

SCREENING TOMATO GLUTATHIONE S-TRANSFERASE DIVERSITY FOR ASSOCIATION TO DROUGHT STRESS

CASTALDO C.*, PANE M.*, GENTILE D.*, CIRILLO V.*, D'AGOSTINO N.*, IORIZZO M.**, DI MATTEO A.*

*) University of Naples Federico II - Dept of Agricultural Sciences

**) NC State University - Department of Horticultural Science

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Environmental stresses are one of the main challenges in agriculture, as they significantly limit crop productivity. As a result of global warming and climate change, extreme environmental events and abiotic stresses are expected to increase in intensity and frequency. Selecting tolerant plants for more resilient agro-ecosystems is a promising strategy for mitigating the effects of adverse environments. The cultivated tomato (*Solanum lycopersicum* L.) has a narrow genetic base, and its reduced genetic diversity hampers the progress of breeding. However, the rediscovery and use of local and wild genetic resources allow useful/beneficial alleles to be identified and used in breeding programs. To improve adaptation to harmful conditions, plants have evolved many molecular and genetics mechanisms, including hormone regulation and the activation of specific gene networks. Among others, glutathione S-transferase (GST) genes have been identified in various plant species and found to be involved in different physiological, developmental and stress modulation pathways. The aim of this research is to add insights into the role of tomato GST genes in the plant response to stress by associating allele diversity with drought tolerance.

Tomato GST sequences were characterized *in silico* showing their diffuse clustering pattern across chromosomes with TAU class GSTs duplicated in tandem and mostly aggregated on chromosomes 7 and 9. To characterize the diversity of selected GSTs, a core collection of 75 tomato accessions was selected based on microsatellite screening from a larger collection of

worldwide accessions. The core collection was screened for drought tolerance when 50% of plants showed fruit set on the lower flower truss. At this stage, two different water treatments were applied consisting in the complete restitution of the water lost due to evapo-transpiration (Full Water Restitution FWR) and the restitution of 50% of the water lost (Half Water Restitution HWR). After seven days the plants showed visible sign of drought and the leaves were analyzed for gas exchange and colorimetric variations. A gas exchange analyzer (Licor 6400) was used to measure the assimilation rate of foliar CO₂ ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$) and transpiration ($\text{mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$). The colorimetric analysis was carried out with a portable Chroma Meters (Minolta).

The eco-physiological data showed a continuous variability of the response to drought throughout the collection and allowed to identify eight tomato accessions combining a lower reduction in CO₂ assimilation, transpiration and water use efficiency and higher stability in leaf temperatures and colorimetric variations when challenged with HWR. Enriched libraries for GST loci were prepared based on the specific hybridization of short oligonucleotide primers (20-25 bases). Libraries will be sequenced using next/third generation sequencing technology (Illumina or PacBio). Mapped variants and haplotypes in the GST loci will be integrated with drought tolerant traits and used to identify associations with tolerant phenotypes to additional environmental stresses.