8* (At4g18780), *FRA8* (At2g28110), and *IRX9* (At2g37090). Phenylpropanoid biosynthetic shown are *PAL1* (At2g37040), *PAL2* (At3g53260), *PAL3* (At5g04230), *PAL4* (At3g10340), *C4H* (At2g30490), *4CL1* (At1g51680), *4CL2* (At3g21240), *4CL3* (At1g65060), *C3'H* (At2g40890), *HCT* (At5g48930), *CCoAOMT* (At4g34050), *CCR1* (At1g15950), *CCR2* (At1g80820), *F5H* (At4g36220), *COMT* (At5g54160), *CAD4* (At3g19450), *CAD5* (At4g34230), *LAC1* (At1g18140), *LAC3* (At2g30210), *LAC4* (At2g38080), *REF1* (At3g24503), *SMT* (At2g22990), *BRT1* (At3g21560), *UGT84A1* (At4g15480), and *UGT84A3* (At4g15490).

Supplemental Figure S4. The *pap1-D* mutant allele does not induce the accumulation of anthocyanins when expressed in the *ref4-3* mutant background.

Supplemental Figure S5. An intragenic R387H mutation rescues the dwarf phenotype of *ref4-3* mutant plants. The plants labeled *sup/sup* and *sup/ref4-3* are homozygous and heterozygous for the intragenic suppressor allele *ref4-G383S R387H*.