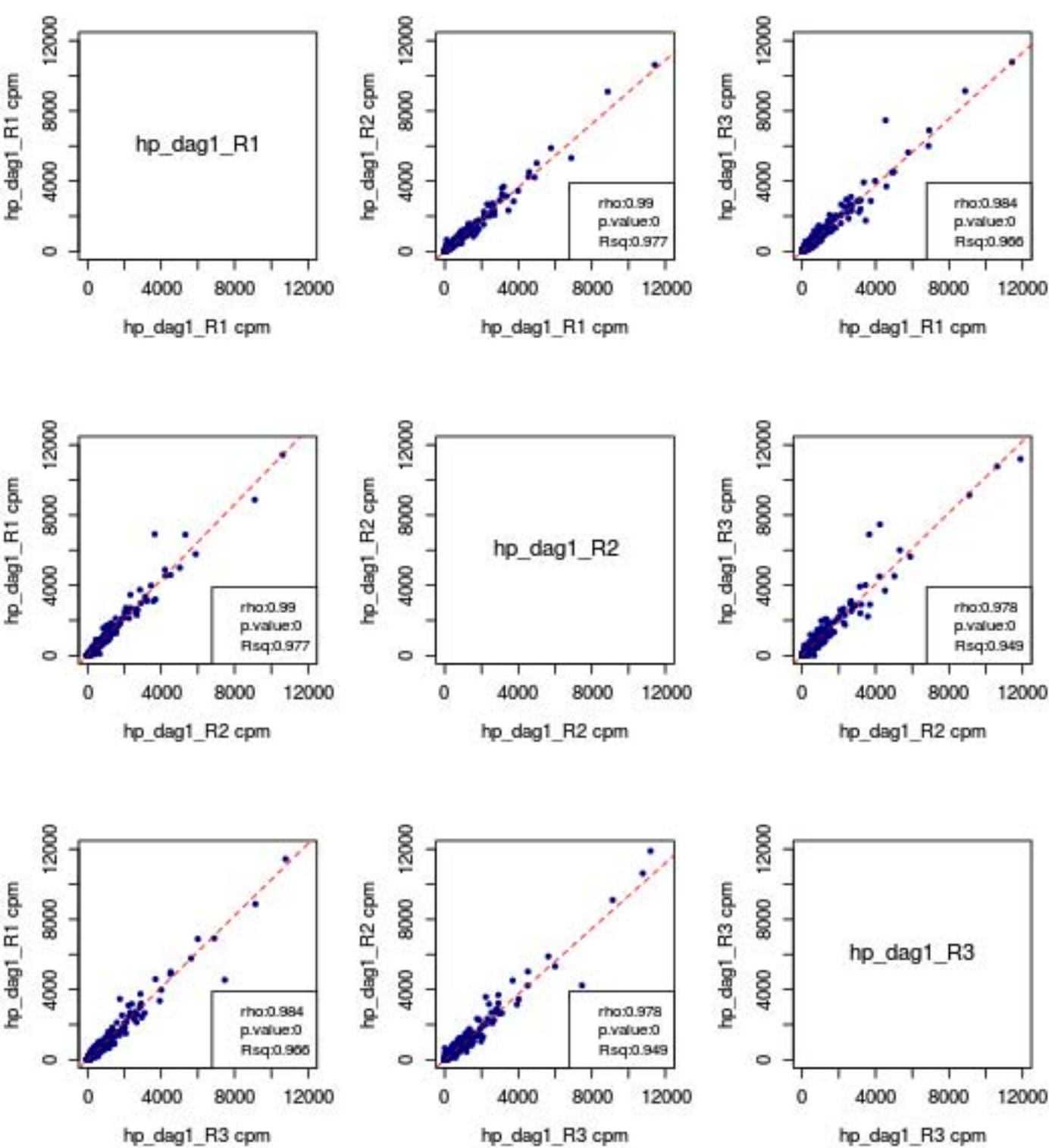
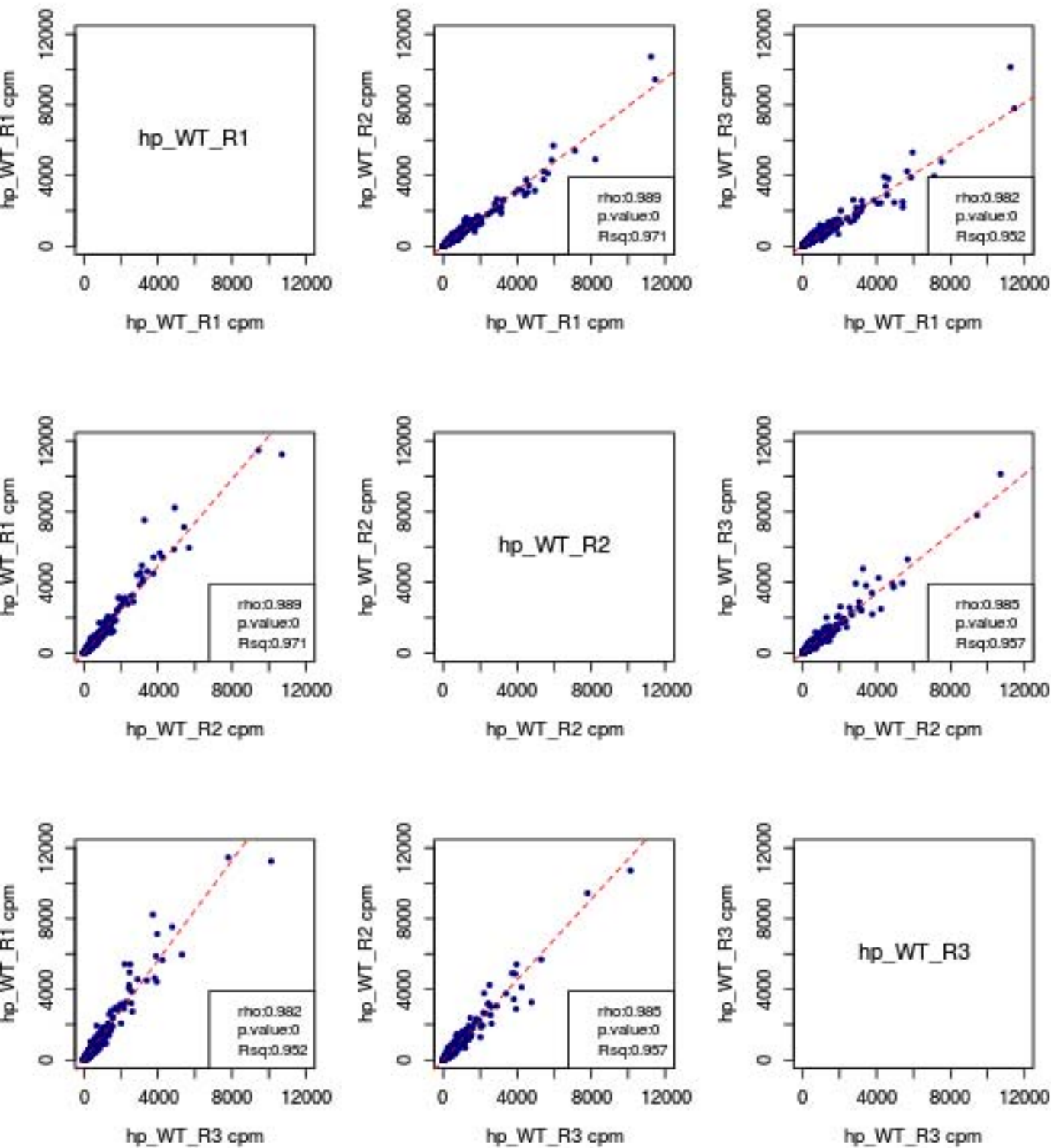


Genome-wide RNA-seq analysis indicates that the DAG1 transcription factor promotes hypocotyl elongation acting on ABA, ethylene and auxin signalling

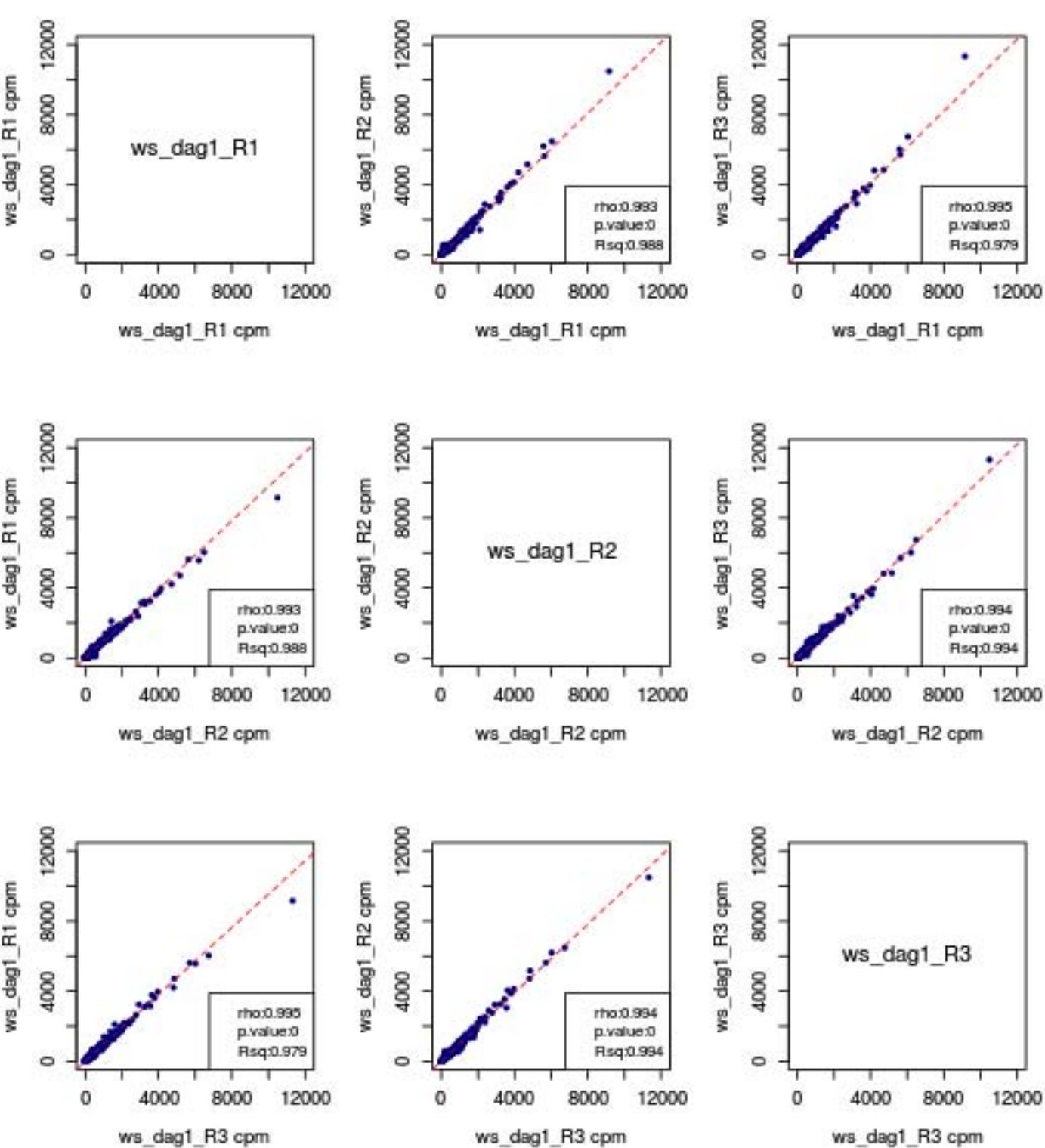
Lorrai Riccardo¹, Gandolfi Francesco², Boccaccini Alessandra¹, Ruta Veronica¹, Possenti Marco³, Tramontano Anna², Costantino Paolo¹, Lepore Rosalba^{2*}, Vittorioso Paola^{1*}



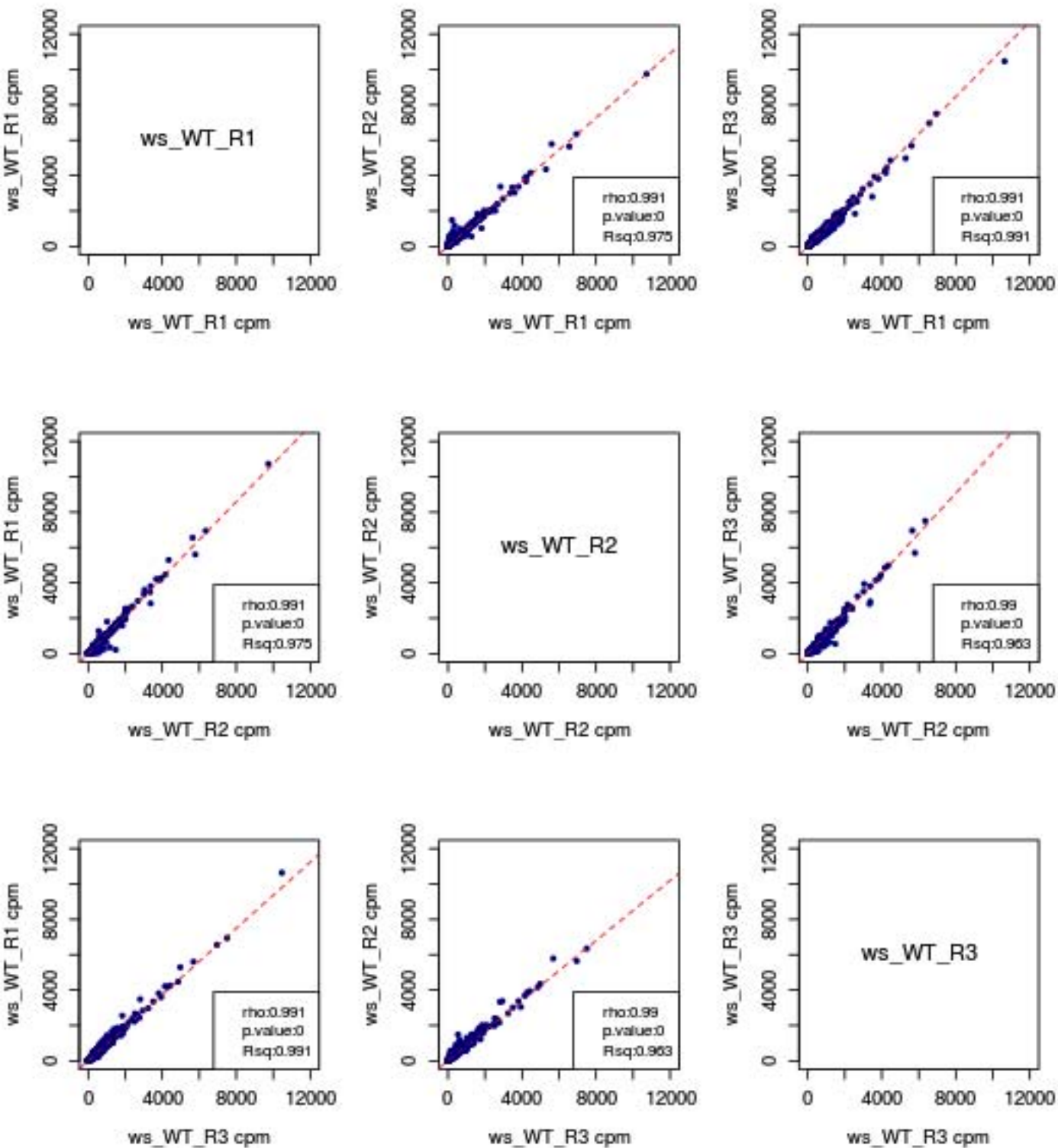
Supplementary Fig S1. Scatter plots of CPM values between replicates and Spearman's correlation coefficients.



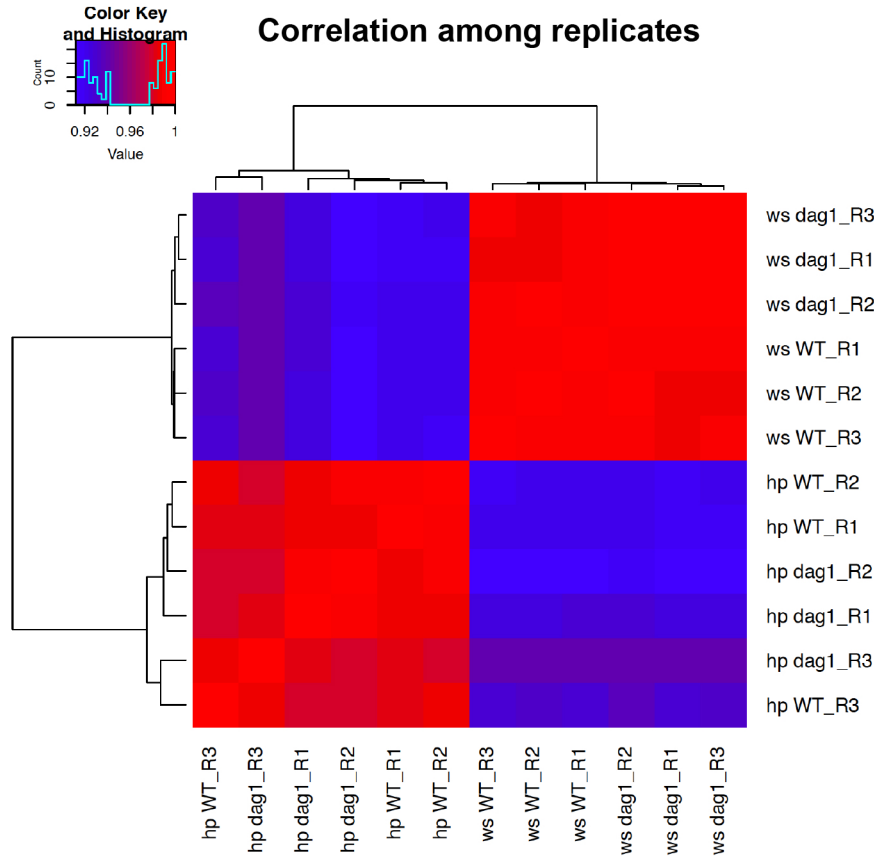
Supplementary Fig S2. Scatter plots of CPM values between replicates and Spearman's correlation coefficients.



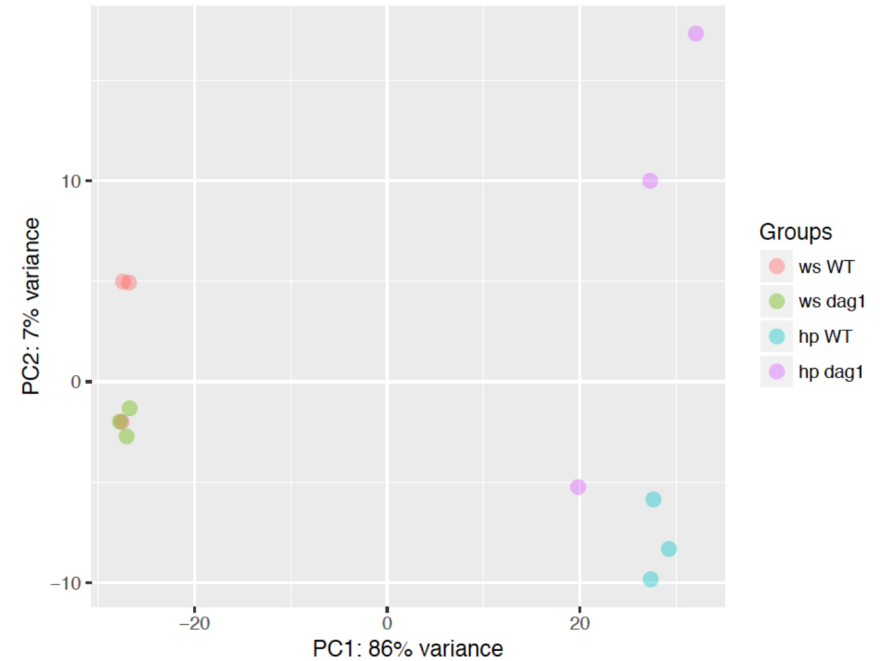
Supplementary Fig S3. Scatter plots of CPM values between replicates and Spearman's correlation coefficients.



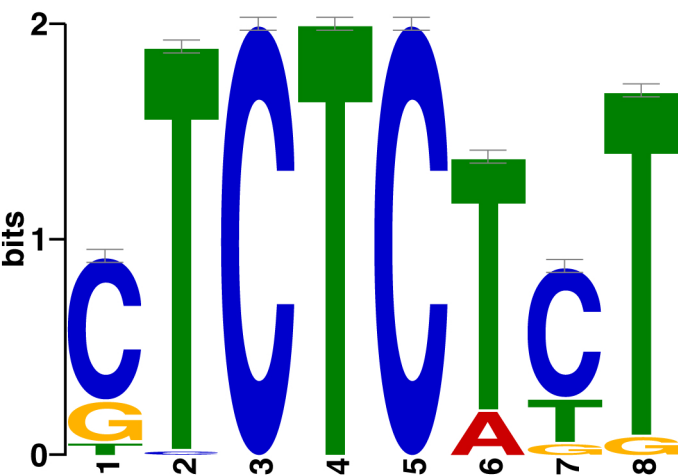
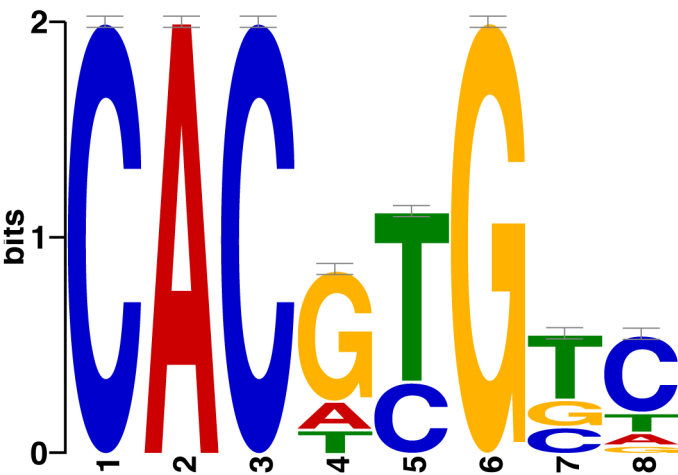
Supplementary Fig S4. Scatter plots of CPM values between replicates and Spearman's correlation coefficients.

a**b**

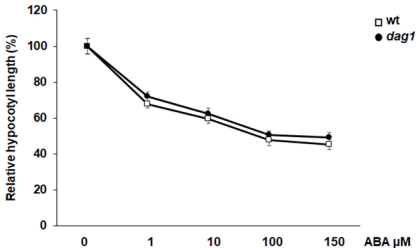
Principal Component Analysis of sample replicates



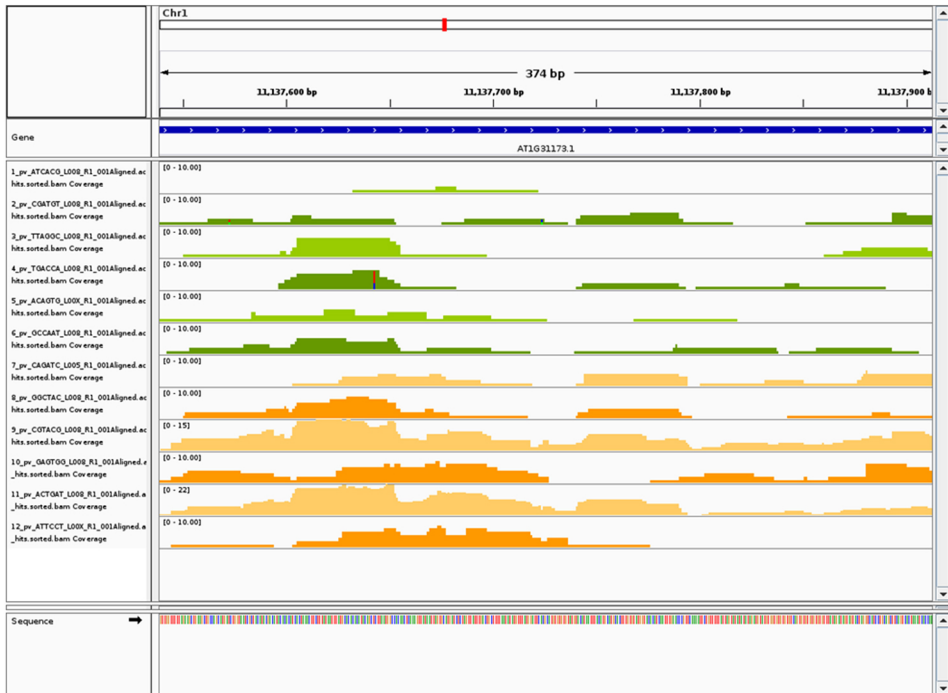
Supplementary Fig S5. Sample clustering and Principal Component Analysis of RNA-seq data. (a) Correlation clustering of RNA-seq replicates according to normalized gene expression values. Spearman correlation was used as a distance metric (defined as $1 - \text{Spearman's correlation coefficient}$) and the “average” method as linkage function. **(b)** Principal Component Analysis (PCA) was performed using the ‘plotPCA’ function provided within the ‘DESeq2’ (v1.18) Bioconductor R package.



Supplementary Fig S6. The PIF binding motif is enriched in DE genes. Hidden Markov model sequence logos obtained using MEME¹/TOMTOM² for significantly enriched motifs found in DE genes promoter sequences.



Supplementary Fig S7. Relative hypocotyl length of *dag1* (black) and wild type (white bar) seedlings grown under red light for five days, with increasing concentration of ABA (0, 1, 10, 100 and 150 μM). The values, normalised to mock treated samples for both genotypes, are the mean of three biological replicates, presented with SD values.



Supplementary Fig S8. Relative sequence read abundance of *miR167D* (Integrative Genome Browser view) as histograms, in *dag1* and wild type hypocotyls and seedlings. The Y-axis indicates read depth with the same scale for all samples (three biological replicates). The green profiles are referred to the whole seedlings, whereas the orange profiles are referred to the hypocotyls (*dag1* in dark green/orange, wild type in light green/orange). The *miR167* locus is shown by the blue track in the top panel.

Supplementary Table S1. Reads mapped to the *Arabidopsis thaliana* genome (TAIR10)

Sample	Tot Reads	% Uniquely mapped	% Multi Mapped	% Unmapped
WT 1 (ws)	18967605	92	7,25	0,75
<i>dagl</i> 1 (ws)	27654782	91	7,9	1,08
WT 1.2 (ws)	25388619	91,25	7,88	0,87
<i>dagl</i> 1.2 (ws)	29184780	91,06	7,74	1,2
WT 1.3 (ws)	21427524	91,98	6,96	1,05
<i>dagl</i> 1.3 (ws)	24563037	91,92	6,84	1,24
WT 1 (hp)	19885967	90,04	5,82	4,05
<i>dagl</i> 1 (hp)	19617630	92,35	6,24	1,4
WT 1.2 (hp)	31727863	93,48	5,42	1,1
<i>dagl</i> 1.2 (hp)	31503405	93,22	5,43	1,36
WT 1.3 (hp)	24355880	92,7	6,52	0,78
<i>dagl</i> 1.3 (hp)	20956766	92,72	5,8	1,47

Supplementary Table S3. List of the seed-specific genes differentially expressed in *dag1* hypocotyls respect to wild-type.

Name	Locus	Function
<i>EM6</i>	AT2g40170	LEA
<i>LEA18</i>	AT2g35300	LEA
<i>M10</i>	AT2g41280	LEA
<i>M17</i>	AT2g41260	LEA
	AT2g23110	LEA
	AT3g17520	LEA
	AT3g53040	LEA
<i>CRA1</i>	AT5g44120	seed storage proteins
<i>PAP85</i>	AT3g22640	seed storage proteins
<i>SESA5</i>	AT5g54740	seed storage proteins

Supplemental Table 3.

Specific primer sets used in the qRT-PCR:

	Forward	Reverse
<i>UBQ10</i>	GGCCTTGTATAATCCTGATGAATAAG	GGCCTTGTATAATCCTGATGAATAAG
<i>RAB18</i>	CGATGGGCGGCATGGGAT	CACCACCGGGAAGCTTTTCCTT
<i>ABR1</i>	TCCGTTGCGTTTCGGTCACA	CTGATTCTGGTGGCGGCTGAA
<i>AT2G23110</i>	ACCGATGCTCCCCTCCTTCA	CGGAACGCCCTGACGGTT
<i>LEA18</i>	CTAAGGAGGCGCAGGCCAAA	CTCGGGTAGGGTAGGTGGAGTG
<i>EM6</i>	GGAGCCGAGGAGGGCAAAC	CTCAGCGTGTCCCCACCAG
<i>M17</i>	TGGTGGTTGTAGGTGGGGATGTT	CAACAGTTTCCACAACCTCGTTTGC
<i>M10</i>	TGGTGGTGGTTGTAGGTTCCGGA	CTTCGTTGGCCTCGGCTTGA
<i>AT3G17520</i>	CGAAAGATAAGGCGTCGCAGAGTT	GCTCAGCATCATCGTCGGTCAA
<i>AT3G53040</i>	GCCCATGGAGACGAGAGCAC	CCGCCTCTTTCAGTACCGTCG
<i>CRA1</i>	GGTGGTGAAACGCGCAACAA	GACTGAGGTTCGTCCCGC
<i>WRKY6</i>	TCTCGGGGCTGCAGTTCTCT	GGTCAGCTGTGAGTGCCGTT
<i>WRKY18</i>	GGGATGCTACAAGCCAGGGTG	TCCCTTTCGTTTTTCTCCAACGCT
<i>WRKY28</i>	GCAGACCTCATGACTCCAAGAAGC	ATAATCCAAAGCCGCCGCCA
<i>WRKY33</i>	ACCATCGGTTGTCCAGTGAGGA	AACCGCTACCACGAGCTGC
<i>WRKY40</i>	GCTTGACTGTGCCGGTGACT	GGAAGAAGCCATTTGCTCCACCA
<i>WRKY46</i>	CTGCACCTGCTGCTGTTGAGAA	CGACCACAACCAATCCTGTCCG
<i>WRKY70</i>	CAGGCCAGTTACGTCAATGGGAAAA	GAAATCGCCGCCACCTCCA
<i>SAUR50</i>	GAGATGGTTAAAGAAGAAGCGG	CCTAGATGGGCTTAAAAATAAAGG
<i>SAUR63</i>	TGTAGTACTACGCAACAGTTGC	GAAATGATCTGTCTATCTAATCCAC
<i>SAUR65</i>	CCACAAGAACAACAGAGTGGTT	ACAGAGGAAAAAATGTCTACTC
<i>SAUR67</i>	CAGAAAAGGCTCTGTTGATGTC	GTTGAGTACTCTGTTCTTGCTG
<i>ERF2</i>	AATTCGGTGAACCTGACCC	ACGACGTAGAAGAGGACGAC
<i>ERF5</i>	CCGATGAAGGTGAGAAGAAAC	GTCAACGCTCTGTTCCGTCT
<i>ERF11</i>	TAAGGCTGGGATGATGGTGT	AAAACCACACGTCGTCCTTC
<i>ERF105</i>	CCGGTGGTTCAAGAGAATGAT	CCTAACCCAGACACGAACACC
<i>ERF109</i>	CAGTTGAAGCAGAGCAATGG	CCATTCCTCCAAAATCCATCATC

Specific primer sets used for chIP:

	Forward	Reverse
<i>WRKY18 c</i>	CAGCTTCTATGTCTTGTGTGC	AAAGCTGTCTTCTATAAAGTCCAG
<i>WRKY18 b</i>	CTGGACTTTATAGAAGACAGC	AGATAGGTTTGGGATATGTTAGG
<i>WRKY18 a</i>	CGACAGTAAATTTAACCCATGCAT	ACACTGATTGGCTAAAAATATGGG
<i>ERF2 c</i>	CCCCTAACTAAAACAAATTTGG	ATGTGGAAGGTGGTTAAGAGG
<i>ERF2 b</i>	GACATTGGCAATTAGGAAAATTCT	ATAATTATATGAATGGGACACTG
<i>ERF2 a</i>	CCAAATCATTGGTGTACAAAAAG	GGAGTAGTGAAATCATAGTGATA
<i>SAUR67 c</i>	CTCTTCTTTCTCCACACTTTGC	CTTCATAAGCTTCTTTGCGTTTATC
<i>SAUR67 b</i>	CAACCACAACCTTTCATTCAAAGAG	GGAGAGTTAAAGCTTGAGATTGG
<i>SAUR67 a</i>	TTTACCTGATGGACTACTAACCAA	AAGCCAAAAATACCAACTCATGC

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- 2 Gupta, S., Stamatoyannopoulos, J. A., Bailey, T. L. & Noble, W. S. Quantifying similarity between motifs. *Genome Biol* **8**, R24, doi:10.1186/gb-2007-8-2-r24 (2007).