SUPPLEMENTAL DATA

SUPPLEMENTAL FIGURE LEGENDS

Fig. S1. Seed metal concentrations, leaf weight, and yield of *pez1* mutants

Zinc (A), manganese (B), iron (C) and copper (D) concentrations in the seeds of WT, *pez1-1* (*1-1*), and *pez1-2* (*1-2*) grown in soil. (E) Yield of WT, *pez1-1* (*1-1*), and *pez1-2* (*1-2*) grown in soil. Columns (means ± SD) with different letters are significantly different from each other according to a one-way ANOVA followed by a Student–Newman–Keuls test: $P < 0.05$; n = 3 each.

Fig. S2. T-DNA insertions of *pez1* mutants.

PEZ1 expression in the leaves (A) and roots (B) of WT, *pez1-1* (*1-1*), and *pez1-2* (*1-2*). (C) T-DNA insertions in *pez1*. Blue box: exon, green box: T-DNA, light-blue bars: primers for T-DNA check in Fig. S2, D, E. (D, E) T-DNA insertion check. *α-Tubulin* was used as a positive control. Columns (means ± SD) with different letters are significantly different from each other according to a one-way ANOVA followed by a Student–Newman–Keuls test: $P < 0.05$; n = 3 each.

Fig. S3. Subcellar localization of PEZ1 in onion epidermal cells and the growth of yeast cells expressing PEZ1.

Subcellar localization of PEZ1-GFP (A), and GFP (B). Scale bars, 20 μm. Growth of yeast cells expressing *PEZ1*. A Cd-sensitive mutant, *ycf1*, was transformed with the empty vector (V.C.) or with the plasmid containing *PEZ1*. Yeast cell suspensions were spotted on synthetic defined medium without (C) or with 10 μM Cd (D).

Fig. S4. Metal concentration of *pez1* mutant

Zinc (A), manganese (B), and copper (C) concentrations in the leaf of WT, and *pez1-2* (*1-2*) without or with 10 μM cadmium. Zinc (D), manganese (E), and copper (F) concentrations in the root of WT, and *pez1-2* (*1-2*) without or with 10 μM cadmium. Columns (means ± SD) with different letters are significantly different from each other according to a one-way ANOVA followed by a Student–Newman–Keuls test: $P < 0.05$; n = 3 each.

Fig. S5. Characterization of *PEZ1* over-expression lines.

PEZ1 expression in WT, over-expression (OX) 1, OX2 and OX3 plants in the leaf (A) and the root (B) under normal nutrient conditions. Leaf dry weight (C), root dry weight (D), Leaf Zn concentration (E), root Zn concentration (F), leaf Mn concentration (G), root Mn concentration (H), leaf Cu concentration (I), root Cu concentration (J) and SPAD value of WT, and

over-expression plants Columns (means ± SD) with different letters are significantly different from each other according to a one-way ANOVA followed by a Student–Newman–Keuls test: $P < 0.05$; n = 3 each.

Fig. S6. Iron accumulation in the roots of WT and OX1 on MS media containing 1.0 mM iron.

Fig. S7. Plant height of WT and over-expression lines (OX1, OX2 and OX3) (A). SPAD data of WT and over-expression (OX1, OX2 and OX3) plants (B). One week old plants were transferred to calcareous soil and data was recorded for ten days. Means ± SD. n=3 each

Fig. S8. Expression of PEZ1 in rice roots and leaves. Four week old plants were grown with or without 100 µM Fe for one week. Columns (means ± SD) with different letters are significantly different from each other according to a one-way ANOVA followed by a Student–Newman–Keuls test: $P < 0.05$; n = 3 each.

Fig. S9. The apparent binding assay of PCA to cadmium.
(A) Titration by added cadmium of the fluorescence of 1 µM FluoZin-1 in the presence of 200 mM HEPES–NaOH buffer (25°C, pH 7.4) with excitation at 495 nm and emission at 517 nm. After 2 µM, total cadmium was first added to 1 µM FluoZin-1; increasing concentrations of PCA (B) or glutathione (C) were then added in the same buffer described previously (28).

Figure S1

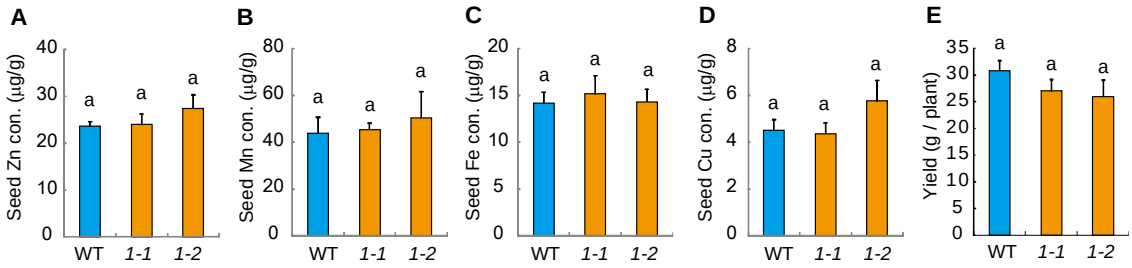

A
Seed Zn con. (μg/g)
40
30
20
10
0
a
a
a
WT
1-1
1-2
B
Seed Mn con. (μg/g)
80
60
40
20
0
a
a
a
WT
1-1
1-2
C
Seed Fe con. (μg/g)
20
15
10
5
0
a
a
a
WT
1-1
1-2
D
Seed Cu con. (μg/g)
8
6
4
2
0
a
a
a
WT
1-1
1-2
E
Yield (g / plant)
35
30
25
20
15
10
5
0
a
a
a
WT
1-1
1-2

Figure S2

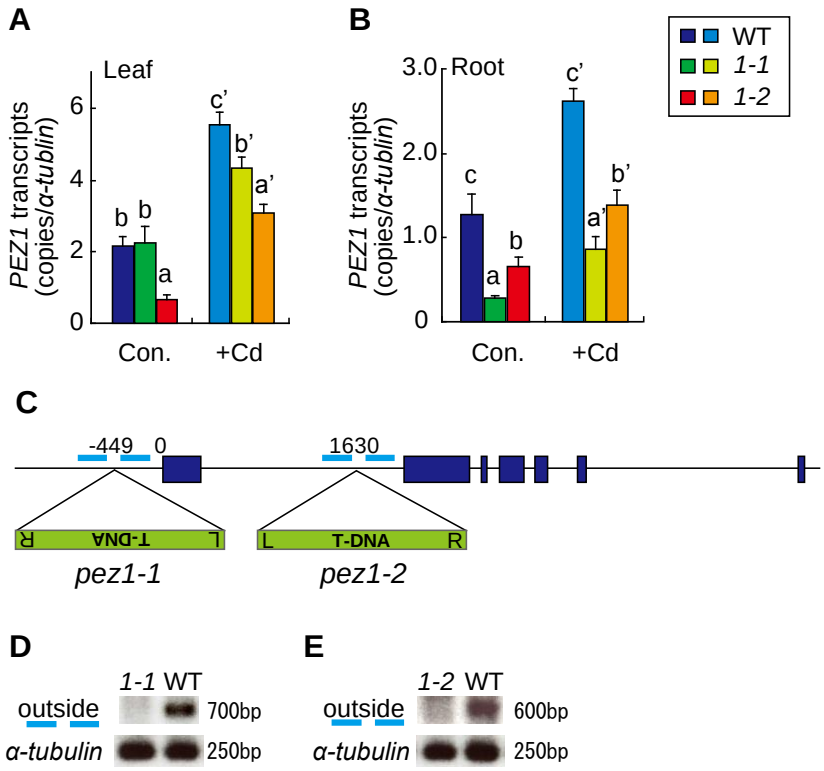

A
Leaf
PEZ1 transcripts (copies/α-tublin)
6
4
2
0
b
b
a
c'
b'
a'
Con.
+Cd
B
Root
3.0
2.0
1.0
0
c
a
b
c'
a'
b'
Con.
+Cd
WT
1-1
1-2
C
-449
0
1630
T-DNA
L
R
pez1-1
pez1-2
D
1-1 WT
outside
700bp
α-tubulin
250bp
E
1-2 WT
outside
600bp
α-tubulin
250bp

Figure S3

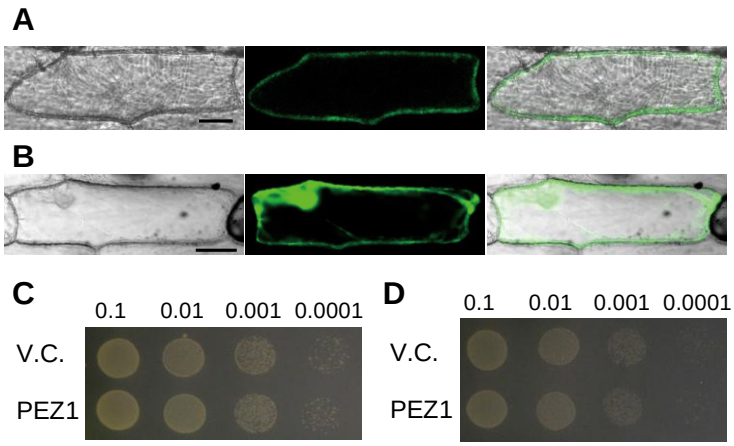


Figure S4

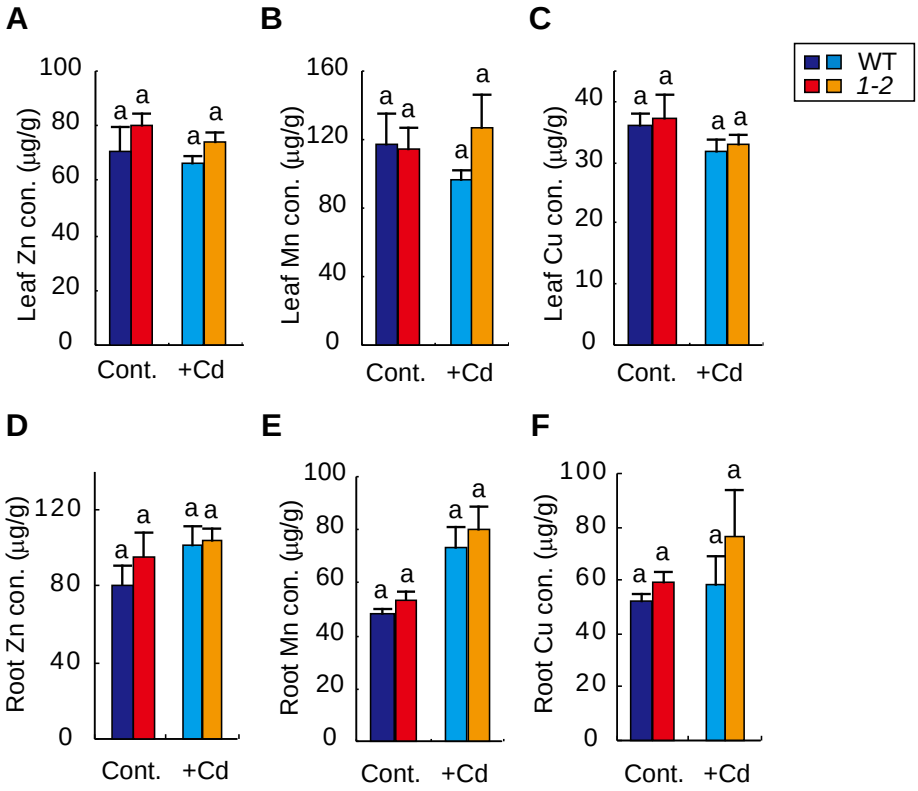

A
B
C
D
E
F
WT
1-2
Leaf Zn con. (μg/g)
Leaf Mn con. (μg/g)
Leaf Cu con. (μg/g)
Root Zn con. (μg/g)
Root Mn con. (μg/g)
Root Cu con. (μg/g)
Cont.
+Cd
a

Figure S5

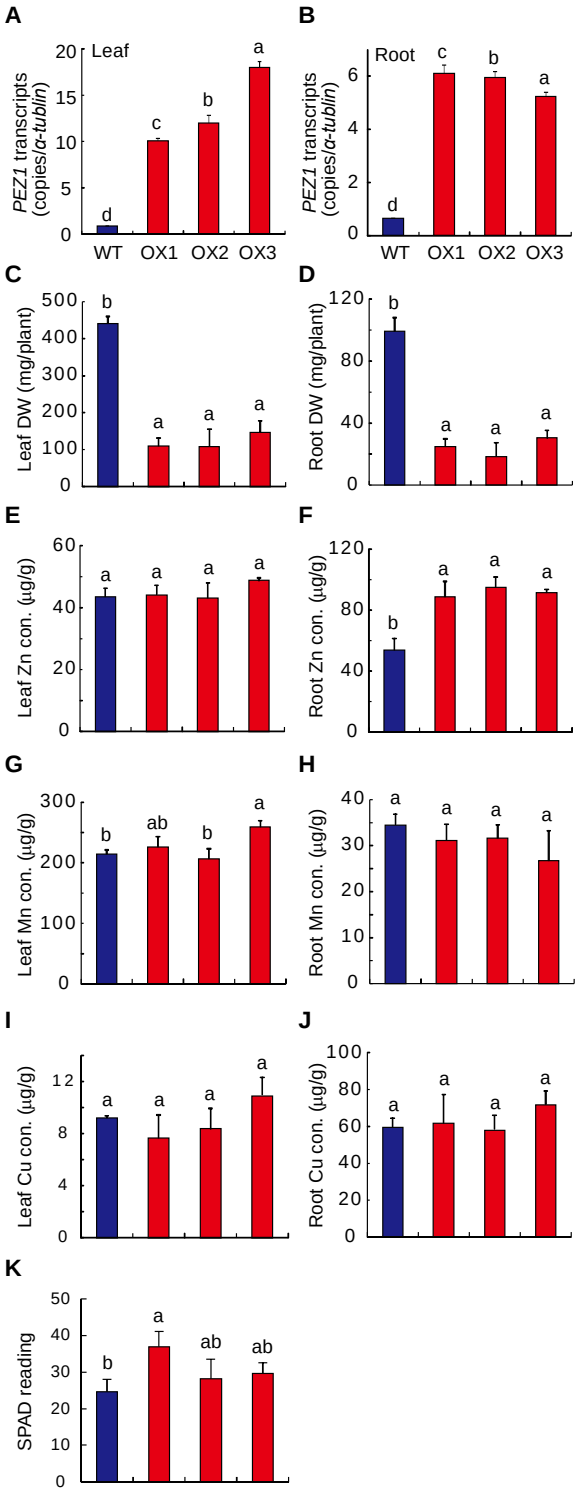

A
Leaf
PEZ1 transcripts (copies/α-tublin)
20
15
10
5
0
d
c
b
a
WT
OX1
OX2
OX3
B
Root
PEZ1 transcripts (copies/α-tublin)
6
4
2
0
d
c
b
a
WT
OX1
OX2
OX3
C
Leaf DW (mg/plant)
500
400
300
200
100
0
b
a
a
a
D
Root DW (mg/plant)
120
80
40
0
b
a
a
a
E
Leaf Zn con. (μg/g)
60
40
20
0
a
a
a
a
F
Root Zn con. (μg/g)
120
80
40
0
b
a
a
a
G
Leaf Mn con. (μg/g)
300
200
100
0
b
ab
b
a
H
Root Mn con. (μg/g)
40
30
20
10
0
a
a
a
a
I
Leaf Cu con. (μg/g)
12
8
4
0
a
a
a
a
J
Root Cu con. (μg/g)
100
80
60
40
20
0
a
a
a
a
K
SPAD reading
50
40
30
20
10
0
b
a
ab
ab

Figure S6

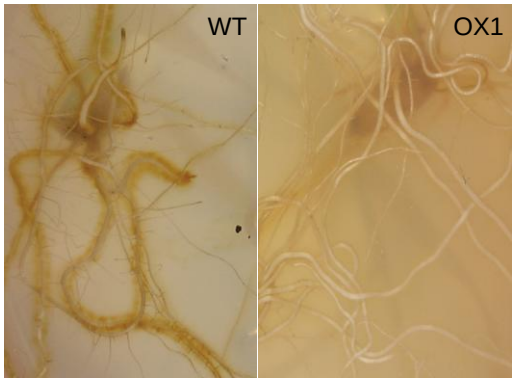


Figure S7

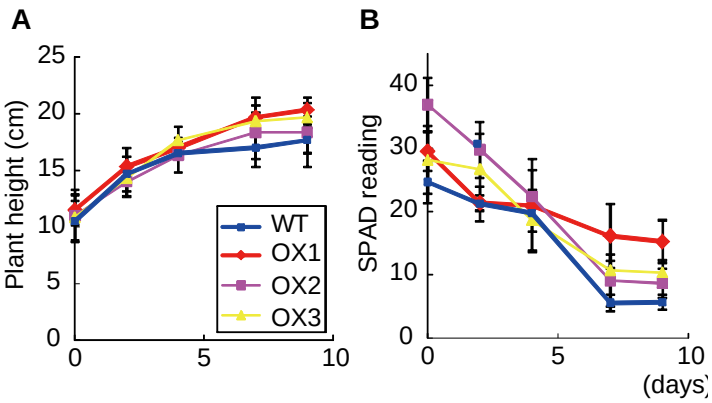


Figure S8

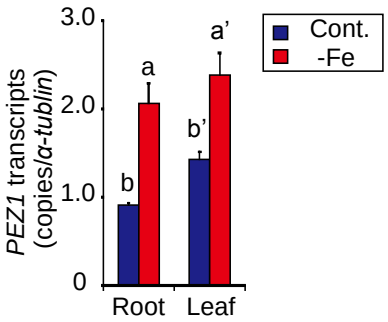
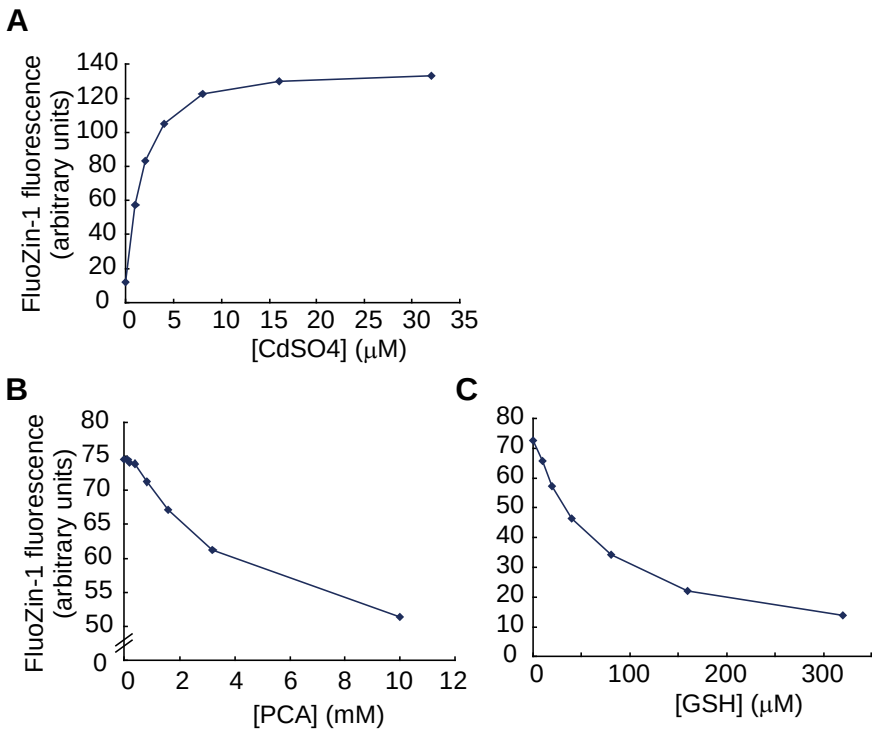

3.0
2.0
1.0
0
PEZ1 transcripts
(copies/α-tublin)
a
b
a'
b'
Cont.
-Fe
Root
Leaf

Figure S9


A
FluoZin-1 fluorescence (arbitrary units)
140
120
100
80
60
40
20
0
0 5 10 15 20 25 30 35
[CdSO4] (μM)
B
FluoZin-1 fluorescence (arbitrary units)
80
75
70
65
60
55
50
0
0 2 4 6 8 10 12
[PCA] (mM)
C
80
70
60
50
40
30
20
10
0
0 100 200 300
[GSH] (μM)