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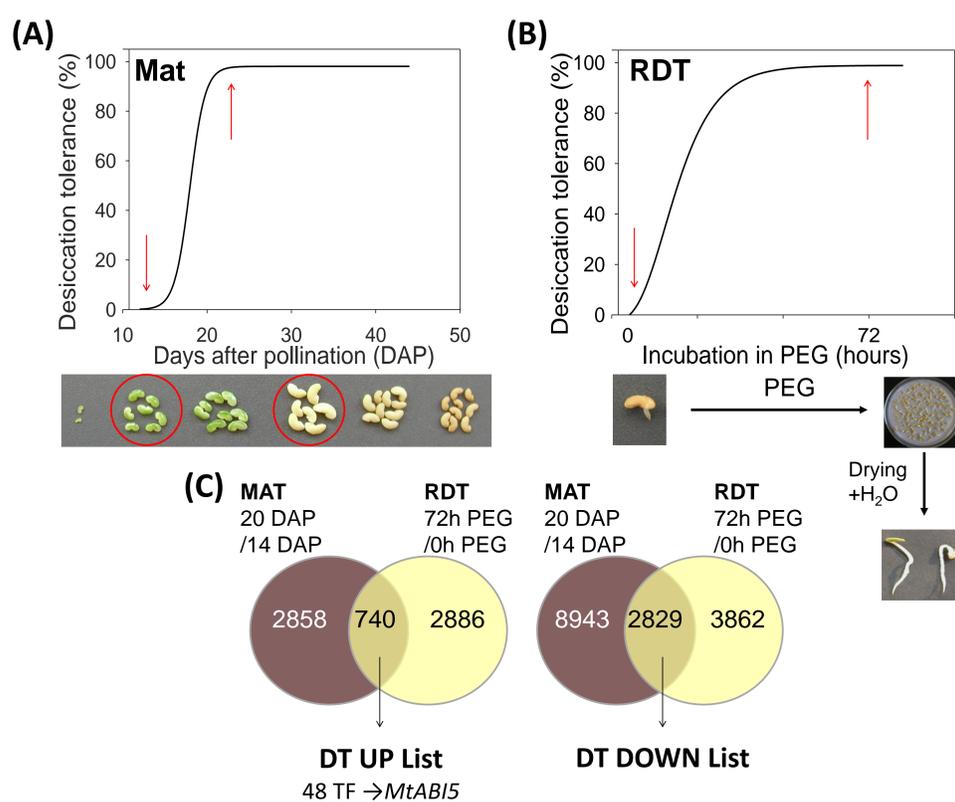
# Involvement of *MtABI5* in the acquisition of desiccation tolerance in *Medicago truncatula* seeds and seedlings: identification of potential targets



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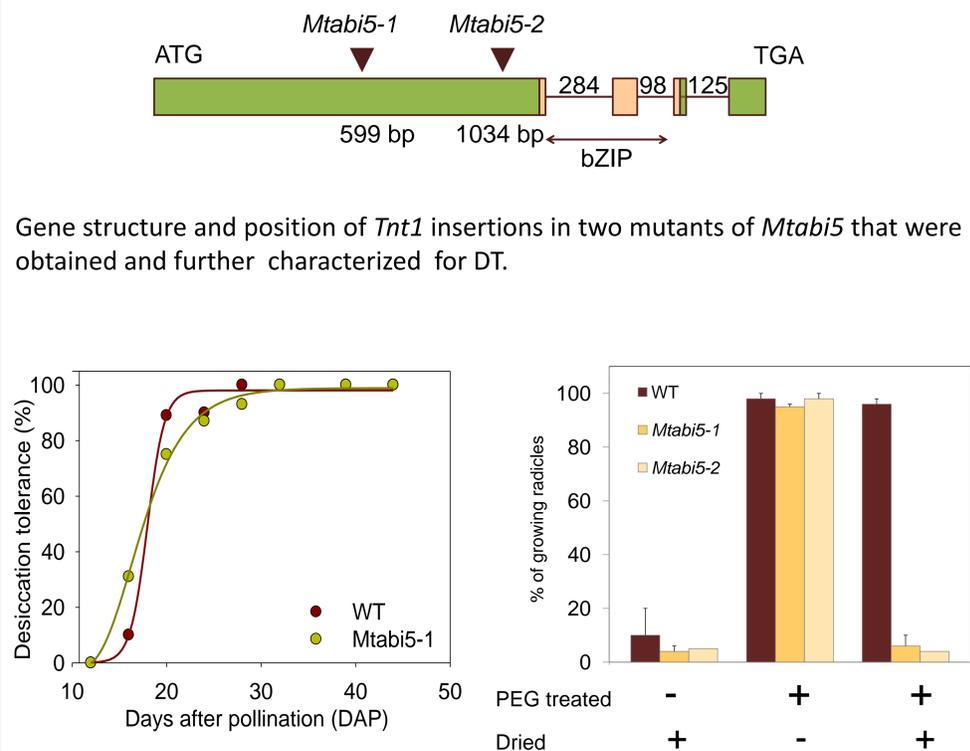
*ABI5*, a bZIP family transcription factor (TF) is one of the components of the abscisic acid (ABA) signalling pathway in seeds. In *Arabidopsis thaliana*, it is also involved in tolerance against water stress that induces growth arrest after germination. Our purpose is to investigate the role of *MtABI5* in desiccation tolerance (DT) of *Medicago truncatula* seeds using insertion mutants and transcriptomic analysis.

## Identification of transcripts linked to DT



A transcriptomic analysis was performed on the acquisition of DT during seed development (A) and the re-establishment of DT by an osmotic treatment with polyethylene glycol (PEG) (B). (C) A comparative analysis between both datasets revealed 740 genes that are up-regulated (DT-UP) and 2829 genes that are down-regulated (DT-DOWN) in relation to DT. *MtABI5* is one of the 48 TF present in the UP list.

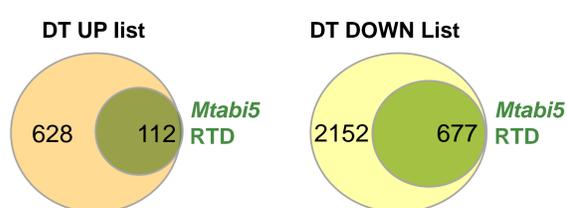
## Involvement of *MtABI5* in DT



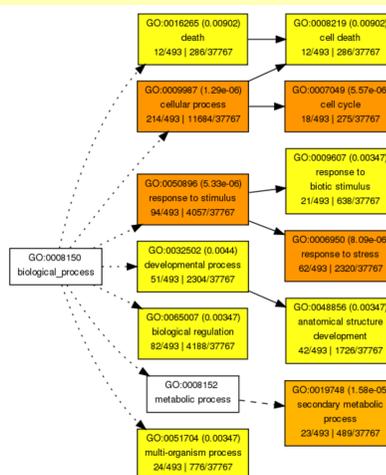
During seed development, no difference in the acquisition of DT was found between wild-type and *MtABI5-1* line. DT was scored as germination after rapid drying at 43% RH and rehydration.

However, germinated radicles of *MtABI5* mutants were unable to regain DT during PEG treatment, suggesting that *MtABI5* is a key component in the re-establishment of DT after germination, but not during development.

## *MtABI5* affects transcript levels related to biotic defense response during the re-establishment of DT



A transcriptome analysis was performed on protruded radicles of *MtABI5* mutants before and after PEG treatment compared to WT radicles (*MtABI5* RTD). Comparison of these data with the DT-UP and DOWN list revealed that 112 of the DT-UP list (15%) were no longer up-regulated in the *MtABI5* mutants whereas 23% (677 transcripts) of the DT-DOWN list were no longer down-regulated in the mutant radicles.



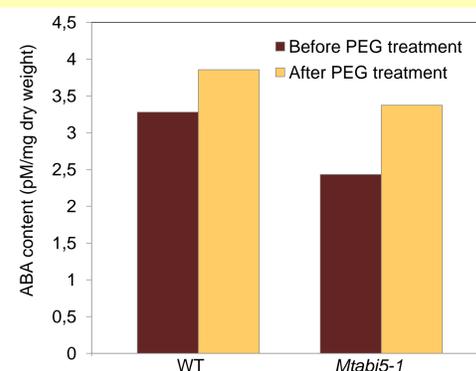
To identify biological processes that are (in)directly repressed by *MtABI5* during re-establishment of DT, a GO analysis was performed. Over-represented GO terms were linked with cell cycle and development. Surprisingly, many genes related to biotic defense were also found, including nucleotide-binding site (NBS)-leucine-rich repeat (LRR) genes, being the largest class of known disease resistance genes effective against pathogens. This suggests that *MtABI5* could be involved in a cross-talk between DT and biotic stress tolerance.

## *MtABI5* and ABA synthesis during the re-establishment of DT

Locus Id <i>Medicago</i>	Identification	WT PEG/Cntrl	<i>MtABI5-1</i> PEG/Cntrl	<i>MtABI5-2</i> PEG/Cntrl
MEDTR5G017330	ZEP	1.48 (3.535E-05)	-1.75 (0.00408919)	-1.13 (0.02020474)
MEDTR5G017350	ZEP	2.53 (0.00052403)	-1.32 (5.6314E-05)	-1.29 (0.00021847)
MEDTR5G025610	CYP707A1	-2.43 (1.7196E-05)	-0.21 (0.32395546)	-0.29 (0.45585037)
MEDTR8G072260	CYP707A1	-1.16 (0.0090278)	-1.65 (0.00727904)	-1.07 (0.20385142)

Ratios indicate the difference in transcript levels between PEG treated and untreated radicles (Cntrl). P-value is indicated in brackets.

Genes coding for zeaxanthine epoxidases (*ZEP*, *ABA1*) that are involved in ABA synthesis were upregulated upon the re-establishment of DT, whereas genes involved in ABA catabolism (*CYP707A1*, abscisic acid 8'-hydroxylase) were downregulated. Transcript levels of these genes were deregulated in *MtABI5* mutants after PEG treatment



ABA contents increased in emerged radicles of both WT and *MtABI5-1* mutant emerged radicles after PEG treatment. However, contents were 1.2 fold lower in *MtABI5-1* radicles that remain sensitive to desiccation after PEG treatment.