

Supplementary data for

Overexpression of the *Arabidopsis* NF-YA transcription factor family leads to opposing abscisic acid responses during seed germination

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Table S1 List of homozygous and hemizygous *p35S::NF-YA* plant lines.

AGI#	Construct	Plant line#	Homozygous or Hemizygous
At5g12840	<i>p35S::NF-YA1-CFP/HA</i>	1	Homozygous
		2	Homozygous
At3g05690	<i>p35S::NF-YA2-CFP/HA</i>	1	Homozygous
		2	Homozygous
At1g72830	<i>p35S::NF-YA3-CFP/HA</i>	1	Homozygous
		2	Homozygous
At2g34720	<i>p35S::NF-YA4-CFP/HA</i>	1	Homozygous
		2	Hemizygous
At1g54160	<i>p35S::NF-YA5-CFP/HA</i>	1	Hemizygous
		2	Hemizygous
At3g14020	<i>p35S::NF-YA6-CFP/HA</i>	1	Hemizygous
		2	Hemizygous
At1g30500	<i>p35S::NF-YA7-CFP/HA</i>	1	Homozygous
		2	Homozygous
At1g17590	<i>p35S::NF-YA8-CFP/HA</i>	1	Homozygous
		2	Homozygous
At3g20910	<i>p35S::NF-YA9-CFP/HA</i>	1	Homozygous
		2	Homozygous
At5g06510	<i>p35S::NF-YA10-CFP/HA</i>	1	Hemizygous
		2	Homozygous

a

	AtNF-YA2	AtNF-YA10	AtNF-YA1	AtNF-YA9	AtNF-YA5	AtNF-YA6	AtNF-YA3	AtNF-YA8	AtNF-YA4	AtNF-YA7	HsNF-YA
AtNF-YA2		63.2%	23.0%	24.0%	22.3%	21.4%	21.6%	19.4%	27.7%	26.5%	19.6%
AtNF-YA10	63.2%		21.5%	20.9%	18.8%	18.5%	20.6%	18.5%	27.5%	27.0%	20.2%
AtNF-YA1	23.0%	21.5%		41.6%	22.2%	18.8%	22.4%	20.0%	26.8%	28.6%	20.4%
AtNF-YA9	24.0%	20.9%	41.6%		22.8%	25.0%	23.1%	21.8%	29.8%	31.0%	20.3%
AtNF-YA5	22.3%	18.8%	22.2%	22.8%		56.3%	34.4%	33.7%	30.5%	32.1%	21.3%
AtNF-YA6	21.4%	18.5%	18.8%	25.0%	56.3%		35.9%	33.7%	30.1%	30.6%	18.6%
AtNF-YA3	21.6%	20.6%	22.4%	23.1%	34.4%	35.9%		63.9%	33.3%	33.0%	21.3%
AtNF-YA8	19.4%	18.5%	20.0%	21.8%	33.7%	33.7%	63.9%		32.3%	31.5%	19.7%
AtNF-YA4	27.7%	27.5%	26.8%	29.8%	30.5%	30.1%	33.3%	32.3%		64.5%	23.0%
AtNF-YA7	26.5%	27.0%	28.6%	31.0%	32.1%	30.6%	33.0%	31.5%	64.5%		25.7%
HsNF-YA	19.6%	20.2%	20.4%	20.3%	21.3%	18.6%	21.3%	19.7%	23.0%	25.7%	

b

	AtNF-YA1	AtNF-YA9	AtNF-YA4	AtNF-YA7	AtNF-YA3	AtNF-YA8	AtNF-YA5	AtNF-YA6	AtNF-YA2	AtNF-YA10	HsNF-YA
AtNF-YA1		78.9%	64.9%	70.2%	66.7%	64.9%	70.2%	66.7%	57.4%	63.2%	64.9%
AtNF-YA9	78.9%		71.9%	77.2%	73.7%	75.4%	73.7%	75.4%	59.0%	63.2%	68.4%
AtNF-YA4	64.9%	71.9%		86.0%	74.1%	75.9%	74.1%	72.4%	61.3%	63.8%	63.8%
AtNF-YA7	70.2%	77.2%	86.0%		75.9%	77.6%	77.6%	75.9%	61.3%	63.8%	65.5%
AtNF-YA3	66.7%	73.7%	74.1%	75.9%		93.1%	82.8%	81.0%	64.5%	63.8%	63.8%
AtNF-YA8	64.9%	75.4%	75.9%	77.6%	93.1%		86.2%	84.5%	61.3%	60.3%	65.5%
AtNF-YA5	70.2%	73.7%	74.1%	77.6%	82.8%	86.2%		91.4%	64.5%	63.8%	67.2%
AtNF-YA6	66.7%	75.4%	72.4%	75.9%	81.0%	84.5%	91.4%		61.3%	60.3%	65.5%
AtNF-YA2	57.4%	59.0%	61.3%	61.3%	64.5%	61.3%	64.5%	61.3%		83.9%	51.6%
AtNF-YA10	63.2%	63.2%	63.8%	63.8%	63.8%	60.3%	63.8%	60.3%	83.9%		55.2%
HsNF-YA	64.9%	68.4%	63.8%	65.5%	63.8%	65.5%	67.2%	65.5%	51.6%	55.2%	

Fig. S2 Identity matrix of the NF-YA proteins. a) Full-length protein. b) Conserved domain. At, *Arabidopsis thaliana*; Hs, *Homo sapiens*. Note that pairs of Arabidopsis NF-YA paralogs demonstrate high identity across the full-length protein sequence and the conserved domain.

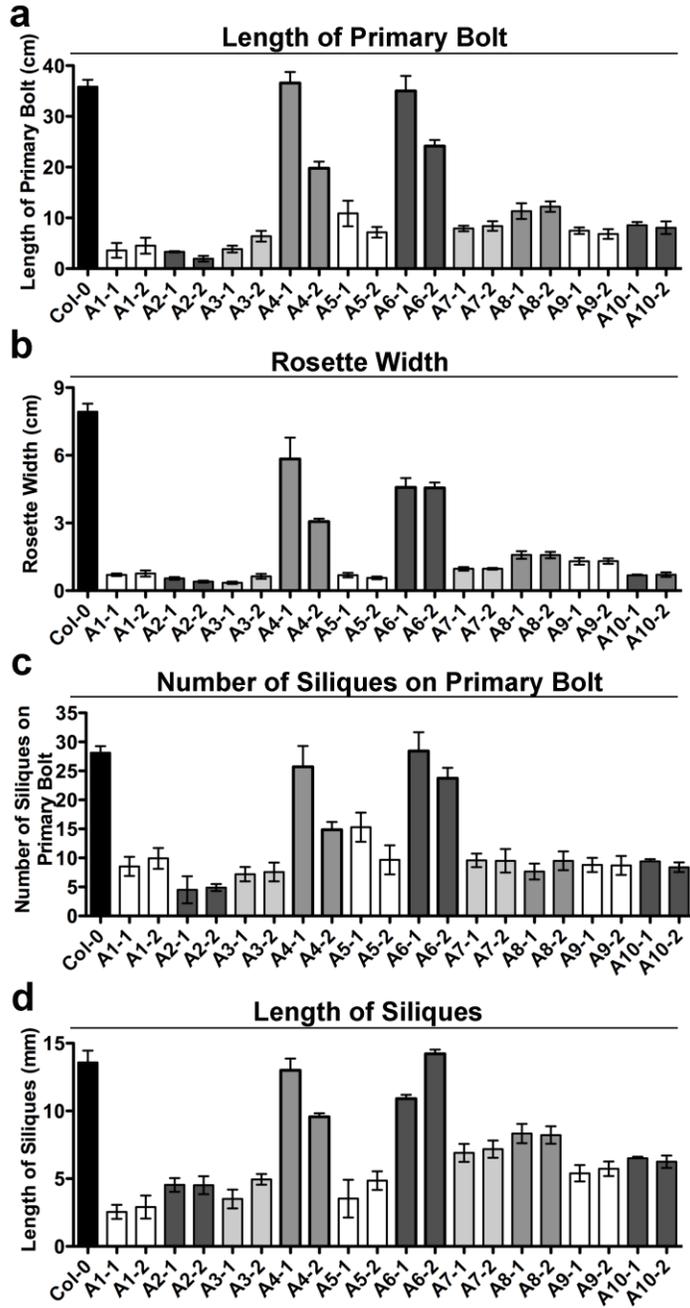


Fig. S3 Overexpression of the *NF-YA* gene family leads to growth defects. Quantitative measurements of (a) plant height (b) rosette width (c) number of siliques on primary bolt (d) length of siliques.

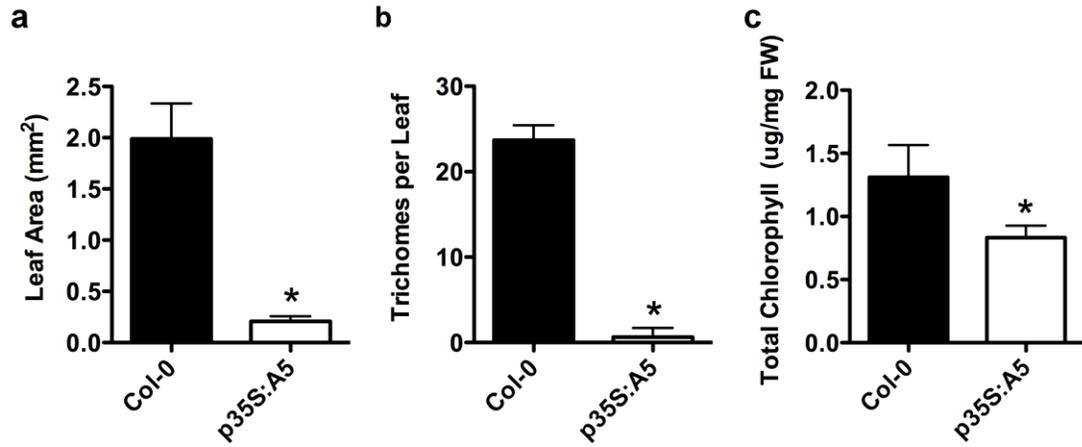


Fig. S4 Quantifications of the cotyledon-like leaves in *p35S::NF-YA5*. (a) Leaf area (b) number of trichomes (c) chlorophyll content. Asterisks represent significant differences derived from Student's t-tests.

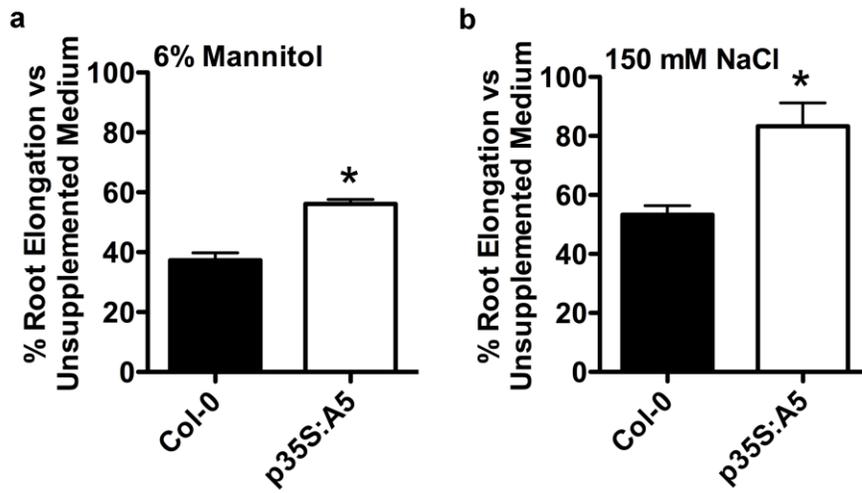


Fig S5. Abiotic stress responses of *p35S::NF-YA5* seedlings. Response to a) 6% mannitol b) 150 mM NaCl. Asterisks represent significant differences derived from Student's t-tests.

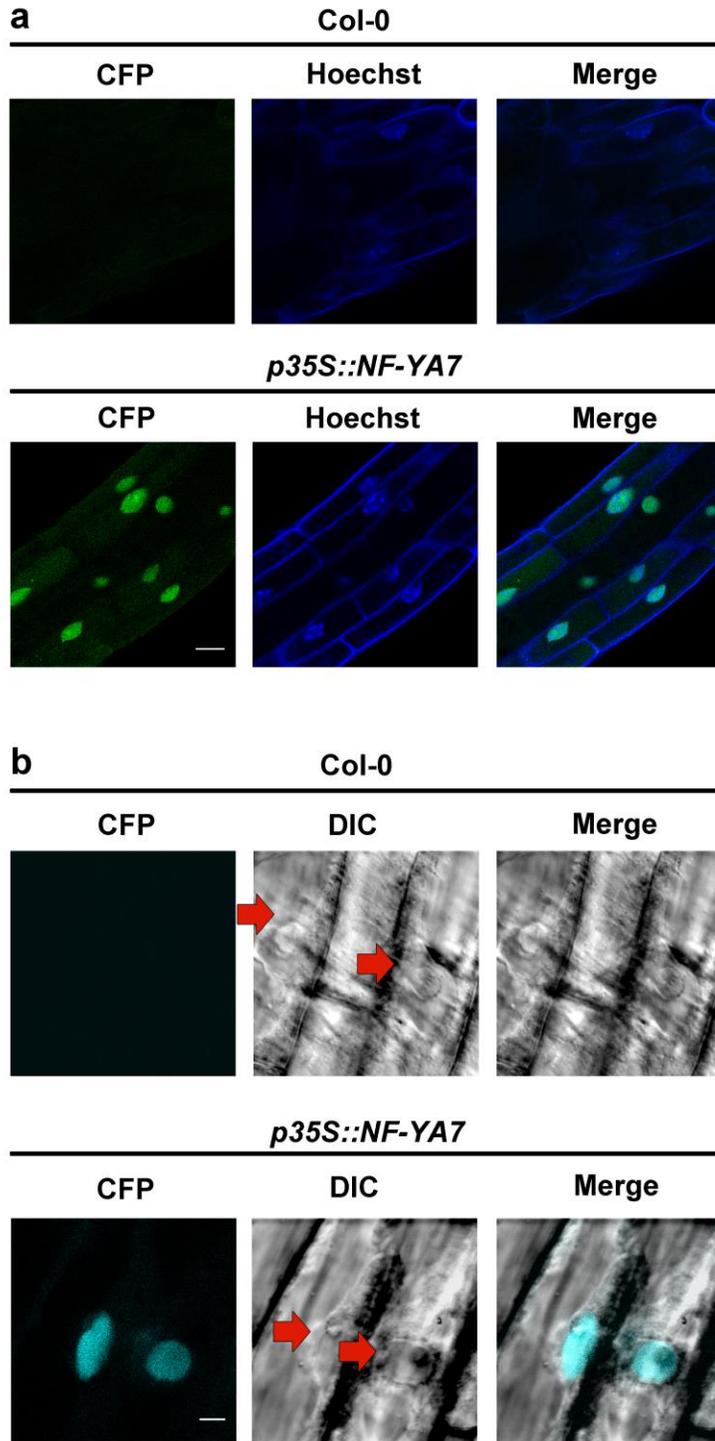


Fig. S6 Confirmation of nuclear localization of NF-YA proteins. (a) Hoechst 33342 (blue) labels nuclei and CFP signal (green) shows localization of NF-YA7. Merged images demonstrate co-localization of DNA label and CFP fluorescence. Scale - 15 μ m. (b) Differential Interference Contrast (DIC) imaging (grey) of root cell nuclei (red arrows point to the nuclei) with CFP signal (cyan) showing localization of NF-YA7. Merged images indicate CFP fluorescence is localized to the root cell nuclei. Scale - 5 μ m.

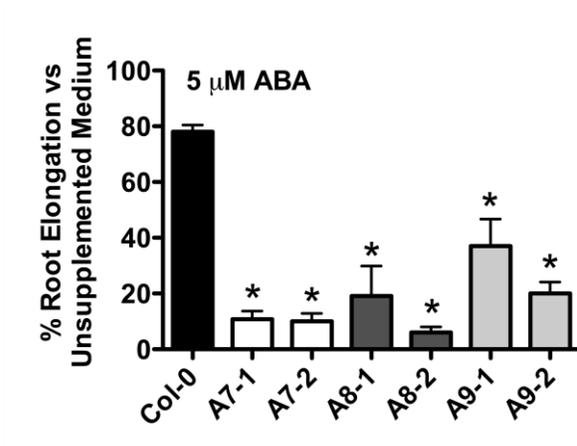


Fig. S7 Overexpression of select *NF-YA* genes leads to ABA hypersensitivity during root elongation. The percent root elongation in growth medium with ABA was compared to the unsupplemented medium for Col-0, *p35S::NF-YA7*, *p35S::NF-YA8*, and *p35S::NF-YA9*. Asterisks represent significant differences derived from one-way ANOVA ($P < 0.05$) followed by Dunnett's multiple comparison post hoc test against Col-0

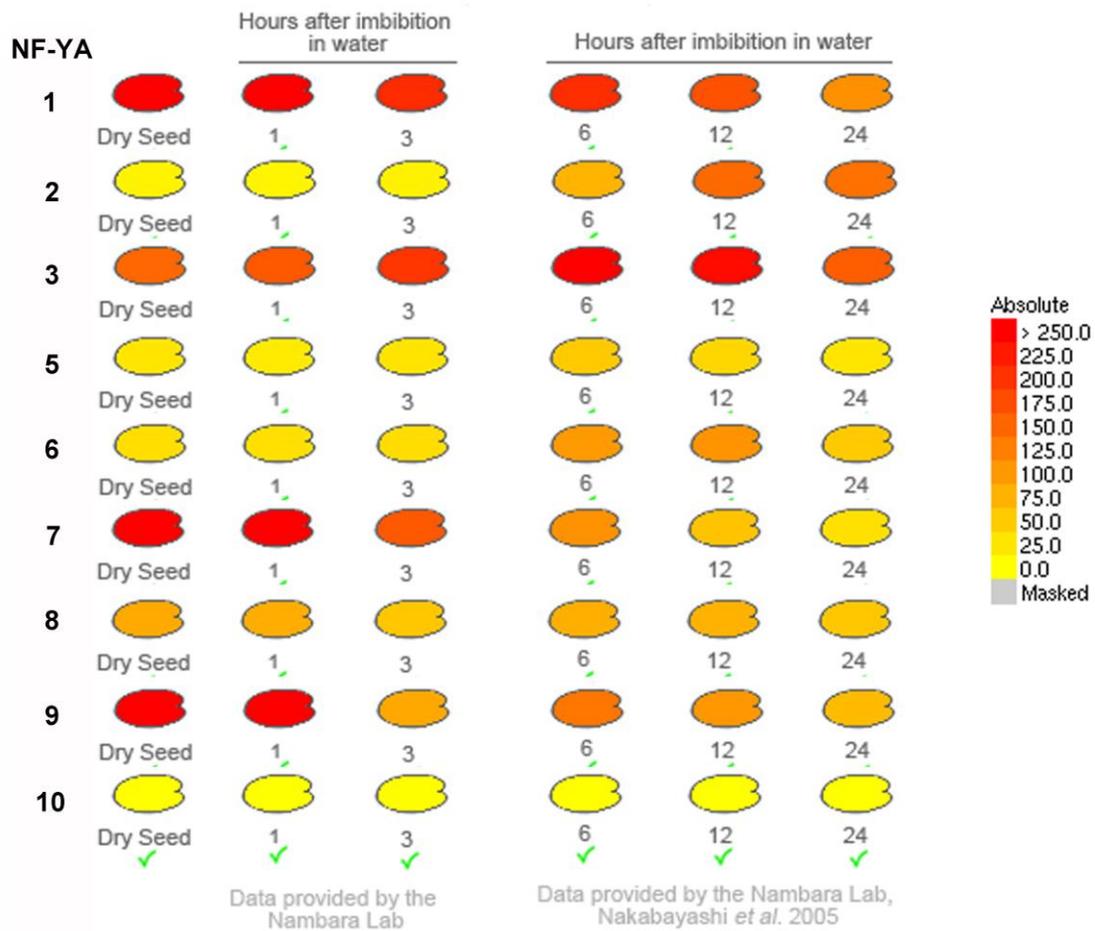


Fig. S8 Publicly available microarray data demonstrate that *NF-YA* genes are expressed in seeds. Data was obtained from the Arabidopsis e-FP browser at <http://bar.utoronto.ca/efp/cgi-bin/efpWeb.cgi>.

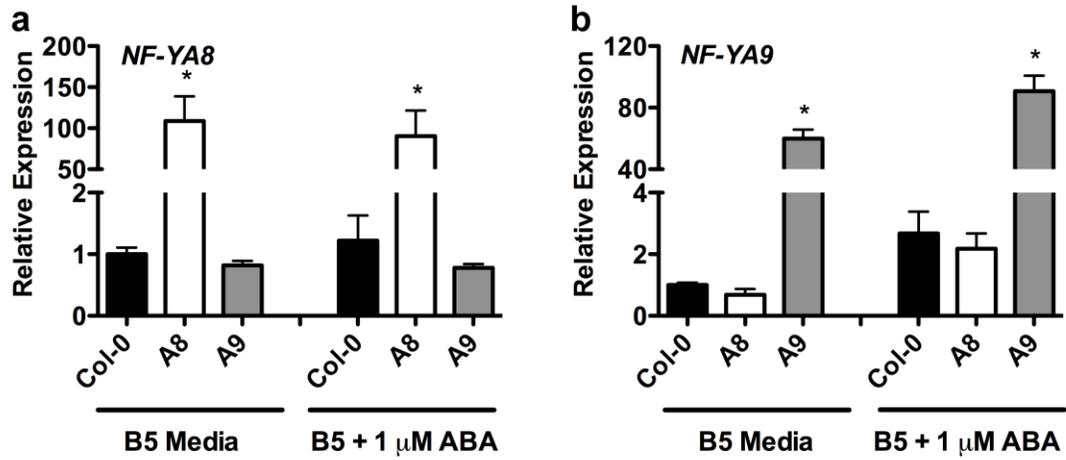


Fig. S9 qPCR analysis of gene expression in *p35S::NF-YA8* and *p35S::NF-YA9* seeds. (A) *NF-YA8* (B) *NF-YA9*. Asterisks represent significant differences derived from two-way ANOVA ($p < 0.05$), in which genotype and seed growth media are the two variables, followed by Bonferroni multiple comparisons post hoc test against Col-0 on B5 media or on B5 + 1 μ M ABA.

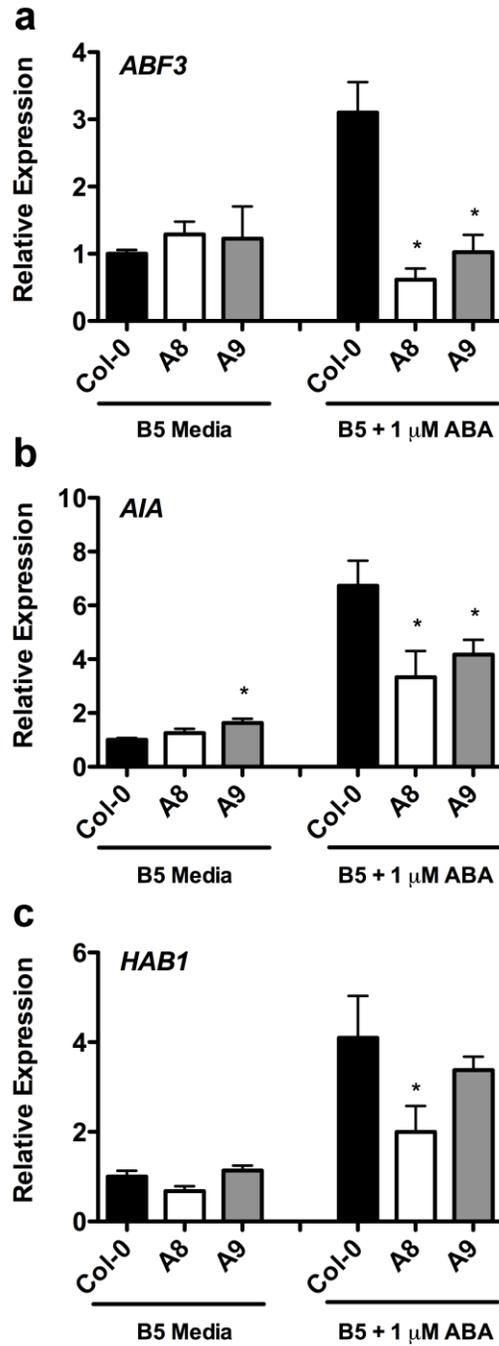


Fig. S10 ABA-induction of gene expression in *p35S::NF-YA8* and *p35S::NF-YA9* seeds. Expression of (A) *ABF3* (B) *AIA* (C) *HAB1*. Asterisks represent significant differences derived from two-way ANOVA ($p < 0.05$), in which genotype and seed growth media are the two variables, followed by Bonferroni multiple comparisons post hoc test against Col-0 on B5 media or B5 + 1 μ M ABA.