## **Supplementary Information**

Target Gene	Primer Name	Forward Primer	Reverse Primer	GenBank
Globulin	pANDA-Glb	AAAAAGCAGGCTGAGAGGTTCCAGCCGA	AGAAAGCTGGGTCTCGCCCTGGTCAGC	GQ848069
	Glb	AGTCGGAGATGAGGTTCAGG	GAACATCGGCTGGAACCTC	
Prolamin 10 kDa	10	TTATTTGTGCTGGACTCGGG	GAGAGTTGGAAGTTGACAGGG	EF122448
Prolamin 13 kDa-I	13 I	CAACTACAGTCGCATCTCCTAC	GGGTTGCCACTATGCTATACTG	EF122447
Prolamin 13 kDa-IIa	13 IIa	GCTCTGTTGGCTTTTAACGTG	ACTCATTACAAGACACCGCC	GU120358
Prolamin 13 kDa-III	13 III	TCACCCGTGTTTCAACTGAG	CACAATAGCCTGAACACTGC	FJ940200
Prolamin 16 kDa	16	CTCAATTTGCCCTCCATGTG	AGAACCGCAATGACCAGTAG	EF122449
Glutelin A	GluA	AATGATGGTGAAGTGCCGGT	TCACGCCTGTATGCTTGAGG	EF122456
Glutelin B	GluB	ATTGAGCAACACTCTGGGCA	TGGCTCTGTAGCCTCTTTGC	EF122460
Glutelin C	GluC	CACAAGGGCCAATAGCCAGA	GGTCACGTACATCACCGTGT	EF122465
Glutelin D	GluD	AAGACAGAGCGACCAAGCTC	ATGTGCAACACTAGCCGGAA	EF122464
Binding protein	BiP	AGGACATCAGCAAGGACAAC	GGACTCAATCTCAACACGGAC	AK065743
Protein disulfide isomerase like 1;1	PDIL1;1	AACGATGTGCCAAGCGAGTTCGAT	TTAGAGCTCATCCTTGAGAGGCTC	AB373950
GTP-binding protein Sar1a	Sarla	AGTGTTGTCCGCAAGATGGG	CGCTGGGCAGAGTATGCAAG	AK112012
GTP-binding protein Sar1b	Sar1b	GCAAGATGGGCTATGGGGA	TGGTAAGGTGAAACAGGAGTATGAAC	AK099149
GTP-binding protein Sar1c	Sarlc	GCGTCGTCCGCAAGATG	AGGAGAGTTGATAAACAGAACCAGAG	AK119548
Calnexin	CNX	TCGACAACCCCAACTACAAAG	ATCTCAATCCCAATAGCGGC	AK069118
Protein disulfide isomerase like 2;3	PDIL2;3	ATAAGAGGATTTCCAACTATTAAG	TGCTCCTTGATAATCTACTG	AP005559

 Table S1. Primer list used in this study.





**Figure S1.** Phenotype analysis of seed endosperm by scanning electron microscopy. Seeds were transversely sectioned using a sharp knife and prepared on a specimen slice. Aleurone layers as well as starch granules were observed in wild type (WT) and Glb-RNAi lines. Comparative analysis of endosperm morphology indicates no significant differences between wild type and transformants.



**Figure S2.** Measurement of starch content in wild type (WT) and Glb-RNAi line. An equal amount (100 mg) of seed powder was prepared from WT and Glb-RNAi 1–6. Total starch measurement was performed using TOTAL STARCH Kit, as described by the manufacturer (Megazyme). This analysis indicates that starch content was similar between the wild type (71.059  $\pm$  0.944) and Glb-RNAi 1–6 lines (71.004  $\pm$  1.959).



**Figure S3.** Phenotypic analysis of seed growth. (**a**) The photos indicate comparative grain sizes for the wild type (WT) and Glb-RNAi transformants; (**b**) Ten seeds were randomly selected from the WT and Glb-RNAi lines, and grain size was measured using ImageJ.



**Figure S4.** Immunoblotting analyses using anti-glutelin antibodies. (**a**) Total protein was extracted from one seed which was selected at random from both wild type (WT) and Glb-RNAi lines. Proteins were loaded onto SDS-PAGE and further transferred onto PVDF membranes. These membranes were incubated with diverse anti-glutelin antibodies. The signal was detected and recorded by a luminescent image analyzer (LAS-4000, Fujifilm); (**b**) Proteins used in this analysis were isolated from dry seeds of T<sub>1</sub> generation of WT and Glb-RNAi transformants. The experiment process was identical to the method described previously.