The Real Sour Grapes: Genetic Loci, Genes, and Metabolic Changes Associated with Grape Malate Levels

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Abstract:

Context and purpose of the study - Insufficient levels of malate and lack of acidity in commercial grape cultivars (*V.vinifera*) hinders the quality of fruit grown in warm climates. Conversely, excessive levels of malate and sourness in wild *Vitis* grape, leads to unpalatable fruit and complicates the introgression of valuable disease resistant alleles through breeding. Nonetheless, albeit decades of research, knowledge regarding the molecular regulation of malate levels in grape remains limited.
While malate dissimilation is a hallmark of grape ripening, it was found to be absent or limited in wild *Vitis* fruit (*riparia, cinerea*). Hence, these genotypes serve as unique resources to deepen our understanding of malate regulation, with the overarching goal of controlling fruit acidity by breeding.
Our research aimed to (i) Identify genetic loci tightly associated with fruit malate levels in interspecific families, and (ii) highlight differences in metabolism and gene expression, associated with contrasting malate behavior between wild and commercial genotypes.

Material and methods - QTL mapping was performed using a novel set of amplicon-based markers (rhAmpSeq) and six years of phenotyping of a complex interspecific F1 family with strong and stable variation in malate at ripeness. In addition, a comparative RNAseq and primary metabolite profiling was performed during fruit development in *riparia* and *cinerea* accessions, and commercial *vinifera* cultivars.

Results - Three significant QTL for ripe fruit malate on chromosomes 1, 7, and 17, accounted for over two-fold and 6.9 g/L differences, and explained 40.6% of the phenotypic variation. QTL on chromosomes 7 and 17 were stable in all and in three out of five years, respectively. Lack of malate degradation in wild genotypes was associated with higher fruit respiration rates, higher levels of amino acids, TCA and fermentation metabolites, and higher expression of their corresponding genes, some of which positioned within the identified QTL in the studied population.
The developmental pattern and inter-specific differences in the expression of genes presumably involved in malate biostynthesis, degradation, and transport, allowed us to highlight major candidate genes involved in malate regulation across *Vitis* species. These results advance current knowledge regarding the regulation of malate at the mechanistic and metabolic levels, and highlight genetic markers and candidate genes associated with grape acidity.

Keywords: Fruit acidity, Wild *Vitis*, marker-assisted breeding, rhAmpSeq, vacuolar transport, climate-change adaptation, disease resistance