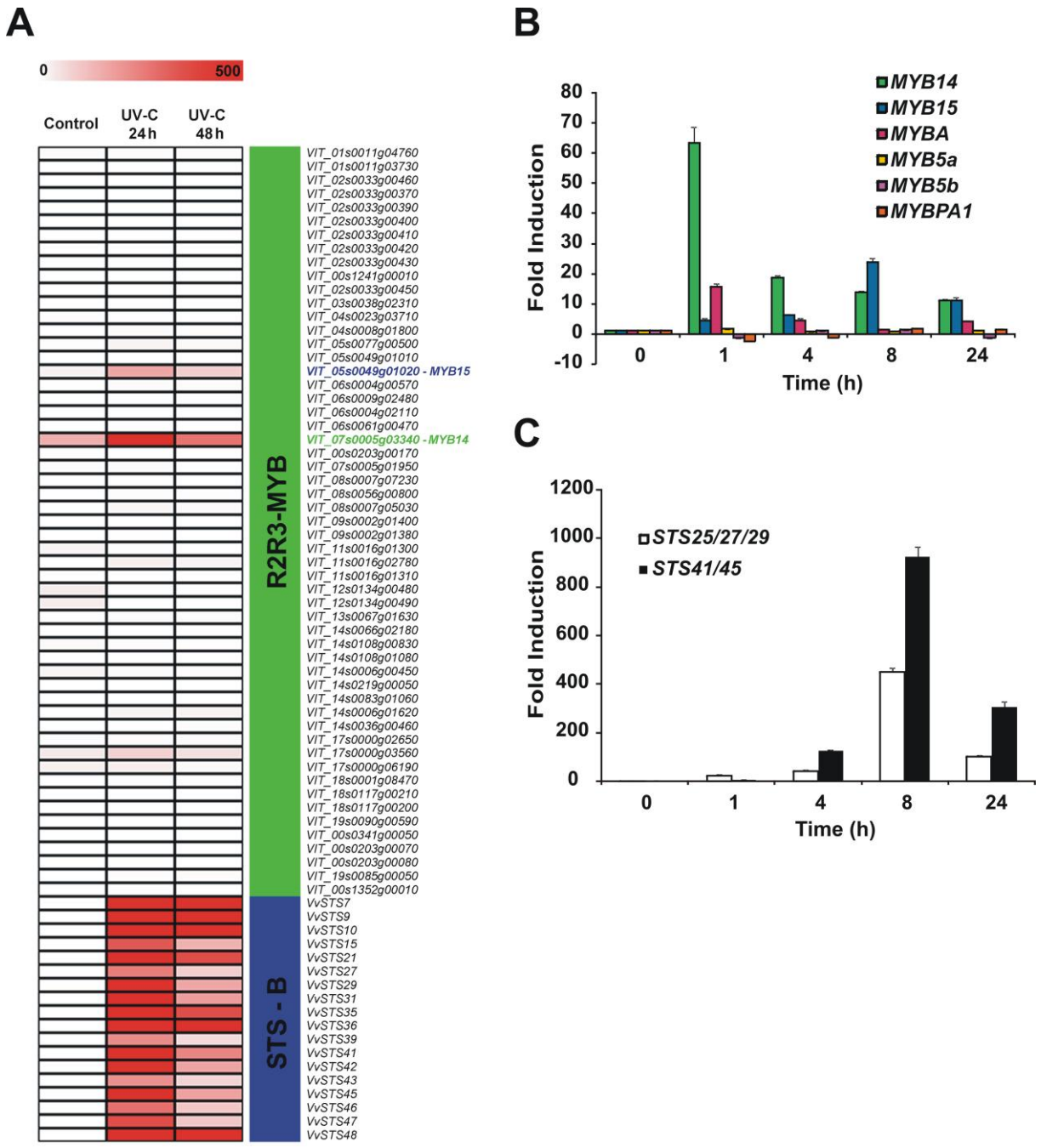
Supplemental Data

Supplemental Figures



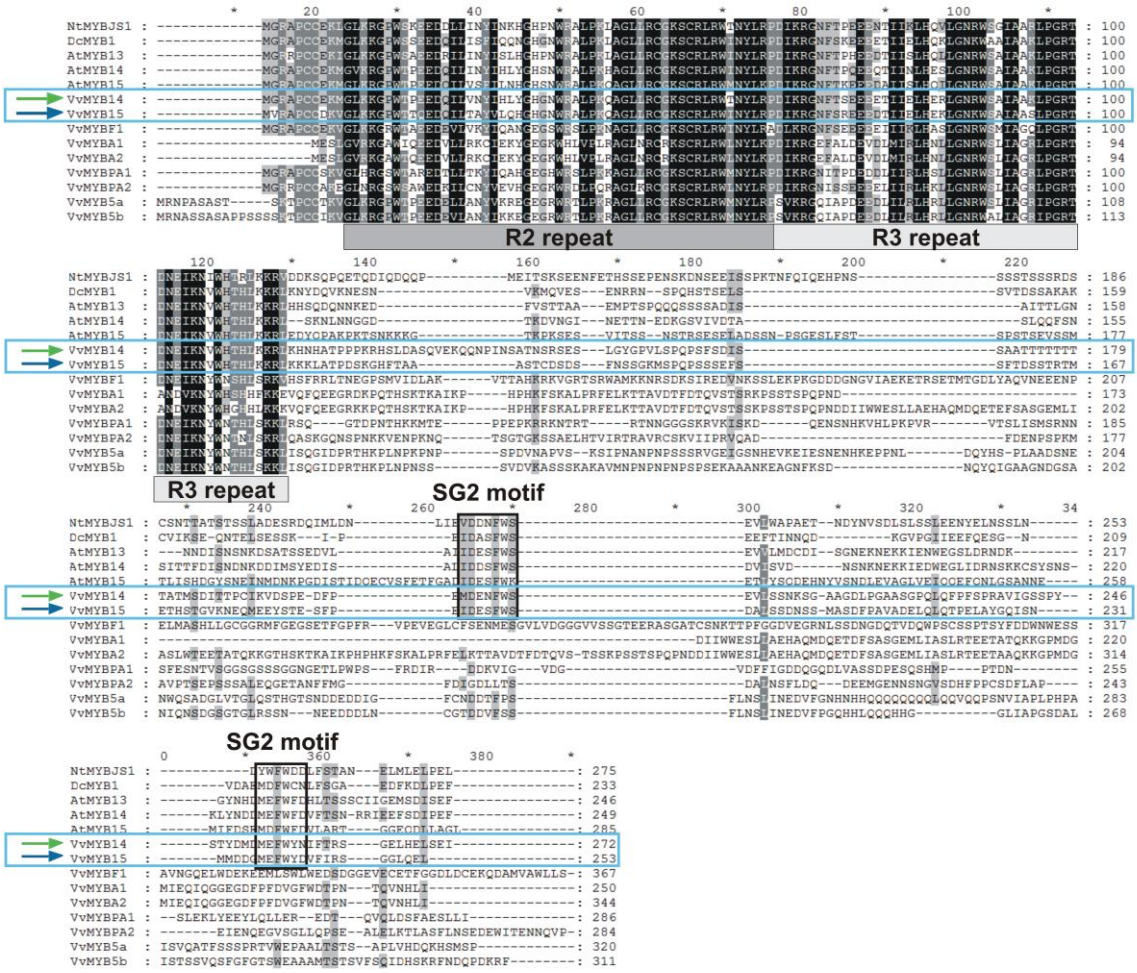
Supplemental Figure 1. Co-induction of selected *MYB* TFs and *STS* genes in grapevine leaves in response to abiotic stress.

Supplemental Figure 1. (continued).

(A) Expression image representation of transcription of the complete grapevine R2R3-MYB TF gene family together with *STS* group B genes obtained from mRNA-seq analysis of UV-C treated *V. vinifera* cv. Pinot noir leaf discs. Discs were sampled at 0 (control), 24 and 48 h after UV-C irradiation. Annotations are displayed to the right of each row. Expression data are expressed in reads per kb million mapped reads (RPKM). Unexpressed members with a value < 0.01 RPKM were excluded from the figure to simplify. Gene predictions coding for Vv-*MYB14* and Vv-*MYB15* are highlighted in bold green and blue respectively, and are the only TFs co-expressed with the majority of *Vv-STS* genes.

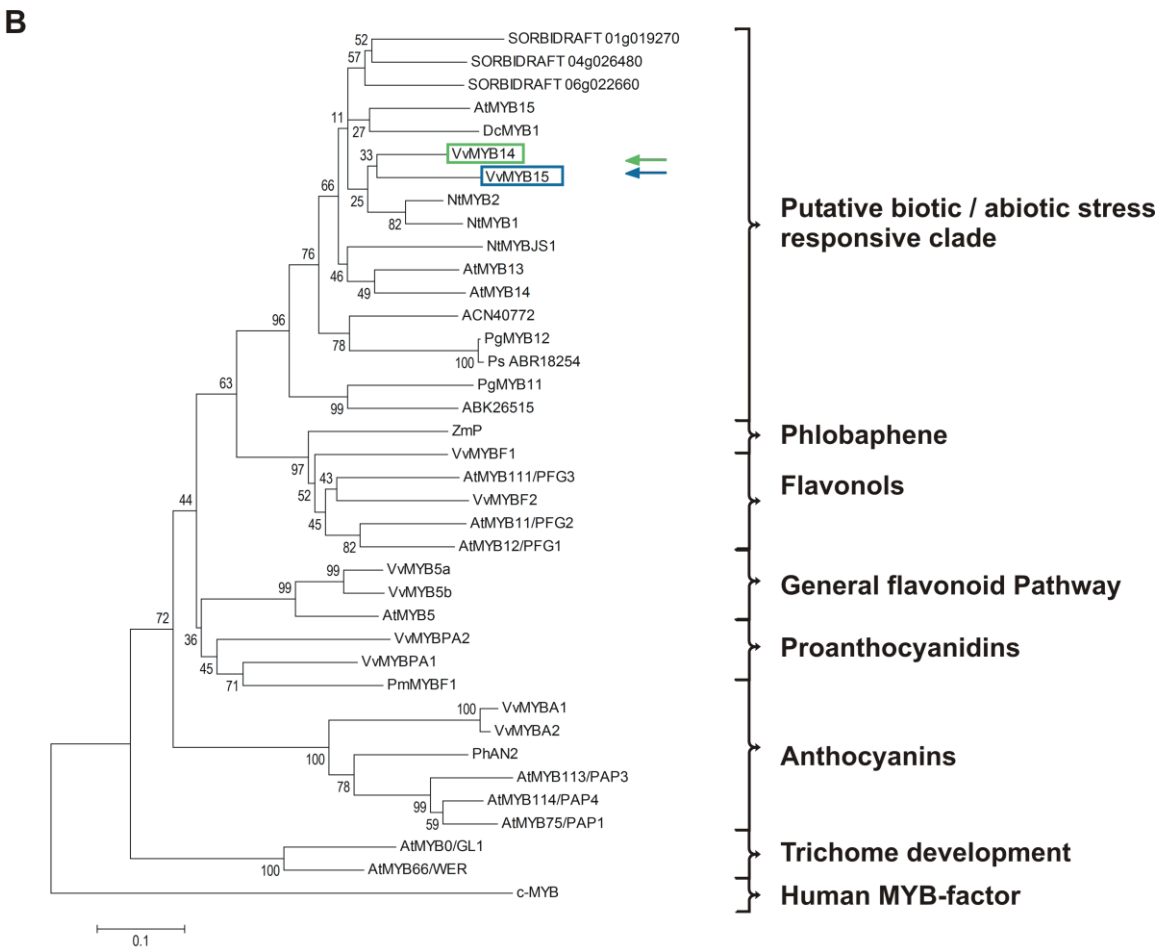
(B) Quantitative RT-PCR analysis of the transcription of Vv-*MYB* and Vv-*STS* genes in *V. vinifera* cv. Shiraz discs following wounding. Leaf discs were harvested at different time points following treatment. Transcript levels were normalized to the expression of elongation factor Vv-*EF1-α* and plotted as fold change relative to the control (0 h) sample. Bars indicate SEM of three technical replicates.

A



Supplemental Figure 2. Phylogenetic analysis of putative phenylpropanoid transcriptional regulators.

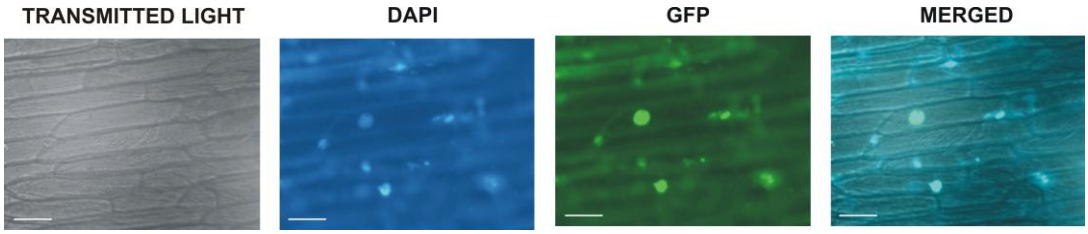
(A) Alignment of R2R3-MYB type TFs putatively regulating stilbene and flavonol synthesis in various plant species: *A. thaliana* (At-MYB13, At-MYB14 and At-MYB15), *Daucus carota* (Dc-MYB1), *N. tabacum* (Nt-MYBJS1) and *V. vinifera* (Vv-MYB14 green arrow; Vv-MYB15 blue arrow, Vv-MYBF1, Vv-MYBA1/A2, Vv-MYBPA1/PA2, Vv-MYB5a/5b). The R2R3-type domain of the MYB factors is indicated below the alignment. Black boxes show the putative C-terminal SG2 amino acid motif, previously described as the stress response motif (Stracke et al., 2001).



Supplemental Figure 2. (continued).

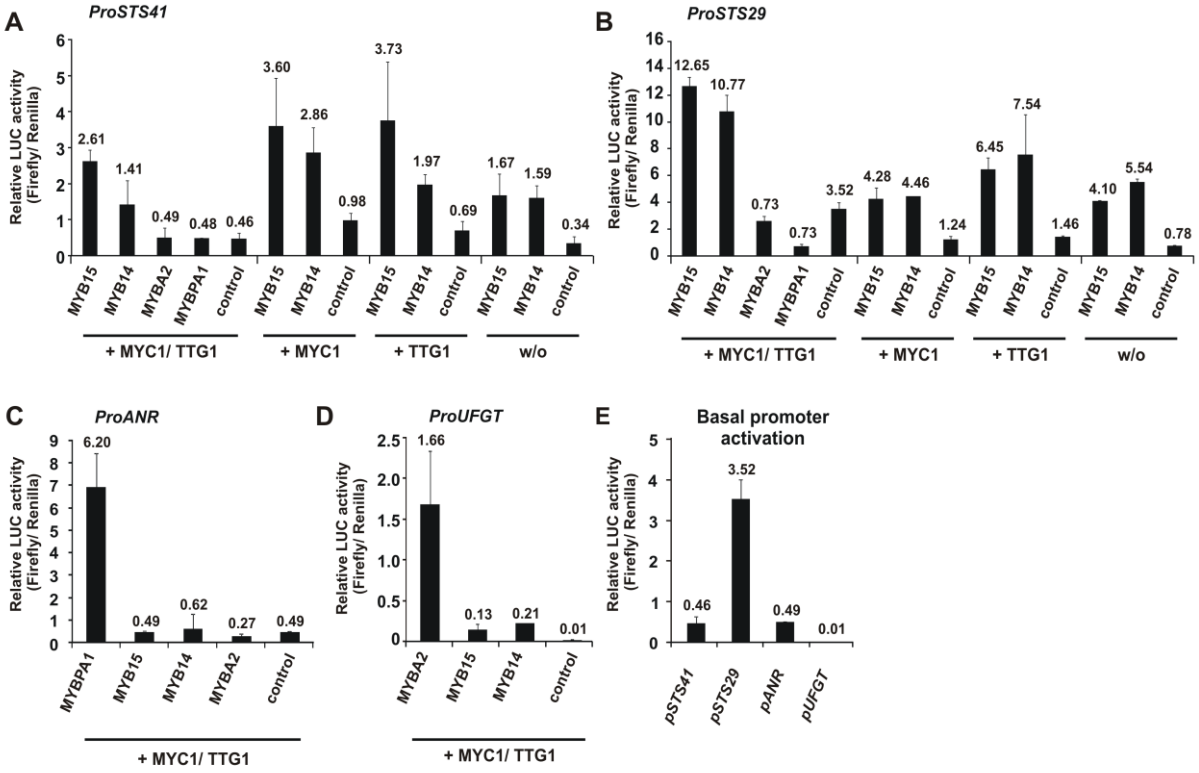
(B) The tree was constructed with MEGA 3.1 alignment software based on the neighbour-joining (NJ) method. The scale bar represents 0.1 substitutions per site and the numbers next to the nodes are bootstrap values from 100000 replicates. The GenBank accession numbers of the MYB proteins are shown in the Methods.

(A-B) Phylogenetic tree and alignment of predicted translated sequences of selected plant MYB transcription factors. Predicted functions of some of the proteins are given in bold. The phylogenetic analysis of the putative phenylpropanoid regulators is based on an out grouping mammalian c-MYB factor. The multiple sequence alignment was performed using the domain of the MYB proteins and the default parameters of the MEGA 3.1 package (Kumar et al., 2004).



Supplemental Figure 3. Targeting of a MYB14:GFP fusion protein within onion cells.

A MYB14:GFP fusion construct was bombarded into onion epidermal cells and cells were analyzed for GFP fluorescence after 48 h. The location of the nucleus is indicated by DAPI staining. The pattern shown is typical of that observed in multiple bombarded cells. Bar = 100 µm.



Supplemental Figure 4: MYB14 and MYB15 specifically induce the activity of *STS41* and *STS29* promoters.

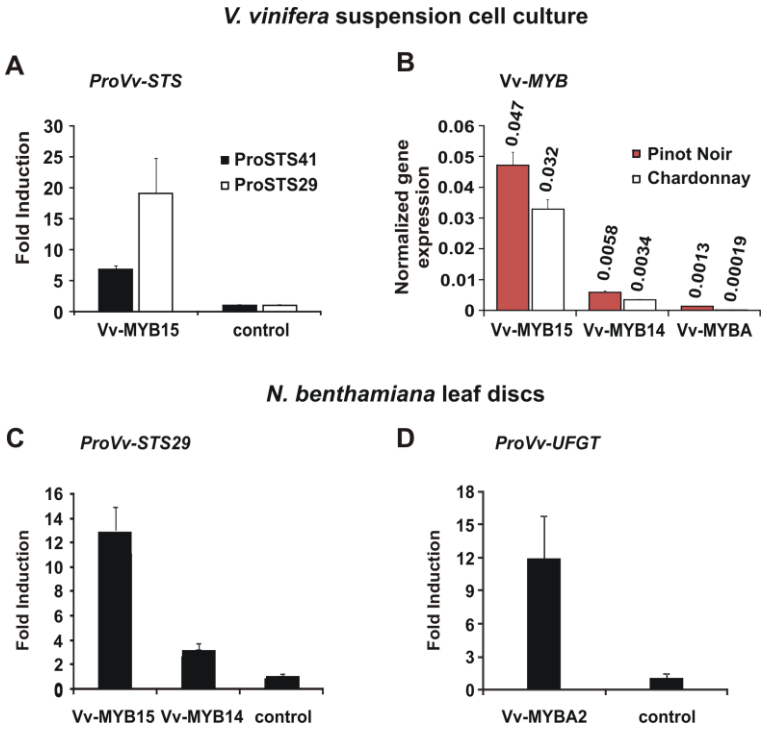
(A-B) *STS* promoter activity in the presence of different MYB TFs and different combinations of the co-factors Vv-MYC1 and At-TTG1. Control indicates promoter activity in the absence of MYB TFs.

(C-D) *ANR* and *UFGT* promoter activity in the presence of different MYB TFs. The co-factors Vv-MYC1 and At-TTG1 were present in all bombardments. Control indicates promoter activity in the absence of MYB TFs.

(E) Basal activity of *V. vinifera* promoters in suspension cell culture in the absence of added MYB TFs.

Supplemental Figure 4. (continued).

(A-E) Transient expression in *V. vinifera* cv. Chardonnay suspension cell culture following particle bombardment. Specific promoters linked to a firefly luciferase were co-bombarded into cells with pART7-VvMYB TF constructs or with a pART7 (empty vector) control. Each transfection contains as an internal control, i.e. the *Renilla* luciferase plasmid pRluc (Horstmann et al., 2004). Where indicated, the co-factors Vv-MYC1 (bHLH type) and At-TTG1 (WDR type) were also co-bombarded. The columns represent the mean promoter activity of six independent experiments with error bars indicating standard error.



Supplemental Figure 5. STS promoter induction and expression analysis of MYBs in *V. vinifera* cell culture and promoter induction analysis in *N. benthamiana* leaf discs.

(A) Fold induction of *STS29* and *STS41* promoter activity in *V. vinifera* cv. Pinot Noir cell culture in the presence of MYB15 relative to promoter activity in the absence of MYB15. Control columns show a fold induction of 1.0, indicating no effect on promoter activity.

(B) Expression of Vv-*MYBs* in grapevine *V. vinifera* cv. Pinot Noir cell culture.

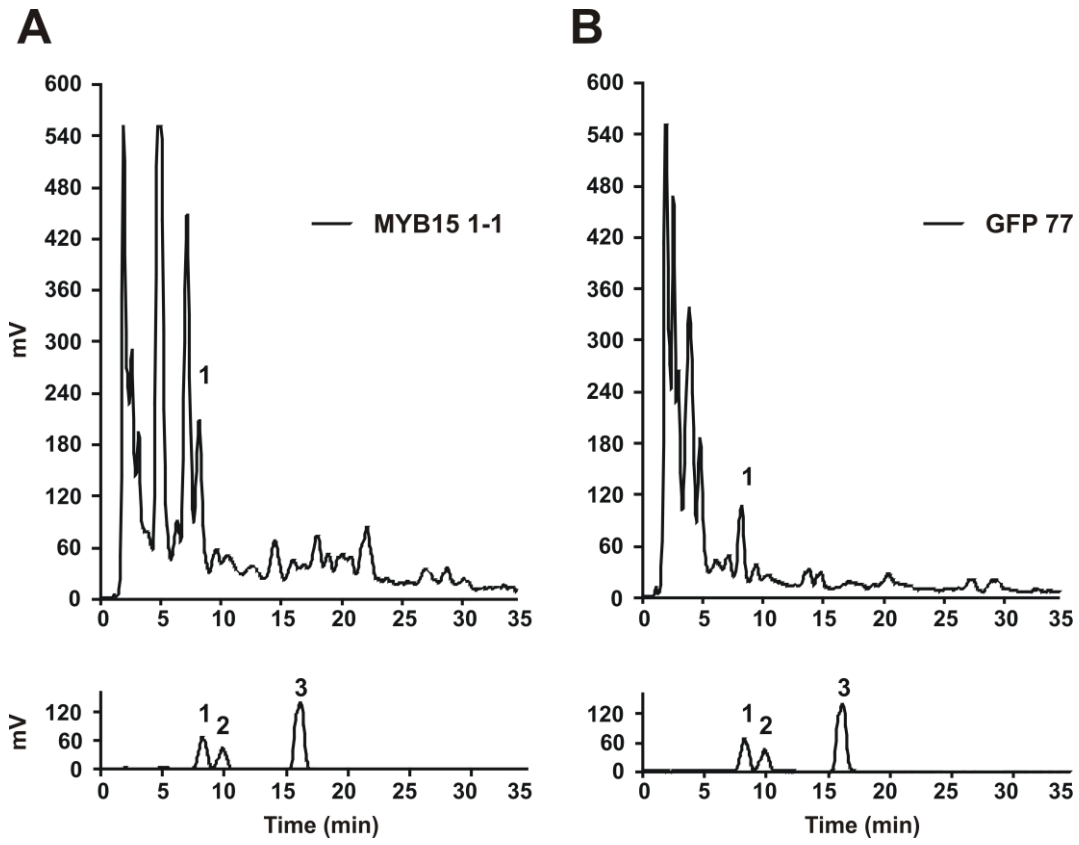
(C) Induction of the *Vv-STS29* promoter by Vv-MYB14 and Vv-MYB15 in *N. benthamiana* leaf discs.

(D) Induction of the *Vv-UFGT* promoter by Vv-MYBA2 in *N. benthamiana* leaf discs.

Supplemental Figure 5. (continued).

(A, C, D) Specific promoters linked to a firefly luciferase gene were co-bombarded into cells with pART7-VvMYB TFs constructs or with a pART7 (empty vector) control. Each transfection contains as an internal control i.e. the *Renilla* luciferase plasmid pRluc (Horstmann et al., 2004), and the co-factors Vv-MYC1 (bHLH type) and At-TTG1 (WDR type). The columns represent relative LUC activity (Firefly/Renilla) of the corresponding promoter plus MYB factor relative to the respective control (without MYB factor) of six independent experiments with error bars indicating standard error.

(B) Transcript levels of Vv-*MYB15,* Vv-*MYB14*, Vv-*MYBA* and Vv-*MYBPA1* in *V. vinifera* cv. Pinot Noir (red column) and cv. Chardonnay (white column) suspension cell cultures as determined by qPCR. Transcript levels were corrected to *Vv-Ubiquitin1* expression and represent mean values of three replicate PCRs (n=6) and error bars indicating standard error.



Supplemental Figure 6. **Chromatogram of putative stilbenoids extracted from *MYB15* overexpressing hairy root line 1-1 compared to GFP:GUS-control line 77.**

(A-B) HPLC Chromatogram shows stilbenes extracted from *MYB15* overexpressing hairy root line 1-1 (**A**) compared to the GFP:GUS-control line 77 (**B**). Note that data shown are typical of that obtained from extraction of the other Vv-MYB15- and GFP-transformed roots. Bottom panel: Chromatogram showing stilbene standards used for HPLC analysis. 1: *Trans*-piceid, 2: *2,3,4',5*-Tetrahydroxystilbene-2-glucoside, 3: *Trans*-resveratrol. Data shown are from one representative experiment, which was repeated three times with similar results. Note, that each line is a pool of several hairy roots. FW, Fresh weight; mV, milli volt.

Supplemental Table

Supplemental Table 1. Identification of stilbenoids in *V. vinifera* tissues via LC-QTOF-MS

t (min) [a]	Δ m/z[b]	Putative identified compound[c]	HRs[d]	Seed[e]	Skin[f]
2,15	243,102	4-Methoxyresveratrol	**y**[g]	**y**	
7,01	421,128	Rhapontigenin glucoside	**y**		
9.14	389,124	*trans*-piceid	**y**	**y**	**y**
10,41	333,132	Combrestatin A-1	**y**		
10,83	245,081	2,3',4,5'-Tetrahydroxystilbene (Oxyresveratrol)	**y**	**y**	**y**
11,34	257,117	Pterostilbene		**y**	
13,06	455,149	Viniferin, Ampelopsin B,D,F, Leachinol F, Pallidol, Parthenocissin A, Quadrangularin A	**y**		
13,22	453,153	Amurensin H	**y**		
13,22	471,144	Ampelopsin A	**y**		
18,36	681,213	Resviniferin A,B, Amurensin B,G, Miyabenol C, Malaysianol A, trans-Diptoindonesin B	**y**		
18,6	907,275	Vitisin A,B,C, Vaticanol B, Hopeaphenol	**y**		
19,09	907,273	Vitisin A,B,C, Vaticanol B, Hopeaphenol	**y**		

Supplemental Table 1. (continued).

a Retention time in min

b Mass to charge ratio

c Putative identified compounds, for some identified ∆m/z values more than one compound has a matching mass

d Analyzed Vv-MYB15 hairy root lines 2-2, 2-3 and controls GFP77, GFP78, GFP79,GFP80

e Analyzed seed samples (0, +4 weeks after véraison)

f Analyzed skin samples (0, +4 weeks after véraison)

g Sample contain the identified compound. y: yes.

Supplemental Methods

Accession numbers used for phylogenetic analysis

Accession numbers of selected plant MYB transcription factors obtained from the Arabidopsis Genome Initiative (AGI, http://www.arabidopsis.org), GenBank or EMBL database (The EMBL Nucleotide Sequence Database) used for phylogenetic analysis (Supplemental Figure 2A/B) are: AAB41101 (Nt-MYB1), BAA88221 (Nt-MYB2), BAE54312 (Dc-MYB1), BAE93149 (Nt-MYBJS1), AT2G31180 (At-MYB14; AAD20663), ABW34392 (Vv-MYB14), AT1G06180 (At-MYB13; AAF80215), KC514110 (Vv-MYB15), XP_002448188 (hypothetical protein SORBIDRAFT_06g022660 [*Sorghum bicolor*]), AT3G23250 (At-MYB15; NP_188966), XP_002454197 (hypothetical protein SORBIDRAFT_04g026480 [*Sorghum bicolor*]), XP_002464484 (hypothetical protein SORBIDRAFT_01g019270 [*Sorghum bicolo*r]), ACN40772 (unknown [*Picea sitchensis*]), ABQ51228 (Pg-MYB12), ABR18254 (unknown [*Picea sitchensis*]), ABK26515 (unknown [*Picea sitchensis*]), ABQ51227 (PgMYB11), ACK56131 (Vv-MYBPA2), Q9FJA2 (At-MYB123/TT2), AM259485 (Vv-MYBPA1), AAA82943 (Pm-MYBF1), P27898 (Zm-P), AAS68190 (Vv-MYB5a), Q58QD0 (Vv-MYB5b), AT3G13540 (At-MYB5; U26935), AAF66727 (Ph-AN2), BAD18977 (Vv-MYBA1), BAD18978 (Vv-MYBA2), AT1G66370 (At-MYB113/PAP3; NP_176811), AT1G56650 (At-MYB75/PAP1; AAG42001), AT1G66380 (At-MY114/PAP4; NP_176812), AT3G27920 (At-MYB0/GL1; P27900), AT5G14750 (At-MYB66/WER; CAC01874), FJ948477(Vv-MYBF1), DQ074470 (Vv-MYBF2), AT5G49330 (At-MYB111/PFG3; AAK97396), AT3G62610 (At-MYB11/PFG2; NP_191820), AT2G47460 (At-MYB12/PFG1; CAB09172), AT5G35550 (At-TT2; Q9FJA2), NP_001123644 (c-MYB); EC959059 (Vv-EF1-α), EU447172 (Vv-MYC1), TC32075 (Vv-Ubiquitin1).

Supplemental References

Horstmann, V., Huether, C., Jost, W., Reski, R., and Decker, E. (2004). Quantitative promoter analysis in *Physcomitrella patens*: a set of plant vectors activating gene expression within three orders of magnitude. BMC Biotechnology **4,** 13.

Kumar, S., Tamura, K., and Nei, M. (2004). MEGA3: Integrated software for Molecular Evolutionary Genetics Analysis and sequence alignment. Briefings in Bioinformatics **5,** 150-163.

Stracke, R., Werber, M., and Weisshaar, B. (2001). The R2R3-MYB gene family in *Arabidopsis thaliana*. Current Opinion in Plant Biology **4,** 447-456.

The EMBL Nucleotide Sequence Database. http://www.ensembl.org/index.html)