Supplimentary figures

Mechanisms Underlying the Enhanced Biomass and Abiotic Stress Tolerance Phenotypes of an Arabidopsis MIOX Over-expresser

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Supplementary Figure 2. A) UV-spectra of glucoerucin at 229nm B) Chromatogram of glucoerucin detected at 229nm.

| -2.5 -2.0 -1.5 -1.0 -0.5 0.0 0.5 1.0 1.5 2.0 2.5 | | | | | | | | | | | | | | | | | | | | | | | | | | |
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| Down-regulated Up-regulated | | | | | | | | ~ | | | | | | | | | | | | | | | | | | |
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| | G16 G21 G51 | G28 G67 G67 | G666 G75 | G41 G26 G26 | C20 | C 47 | C26 | C14 | C 76 C 76 C 48 | G10 C28 | G24 G09 G55 | G14 | | | 8688 | C07 | C23 | G17 G50 G50 G50 | C07 C05 C05 | G24 G15 | 015 | C13 C13 C13 C13 | G74 G38 G24 | G52 G53 G52 G53 | C03 C03 C33 C33 | C03 C17 C17 |
| | AT4 AT5 AT3 | ATA ITA ITA ATI | LTA LTA | AT5 AT5 AT4 | 4T2 4T4 4T4 | AT2 AT2 AT3 | TA STA STA | AT5 AT5 AT2 | LTA LTA | TTP LTTP | AT5 AT3 AT3 | ATA AT4 | AT2 ATC AT4 AT4 | ETA ETA | 414 412 412 | AT2 | AT1 AT4 AT4 | AT4 AT2 AT5 AT5 | AT5 AT3 AT5 | ATC AT4 AT2 | AT3 AT4 AT5 | AT5 AT5 AT5 AT2 | LTA LTA LTA | AT5 AT5 AT5 AT1 | ATA AT4 AT4 | AT3 AT3 AT3 AT3 |
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| Arabidopsis thaliana (Top 50 most similar Perturbations) | 24 249 252 | 255 260 260 | 256 249 262 | 249 248 254 | 252 | 266 | 266 | 25(25(265 | 255 | 24 26 26 | 24 258 25 | 26 ² 253 | 266 242 252 254 | 249 | 245 | 25: 266 | 252 | 249 266 248 248 | 25(25) | 244 254 265 | 259 | 249 25(25(26(| 26/ 26/ | 25 24 24 26(| 265 255 255 | 255 263 263 263 263 |
| myb21-5 myb24-5 / Col 🚯 | | | | | | | | | | | | | | | | | | | | | | | | | | |
| CS57674 / Bay-0 parent | | | | | | | | | | | | | | | | | | | | | | | | | | |
| flu1-1 / Col-0 🕦 | | | | | | | | | | | | | | | | | | | | | | | | | | |
| salicylic acid study 13 (CS57666) / mock treated rosette leaf samples (CS57666) | | | | | | | | | | | | | | | | | | | | | | | | | | |
| osmotic study 4 (shoot) / mock treated Col-0 shoot samples | | | | | | | | | | | | | | | | | | | | | | | | | | |
| ARR22ox / Col-0 🕕 | | | | | | | | | | | | | | | | | | | | | | | | | | |
| drought study 13 (Tamm-2) / mock treated Tamm-2 rosette leaf samples | | | | | | | | | | | | | | | | | | | | | | | | | | |
| wounding (late) / untreated green tissue samples (late) | | | | | | | | | | | | | | | | | | | | | | | | | | |
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| SOS1 OX / Col(all) | | | | | | | | +++ | | | | | | | | | | | | | | | ┝┼╾┼╴┤ | | | |
| nad4-5 / Ws-0 6 | | | | ╞╼┼╼┼ | | | ┼┼┼┼ | | | | | | | | | ╉╋╋ | | | | | | | ┼┼┼┼┤ | | | |
| HSP90(RNAI-C1) / Col-0 | | | | | | | | | | | | | | | | | | | | | | | | | | |
| atcesA6 / atcesA6 het. | | | | | | | | | | | | | | | | | | | | | | | | | | |
| drought study 16 (Col) / mock treated Col whole plant samples | | | | | | | | | | | | | | | | | | | | | | | | | | |
| bhlh100 bhlh101 / Col-0 🖲 | | | | | | | | | | | | | | | | | | | | | | | | | | |
| 35S::amiRAP2.2-12 / Col-0 📵 | | | | | | | | | | | | | | | | | | | | | | | | | | |
| low R/FR + MeJA (2h) / low R/FR (2h) | | | | | | | | | | | | | | | | | | | | | | | | | | |
| osmotic study 4 (root) / mock treated Col-0 root samples | | | | | | | | | | | | | | | | | | | | | | | | | | |
| JA (Sha) / mock treated rosette leaf samples (Sha) | | | +++- | | | | | | | | | | | | | | | | | | | | | | | |
| JA (Bay-0) / mock treated rosette leaf samples (Bay-0) | | | +++ | | | | | | | | | | | | | | | | | | | | | | | |
| P. syringae pv. tomato study 4 (DC3000 avrRpm1) / P. syringae pv. tomato stu | | | | | | | +++ | | | | | | | | | ++++ | | | | | | | | | | |
| Tamm-2 / Omo2-5 V | | | | | | | | | | | | | | | | | | | | | | | | | | |
| ARA study 4 (Col=0) / mock treated leaf samples (Col=0) | | | +++- | | | | | +++ | | | | | | | ╉╋╋ | | | | | | | | | | | |
| anoxia study 2 / dark grown Col-0 seedling samples | | | | | | | | | | | | | | | | | | | | | | | | | | |
| elevated CO2 study 3 (mature leaf 10) / elevated CO2 study 3 (expanding leaf | | | | | | | | +++ | | | | | | | | | | | | | | | | | | |
| atcesA4 / atcesA4 het. | | | | | | | | | | | | | | | | | | | | | | | | | | |
| ABA study 8 (agb1-2) / solvent treated leaf samples (agb1-2) | | | | | | | | | | | | | | | | | | | | | | | | | | |
| drl1-2 / Ler-0 | | | | | | | | | | | | | | | | | | | | | | | | | | |
| salt study 4 (Ws) / Hoagland solution watered Ws leaf samples | | | | | | | | | | | | | | | | | | | | | | | | | | |
| MeJa study 2 (1h) / mock treated seedlings (1h) | | | | | | | | | | | | | | | | | | | | | | | | | | |
| B. cinerea / non-infected rosette leaf samples | | | | | | | | | | | | | | | | | | | | | | | | | | |
| ABA Study 8 (agb1-2 gpa1-4) / solvent treated lear samples (agb1-2 gpa1-4) | | | | | | | | | | | | | | | | | | | | | | | | | | |
| R solani (AC2-1) / mock inoculated whole plant samples | | | +++ | | | | | +++ | | | | | | | | | | | | | | | | | | |
| drought study 3 (late) / untreated green tissue samples (late) | | | | | | | | | | | | | | | | | | | | | | | | | | |
| elo2 / Ler-0 | | | | | | | | | | | | | | | | | | | | | | | | | | |
| high light study 3 (3h) / untreated leaf samples (CAT2HP1) | | | | | | | | | | | | | | | | | | | | | | | | | | |
| myb21-5 myb24-5 / Col 🖲 | | | | | | | | | | | | | | | | | | | | | | | | | | |
| high light (Col-0) / low light grown seedlings (Col-0) | | | | | | | | | | | | | | | | | | | | | | | | | | |
| lcr-3P77 / Col-0 | | | | | | | | | | | | | | | | | | | | | | | | | | |
| elo3 / Ler-0 | | | | | | | | | | | | | | | | | | | | | | | | | | |
| CS57666 / Bay-U parent | | | | | | | | | | | | | | | | | | | | | | | | | | |
| IUII-394031 / COI-U ELC22 study 9 (0b) / H2O protroated loaf disc camples (Ler) | | | | | | | | | | | | | | | | | | | | | | | | | | |
| Mela study 2 (3h) / mork treated seedlings (2h) | | | | | | | | | | | | | | | | | | | | | | | | | | |
| ARA study 8 (ma1-4) / solvent treated leaf samples (ma1-4) | | | | | | | | | | | | | | | | | | | | | | | | | | |
| osmotic study 2 (late) / untreated root samples (late) | | | | | | | | | | | | | | | | | | | | | | | | | | |
| OPDA study 2 (tga2-5-6) / solvent treated (tga2-5-6) seedlings | | | | | | | | | | | | | | | | | | | | | | | | | | |
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Supplementary Figure 3. GeneVestigator analysis of top 200 up-regulated transcripts (\log_2 fold > 1.75) were compared with signature expression transcripts from 50 abiotic and biotic stress perturbations. *ATCAD8*, *GLYI7* and *NAC019* were also highly expressed in most of the abiotic stress (heat, salt, drought cold) tolerance experiments.



Supplementary Figure 4. Down-regulation of cell wall metabolism transcripts. Validation of down-regulation of plant invertase/ pectinmethylestarase superfamily protein and Laccase 2 in *At*MIOX4 OE line using RT-qPCR. * indicated p<0.05 at 0.05 significance level. Data was mean ± SEM (n=3).





Supplementary Figure 5. ATTEDII analysis of up-regulated transcripts. A) Mapping of up-regulated transcripts in plant hormone signaling pathway, B) Mapping of up-regulated transcripts in glucosinolates biosynthesis pathway.

В

O 3-Methylthio-

propyl-glucosinolate

 4-Methylthiobutyl-glucosinolate

-O 5-Methylthio-

6-Methylthio

O 7-Methylthio

 S-Methylthiooctyl-glucosinolate

heptyl-glucosinolate

hexyl-glucosinolate

pentyl-glucosinolate



Supplementary Figure 6. Pageman analysis of differentially expressed transcripts. The transcripts associated with light reactions of photosynthesis, lipid metabolism, secondary metabolism and hormone metabolism are up-regulated whereas transcripts related to cell wall metabolism are down-regulated in *At*MIOX4 transcriptome. The functional enrichment analysis was performed using binwise wilcoxon test, no multiple corrections, and over representation value 3.



Supplementary Figure 7. Schematic representation of RNA-Seq data analysis. Raw RNA Sequence data was analyzed by using TopHat, Cufflink, Cuffmerge, Cuffdiff and CummeRbund algorhitms. Functional annotation and metabolic pathways of transcriptomics datasets were analyzed by BLAST2gOv3.1 and Mapman. RT-qPCR validation of hallmark genes expression, quantification of metabolites by LC-MS/MS and completion of data analysis.