**Table S1.** Sequences and reaction efficiencies of quantitative RT-PCR primer pairs.

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Gene** | **Primer** | **Sequence 5'=>3'** | **Reaction efficiency**  | **Standard deviation RE** |
| *At1g18050* | Fwd | CCATTCTACTTTTTGGCGGCT | 1.964 | 0.020 |
| Rev | TCAATGGTAACTGATCCACTCTGATG |
| *UBQ10* | Fwd | GGCCTTGTATAATCCCTGATGAATAAG | 1.936 | 0.028 |
| Rev | AAAGAGATAACAGGAACGGAAACATAGT |
| *AtEF1α* | Fwd | TGAGCACGCTCTTCTTGCTTTCA | 1.939 | 0.023 |
| Rev | GGTGGTGGCATCCATCTTGTTACA |
| *AtZIP9* | Fwd | CCATCACTACTCCGATCGGTGT | 1.869 | 0.018 |
| Rev | CACCAATGCTGCAACGCTATAA |
| *AtIRT3* | Fwd | AGTCATCCTCCTGGTCATGATT | 1.967 | 0.023 |
| Rev | GAGCATGACCAATGTCGAT |
| *AtIRT1* | Fwd | CCCCGCAAATGATGTTACCTT | 1.951 | 0.024 |
| Rev | GGTATCGCAAGAGCTGTGCAT |

RE: reaction efficiency

**Table S2.** Elemental profiling of the root and shoot tissues of wild-type (Col-0), *hma2hma4* mutant and *hma2hma4* expressing AtHMA4 or AtHMA4CCAA plants upon growth in control conditions (Ctrl, 1 µM Zn, no Cd) or exposed for 3 weeks to 0.05 µM Cd. Values (in µg/g DW) are mean±SD of 3 biological replicates each consisting of pools of 3 plants from 2 independent lines per genotype. The data were analyzed with one-way ANOVA followed by Tukey’s multiple comparison test. Statistically significant differences (P<0.05) between means within tissues and treatments are indicated by different letters. DW: dry weight; CCAA: di-Cys --> di-Ala motifs; n.d.: not detected.

| **Samples** | **Ca** | **Cd** | **Cu** | **Fe** | **K** | **Mg** | **Mn** | **Mo** | **Zn** |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **Ctrl - Shoot** |  |  |  |  |  |  |  |  |  |
| Col-0 | 41590 ±1856a | n.d. | 36±1.5a | 99±9.0a | 48807±3231a | 7011±178a | 181±16.3a | 9±6.2a | 104±4.8a |
| *hma2hma4* | 45193±1170b | n.d. | 41±1.0a | 98±5.5a | 45016±5449b | 7220±157b | 200±12.3a | 30±6.1b | 37±6.5c |
| AtHMA4 | 45932±2080b | n.d. | 27±0.7b | 77±4.6b | 49394±1303a | 7042±287a | 205±16.5a | 7±4.5a | 93±2.6a |
| AtHMA4CCAA | 47750±3246c | n.d. | 25±1.7b | 88±7.3a | 47674±2205a | 7041±395a | 219±22.0a | 3±3.0a | 56±2.5b |
|  |  |  |  |  |  |  |  |  |  |
| **Ctrl - Root** |  |  |  |  |  |  |  |  |  |
| Col-0 | 7964±602a | n.d. | 192±8.9a | 2041±145a | 32105±3721a | 2453±82a | 108±12.4a | 71±15a | 540±28a |
| *hma2hma4* | 7060±122a | n.d. | 251±3.1b | 1932±24.2a | 53266±6708b | 2590±77b | 75±6.7a | 175±2.6b | 782±5.7b |
| AtHMA4 | 6529±234a | n.d. | 229±15b | 1677±67a | 39177±2593a | 2418±37a | 167±19.9b | 85±9.6a | 495±23a |
| AtHMA4CCAA | 6986±764a | n.d. | 222±25b | 1833±186a | 48340±2555b | 2729±139c | 292±48.8c | 89±11.4a | 971±61c |
|  |  |  |  |  |  |  |  |  |  |
| **0.05 µM Cd - Shoot** |  |  |  |  |  |  |  |  |  |
| Col-0 | 46605±1639a | 23±1.9a | 35±2.1a | 99±9.4a | 48865±2057a | 7317±204a | 178±13.1a | 21±4.3a | 112±4.6a |
| *hma2hma4* | 54468±2999b | 6±0.1b | 45±0.2b | 79±0.6b | 48844±971a | 7848±315b | 206±14.1a | 11±5.9b | 53±3.0b |
| AtHMA4 | 55345±15176b | 27±7.8a | 43±10b | 99±8.7a | 51688±12559b | 8599±2360c | 215±63a | 31±8.7c | 114±27a |
| AtHMA4CCAA | 53038±3049b | 19±2.9c | 34±4.2a | 80±5.6b | 45200±2177c | 7651±456b | 226±15.7a | 27±2.9c | 65±7.2b |
|  |  |  |  |  |  |  |  |  |  |
| **0.05 µM Cd - Root** |  |  |  |  |  |  |  |  |  |
| Col-0 | 6428±155a | 32±1.1a | 211±7.3a | 1827±111a | 48655±2553a | 2213±65a | 110±4.1a | 129±14a | 640±32a |
| *hma2hma4* | 9037±333b | 204±7.0b | 256±34a | 3129±270b | 27152±9779b | 2605±263a | 50±8.3b | 139±11a | 1229±34b |
| AtHMA4 | 8369±397b | 47±3.4a | 243±6.5a | 2707±309b | 47657±4467a | 2589±187a | 110±6.7a | 109±23b | 830±58a |
| AtHMA4CCAA | 7451±383b | 148±11b | 204±13.5a | 2681±196b | 39013±2558a | 2700±80a | 172±40.2c | 126±6.2a | 1548±69c |



**Figure S1.** Zinc and cadmium accumulation in shoot (A, C) and root (B, D) tissues of wild-type (Col-0), *hma2hma4* mutant and *hma2hma4* expressing AtHMA4 or AtHMA4CCAA plants upon hydroponic growth exposed for 3 weeks to 1 µM Cd. Values (in µg/g DW) are means ± SD of 3 biological replicates each consisting of pools of 3 plants from 2 independent lines per genotype. The data were analyzed with one-way ANOVA followed by Tukey’s multiple comparison test. Statistically significant differences (P<0.05) between means are indicated by different letters. DW: dry weight; CCAA: di-Cys --> di-Ala motifs.