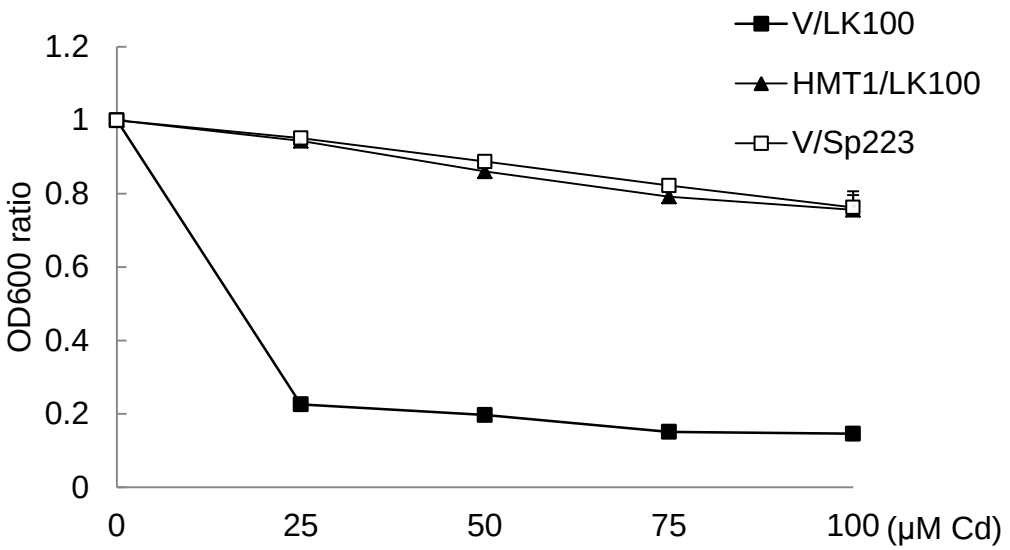
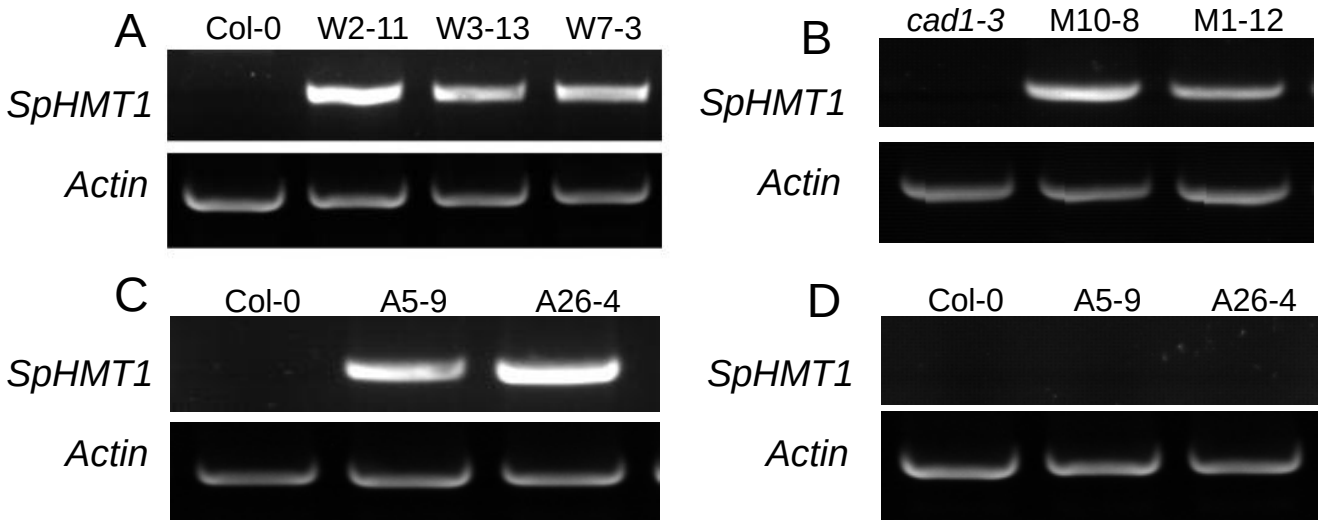
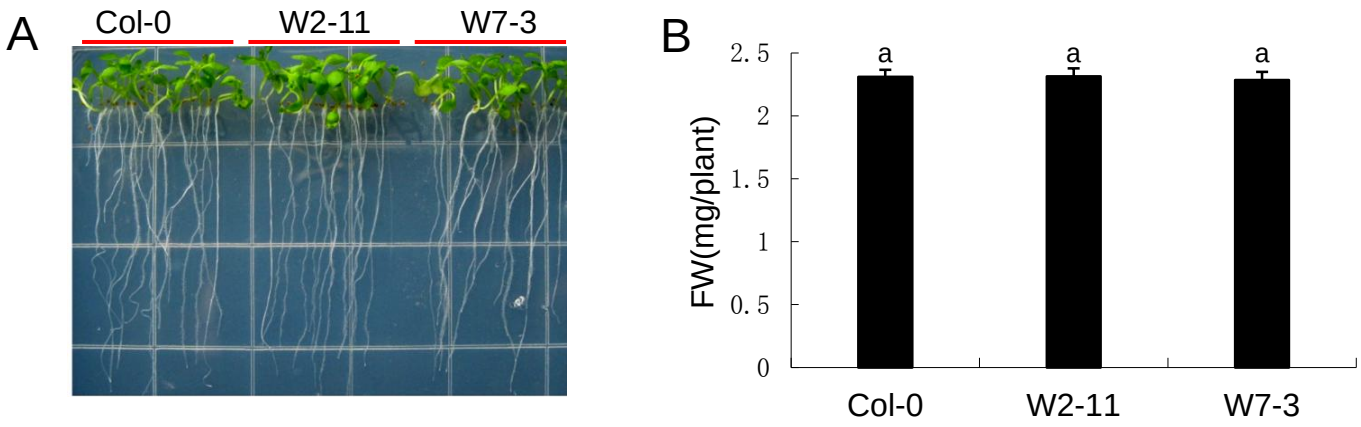


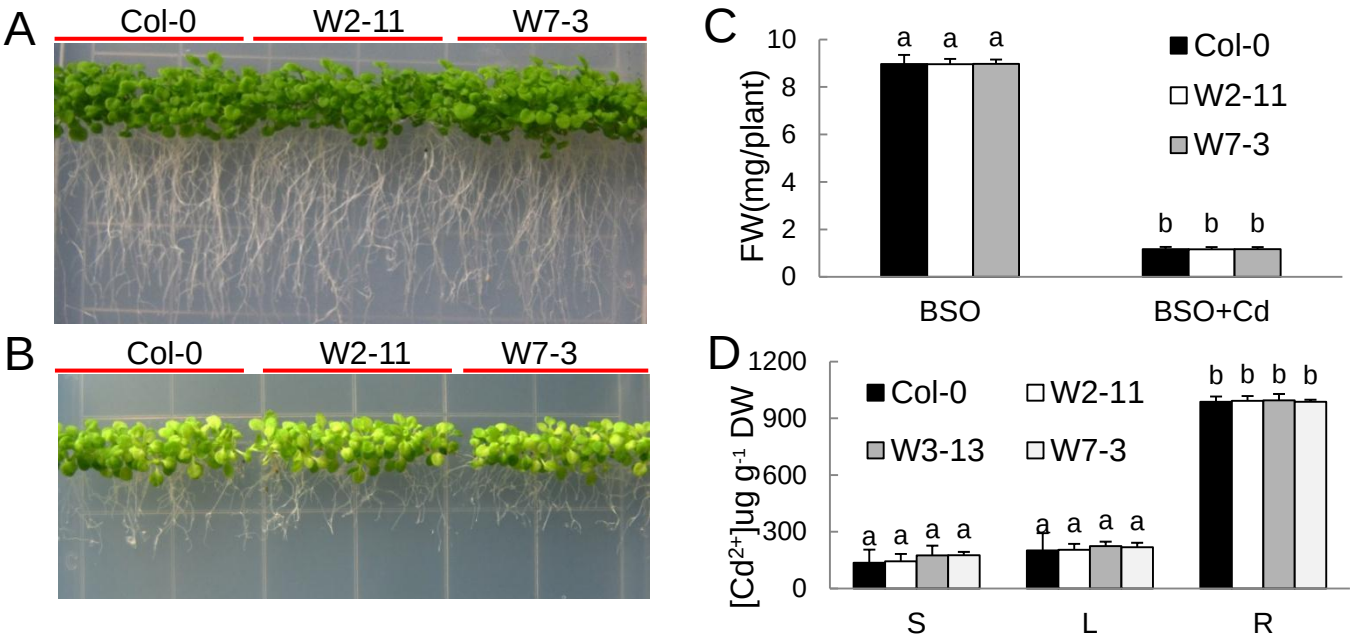
Supplemental Figure S1. Rescue of Cd^{2+} sensitivity in *hmt1*$^{-}$ mutant LK100 by c-myc tagged *SpHMT1*. Relative growth rate of yeast strains cultivated in selective EMM media supplemented with different levels of Cd^{2+}. Metal sensitivity assay described in Materials & Methods. Data are mean ± SD from 3 independent experiments.



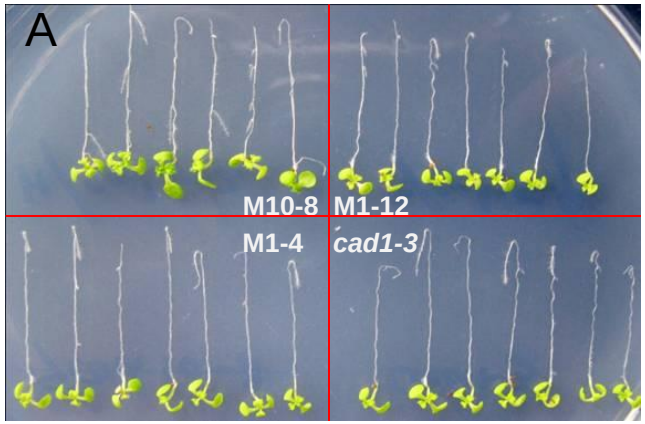
Supplemental Figure S2. RT-PCR analysis of *SpHMT1* expression. A and B, mRNA from 4-week-old plants used to determine the expression of *SpHMT1* driven by the *35S* promoter (W2-11, W3-13, W7-3, M10-8, M1-12). Root (C) and shoot (D) expression by the *Adh* promoter (A5-9, A26-4). Control plants are Col-0 and *cad1-3* as indicated. *SpHMT1*-specific primers used: 5'-TATCTTAAGCAAGAGCGGAAGG -3' and 5'-CTTTAAGCCTCTTTCTCCGACA-3'. *Actin* used as loading controls.



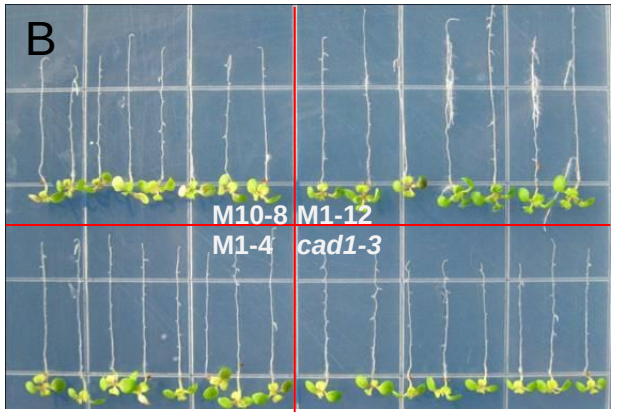
Supplemental Figure S3. Transgenic lines harboring *SpHMT1* showed similar growth with the wild type Col-0 under control condition. A, Wild type (Col-0) and transgenic lines (W2-11, W7-3) germinated on one-half MS plates, and grown vertically for 7 days. B, Fresh weight of plants grown under condition in (A). Values are mean±SD, n=12, each containing > 5 plants. Lowercase letters indicate whether averages were statistically different at $P < 0.05$ by *t* tests.



Supplemental Figure S4. Enhanced Cd^{2+} tolerance and accumulation aborted by BSO. Wild type Col-0 and transgenic lines W2-11 and W7-3 were germinated and grown vertically for about 20d on one-fourth MS plates containing 0.5 mM BSO plus 0 μM (A) or 10 μM $CdCl_2$ (B). C, Fresh weight determined on plants grown under conditions in (A) and (B). D, Cd^{2+} in stems (S), rosette leaves (L) and roots (R) determined on four-week-old hydroponically grown plants (Col-0 and transgenic lines W2-11, W7-3, W3-13) exposed to 10 μM $CdCl_2$ plus 0.25 mM BSO for 3d. Values are mean±SD, n=12 for C or n=4 for D , each containing > 5 plants. Lowercase letters indicate whether averages were statistically different at $P < 0.05$ by *t* tests.

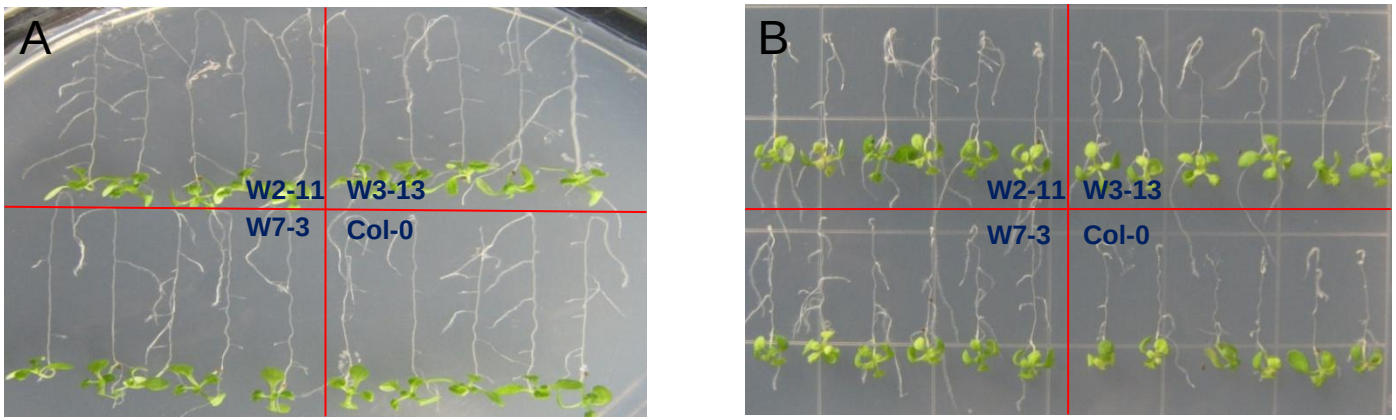


Cd^{2+} (20 μM)



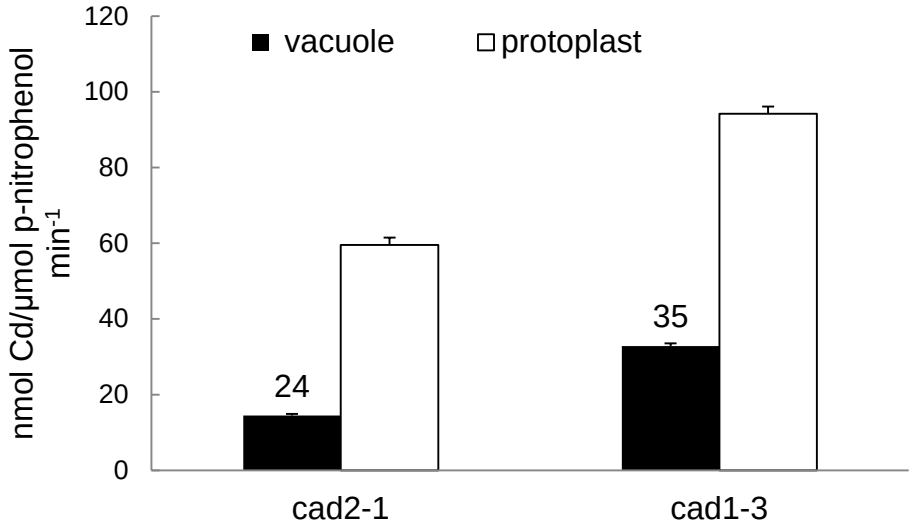
Cd^{2+} (30 μM)

Supplemental Figure S5. *SpHMT1* fails to rescue Cd^{2+} sensitivity in *cad1-3* mutant. 6-day-old seedlings transferred to plates supplemented with 20 μM (A) or 30 μM (B) $CdCl_2$, and grown further for 5d for root-bending assay . M10-8, M1-12 and M1-4 represented independent lines harboring *SpHMT1* in *cad1-3* background.

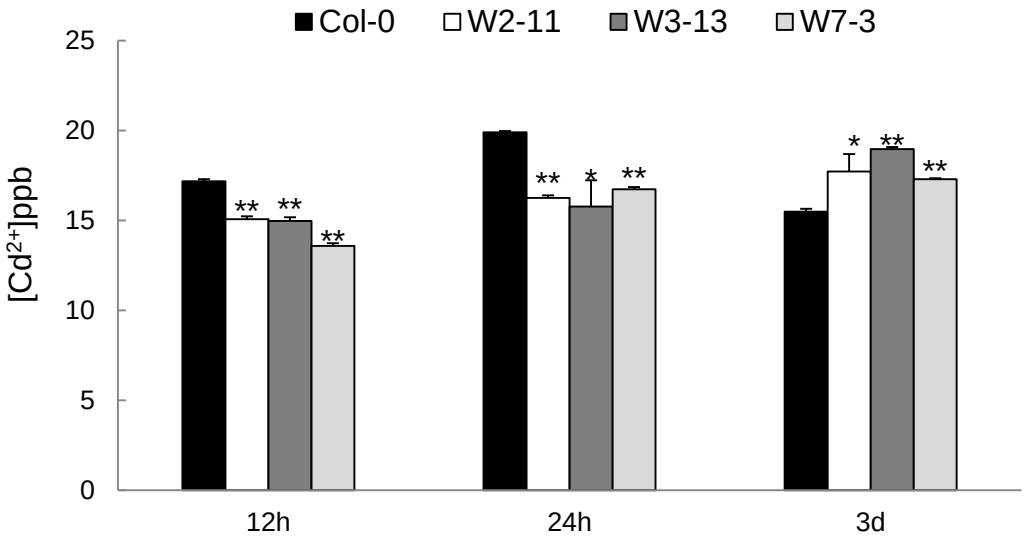


control Cd^{2+}(150 μM)

Supplemental Figure S6. *SpHMT1* enhanced cadmium tolerance in Col-0. 6-day-old seedlings were transferred to plates supplemented with 0 μM (A) or 150 μM (B) $CdCl_2$, and grown for another 5d for root-bending assay. W2-11, W3-13 and W7-3 represented independent lines harboring *SpHMT1* in Col-0 background.



Supplemental Figure S7. Subcellular localization of Cd^{2+}. Cd^{2+} contents in protoplasts and vacuoles isolated from leaves of soil grown *cad2-1* and *cad1-3.* Values above the bars represent the percentage of vacuolar Cd^{2+} relative to the total in the protoplasts. Acid phosphatase (ACP) activity specific to vacuoles was used to normalize Cd^{2+} accumulation. Data are mean±SE from 3 independent experiments.



Supplemental Figure S8. Cd^{2+} concentration in xylem sap. Plants were grown hydroponically for 4 weeks before treatment with 10 μM $CdCl_2$ for 12h, 24h and 3d. Cd^{2+} concentrations in xylem sap of wild type Col-0 and transgenic lines (W2-11, W7-3 and W3-13) were determined. Values are mean±SD, n=3, each containing > 8 plants. Asterisks indicate significant difference between wild type and the transgenic lines at $P < 0.05$ (*) and 0.01(**) by *t* tests.

Supplemental Table S1. Determination of vacuole purity. Levels of marker enzymes, chlorophyll (Chl), and proteins in protoplasts and vacuoles from Arabidopsis Leaves were analyzed to determine vacuole purity in addition to microscopic observation. Data represent mean ± SD, n = 3.

Marker	protoplast	vacuole	% in vacuole
acid phosphatase (nmol p-nitrophenol /min)	2.95±0.6	2.97±0.72	100.7
Cyt c oxidase (nmol cytochrome C /min)	113.92±6.83	ND[a]	0
Chl (μg)	210.35±26.64	ND[a]	0
Protein(μg)	2226.17±117.20	47.78±9.47	2.15

[a] Not detectable