# **Supporting Information**

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### **SI Materials and Methods**

Plant Materials and Growth Conditions. The prematurely senile 1 (ps1-D) mutant was identified by screening our transferred-DNA (T-DNA) population in the japonica Nipponbare background (1). For expression analysis, various organs were collected starting from the heading stage. To determine the fine expression pattern of Oryza sativa NAC (no apical meristem, Arabidopsis ATAF1/2, and cup-shaped cotyledon2)-like, activated by apetala3/pistillata (OsNAP), flag leaves were harvested every 7 d after leaf emergence, and seeds were collected every 2 d after flowering. For PS1 induction analysis, wild-type plants were hydroponically cultured with normal half-strength modified Hoagland medium after germination in the greenhouse for 10 d at 30 °C and with a 14-h light/24 °C, 10-h dark cycle. Whole 10-dold Nipponbare plants were immersed in nutrition solutions containing different kinds of phytohormones, including brassinosteroid (50 µm), gibberellin (50 µm), auxin (50 µm), 6-benzylaminopurine (50 µm), salicylic acid (100 µm), 1-aminocyclopropane-1-carboxylic acid (50 µm), jasmonic acid (50 µm), and abscisic acid (ABA) (50 µm), and were maintained for 2 h in the dark under cold (4 °C) and alkaline (pH = 11) conditions with 200 mM mannitol and 200 mM NaCl. Twenty plants were collected for RNA isolation. All experiments were repeated three times independently.

**Cloning PS1.** Genetic linkage analysis was performed with the T-DNA insertion using the  $T_1$  progenies of the *ps1-D* mutant. The flanking sequence of the T-DNA in *ps1-D* was isolated by SiteFinding PCR (2). (The primers used for SiteFinding PCR are listed in Table S5.)

**Constructions for Rice Transformation.** For the *OsNAP* overexpression construct, a 1,179-bp section of full-length cDNA was amplified by PCR from the cDNA library of Nipponbare and was subcloned into the XmaI and XbaI sites of the binary vector pCAMBIA2300-*Actin1* to generate the pAOS construct. To make the *OsNAP* RNAi construct, a 274-bp gene-specific fragment from bp 457–730 of the *OsNAP* ORF was amplified, and cloned into the SaII and PstI sites of the binary vector pCAMBIA2300-35S as described previously (5). For promoter analysis, the fragment ~1.9 kb upstream of the *OsNAP* 5' region was amplified and ligated into the pCAMBIA2391Z vector between SaII and EcoRI. The wild-type rice variety Nipponbare, was used as the recipient for all transformations by the *Agrobacterium tumefaciens*-mediated method (3). The primer sequences are listed in Table S6.

**Constructions for Yeast One-Hybrid Assay.** To generate activation domain OsNAP (AD-OsNAP), the full-length *OsNAP* was amplified using thermostable DNA polymerase from *Thermococcus kodakaraensis* (KOD) polymerase (Toyobo) and was subcloned into the EcoRI and XhoI sites of the binary vector pJG4-5 vector (Clontech). To generate *SGRp::LacZ*, *NYC1p::LacZ*, *NYC3p::LacZ*, *RCCR1p::LacZ*, *Osh36p::LacZ*, *Osh69p::LacZ*, and *OsI57p::LacZ* reporter constructs, the promoter fragments were amplified by PCR and then cloned into the pLacZi2µ vector. The primer sequences are listed in Table S6.

**Expression Analysis.** Total RNA was extracted using a TRIzol kit (Invitrogen) according to the manufacturer's instructions. RNA was reverse-transcribed using the ReverTra Ace quantitative PCR RT Master Mix Kit with gDNA remover (Toyobo). Quanti-

tative real-time PCR was performed with a Chromo4 real-time PCR detection system (Bio-Rad) according to the manufacturer's instructions. Rice *OsActin1* was used as an internal control. The primers used for quantitative RT-PCR (qRT-PCR) are listed in Table S6. Values are means  $\pm$  SD of three biological repeats. The Student *t* test was used for statistical analysis.

 $\beta$ -Glucuronidase Staining.  $\beta$ -Glucuronidase (GUS) staining was performed as described previously (4). Images were taken directly or with a stereomicroscope (SZX16; Olympus).

**Chlorophyll Measurements.** Chlorophyll was extracted from 50 mg of leaf tissue (fresh weight), and its content was determined by measuring the absorbance at 652 nm using a Tecan Infinite M200 multimode reader (Tecan Group Ltd) as described previously (5).

**Transactivation Activity Assay in Yeast.** The coding sequences of *OsNAP* and *OsNAP* fragments were obtained by PCR. Primers used are included in Table S6. The PCR products were digested by NdeI/EcoR, and then cloned into the vector pGBKT7. Three constructs with deletions in middle of the gene were made also, and the N-terminal parts (amino acids 1–64, 1–100, and 111–142) of these constructs were fused to the C-terminal fragment (amino acids 181–392). The Matchmaker Gold Yeast Two-Hybrid system (Clontech) was used for the transactivation activity assay according to the manufacturer's protocol. Each yeast liquid culture was diluted serially to  $OD_{600} = 0.6$ , and 3 µL of each dilution was inoculated onto tryptophan-, histidine-, and adenine-negative synthetic dropout medium.

**Dark-Induced Leaf Senescence.** Fully expanded flag leaves were excised carefully. Detached leaves were cut into  $\sim$ 3-cm pieces and floated on 25 mL of water or 50  $\mu$ M of ABA solution in Petri dishes with the adaxial side up. The samples were incubated at 28 °C in darkness for 5–10 d (5).

Yeast One-Hybrid Assay. Plasmids for the GAL DNA-binding domain fused with *OsNAP* were cotransformed with the *LacZ* reporter genes driven by *SGR*, *NYC1*, *NYC3*, *RCCR1*, *Osh36*, *OsI57*, and *Osh69* promoter fragments (Clontech) into the yeast strain EGY48 using a standard transformation technique. Transformants were grown on proper drop-out plates containing X-Gal for blue color development (6). Representative data are shown from one of the three biological replicates, which yielded similar results.

**ABA Analysis.** Extraction and determination of ABA from agematched flag leaves at the heading stage was performed as described (7). Four independent biological repeats were performed.

**Microarray Hybridization and Data Analysis.** For the microarray experiment, RNAs from fully expanded flag leaves of wild-type and *ps1-D* plants were used for microarray hybridization (CapitalBio Corporation). Probe labeling and chip hybridization were carried out through the Affymetrix custom service following the standard protocol (www.affymetrix.com/). Normalization was performed according to the standard Affymetrix protocol to allow the comparison of the samples for each set of experiments. The differently expressed genes (DEGs) in wild-type and *ps1-D* plants were classified functionally using the biological process category of Rice Gene Ontology (www.geneontology.com). The percentage shown in Table S1 refers to the ratio of genes relative

to the total up-regulated or down-regulated DEGs in each functional category. Three independent biological replicates were conducted for the wild-type and *ps1-D* plants, and DEGs with  $log_2$  ratios  $\geq 1.00$  or  $\leq -1.00$  (only GO Slim IDs with *P* values  $\leq 0.5$ ) were analyzed.

**Element and Protein Measurements.** Element and protein measurements were performed as described (8, 9).

**ChIP-PCR Assay.** The wild-type rice (Nipponbare) and its transgenic calli harboring the *Pro35S:OsNAP-MYC* construct were used for ChIP assays according to the method described previously (10). Antibody to the MYC tag was used to immunoprecipitate OsNAP-MYC and associated DNA fragments. Immunoprecipitation was performed with anti-MYC antibody (A-MYC) or without antibody (No Ab). Fragments (2,000 bp) of sequences upstream of ATG were chosen as the promoter regions for designing primers. The promoter of *Ubiquitin* was used as a negative control. Three independent biological repeats were performed.

**Phylogenetic Analysis.** Protein sequences showing similarity to OsNAP were retrieved using the public BLAST on the National Center for Biotechnology Information Website (www.ncbi.nlm. nih.gov/). Multiple alignments of the homologs were performed by Clustal X version 2.0 with the default parameters and manually adjusted (11).

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**Field Cultivation of Rice.** The rice plants were grown in a standard paddy field at the Experimental Station of the Institute of Genetics and Developmental Biology. The field planting followed a randomized complete-block design with three replicates; the plants were transplanted at a spacing of  $20 \times 20$  cm. The area per plot was 4 m<sup>2</sup>. These plants then were grown under conventional cultivation conditions. Agronomic traits were measured using at least 10 plants randomly obtained from the center of the plot to avoid any irregularities present at the plot margins.

Accession Codes. Genes and their associated accession codes from GenBank/EMBL are as follows: Osh36 (AF251070), OsI57 (AF251076), Osh69 (AF251068), OsI85 (AF251075), OsNCED1 (AY838897), OsNCED2 (AY838898), OsNCED3 (AY838899), OsNCED4 (AY838900), and OsNCED5 (AY838901). Genes and their associated accession codes from Rice Annotation Project Database are as follows: OsNAP (LOC\_Os03g21060), G1 (LOC\_ Os03g21020), G2 (LOC\_Os03g21030), G3 (LOC\_Os03g21040), G4 (LOC\_Os03g21050), G5 (LOC\_Os03g21070), G6 (LOC\_ Os03g21080), G7 (LOC\_Os03g21090), G8 (LOC\_Os03g21100), G10 (LOC\_Os03g21110), SGR (LOC\_Os09g36200), NYC1 (LOC\_Os01g12710), NYC3 (LOC\_Os06g24730), RCCR1(LOC\_ Os10g25030), OsZEP (LOC\_Os04g37619), OsABA80X1 (LOC\_ Os02g47470), OsABA80X2 (LOC\_Os08g36860), and OsABA80X3 (LOC\_Os09g28390).

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**Fig. S1.** Characterization of *ps1-D* mutants at the vegetative growth stage. (*A*) Phenotypes of the *ps1-D* mutant in the vegetative growth stage. (Scale bar: 20 cm.) (*B*) The upper five leaves from the main culm of *ps1-D* mutants and wild-type plants 60 d after transplanting. (*C*) Chlorophyll content of the leaves shown in *B*. Values are means  $\pm$  SD of 10 measurements. (*D*) Chlorophyll content of the leaves shown in Fig. 1*D*. Values are means  $\pm$  SD of 10 measurements. (*E* and *F*) Expression of the senescence markers *Osh36* (*E*) and *Osl57* (*F*) in the detached leaves of *ps1-D* mutants and wild-type plants incubated with water for 5 d in darkness. \*\* $P \le 0.01$ ; Student *t* test.



**Fig. 52.** Cloning of *PS1*. (*A*) Schematic representation of the T-DNA flanking region of the *ps1-D* mutant. LB and RB represent the left and right borders of the T-DNA, respectively. (*B*) Flanking sequence was confirmed by PCR using the primers indicated in *A*. Heter, heterozygous *ps1-D* mutant; Homo, homozygous *ps1-D* mutant. (C) The expression of *PS1* was specifically activated in *ps1-D* mutants. G1–G4 are four genes located in the 50-kb region upstream of the T-DNA insertion site; G5–G10 are six genes located in the 50-kb region downstream of the *PS1* gene. \*\* $P \le 0.01$ ; Student *t* test. (*D*) Overexpression of *PS1* minics the premature senescence phenotype of *ps1-D* mutants. (Scale bar: 20 cm.) (*E*) qRT-PCR analysis of *PS1* transcript levels in wild-type plants and three lines over-expressing *PS1* (OE1–OE3). (*F*) Protein sequence alignment of PS1 and homologs in *Arabidopsis* and wheat using Clustal W. Conserved amino acids are highlighted: White letters on a black background indicate amino acids that are conserved across all three samples (100% conservation); black letters on a gray background indicate amino acids that are conserved ion).



Fig. S3. Transcriptional analysis of OsNAP. (A) Schematic structure of Va (1)/ NAP members in rice. Numbers indicate the position of amino acids. (B) (Left) Schematic structure of OsNAP protein. Numbers indicate the position of amino acids. (Right) Truncation analysis of transcriptional activation activity of OsNAP. Various truncated OsNAPs were constructed as bait vectors. BD, GAL4 DNA-binding domain. Empty control vector (BD) and GAL4 genes were used as negative and positive controls, respectively. All bait vectors were transformed into yeast strain Y2HGold (Invitrogen). Yeast was grown at 30 °C for 3 d on the synthetic dextrose agar plates lacking leucine and histidine and containing different concentrations of 3-amino-1,2,4-triazole, as indicated.

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**Fig. 54.** Analysis of *OsNAP* expression. (*A*) Expression of *OsNAP* in various organs, including root, culm, leaf blade, leaf sheath, and panicle at the heading stage and endosperm 7 d after flowering. (*B*) Expression of *OsNAP* in leaves at different developmental stages. LS, leaf at the late stage of senescence, with >50% leaf area yellowing; NS, fully expanded, nonsenescent leaf; OS, leaf at the onset of senescence, with <5% leaf area yellowing; YL, young leaf approximately half the size of a fully expanded leaf. (*C*) Change over time in the *OsNAP* transcription levels of the seeds. Y1, young panicle (1–2 cm). (*D*) *OsNAP* expression in different leaves at the heading stage. L1–L5, leaf of plants numbered from top to bottom.



**Fig. S5.** Expression of ABA-related genes. \*\* $P \le 0.01$ ; Student *t* test. (*A*) Expression of ABA biosynthetic genes in wild-type and *ps1-D* mutant plants. (*B*) Expression of ABA metabolism genes in wild-type and *ps1-D* mutant plants.



**Fig. S6.** Confirmation of *OsNAP* function by RNAi (Ri) transgenic lines. (*A*) Expression analysis of five orthologous members of *OsNAP* among wild-type, Ri1, and Ri2 transgenic lines. *LOC\_Os05g34310* was not detected in leaves. (*B* and *C*) Expression analysis of four chlorophyll degradation-related genes (CDGs) (*SGR*, *NCY1*, *NCY3*, and *RCCR1*) (*B*) and four other senescence-associated genes (SAGs) (*Osh39*, *Osl57*, *Osh69*, and *Osl85*) (*C*) in wild-type and *OsNAP* Ri transgenic lines. (*D*) Delay in the dark- and ABA-induced senescence phenotype of the *OsNAP* RNAi leaves. Detached flag leaves from the wild-type and *OsNAP* RNAi lines at the heading stage were incubated with continuous light (H<sub>2</sub>O), darkness (H<sub>2</sub>O), and ABA (continuous light). PT, Pretreatment. (Scale bar: 2 cm.) (*E*) Chlorophyll content of the detached leaves; labels are as in *D*. Values are means  $\pm$  SD of five measurements. (*F–H*) Expression of *OsNAP* (*F*), *Osh36* (*G*), and *Osl57* (*H*) in detached leaves; labels are as in *D*. \**P*  $\leq$  0.01; Student *t* test.



**Fig. 57.** OsNAP binds directly to the promoter regions of nutrient transport-related genes. ChIP-PCR analysis of the promoter region of SAGs. The ChIP assays were performed using calli expressing the OsNAP-MYC fusion protein. Antibody to the MYC tag was used to immunoprecipitate OsNAP-MYC and associated DNA fragments. Immunoprecipitation was performed with or without (No Ab) anti-MYC antibody. Fragments (2,000 bp) of sequences upstream of ATG were chosen as the promoter regions for designing primers. The promoter of *Ubiquitin* was used as a negative control.

## Table S1. Functional classification of DEGs in ps1-D

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	No. of up-regulated DEGs				No. of down-regulated DEGs			
Category	≥2	1 or 2	Total	Percentage	≤2	2 or 1	Total	Percentage
Regulatory genes								
Transcription factors and nucleic acid-binding proteins	7	18	25	8.2	2	15	17	5.7
Ubiguitination control	1	3	4	1.3	0	7	7	2.4
Protein kinase and phosphatases	0	9	9	3.0	3	14	17	5.7
Signal transduction	1	5	6	2.0	2	12	14	4.7
Macromolecule degradation and mobilization								
Amino acid metabolism and N mobilization	2	15	17	5.6	2	11	13	4.4
Lipid metabolism and mobilization	0	9	9	3.0	0	4	4	1.4
Nucleic acid metabolism and phosphate mobilization	1	7	8	2.6	0	6	6	2.0
Carbohydrate metabolism	2	6	8	2.6	1	15	16	5.4
Growth and development								
Embryo and seed development	0	2	2	0.7	0	2	2	0.7
Flavonoid/anthocyanin pathway	0	5	6	2.0	0	4	4	1.4
Stress/defense response	1	22	23	7.6	3	21	24	8.1
Transport facilitation	2	12	14	4.6	0	16	16	5.4
Electron transport	1	9	10	3.3	1	2	3	1.0
Biosynthesis	0	6	6	2.0	1	9	10	3.4
Hormone	1	3	4	1.3	0	3	3	1.0
Chlorophyll degradation	0	5	5	1.6				
Metal binding	0	3	3	1.0	0	1	1	0.3
Structural	3	2	5	1.6	0	0	0	0
Senescence	0	0	0	0	1	1	2	0.7
Unknown function	23	83	106	34.9	5	91	96	32.4
Others	4	29	33	10.9	4	37	41	13.9

DEGs, differentially expressed genes. Functional classification of the DEGs was performed using the biological process category of the Rice Gene Ontology (http://www.geneontology.com). Percentage refers to the ratio of genes in each functional category to the total up-regulated or down-regulated DEGs identified in the microarray experiment.

Table S2.	Transport-associated	genes identified	in the microarray	<sup>,</sup> data as being	differentially	expressed in	ps1-D
mutant pl	ants						

Locus	Annotation	Fold change
Up-regulated		Log2 ratio
LOC_Os07g33910	Transporter family protein	3.70
LOC_Os04g38680	Transmembrane amino acid transporter protein	2.52
LOC_Os12g02310	Lipid transport	1.98
LOC_Os06g05160	Sulfate transporter	1.77
LOC_Os01g65110	Proton-dependent oligopeptide transporter family protein	1.38
LOC_Os11g02389	Lipid transport	1.37
LOC_Os04g32920	Potassium transporter	1.21
LOC_Os02g57240	Probable voltage-gated potassium channel subunit beta	1.15
LOC_Os01g31940	Purine permease	1.14
LOC_Os03g40780	Transport protein-related	1.08
LOC_Os12g44100	Peptide transporter PTR2	1.06
LOC_Os12g02320	Transport	1.05
LOC_Os07g15460	Metal transporter Nramp6	1.05
LOC_Os10g22039	Mitochondrial import inner membrane translocase subunit Tim17	1.03
Down-regulated		
LOC_Os06g22960	Aquaporin protein	-1.82
LOC_Os03g10640	Calcium-transporting ATPase, plasma membrane-type	-1.76
LOC_Os02g07830	OsHKT6, high affinity potassium transporter	-1.63
LOC_Os06g38120	Low-affinity cation transporter	-1.54
LOC_Os07g26660	Aquaporin protein	-1.53
LOC_Os12g12934	Peptide transporter PTR3-A	-1.46
LOC_Os02g21340	ABC-2 type transporter family protein	-1.41
LOC_Os02g27490	Bile acid sodium symporter family protein	-1.39
LOC_Os04g37990	Transporter family protein	-1.27
LOC_Os05g07870	Triose phosphate/phosphate translocator	-1.22
LOC_Os06g29790	Phosphate transporter 1	-1.20
LOC_Os10g28240	Calcium-transporting ATPase, plasma membrane-type	-1.18
LOC_Os12g08130	Amino acid transporter	-1.16
LOC_Os04g12499	Amino acid transporter protein	-1.08
LOC_Os04g55260	Thiamine-repressible mitochondrial transport protein THI74	-1.03
LOC_Os02g37160	Heavy metal transport/detoxification protein	-1.02

Table S3. Residual nitrogen, P, S, K, Mg, Fe, and Zn in flag leaves among wild type, *ps1-D*, *OsNAP* OE, and RNAi transgenic lines

	N, %	P, ppm	S, ppm	K, ppm	Mg, ppm	Fe, ppm	Zn, ppm
WT	1.3	607.9	927.0	4,866.5	1,326.1	822.9	20.3
ps1-D	1.2	438.8	683.4	4,320.3	877.2	543.2	15.7
Δ, %	-9.4	-27.8	-26.3	-11.2	-33.9	-33.9	-22.3
OE1	1.1	531.1	688.4	4334.9	907.6	577.7	14.7
Δ, %	-14.9	-12.6	-25.7	-10.9	-31.6	-29.8	-27.4
OE2	1.2	447.1	790.2	4,774.8	1,187.2	603.7	16.5
Δ, %	-7.8	-26.5	-14.8	-1.9	-10.5	-26.6	-18.7
Ri1	1.6	707.6	1,146.2	5,971.8	1,773.1	1,040.5	25.7
Δ, %	21.1	16.4	23.6	22.7	33.7	26.4	26.9
Ri2	1.4	806.1	1,240.7	6,124.9	1,623.2	1,066.5	22.5
Δ, %	11.7	32.6	33.8	25.9	22.4	29.6	11.1

Ri1 and Ri2 represent two independent *OsNAP* RNAi lines. Differences between the means of wild type and *ps1-D*, OE, or RNAi transgenic plants ( $\Delta$  %) are reported in terms of change with respect to the wild type. Values are means  $\pm$  SD of ten biological replicates.

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Table S4. Agronomic traits of ps1-D and OsNAP RNAi transgenic lines

WT	ps1-D	Ri1	Ri2
96.2 ± 1.8	75.1 ± 2.2*	98.3 ± 0.7*	98.5 ± 1.6*
25.7 ± 1.4	18.9 ± 3.2*	28.0 ± 2.0*	29.0 ± 2.6*
12.5 ± 0.8	10.3 ± 0.8	12.6 ± 1.6	12.7 ± 1.7
6.2 ± 0.2	$4.5 \pm 0.2*$	6.9 ± 0.3*	$7.5 \pm 0.8^{*}$
	WT 96.2 ± 1.8 25.7 ± 1.4 12.5 ± 0.8 6.2 ± 0.2	WT ps1-D   96.2 $\pm$ 1.8 75.1 $\pm$ 2.2*   25.7 $\pm$ 1.4 18.9 $\pm$ 3.2*   12.5 $\pm$ 0.8 10.3 $\pm$ 0.8   6.2 $\pm$ 0.2 4.5 $\pm$ 0.2*	WTps1-DRi1 $96.2 \pm 1.8$ $75.1 \pm 2.2^*$ $98.3 \pm 0.7^*$ $25.7 \pm 1.4$ $18.9 \pm 3.2^*$ $28.0 \pm 2.0^*$ $12.5 \pm 0.8$ $10.3 \pm 0.8$ $12.6 \pm 1.6$ $6.2 \pm 0.2$ $4.5 \pm 0.2^*$ $6.9 \pm 0.3^*$

Ri1 and Ri2 represent two independent *OsNAP* RNAi lines. DWV, dry weight of vegetative organs. NPR stands for the rate of net photosynthesis (400  $\mu$ mol•CO2•m<sup>-2</sup>•s<sup>-1</sup>), measured at the heading stage (0 DAF) and 15 d after flowering (15 DAF). Values are means  $\pm$  SD from at least 10 plants in Beijing. Asterisks indicate a statistically significant difference in comparison to the wild type (\*P  $\leq$  0.05).

Table S5.	Primers used	in this study	for molecular	cloning of PS1

SF SEP1	CACGACACGCTACTCAACACCACCTCGCACAGCGTCCTCAAGCGGCCGCNNNNNNGCCT
SFP2	ACTCAACACCACCTCGCACAGC
SP1 SP2	AGGCATCGATCGTGAAGTTTCTCATC
SP3	GCTTTCGCCTATAAATACGACGGATCG

Table S6. Primers used in this study for qRT-PCR, to generate DNA constructs, for ChIP-PCR, and for yeast one-hybrid assays

	Forward primers	Reverse primers
Primers used for qRT-P	PCR	
ACT1	TCCATCTTGGCATCTCTCAG	GTACCCTCATCAGGCATCTG
PS1	CAAGAAGCCGAACGGTTC	GTTAGAGTGGAGCAGCAT
G1	TAACGAGAAGCAACAGGAG	ATTGGTTCAGCAGAAGATATG
G2	CAGGTCATGAGCTACCAGCA	CGGGAGCGAGAAGTACTGAC
G3	GATCAAGGACATGAGGCACA	CCTGGTATCCGGGGTTGTA
G4	TGCAGGGCAGACCTCTAAGT	CGTGCCAACACCATAAACAG
G5	CATCCCTTTTTGGAGTGGAA	CACGGGATTCTTGGTCACTT
G6	TGGATGGCAATCACACCTTA	GCCCGGACAGTACCAGTAGA
G7	CGTACCCTTGGTGGAAAGAA	TTGTTTCTCCGGGTGTTAGG
G8	ATGTCATAAAGCCGGCTGAC	TAGCCCCTATGCAATCCTTG
G9	TAGATGCACACGCTTCTTCG	TCCGTCGGAGAGAAATCATC
G10	GGTGCATTCCAGTGGGTTAC	AGCCATTTGGACAGAACCAC
NYC1	CATGCAACACCAACAAAAGG	GACCATTCCAGGAGAAGCAG
NYC3	TGTCGTTGCCATGTGAAGAT	TTGGTCACGCCACAAATCTA
SGR	AGGGGTGGTACAACAAGCTG	GCTCCTTGCGGAAGATGTAG
RCCR1	CGCATTTCCTCATGGAATTT	CTTCTCACGCTGTTTGTCCA
Osh36	GCACGGAGGCGAACGA	TTGAGCGGTAGCACCCATT
Osl57	ACCCTAAAGTAAATGAAGTC	CCTGCTCTTGTCTTGTTA
Osh69	CCACAACACGGATAACTT	GGTGAACACTATGGAACA
Osl85	GAGCAACGGCGTGGAGA	GCGGCGGTAGAGGAGATG
OsNCED1	ACCATGAAGTCCATGAGGCT	TCTCGTAGTCTTGGTCTTGG
OsNCED2	ATGGAAACGAGGATAGTGGT	CTTATTGTTGTGCGAGAAGT
OsNCED3	CTCCCAAACCATCCAAACCG	TGAGCATATCCTGGCGTCGT
OsNCED4	ATCTCCTTCTCCCTCCCCA	TCGCACCCTGCTTGATCTTGC
OsNCED5	TCCGAGCTCCTCGTCGTGAA	AGGTGTTTTGGAATGAACCA
OsZEP	GGATGCCATTGAGTTTGGTT	TGGCTGACTGAAGTCTCTCG
OsABA8ox1	AAGCTGGCAAAACCAACATC	CCGTGCTAATACGGAATCCA
OsABA8ox2	CTACTGCTGATGGTGGCTGA	CCCATGGCCTTTGCTTTAT
OsABA8ox3	AGTACAGCCCATTCCCTGTG	ACGCCTAATCAAACCATTGC
Os01g01430	CAACGATGACGAACACAACC	GCAGCATATCTGCAGTGGAA
Os05g34310	GCCCATGGAAACCAAGACTA	CGAGCACTGTAACCGTGAGA
Os07g48450	GAGTCCTTGACGACGCCTAC	ACTCCGCGAGGAAGTTGTC
Os11g03300	TCAGCTAACAGCACCACCAC	ATGGGCTGGAAATCTGACTG
Os12g03040	CCTCACAGGCACATCAGCTA	ATGGGCTGGAAATCTGACTG
Primers used to genera	ate DNA constructs	
PS1-OE	CCCCCCGGGATGGTTCTGTCGAACCCGGC	TAGTCTAGATCAGTTCAGTCCCCATGTTAG
PS1-RNAi	GTCACTCGAGATGAAGTTCCGCAACACCTC	GTCAGGATCCTGGAAGGGATCTTCTGCATC
PS1-GFP	<u>ATCTCGAGG</u> ATGGTTCTGTCGAACCCGGC	ATCCCGGGGTTCATCCCCATGTTAGAGTG
PS1-GUS	TCGGTCGACGGCCTCAGTCCCTAAAAGGT	<u>CCGGAATTC</u> GGTTCGACAGAACCATCGTC

ZANG SANAS

## Table S6. Cont.

PNAS PNAS

	Forward primers	Reverse primers
PS1-BD-FULL	CGCTCATATGATGGTTCTGTCGAACCCGGCGA	GGTGGAATTCTCAGTTCATCCCCATGTTAGAG
PS1-BD-1/190	CGCTCATATGATGGTTCTGTCGAACCCGGCGA	GGTGGAATTCTCACTGCTCGTGGTCGGAGAGC
PS1-BD-181/392	CGCTCATATGGCCGTGCCGCCGCTCTCCGACC	GGTGGAATTCTCAGTTCATCCCCATGTTAGAG
PS1-BD-298/392	CGCTCATATGGCGAACAACAGCATGGTTCAG	GGTGGAATTCTCAGTTCATCCCCATGTTAGAG
PS1-BD-157/392	CGCTCATATGAACACCTCCATGAGGCTGGATG	GGTGGAATTCTCAGTTCATCCCCATGTTAGAG
PS1-BD-111/392	CGCTCATATGGTCGGCGTCAAGAAGGCGCTCG	GGTGGAATTCTCAGTTCATCCCCATGTTAGAG
PS1-BD-62/392	CGCTCATATGGACAGGGAGTGGTACTTCTTC	GGTGGAATTCTCAGTTCATCCCCATGTTAGAG
PS1-BD-25/392	CGCTCATATGCACTACCTCCGCAACCGGGC	<b>GGTGGAATTC</b> TCAGTTCATCCCCATGTTAGAG
PS1-BD-1/64	CGCTCATATGATGGTTCTGTCGAACCCGGCGA	GGTGGAATTCTCACTCCCTGTCCCCGTAATTCT
PS1-BD-1/100	CGCTCATATGATGGTTCTGTCGAACCCGGCGA	GGTGGAATTCTCAGTGGATGGGCTTGTCGGTG
PS1-BD-1/142	CGCTCATATGATGGTTCTGTCGAACCCGGCGA	<u>GGTGGAATTC</u> TCATGCGGCGGCGAGGCGGTAC
Primers used for ChIP-PCR		
SGR	CTCCCATTTTCGTCTTCCAA	GGCAGCTGACACACTCACAT
NYC1	TCGTCGTCATTATCCAGCAG	GCGTAGGAGGTGTGGTCATT
NYC3	CCCGATAAAATGGCTCTGAA	TTTGCGGAGAATTCCAGGTA
RCCR1	TCTGGCCTTTTGTGAGCTTT	TCGTTTTGCGTGAAACACTT
Osh36	CGTCTCACACTAGCCTGGTT	TGCAGCTGCTACGGTGACTA
Osh69	AGCGTCCGACTCAGATCCTA	CAACTAAGATCCACGCGTATG
OsI57	ACAAGTGGAAGACCCTGGTG	CAGGTAGGGCAGATTTCAGG
Osl85	CACGCATGCATGTTTCTTCT	GCTGCATCAAAGACCACAAG
Os02g07830	AACCGCTTGCAACTGTATATG	TCGGTGAGATGATTTGGTCA
Os02g37160	AGCGCAAGCAAGATAACCAT	ACGGCGGAGTTAAAAAGCAT
Os03g10640	GTGGCCTCGTTGTTTCTTTC	GTGGTTAGAGCACCGATTCC
Ubiquitin	TCGGAGACCGTGCTAGGTTT	GCCAGCGCCCATCGATT
Primers used for yeast one	-hybrid assays	
PS1-Y1H	GGTGGAATTCATGGTTCTGTCGAACCCGGCGA	GGTGCTCGAGTCAGTTCATCCCCATGTTAGAG
SGRp	TTGAATTCGAGCTCGGTACCGCCGTAACCGAATGTCAAAT	TCGACAGATCCCCGGGTACCGAGAGCGGGTTAAGTGAGAAGA
NYC1p	TTGAATTCGAGCTCGGTACCGCAATACGGGAAGGAAAAGA	TCGACAGATCCCCGGGTACCCGTCGCTTTGCTTTAGCTTT
NYC3p	TTGAATTCGAGCTCGGTACCGGGACACCCAGATCTAAGCA	TCGACAGATCCCCGGGTACCGTCTGCCGGACTAGCGTAAG
RCCR1p	TTGAATTCGAGCTCGGTACCAAGGCGAGAGCTGATGTGAT	TCGACAGATCCCCGGGTACCTTTGACACCGTTACGGCTAA
Osh36p	TTGAATTCGAGCTCGGTACCGGGACATGGTGTGTAGCTGA	TCGACAGATCCCCGGGTACCAGGGAGGAAGAAAGGGTGAA
Osh69p	TTGAATTCGAGCTCGGTACCGCCGATGTGGCATTATACCT	TCGACAGATCCCCGGGTACCAGTTAGCTACGCAGGGATGG
Osl57p	TTGAATTCGAGCTCGGTACCTCAACGTTGTTGTTTTGCAG	TCGACAGATCCCCGGGTACCCAGGTAGGGCAGATTTCAGG

The underlined nucleotides indicate the restriction sites for cloning.