A versatile genome editing platform for grapevine: improving biotic and abiotic stress resilience

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Abstract (250 words)

New Plant Breeding Techniques (NPBTs) have arisen with the objective of surmounting the constraints inherent in conventional breeding methodologies, thereby enhancing plant resilience against both biotic and abiotic stresses. To date the application of genome editing in grapevine is still limited by the necessity to overcome recalcitrance to produce embryogenic *calli* and to regenerate plants. In our studies, we developed a smart and versatile genetic transformation system carrying all the most promising features of different genome editing approaches. In specific, we joined the GRF-GIF expression to improve regeneration, the systemic movement of the editing transcripts through tRNA-like sequences (TLS) and the cisgenic-like approach to remove transgenes. In parallel, we were able to assess the efficiencies of several guide RNAs (gRNAs) targeting genes with fall-out on drought stress and pathogen resilience. To the first aim, a gene belonging to glutathione S-transferase (*VvGST40*) and two gene belonging to pectin-methyl esterase (*VvPME1* and *VvPME3*) have been targeted. Several edited lines were acclimatized and are currently under evaluation. In parallel, two genes belonging to the Mildew Locus-O (*VvMLO6* and *VvMLO7*) and a non-expressor pathogenesis related gene (*VvNPR3*) were targeted to improve pathogen resilience. Acclimatized plants edited for *MLO* genes resulted almost resistant to *Erysiphe necator* (disease incidence reduction up to 80%), whereas NPR3-edited vines showed a significant reduction in disease severity (up to 70%). In conclusion, our approaches allowed to improve stress resilience of several economically-important genotypes such as Pinot noir, Chardonnay, Sangiovese and Glera but also widely used rootstocks (*e.g.*, 110R and K5BB).

Keywords: powdery mildew, climate change, recalcitrance, cisgenic-like approach, New Plant Breeding Techniques.