

Supplemental Figure.

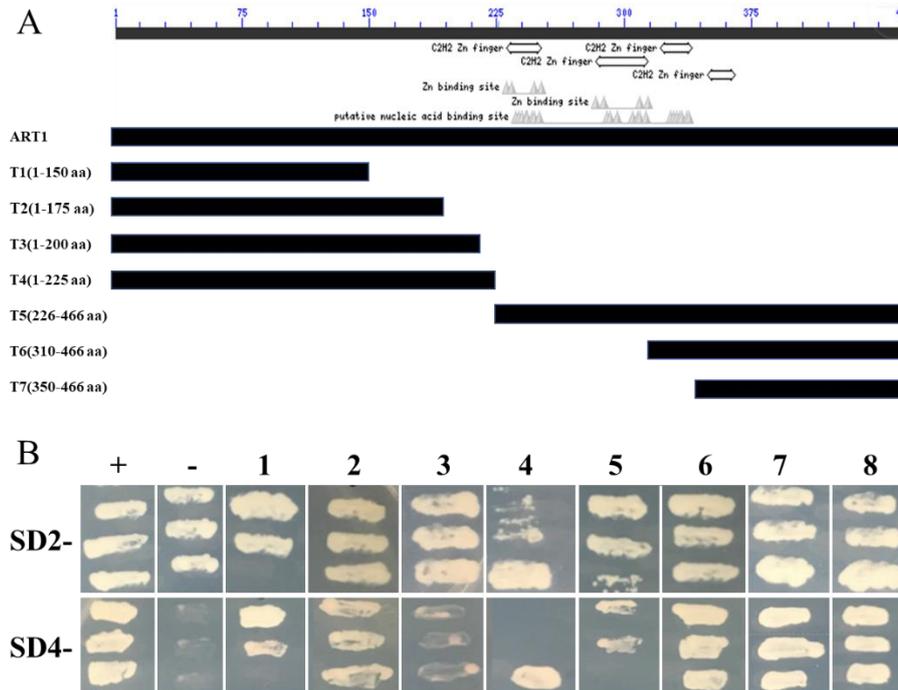
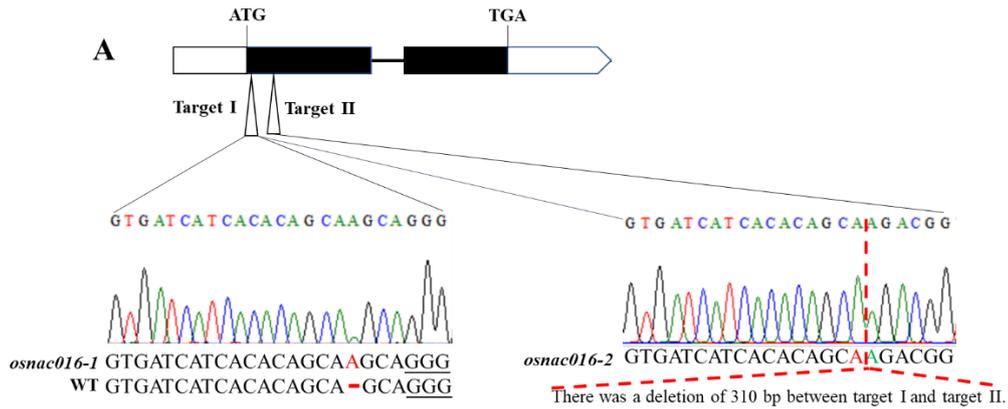


Figure S1. Transcriptional activation analysis of various truncated ART1s. (A) ART1 was truncated into different fragments, including T1 (1-150 aa), T2 (1-175 aa), T3 (1-200 aa), T4 (1-225 aa), T5 (226-466 aa), T6 (310-466 aa) and T7 (350-466 aa). (B) Identification of self-activation domain of ART. + : AD-T7 + BK-53, - : AD + BK, 1: ART1-BK + AD, 2: ART1-T1-BK + AD, 3: ART1-T2-BK + AD, 4: ART1-T3-BK + AD, 5: ART1-T4-BK + AD, 6: ART1-T5-BK + AD, 7: ART1-T6-BK + AD, 8: ART1-T7-BK + AD, the above pair of plasmids were co-transformed into yeast cells(AH109). The yeast colonies were grown on SD2- and SD4- medium plate. The plates were incubated at 30 °C for 3 d.



B

ATGATGACCATCGACCTGCAGCTTCCGGCGGGCGGTGCGGTGATCATCACACAGCAAGCAGGGGCAGGGCTGCCCGGGGATTCCGGTTCCACCCGACGGACGAGGAGTCTCTCCTCACTACCTCGGCAAGCGCGCCGCGGCGCCCTGCCCGCACCGGTGATCGCCAGGTGGACATTACAAGTACAACCCGTGGGAGCTGCCGGCCATGGCGGTGTTCCGGGAGTGGATGCCAGTGGTACTTCTCAGCCACCGCACCCGAAATCCCAACGGCGTCCGTCCAAACCGCGCCGGCTCCGGTACTGGAAAGGCCACCGAAACCGACAAGCCCATCTCCATCTCGGAGACGACGAGCGGTACTGTGGCGTGAAGAAGGCCCTCGTTCTACCGGGTCCCGCCCAAGGGCACAAGACCAGTGGATCATGACGAGTACCGCCTCGCAACGCCCGCTCTCTCTCTCTATACTAGTAACATGAAGCAGCTGGCCTCATCATCGTCCAGCTCAGCAGCGCCAGCATGAGGTGGACGAGTGGGTGTGTCAGAAATCTACAAGAAGAAGGAGGCCAACCCAGCAGCTGCAGCACTACATCGACATGATGATGACGACACAACGATGACGACAACAACCTGCAGGTGCAGCAGCAGCAGCAGCAGCAGGCTCAAAGCCACCGGATGCCAAGGCCCTCATCCATCTCAGACTACCTTCGACTACTCCGACGACCTCCCGCCGTCACCGATCAGACGCCATCTCTGACCTGGATTCACGGCGGTGAACGAGGGCAACAACAAGAGGCCAAAACTATGGAGGAATATTCTATTTCATTTCCTGAGATATGCTGATGCGTCTGTCACGTCACAACAACAAGTCGACGAAATCAACTTCTCTCCATATTTGAGCCGACAGCCTGCAGTCCGGGCCATCAGCTGATGTTCTCAACAACGACGACATCCATATGA.

C

ATGATGACCATCGACCTGCAGCTTCCGGCGGGCGGTGCGGTGATCATCACACAGCAGCAGGGGCAGGGCTGCCCGGGGATTCCGGTTCCACCCGACGGACGAGGAGTCTCTCTCCACTACCTCGGCAAGCGCGCCGCGGCGCCCTGCCCGCACCGGTGATCGCCAGGTGGACATTACAAGTACAACCCGTGGGAGCTGCCGGCCATGGCGGTGTTCCGGGAGTGGATGCCAGTGGTACTTCTCAGCCACCGCACCCGAAATCCCAACGGCGTCCGTCCAAACCGCGCCGGCTCCGGTACTGGAAAGGCCACCGAAACCGACAAGCCCATCTCCATCTCGGAGACGACGAGCGGTACTGTGGCGTGAAGAAGGCCCTCGTTCTACCGGGTCCCGCCCAAGGGCACAAGACCAGTGGATCATGACGAGTACCGCCTCGCAACGCCCGCTCTCTCTCTCTATACTAGTAACATGAAGCAGCTGGCCTCATCATCGTCCAGCTCAGCAGCGCCAGCATGAGGTGGACGAGTGGGTGTGTCAGAAATCTACAAGAAGAAGGAGGCCAACCCAGCAGCTGCAGCACTACATCGACATGATGATGACGACACAACGATGACGACAACAACCTGCAGGTGCAGCAGCAGCAGCAGCAGGCTCAAAGCCACCGGATGCCAAGGCCCTCATCCATCTCAGACTACCTTCGACTACTCCGACTACTCCGACGACCTCCCGCCGTCACCGATCAGACGCCATCTCTGACCTGGATTCACGGCGGTGAACGAGGGCAACAACAAGAGGCCAAAACTATGGAGGAATATTCTATTTCATTTCCTGAGATATGCTGATGCGTCTGTCACGTCACAACAACAAGTCGACGAAATCAACTTCTCTCCATATTTGAGCCGACAGCCTGCAGTCCGGGCCATCAGCTGATGTTCTCAACAACGACGACATCCATATGA.

Figure S2. Mutated sequence of two independent *OsNAC016*-knockout lines generated by CRISPR/Cas9 mutagenesis. (A) Black box indicates the exon. The *osnac016-1* mutant has a base A insertion at the position of target one. The *osnac016-2* mutant had a 307 bp deletion between target I and target II. (B) Open reading frame analysis of mutated *OsNAC016* in the *osnac016-1* line. (C) Open reading frame analysis of mutated *OsNAC016* in the *osnac016-2* line. a premature stop codon was produced at 502 bp in the *osnac016-1* line and at 386 bp in the *osnac016-2* line, respectively.

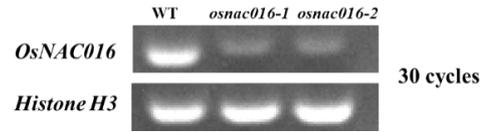


Figure S3. Transcript level of *OsNAC016* in roots of the wild-type and *osnac016* plants. The expression level of *OsNAC016* in the roots of 5-d-old rice seedlings were determined by semi-quantitative RT-PCR. *Histone H3* was used as an internal standard.

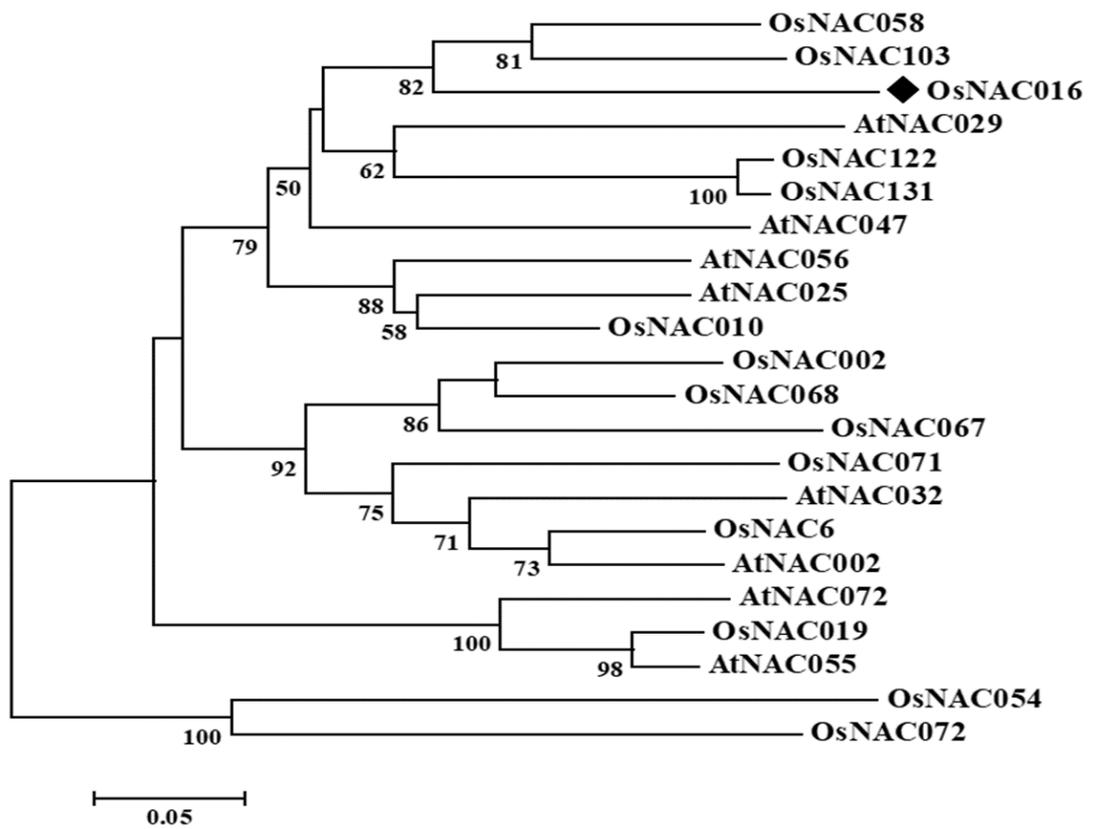


Figure S4. Phylogenetic tree analysis of *OsNAC016*. Homologs sharing with more than 40% identity to *OsNAC016* were used to construct the phylogenetic tree. Multiple sequence alignment was performed using the MEGA 6.0 and the phylogenetic tree was created.

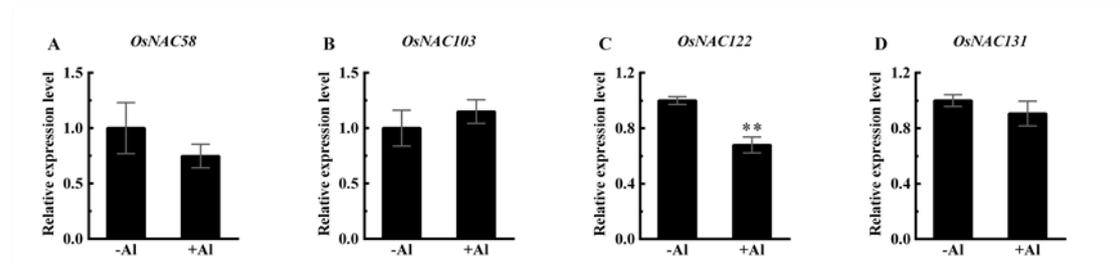


Figure S5. Expression analysis of four OsNAC016 homologous genes in roots. (A-D) Rice seedlings (5-d-old) were subjected to a solution containing 0 or 30 μM AlCl₃ for 8 h. Histone H3 was used as an internal standard. Data are means ± SD (n = 3). Asterisks indicates significant differences (P < 0.01, Tukey's test).

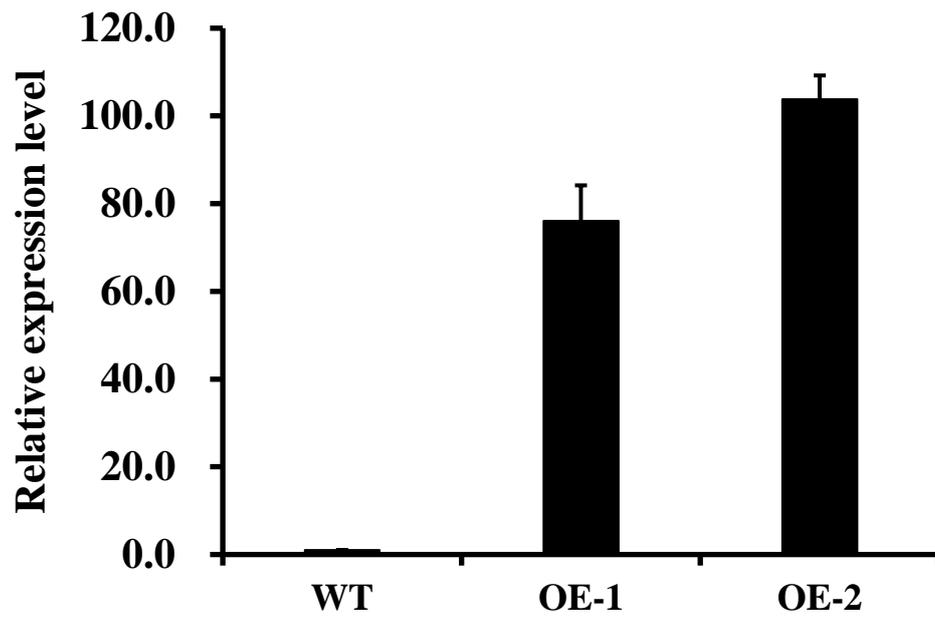


Figure S6. Relative expression levels of *OsNAC016* in the *OsNAC016*-overexpression lines (OE-1, OE-2) in roots. Data are means \pm SD of three biological replicates.

ATCTAATCATTAAATTGTCAAAAGACATTGAGCACATCATCCTCCCTCGTTGCTGCACATAACAATT
GAGATATTTTTATTTTCTTCAACTCTAAACTACAATTTTCTCATTTCGTTAGCACACTATACAAA
TTGTTAAATAGTATTATGTTTTTTTTTCAAAAACCTGTTTATATATAAAATTTCTTTAAATAAATTA
TCTTTAAAGTTTATTATAATTAATTATATGCTAATGTCTTATCTCACTTTATGTGTGCTTGAAAAG
CTAAACCAAATTGTGCTGATTTGAATTAAGCCCGAGTCACTCGCATATGAGTAGTCCTATTTGTT
ACGGTCTCACATGCGAATGATTTAGCTTCCTTATACTATACTCTTTTCGTAACAAAATATAATAA
CCTACTACTGGAACATAAAACCTGTTTAGATTCTTAATACTTGACGGAGCGTGGGGCTCCTCAC
CGGAGACCGCGCGGGCCCCTTTGCCGGTTCGGCCGGGGCGCTAAGTGAGGTTCTAAGCC
CTTGATCTGTGGAATGGTTTCGCGAGAGAGTAAATGCACAAGA CACGGGCGATGTAGACAGGTT
CGGGCCCTGAGAAGCGTAATACCCTACTCCTGTGTTCTGGTGGATCTGTGTATGAAGGAGTTA
CAAAGAGCTGGAGAGCAAGAGAGTTTGAAGTCTAGAGACCCTCTCCCTACTCTTTTTCCCAGTC
CCCTCTTTTTCTTTTCCCCCGTCCGTCAGTCCGTCAGTCCCCCTCTTGGTAGGCATGGTCTC
CTTTTATATCTCAAGGGGATACCACATACACCATTCTACCTACCCTTCTTTGGGTGGGACC
ATCCTACTTTGACCAAAACTTGGCTTGTACCTTCGTCGTGACACCGGGATAACCCTGTCTCCCTC
ATTAATGGGCGGAGATTTGCAATGGCTGCCGTCGAATGGCCCTCCGATGGGATGGGTCATACC
AACCTCCACTCCGCCGAAGCAAGTCAACCTGGAAGCACGGTGTGTTGCTGATGACGCGACC
GGCGTCAGACCAGTCACAGACCGGTATTCTTGTCCAC CACGTGTCAGTTTAGCATGCTA CACG
TCTGCCCTTCTTCATACAATGACTTCCTTGAATGGTTGCGATGAAGCCTGGAATGTATCTAGCC
GGGGTGCAGTGCCAACTCCAGGAGTCCC CACGCCGGCTCCGGCTAGGGACGAGTGCCTGGAG
GCTCTCATATTCCGACGGGACGGGGCGAGGCGTGTGTCA GGGCCCTGTTGCCCCCTAACCCG
CGATCTGACCGGTCTGTGACCGGTACACACTGCGACCGAT CACGGACTAGATAAGCATGTGCT
ACGCTGCATTAATGCGGCGTGAGACGCTCGTCCAACCCGCAATAAACGCGGTTAGGTGAGCG
CCGCTGTGCTCATCTAACCCACA CACGTGGCGCCAAACCCTCAGGGGGTCCGGGGCGCCCCAGC
CCTCGGGGACGAAACGGGAGCGGGCCGACCCCTCGGGGAGATAGAGGGAGGGGGCGGCCTCAT
CACCTC GGGCCC GACCCCCCGAGGGGGTCAAGC CACGTGGGTGACCGCGGCTGCCCAAAC
CTCTAGTCAAGATATCCCCGGTCCCATGTACCCGACAATACTAATAAGATAAATCTTATCCGAC
ACGGAAAGAGATCCTCTTACCTACTTGTGTGCAGTTGAATTACTGATATATAAACCAACA
AGTAGGATTAATTCATATATCAGTGACAACACTACACAGTTAGGTCAAAGGATCCGCTTCCATCCG
GCACTAGGTTGGGAGTATTACCATACAAGCTTCCCTTGCTGCCTGCTGCTGGCCACTCCTTGTGT
CCACTGTCCACAGATACAAACCTCCATTCGTAATTCGTACGGCCGTCGGGTTACATCTAGTGC
AGG CACGAA CACGTACAGACTCCCCTGATTTTTGGATCTATTTAAGCCGAGCTGAGTGAACCAA
TTGGCCACC

Figure S7: Cis-acting element analysis of *OsFRDL4* promoter.

NAC TFs of cis-acting element: CACG.

ART1 TF of cis-acting element: [GGN(T/g/a/C)V(C/A/g)S(C/G)].