

Supplemental Figure S1. Soil compaction inhibits cell elongation in maturation zone. (a) Representative propidium iodide staining of longitudinal sections of root maturation zones of 4-day-old WT seedlings grown in uncompacted or compacted soil. Bars = 100 μ m. (b) Cell length of the root maturation zone of the corresponding seedlings indicated in panel (a). Data are means \pm SD (n \geq 10 independent seedlings). Asterisks indicate significant differences compared with uncompacted at ***P* < 0.01 (Student's *t*-test).



Supplemental Figure S2. ABA inhibits root elongation and promotes root swelling. (a and b) Morphology (a) and primary root length (b) of 4-day-old wild-type (WT) seedlings with or without 0.5 μ M ABA treatment. Bars = 1 cm. Data are means \pm SD (n \geq 30 independent seedlings). (c) Representative propidium iodide staining of longitudinal sections of root tips of 4-day-old WT seedlings with or without 0.5 μ M ABA treatment. White arrows indicate the proximal end of the root meristem. White rectangle insets are an enlargement (3 times magnification) of the regions at the proximal end of the root meristem. Bars = 100 μ m. (d and e) Length (d) and cortical cell number (e) of the root meristem zones of the corresponding seedlings indicated in panel (c). Data are means \pm SD (n \geq 10 independent seedlings). (f) Representative propidium iodide staining of radial sections of root elongation zone of 4-day-old WT seedlings with or without 0.5 μ M ABA treatment. Bars = 100 μ m. (g) Root diameter of the corresponding seedlings indicated in panel (f). Data are means \pm SD (n \geq 10 independent seedlings). Asterisks in (b), (d), (e) and (g) indicate significant differences compared with mock at ***P* < 0.01 (Student's *t*-test).



Supplemental Figure S3. ABA inhibits cell elongation in maturation zone. (a) Representative propidium iodide staining of longitudinal sections of root maturation zones of 4-day-old WT seedlings with or without 0.5 μ M ABA treatment. Bars = 100 μ m. (b) Cell length of the root maturation zone of the corresponding seedlings indicated in panel (a). Data are means \pm SD (n \geq 10 independent seedlings). Asterisks indicate significant differences compared with mock at ***P* < 0.01 (Student's *t*-test).



Supplemental Figure S4. YUC8/REIN7-mediated auxin biosynthesis is required for ABAmodulated root elongation and root swelling. (a) Phenotypes of the primary roots of 4-day-old Kitaake (KT, a wild-type strain) and *rein7-1* seedlings with or without 0.5 μ M ABA treatment. Bar = 1 cm. (b) Primary root length of 4-day-old KT and rein7-1 seedlings treated with various concentrations of ABA. The data are shown as mean \pm SD, n \geq 30 independent seedlings. (c) Representative propidium iodide staining of longitudinal sections of root tips of 4-day-old KT and *rein7-1* seedlings with or without 0.5 μ M ABA treatment. White arrows indicate the proximal end of the root meristem. White rectangle insets are an enlargement (3 times magnification) of the regions at the proximal end of the root meristem. Bars = $100 \,\mu\text{m}$. (d and e) Length (d) and cortical cell number (e) of the root meristem zones of the corresponding seedlings indicated in panel (c). Data are means \pm SD (n > 10 independent seedlings). (f) Representative propidium iodide staining of radial sections of root elongation zone of 4-day-old KT and rein7-1 seedlings with or without 0.5 μ M ABA treatment. Bars = 100 μ m. (g) Root diameter of the corresponding seedlings indicated in panel (f). Data are means \pm SD (n \geq 10 independent seedlings). For (b), (d), (e) and (g), different letters indicate significant differences (P < 0.05, one-way ANOVA with Tukey's test).



Supplemental Figure S5. Soil compaction induces higher expression of auxin biosynthesis genes and auxin accumulation is weakened in *mhz5* mutant roots. (a) Expression of *YUCs*, *TAA1* and *TAR1* in the roots of 4-d-old Nip and *mhz5* seedlings grown in uncompacted or compacted soil. The data are shown as mean \pm SD; n = 3 biological replicates. (b) Indole-3-acetic acid (IAA) contents in 4-d-old Nip and *mhz5* seedlings grown in uncompacted or compacted soil. The data are shown as mean \pm SD; n = 3 biological replicates. Different letters indicate significant differences (*P* < 0.05, one-way ANOVA with Tukey's test).



Supplemental Figure S6. Identification of *OsbZIP46* mutants and overexpression lines. (a) DNA sequences of *OsbZIP46* in *osbzip46* allelic mutants (*osbzip46-1*, *osbzip46-2*, and *osbzip46-3*) generated by CRISPR/Cas9. (b) Amino acid sequences of *OsbZIP46* in *osbzip46* allelic mutants. (c) Expression analysis of *OsbZIP46* in overexpression lines. Values indicate \pm SD (n = 3). ** indicates significant differences by Student's *t*-test compared to Nip at *P* < 0.01.



Supplemental Figure S7. *OsbZIP46* positively regulates ABA response in roots. (a) Phenotypes of the primary roots of 4-day-old Nip, *OsbZIP46* knockout and overexpression seedlings with or without 0.5 μ M ABA treatment. Bar = 1 cm. (b and c) Primary root length (b) and relative root length (c) of 4-day-old Nip, *OsbZIP46* knockout and overexpression seedlings treated with various concentrations of ABA. The data are shown as mean \pm SD, n \geq 30 independent seedlings. Different letters indicate significant differences (*P* < 0.05, one-way ANOVA with Tukey's test).



Supplemental Figure S8. Overexpression of the constitutively active (CA) form of *OsbZIP46* enhanced ABA response in roots. (a) Phenotypes of the primary roots of 4-day-old Zhonghua 11 (ZH11, a wild-type strain) and *OsbZIP46-CA1* overexpression seedlings with or without 0.5 μ M ABA treatment. Bar = 1 cm. (b and c) Primary root length (b) and relative root length (c) of 4-day-old ZH11 and *OsbZIP46-CA1* overexpression seedlings treated with various concentrations of ABA. The data are shown as mean \pm SD, n \geq 30 independent seedlings. Different letters indicate significant differences (*P* < 0.05, one-way ANOVA with Tukey's test).



Supplemental Figure S9. OsbZIP46-mediated pathway is partially required for soil compaction-inhibited primary root growth. (a-d) Morphology (a and c) and primary root length (b and d) of 4-day-old Nip, *OsbZIP46* knockout and overexpression seedlings, Zhonghua 11 (ZH11, a wild-type strain) and *OsbZIP46-CA1* overexpression seedlings grown in uncompacted or compacted soil. Bars = 1 cm. Data are means \pm SD (n \geq 30 independent seedlings). Different letters indicate significant differences (*P* < 0.05, one-way ANOVA with Tukey's test).



Supplemental Figure S10. OsbZIP46 positively regulates the expression of auxin biosynthesis genes in roots. (a and b) Expression of auxin biosynthesis genes in the roots of 4-day-old Nip, *OsbZIP46* knockout and overexpression seedlings, Zhonghua 11 (ZH11, a wild-type strain), and *OsbZIP46-CA1* overexpression seedlings. The data are shown as mean \pm SD, n = 3 biological replicates.



Supplemental Figure S11. *YUC8/REIN7* acts downstream of *OsbZIP46* to regulate ABA response in roots. (a) Phenotypes of the primary roots of 4-day-old Nip, and combinations of *YUC8/REIN7* and *OsbZIP46* knockout and overexpression seedlings with or without 0.5 μ M ABA treatment. Bar = 1 cm. (b and c) Primary root length (b) and relative root length (c) of 4-day-old Nip, and combinations of *YUC8/REIN7* and *OsbZIP46* knockout and overexpression seedlings treated with various concentrations of ABA. The data are shown as mean \pm SD, n \geq 30 independent seedlings. Different letters indicate significant differences (*P* < 0.05, one-way ANOVA with Tukey's test).

Primer	Sequence (5'-3')	
RT-qPCR analysis		
OsActin1-F	GACCTTGCTGGGCGTGAT	
OsActin1-R	GTCATAGTCCAGGGCGATGT	
YUC1-F	TCATCGGACGCCCTCAACGTCGC	
YUC1-R	GGCAGAGCAAGATTATCAGTC	
YUC2-F	CAACTCTGGCATGGAGGTGT	
YUC2-R	GGATCGGCAACCATTTGAGC	
YUC3-F	GTGAGAACGGGCTCTACTCGGTCG	
YUC3-R	GCTTATGCATGACCGATGAACACG	
YUC4-F	CTCGACCTCTGCAACCACAA	
YUC4-R	GGAGTACCAGCCAATCCACC	
YUC5-F	GAGAAATACGGCCTCCGACG	
YUC5-R	CGACCCCATCCTCTGTGAAG	
YUC6-F	CCATTCCCAGATGGTTGGAAGG	
YUC6-R	CATGTTGCGCCTCAAGATATTTG	
YUC7-F	CAACGGATGGAAGGGTGAGT	
YUC7-R	CTTCTTGGTTGGCTTCGTCG	
YUC8-F	CCAACATCTCCTCGGTGTAG	
YUC8-R	GCATCAGACAAGCAACATCC	
YUC9-F	CACGATGTACCCGGTCGTG	
YUC9-R	TCTCCCCCTTCCAGTGATCC	
YUC10-F	TTACCGGAGCACCAAG	
YUC10-R	GTTGCTGATGTGGCCATTGTA	
YUC11-F	ATGCCCAAGAAGGACTTCCC	
YUC11-R	GAAGGCCTTGACGTCATTAGCA	
YUC12-F	CATGCACCGCATCGTTATGG	
YUC12-R	CCGTGCAGTGAGTCTTGCTA	
<i>YUC13-F</i>	CCACACATGGCGTATCCTGT	
YUC13-R	CAGCCCAACGGTTCTTACCT	
YUC14-F	TAGGTTCGAGGGAGAAGCCA	
YUC14-R	GTACCGGGCTACGAACAACT	
TAA1-F	CGCCCTACTACTCGTCATACC	
TAA1-R	GATTGTTCGGGGAGCAGAC	
TAR1-F	GCACCATACTACTCCTCGTACCC	
TAR1-R	GACGAGCTCGACGTAGGTGT	
OsbZIP46-F	AGCAGGTGGAAATGATACAG	
OsbZIP46-R	GGTCCAAGTTGCTGAGTGATTC	
To generate the OsbZIP46 overexpression vectors		
OsbZIP46-		
OE-F	ATGTCGACATGGAGTTGCCGGCGGATGG	
OsbZIP46-		
OE-R	ATGGTACCCCATGGACCAGTCAGTGTTC	

Supplemental Table S1. Primers used in this study.

CRIPSR/Cas9-mediated gene editing of OsbZIP46		
OsbZIP46-		
cas9-F	GGCACATCGACCCGAAATCCTTCT	
OsbZIP46-		
cas9-R	AAACAGAAGGATTTCGGGTCGATG	
For dual-luciferase assays		
YUC8p-LUC-		
F	ATGTCGACCGAGAGAGAGAGAGAGAGAAACA	
YUC8p-LUC-		
R	ATGGATCCCGATCGATCGATGGCCCAAG	
For ChIP-qPCR		
YUC8-P1F	CGAGAGAGAGAGGGAGAAACA	
YUC8-P1R	TTCGAATCCACTCCAAATCA	
YUC8-P2F	ATATAAAATTTAGTACCTCCTG	
YUC8-P2R	TGCATATATCCAGATTTAAGC	
YUC8-P3F	GCACTATTGAGAATCGATTCA	
YUC8-P3R	ACGAATTTAACTCTCAGGAATT	
For EMSA		
OsbZIP46-		
GST-F	ATGGATCCATGGAGTTGCCGGCGGATGG	
OsbZIP46-		
GST-R	ACGTCGACCCATGGACCAGTCAGTGTTC	
YUC8-Probe	GTTGGAAAGCGTATTTTATTCGCAACGTGCGGTTCTTCCAGTTTCTC	
	GGC	
YUC8-Mutant	GTTGGAAAGCGTATTTTATTCGCAAAAAACGGTTCTTCCAGTTTCTC	
Probe	GGC	