Transcriptomic and Metabolomic Analyses of Cabernet Sauvignon Grape Berry Development

Laurent G. Deluc1, Jérôme Grimplet1, Matthew D. Wheatley1, Richard L. Tillett1, David R. Quilici1, Craig Osborne2, David A. Schooley1, Karen A. Schlauch1, John C. Cushman1, Grant R. Cramer*1

1Department of Biochemistry and Molecular Biology, University of Nevada, Reno, Nevada 89557-0014, USA.
2Department of Animal Biotechnology, University of Nevada, Reno, NV 89557-0014

Grape berry development is a dynamic process that involves a complex series of molecular genetic and biochemical changes divided into three major phases. During initial berry growth (Phase I), berry size increases along a sigmoidal growth curve due to cell division and subsequent cell expansion, and organic acids (mainly malate and tartrate), tannins, and hydroxycinnamates accumulate to peak levels. The second major phase (Phase II) is defined as a lag phase in which cell expansion ceases and sugars begin to accumulate. Véraison (the onset of ripening) marks the beginning of the third major phase (Phase III) in which berries undergo a second period of sigmoidal growth due to additional mesocarp cell expansion, accumulation of anthocyanin pigments for berry color, accumulation of volatile compounds for aroma, softening, peak accumulation of sugars (mainly glucose and fructose), and a decline in organic acid accumulation. In order to understand the transcriptional network responsible for controlling berry development, mRNA expression profiling was conducted on berries of V. vinifera Cabernet Sauvignon using the Affymetrix GeneChip® Vitis oligonucleotide microarray ver. 1.0 spanning seven stages of berry development from small pea size berries (E-L stages 31-33 as defined by the modified E-L system), through véraison (E-L stages 34-35), to mature berries (E-L stages 36 and 38). Selected metabolites were profiled in parallel with mRNA expression profiling to understand the effect of transcriptional regulatory processes on metabolite production affecting color, flavor, and aroma characters, which ultimately influence the organoleptic properties of wine.

Results
Over the course of berry development whole fruit tissues were found to express an average of 74.5% of probes represented on the Vitis microarray. Approximately 60% of these transcripts exhibited significant differential expression between at least two out of the seven stages of berry development with more than 28% of transcripts (4,151 Unigenes) showing pronounced (≥ 2 fold) differences in mRNA expression illustrating the dynamic nature of the developmental process. Grouping 4,151 Unigenes having at least two-fold differential expression among two developmental phases revealed twenty well-correlated expression profile groups of interest. Expression profile patterns included those with declining or increasing mRNA expression over the course of berry development as well as transient peak or trough patterns across various developmental stages as defined by the modified E-L system. These detailed surveys revealed the expression patterns for genes that play key functional roles in phytohormone biosynthesis and response, calcium sequestration, transport and signaling, cell wall metabolism mediating expansion, ripening, and softening, phenylpropanoid and flavonoid metabolism, transport, and regulation, aroma biosynthesis, organic and amino acid metabolism, hexose sugar and triose phosphate metabolism and transport, and starch metabolism. In particular, mRNA expression patterns of transcription factor, abscisic acid (ABA) biosynthesis, and calcium signaling genes identified candidate factors likely to participate in the progression of key developmental events such as véraison and potential candidate genes associated with such processes as auxin partitioning within berry cells, aroma compound production, and flavonoid pathway regulation and sequestration of flavonoid compounds. Finally, analysis of sugar metabolism gene expression patterns suggested the existence of an alternative pathway for glucose and triose phosphate production that is invoked from véraison to mature berries.

Conclusions
These results reveal the first high-resolution picture of the transcriptome dynamics that occur during seven stages of grape berry development. This work also establishes an extensive catalogue of gene expression patterns for future investigations aimed at the dissection of the transcriptional regulatory hierarchies that govern berry development in a widely grown cultivar of wine grape. More importantly, this analysis identified a set of previously unknown genes potentially involved in critical steps associated with fruit development that can now be subjected to functional testing.