

Supplemental Figure S1. OXS2 homolog proteins in maize. A, Cladogram shows phylogeny of OXS2-like proteins in *Arabidopsis*, rice and maize. ZmOXS2b and ZmO2L1 indicated in blue lettering. ZmOXS2a not included in the current study indicated in red lettering. Protein information from NCBI. Tree generated with Mega5 (Tamura et al., 2011). B, Gene structures of *AtOXS2*, *ZmOXS2b* and *ZmO2L1* (predicted with SMART, <u>http://smart.embl-heidelberg.de</u>). ANKYRIN repeats indicated in blue; zinc finger domains in red, and polyglutamine stretches in green; inverted triangle: putative NES. There is no intron inside the protein coding regions. Numbers show positions on genomic DNA relative to translation start.



75 µM Cd

ZmOXS2b, ZmO2L1 or DEG23 (*CIMT1*) enhances cadmium tolerance. Plants grown on 1/2MS plates vertically without or with 75 μ M CdCl₂ for 11 (A, C) or 12 days (D), or on 1/2MS plates horizontally without or with 75 μ M CdCl₂ for 11 days (B). Three independent homozygous transgenic lines shown. Representative result from three independent experiments on T3 seedlings.



Supplemental Figure S4. Relative transcription of each DEG transgene compared to its normal expression in WT. 35S::DEG constructs of 8 candidates transformed into WT separately. Three independent T2 transgenic lines of each construct grown on 1/2MS plates for 11 days. Error bars show \pm SD of two independent experiments.

WT(DEG23)

Supplemental Figure S5. Cd accumulation in different genetic backgrounds. A. Cd content measured in plants grown in soil to 3 weeks of age and expose to 75 μ M CdCl₂ for 7 days before sampling. Error bars show \pm SD of three independent experiments. T5 WT(*ZmOXS2b*) and WT(*ZmO2L1*) plants and T3 WT(DEG23) plants were used (DEG23 = *CIMT1*). B. Cd content in shoot, roots, and seeds measured in plants grown in soil to 4 weeks of age and exposed to 5 μ M CdCl₂ until maturity. Error bars show \pm SD of three independent experiments on T4 WT(*ZmOXS2*) and WT(*ZmO2L1*) plants and T2 WT(DEG23) plants. No significant difference detected with Student's *t*-test.

WT(DEG23)

WT(ZmOXS2b)

WT(ZmO2L1)



Supplemental Figure S6. Subcellular localization of AtCIMT1, ZmOXS2b and ZmO2L1 fused to GFP and transiently expressed in *Arabidopsis* protoplasts. Representative fluorescence microscopy photos from 4 independent experiments of protoplasts treated without or with Cd (75 μ M CdCl₂ 2 hr).