

## **THE PROMOTER OF *VACUOLAR METAL TRANSPORTER* IN *ARABIDOPSIS HALLERI*: AN EXAMPLE OF EVOLUTION FOR METAL HYPERTOLERANCE/HYPERACCUMULATION**

FASANI E., DALCORSO G., FURINI A.

Department of Biotechnology, University of Verona

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In the metal hyperaccumulator *Arabidopsis halleri*, the metal vacuolar transporter (VMT) is involved in hyperaccumulation and hypertolerance. Three *VMT* promoter sequences have been identified in the genome of *A. halleri*; expression analysis, conducted by GUS assay, showed that each promoter sequence of *A. halleri* induced higher expression than the promoter sequence of the *VMT* orthologous gene in *A. thaliana*. Moreover, it was observed that *A. halleri* *VMT* members are expressed in leaf trichomes, whereas *VMT* of *A. thaliana* is not present in these organs. *In silico* analysis revealed that the 5' UTR region of *VMT* is highly conserved between the two *Arabidopsis* species, although a dimer of MYB-binding motifs harboured by the *A. halleri* promoters is mutated in a single nucleotide in the sequence of *A. thaliana*. Site-specific mutagenesis of these motifs in the promoter of *A. halleri* indicates that they are likely involved in trichome-specific expression. The role of these MYB-binding motifs in the *A. halleri* *VMT* promoter was tested in *A. thaliana*, considering their effect in both metal tolerance and accumulation. Transgenic *A. thaliana* plants expressing *VMT* under the control of the native *A. halleri* *VMT* promoter are more tolerant, in terms of root length, biomass production and chlorophyll content, to high metal concentrations, than the ones bearing the same construct with the mutated MYB-binding motifs. Differences in shoots and in roots were also observed regarding metal accumulation. These results underline the divergent evolution of the *VMT* promoter, conferring higher metal tolerance, accompanied by expression in trichomes, in the hyperaccumulator *A. halleri* and protection to heavy metal stress in transgenic *A. thaliana*. Furthermore, a possible biotechnological approach might be applied in phytoremediation or biofortification programs to modulate metal content in plants.