

RNA-seq studies using wheat *PHYTOCHROME B* and *PHYTOCHROME C* mutants reveal shared and specific functions in the regulation of flowering and shade-avoidance pathways

Additional file 1

Tables and figures in this additional file:

Table S1: Summary of RNA-seq reads and mapping.

Table S2: TopGO analysis for functional enrichment for genes regulated by *PHYB*, *PHYC* and in concert by both *PHYB* and *PHYC*.

Figure S1: Spike and floral organ phenotype of the *phyB*-null mutant. (a) Whole spike, (b) Single spikelet (c) Separated spikelet and (d) Stamen and stigma. (e) Comparison of internode length between 83-day-old *phyB*-null, *phyC*-null and wild-type Kronos plants. Leaves have been removed to facilitate visualization of internodes. Nodes are indicated by purple arrows. Bar = 10 cm.

Figure S2: Vegetative phenotype of wild-type control, *phyB*-null and *phyC*-null plants. (a) Leaf emergence rate, (b) Leaf length and (c) leaf width at three different timepoints (* $P < 0.05$; ** $P < 0.01$).

Figure S3: Phenotype of four-week old *PHYB* wild-type, *phyB*-null, *PHYC* wild-type and *phyC*-null plants at the stage of harvest for RNA-seq analysis.

Figure S4: PCA plots of normalized expression values of all genes. (a) All libraries, (b) *PHYB* libraries and (c) *PHYC* libraries.

Figure S5: Relative transcript levels of six flowering time genes determined by qRT-PCR in wild-type and *phyB*-null mutants at three stages of development (Leaves from two-week, four-week and six-week-old plants). Expression levels are presented as fold-*ACTIN*.

* $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$.

Table S1: Summary of RNA-seq reads and mapping rates.

Genotype	Experimental replicate	Biological replicate	Raw reads	Trimmed reads	% mapped to gene loci	% uniquely mapped to gene loci
<i>PHYB</i> wild-type	1	1	42,783,726	38,193,836	95.6	58.0
		2	42,254,752	37,228,077	95.4	58.1
		3	58,349,947	50,793,083	95.6	58.1
		4	50,148,260	44,301,543	95.9	56.5
	2	1	57,790,105	53,475,278	95.5	58.4
		2	58,798,668	54,275,607	95.0	58.6
		3	61,694,504	57,095,077	95.6	58.7
		4	64,329,742	59,581,947	95.0	58.8
<i>phyB</i> -null	1	1	48,416,815	43,374,525	94.8	58.5
		2	56,768,866	47,201,620	94.7	58.0
		3	88,913,796	81,109,422	94.9	57.6
		4	53,140,168	47,250,183	93.9	58.5
	2	1	51,385,316	47,550,092	95.0	59.1
		2	54,947,620	50,880,434	94.7	59.2
		3	60,875,008	56,371,576	94.6	59.0
		4	55,147,736	51,113,770	94.7	59.1
<i>PHYC</i> wild-type	1	1	41,280,436	40,263,724	95.7	59.6
		2	41,700,104	40,409,065	95.8	62.3
		3	43,295,783	42,605,342	95.6	59.8
		4	40,970,343	39,796,668	95.7	60.5
	2	1	38,880,169	37,994,979	95.4	60.3
		2	44,203,882	43,262,009	95.5	59.6
		3	42,528,948	41,619,486	95.4	59.7
		4	32,593,169	31,887,538	95.4	59.8
<i>phyC</i> -null	1	1	45,693,558	44,904,496	95.1	58.6
		2	47,618,087	46,776,974	95.2	59.3
		3	44,675,515	43,960,996	95.4	59.5
		4	43,530,452	42,873,364	95.4	59.2
	2	1	36,142,687	35,291,647	95.2	59.7
		2	37,545,579	36,712,107	95.4	58.9
		3	46,594,218	45,656,274	95.1	59.7
		4	39,781,242	38,922,327	96.7	41.0
Average			49,149,350	46,022,908	95.3	58.5

Table S2: TopGO analysis for functional enrichment for genes regulated by *PHYB*, *PHYC* and in concert by both *PHYB* and *PHYC*.

	Accession	Ontology	Annotated	Significant	Expected	<i>P</i>
<i>PHYB</i>-regulated	GO:0009755	Hormone-mediated signaling pathway	1913	85	45.04	2.20E-08
	GO:0032870	Cellular response to hormone stimulus	1928	85	45.39	3.10E-08
	GO:0071495	Cellular response to endogenous stimulus	1930	85	45.44	3.30E-08
	GO:0009725	Response to hormone stimulus	2598	103	61.17	2.10E-07
	GO:0009719	Response to endogenous stimulus	2672	104	62.91	4.20E-07
	GO:0043269	Regulation of ion transport	185	18	4.36	4.60E-07
	GO:0009642	Response to light intensity	337	24	7.93	2.00E-06
	GO:0051049	Regulation of transport	208	18	4.9	2.50E-06
	GO:0006468	Protein phosphorylation	4258	146	100.25	2.90E-06
	GO:0071310	Cellular response to organic substance	2428	93	57.16	3.40E-06
<i>PHYC</i>-regulated	GO:0048586	Regulation of long-day photoperiodism, flowering	12	2	0.03	0.00036
	GO:2000028	Regulation of photoperiodism, flowering	12	2	0.03	0.00036
	GO:0048574	Long-day photoperiodism, flowering	30	2	0.07	0.00233
	GO:0048571	Long-day photoperiodism	38	2	0.09	0.00372
	GO:0015693	Magnesium ion transport	44	2	0.1	0.00496
	GO:0006355	Regulation of transcription, DNA-dependent	3561	16	8.46	0.01022
	GO:2001141	Regulation of RNA biosynthetic process	3561	16	8.46	0.01022
	GO:0051252	Regulation of RNA metabolic process	3587	16	8.53	0.0109
	GO:0048552	Regulation of metalloenzyme activity	6	1	0.01	0.01418
	GO:0048554	Positive regulation of metalloenzyme activity	6	1	0.01	0.01418
<i>PHYB-PHYC</i> regulated	GO:0006355	Regulation of transcription, DNA-dependent	3561	10	2.19	3.30E-05
	GO:2001141	Regulation of RNA biosynthetic process	3561	10	2.19	3.30E-05
	GO:0051252	Regulation of RNA metabolic process	3587	10	2.2	3.50E-05
	GO:0010556	Regulation of macromolecule biosynthetic process	3910	10	2.4	7.20E-05
	GO:2000112	Regulation of cellular macromolecule biosynthetic process	3910	10	2.4	7.20E-05
	GO:0019219	Regulation of nucleobase-containing compound metabolic process	3926	10	2.41	7.50E-05
	GO:0031326	Regulation of cellular biosynthetic process	3950	10	2.43	7.90E-05
	GO:0051171	Regulation of nitrogen compound metabolic process	3955	10	2.43	8.00E-05
	GO:0009889	Regulation of biosynthetic process	4011	10	2.47	9.00E-05
	GO:0006351	Transcription, DNA-dependent	4194	10	2.58	0.00013

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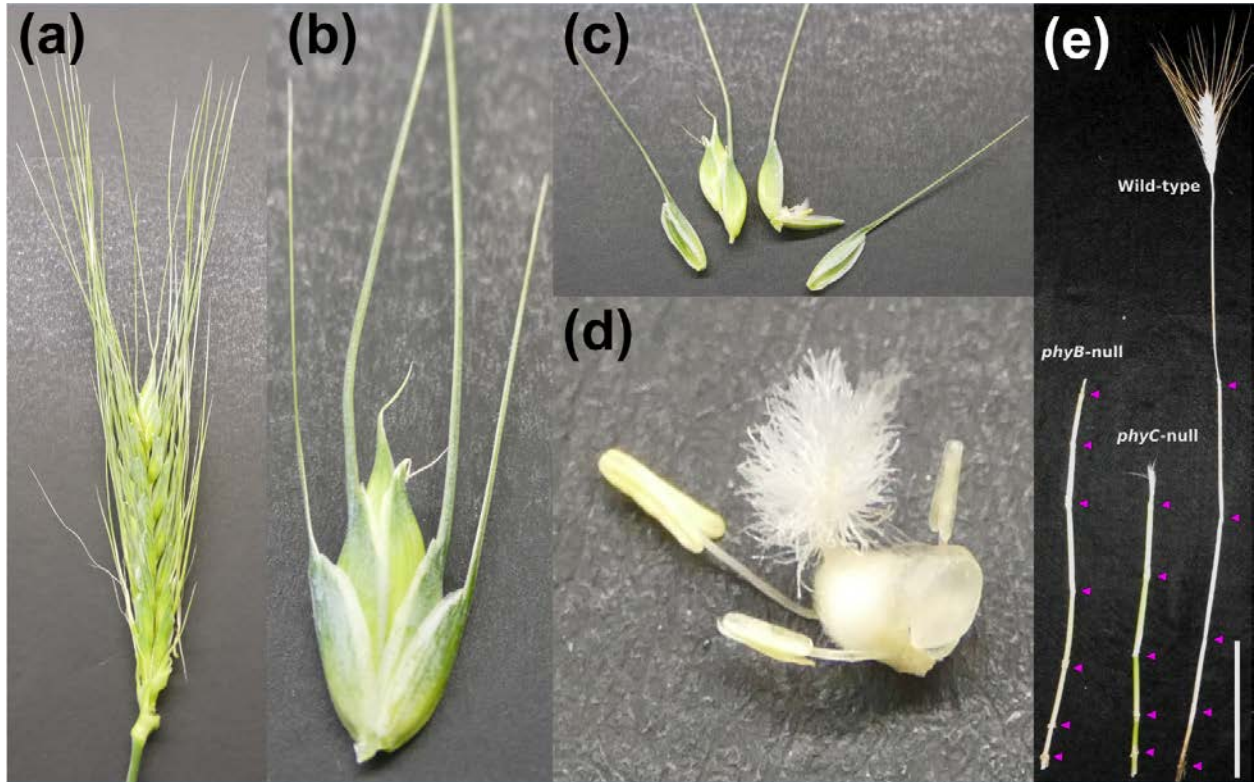


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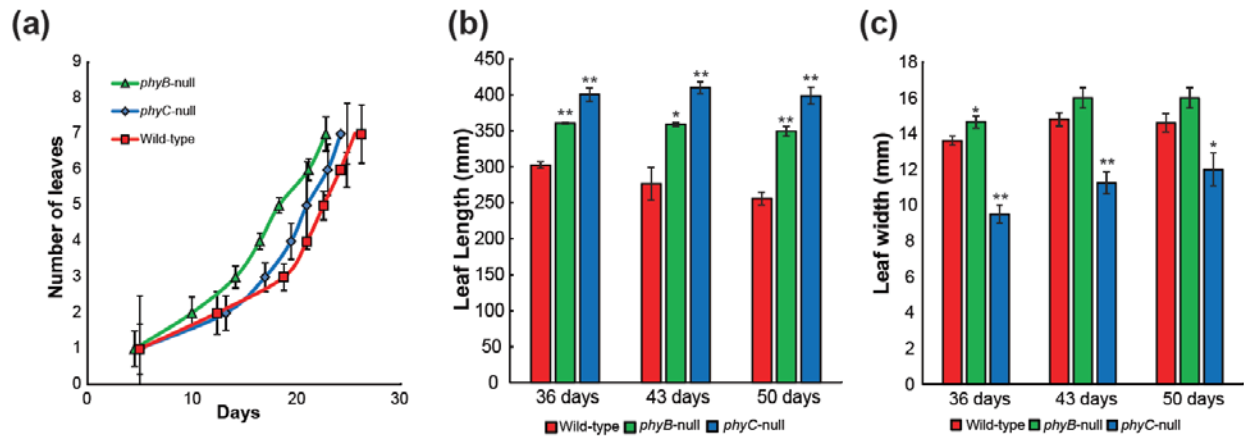


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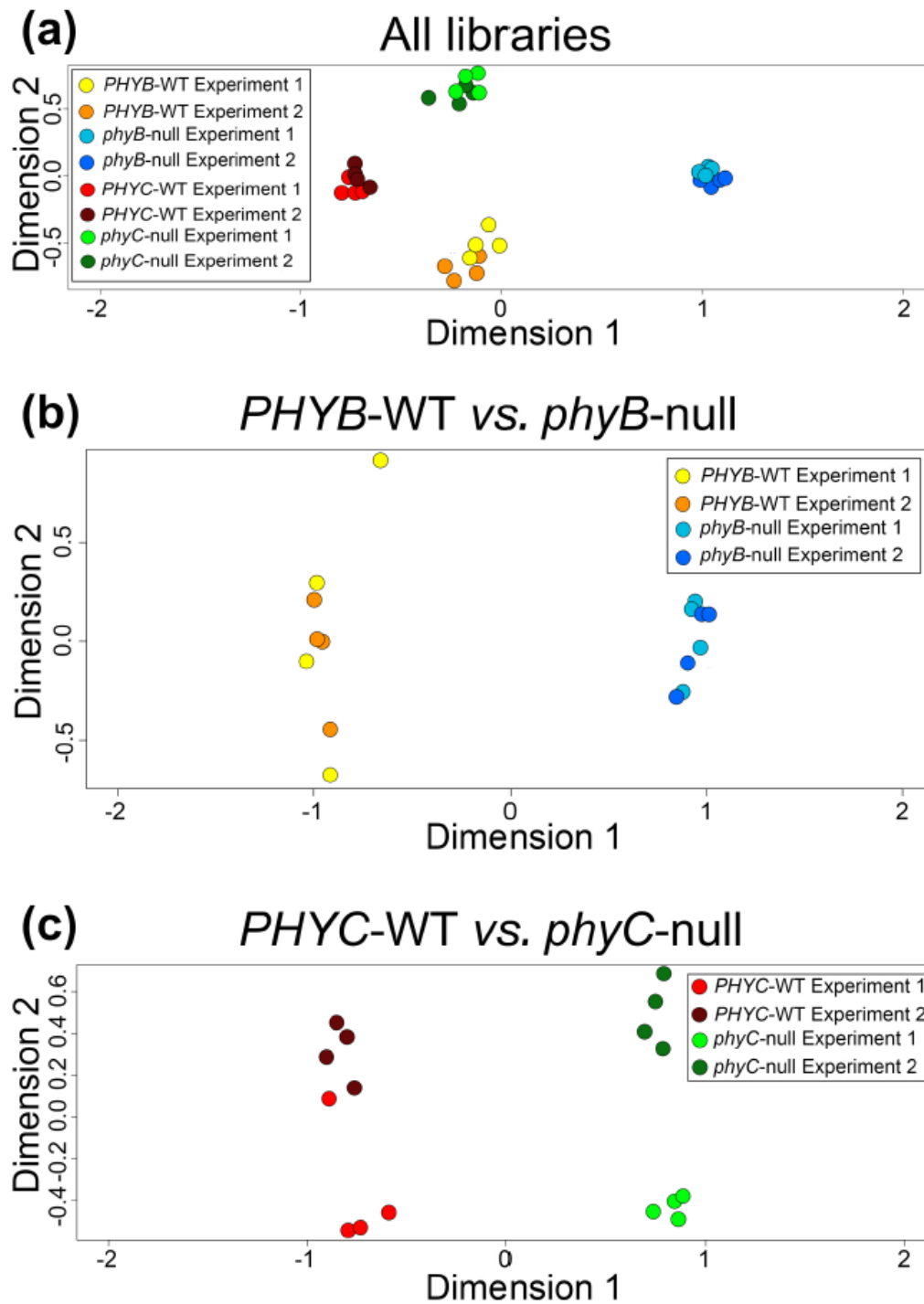


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