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SOMATIC EMBRYOGENESIS IN GRAPEVINE: A MULTILAYERED APPROACH TO DECIPHER MOLECULAR MECHANISMS INVOLVED

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Somatic embryogenesis, a morphogenic process that takes advantage of the regenerative potential of plants to replicate whole plants starting from somatic explants, can be a source of variation with potential applications in plant breeding, i.e. useful for the selection of improved genotypes, and it is one of the most suitable tools to apply functional genomics studies and genetic improvement in plants. However, beyond a few pioneering research works mainly focused on model plants, the molecular characterization of somatic embryogenesis mechanisms is still elusive, especially for woody species. In grapevine, this process is affected by many factors such as explant type, culture conditions and, most importantly, genotype. Many cultivars, in fact, have shown recalcitrance to tissue culture and transformation, and very low somatic embryogenesis competence, thus preventing the widespread application of the so-called "next-generation breeding techniques" such as cisgenesis and genome editing in grapevine.

Here, we explored genetic and epigenetic features of the somatic embryogenesis process in grapevine by investigating the behavior of two genotypes showing opposite somatic embryogenesis competence. Embryogenic tissues were induced from immature stamens excised from field-collected flower clusters of Sangiovese (highly competent for embryogenesis) and Cabernet Sauvignon (poorly competent for embryogenesis). A multilayered approach was used to profile mRNA, smallRNAs and methylated DNA with high-throughput sequencing technologies in the initial explants, on undifferentiated calli induced after 40 days of culture, and in embryogenic and non-embryogenic calli after 3 months of culture.

A comprehensive comparison of transcriptomes of the different types of calli with the grapevine gene expression atlas (Fasoli et al., 2012) revealed that, in grapevine, the dedifferentiation to the callus formation during the embryogenesis process occurs via a berry developmental pathway. This is dissimilar from that shown in Arabidopsis, in which dedifferentiated calli are more similar to the tip of a root meristem. Interestingly, it seems that the more the callus stays in culture without acquiring embryogenecity the more secondary metabolism genes result activated. Indeed, a Gene Ontology (GO) analysis revealed that secondary and carbohydrate metabolisms are the enriched functional categories for gene differentially regulated in embryogenic *vs* non-embryogenic calli, suggesting their pivotal role in grapevine somatic embryogenesis process.