

## **KEY GENES FOR THE ANTIOXIDANT RESPONSE IN PLANT NUCLEUS: ISOLATION AND MOLECULAR CHARACTERIZATION IN THE MODEL LEGUME *MEDICAGO TRUNCATULA***

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Oxidative stress arises from an imbalance between generation and elimination of reactive oxygen species (ROS) and this can lead to specific cellular damages localized in cytoplasm, organelles and in nucleus [1].

Abiotic stresses such as water deficit, heavy metals and UV treatments produce DNA damage (base and sugar lesions, strand breaks, DNA-protein cross-links and base-free sites) and the accurate measurement of these modifications is essential for understanding the mechanisms of oxidative DNA damage and its biological effects. Different mechanisms, such as base-excision repair (BER) and nucleotide-excision repair (NER), are able to remove DNA lesions [2].

The latter can impair the RNA Polymerase II (RNAPII)-mediated transcription and compromise gene expression. In animal cells recent studies demonstrated that transcription elongation factors, required for correct RNAPII function under physiological conditions, play a role in the response to oxidative DNA damage. These factors assist the stalled RNAPII which can bypass the damaged sites and allow correct gene transcription.

In plants, although genes encoding proteins involved in transcriptional repair (such as those belonging to FACT -Facilitates Chromatin Transcription - complex in *Arabidopsis thaliana*) have been characterised, the response to oxidative stress at nucleus level is still poorly explored.

In this study we choose the *top1*, *tdp1* and *TFIIS* genes from the model legume *Medicago truncatula* (R108-1 genotype) as molecular markers to monitor the response to oxidative stress in nucleus. The *top1* gene encoding DNA topoisomerase I plays a key role in the response to UV-C-mediated stress in carrot cells [3,4] while the *tdp1* gene encodes the enzyme Tyrosyl-DNA phosphodiesterase, involved in the repair of DNA topoisomerase I-mediated DNA lesions [5].

Finally, also the *TFIIS* gene coding for the transcription elongation factor TFIIS [6] might represent a valuable marker, as demonstrated in animal systems [7].

We have isolated the cDNA encoding TFIIS from barrel medic. The cloning of *top1* and *tdp1* sequences is still in progress. A preliminary investigation has been also started in order to evaluate the temporal- and tissue-specific expression patterns of those genes in *M. truncatula* plants, in presence/absence of oxidative-stress conditions induced by water deficit and heavy-metal treatments. A future goal will be the production of transgenic barrel medic plants overexpressing the above mentioned genes, to be used as tool for basic and applied research on oxidative stress-related mechanisms.

## References:

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