

The Role of SBI2/ALG12/EBS4 in the Regulation of Endoplasmic Reticulum-Associated Degradation (ERAD) Studied by a Null Allele

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	Ws-2	<i>bri1-5</i>	<i>sbi2 bri1-5</i>	<i>sbi2</i>	Col-0	<i>bri1-9</i>	<i>sbi2 bri1-9</i>	<i>bri1-235</i>	<i>sbi2 bri1-235</i>
Lobe (μm)	18.29 \pm 0.387	11.24 \pm 0.236 ****	14.19 \pm 0.224 ***	17.68 \pm 0.335	19.55 \pm 0.297	13.29 \pm 0.269 ****	14.08 \pm 0.339	14.4 \pm 0.322 ****	14.46 \pm 0.279
Neck (μm)	17.08 \pm 0.296	20.1 \pm 0.34 ****	19.25 \pm 0.283	18.62 \pm 0.312	15 \pm 0.146	17.15 \pm 0.424 ****	18.11 \pm 0.288	19.22 \pm 0.323 ****	18.11 \pm 0.388
Perimeter (μm)	578.8 \pm 15.78	381.3 \pm 12.87 ****	455.6 \pm 14.31 *	619.8 \pm 16.6	492.9 \pm 12.38	308.9 \pm 13.7 ****	358.8 \pm 14.29	407.1 \pm 12.58 ***	453.2 \pm 13.27
Area (μm^2)	5181 \pm 153.4	4101 \pm 171.6 ****	4761 \pm 203.7	5271 \pm 76.45	4687 \pm 116.3	2621 \pm 142.7 ****	3558 \pm 152.7 ***	3930 \pm 153.4 **	4035 \pm 129.6
Circularity	0.199 \pm 0.007	0.361 \pm 0.011 ****	0.292 \pm 0.009 ***	0.179 \pm 0.008	0.251 \pm 0.006	0.356 \pm 0.012 ****	0.362 \pm 0.015	0.302 \pm 0.007 **	0.256 \pm 0.012

Figure S1. The size of cotyledon pavement cells. The measurements of cotyledon pavement cells were performed using ImageJ software. $N \geq 30$ seedlings for measurements. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$, **** $p < 0.0001$ as Tukey's multiple comparisons test.

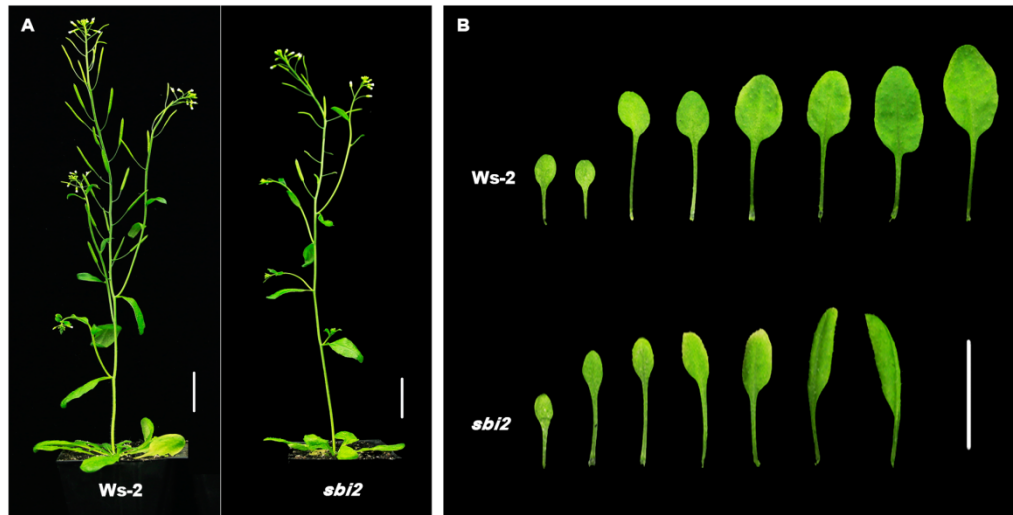


Figure S2. Phenotypes of *Arabidopsis* wild type Ws-2 and mutant *sbi2*. **(A)** The comparison of 2-month-old mature plants of Ws-2 and *sbi2*. Scale bar = 3 cm. **(B)** Fewer leaves were exhibited in *sbi2* than Ws-2. Scale bar = 2 cm.



Figure S3. Three weeks soil-grown plants of *sbi2 bri1-5*, *bri1-5*, three transgenic lines carrying a *SBI2(1-257)* transgene driven by the 35S promoter. Scale bar = 1 cm.

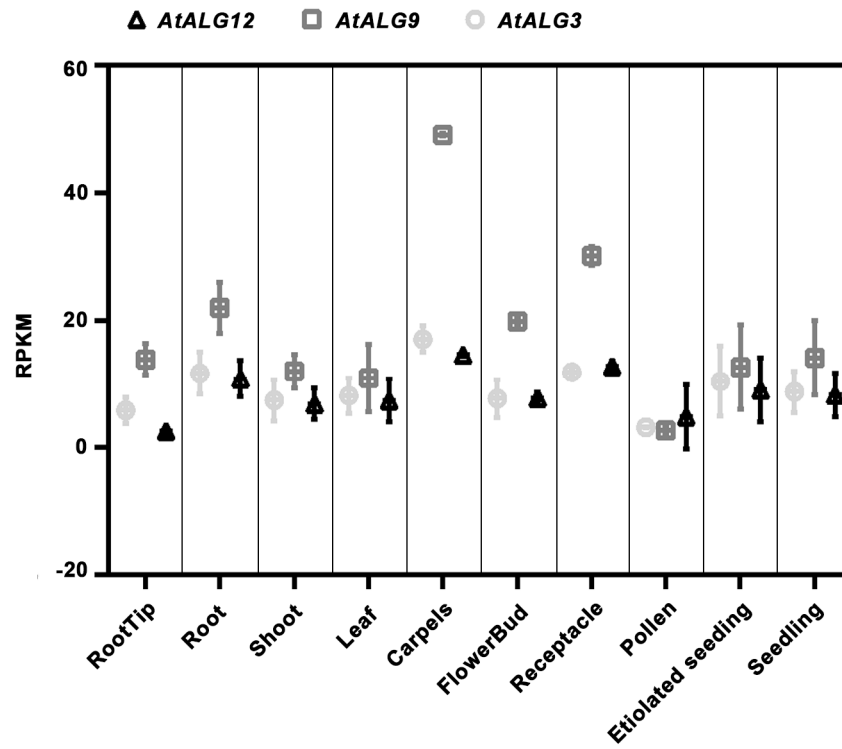


Figure S4. Expression levels of *AtALG12*, *AtALG3* and *AtALG9* were plotted as graph in different tissues (https://bar.utoronto.ca/eFP-Seq_Browser/).

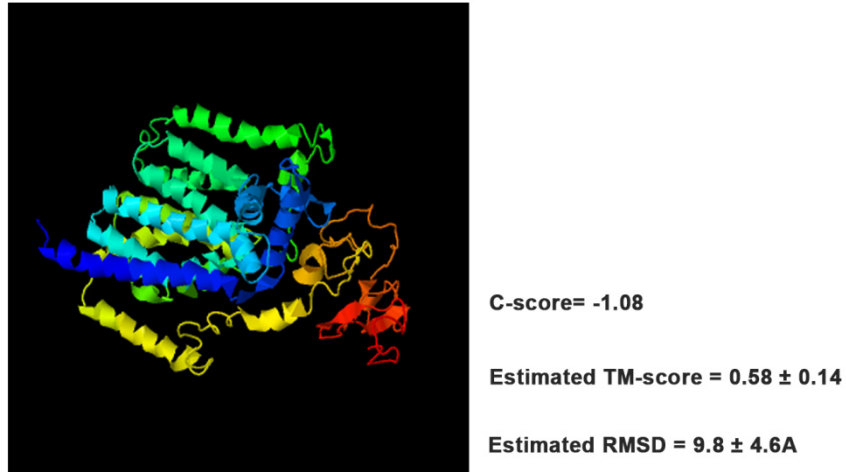


Figure S5. The structure model of SBI2/EBS4 was predicted by I-TASSER. score is in -5, 2 and C-score > -1.5 indicates a model of correct global topology.
(<https://zhanglab.ccmb.med.umich.edu/I-TASSER/>).

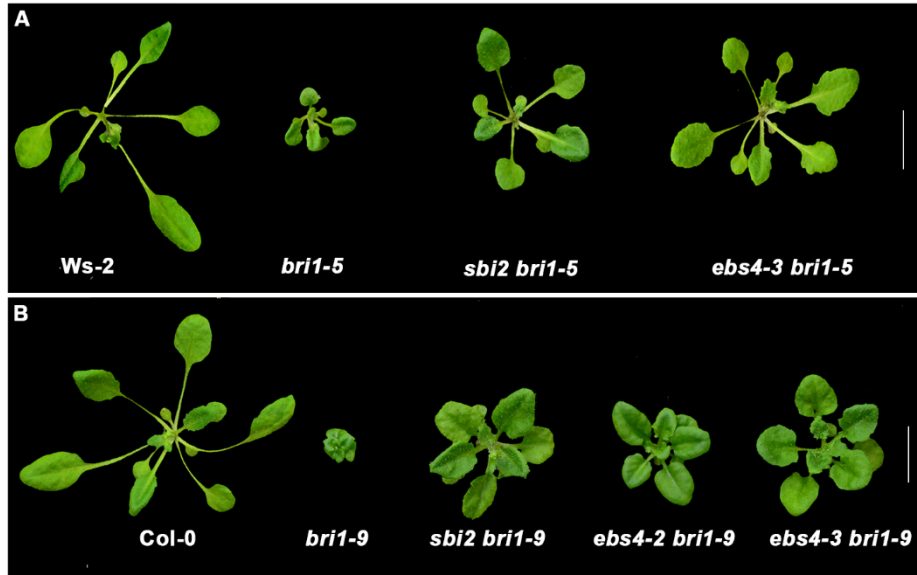


Figure S6. 3-week-old seedlings grown in the soil under a long-day condition (16/8 h, light/dark). (A) Phenotypes of three-week-old soil-grown seedlings of *Ws-2*, *bri1-5*, *sbi2 bri1-5*, and *ebs4-3 bri1-5*. Scale bar, 1 cm. (B) Phenotypes of three-week-old seedlings of *Col-0*, *bri1-9*, *sbi2 bri1-9*, *ebs4-2 bri1-9* and *ebs4-3 bri1-9*. Scale bar, 1 cm.

Table S1. Primers used in this study.

Name	Sequence (5'-3')
Actin2-RT-F	ACTCTCCCGCTATGTATGTCG
Actin2-RT-R	TGGACCTGCCTCATCATACTC
BRI1-RT-F	GGTAGAGAGATGAGGAAGAGA
BRI1-RT-R	CTCATAATTTTCCTTCAGGAAC TTC
CPD-RT-F	GTTCTTATCCTGCTTCCATTTG
CPD-RT-R	AGCCACTCGTAGCGTCTCATT
DWF4-RT-F	CGAAGGAAGGCTCTTTGAATG
DWF4-RT-R	CTTCAACGGCTTTAGGGCAA
BAS1-RT-F	G TTCAGGACATTGTGGAGGAG
BAS1-RT-R	GGATAAAGCAACATAAGGACG
SBI2/EBS4Sac1-F	CGAGCTCATGCCGACGGATTCGAAAATG
SBI2/EBS4BamH1-R	CGGATCCACATCCAGGCCATTTCTTAT
BiP3-RT-F	TTCGAAGTGGACGCAAATGG
BiP3-RT-R	CTATAGCTAACCGACTCAAAGACCT
PDI5-RT-F	CTCGTGAAGCTGAGGGTATTG
PDI5-RT-R	TGTGCGAAATCTAACTCAGAG
ACT2-RT-F	GGTAACATTGTGCTCAGTGGTGG
ACT2-RT-R	AACGACCTTAATCTTCATGCTGC
bri1-5-genotyping-F	AGAATCAAGAGATAGGTGGTTGGG
bri1-5-genotyping-R	GCTGGTTAAAGAAGCAGAGCA
bri1-9-genotyping-F	TAACAATGGTTCGATGATGTTTCTGGACAAGC
bri1-9-genotyping-R	GCATAGTAAGAGCTGACATAGCCTG
bri1-235-genotyping-F	TCTCAACGTCGGATTCAGTGC
bri1-235-genotyping-R	AAGATGTTGCAGAGCAGAGC
bri1-301-genotyping-F	CATCGAAATCTTGTGCCTCTTCTTG
bri1-301-genotyping-R	CTCGGGGTCAAACACATCGCTAATC
bri1-119-genotyping-F	ACCCTTGTAATATCACTAGCAGAGTCTATGAAG
bri1-119-genotyping-R	CGATTTCCGTAAGCATAGTAAGAGC
bri1-116-genotyping-F	TGGCGAGTTACCGATGGATACG
bri1-116-genotyping-R	CTCTTAGATCACCTACCTCATCAGG
det2-1-genotyping-F	AGGATGACTACGAAGACGGAAAC
det2-1-genotyping-R	CAGCCCAGCCCAACCACTCAATAAGCT
LBb1.3	ATTTTGCCGATTTTCGGAAC
cpd-22-LP	TTTCTTTCTCTCCGCTCCTTC
cpd-22-RP	CTACTCCGCCGTACACGTTAC