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# Grape development revisited through the single-berry metabolomic clock paradigm

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## Abstract

Although the ripening process of grapevine berries is well-documented at the vineyard level, pinpointing distinct developmental stages remains challenging. The asynchronous development of berries results in dynamic biases and metabolic chimerism. It is thus crucial to consider individual berries separately and resynchronize their internal clock for deciphering physiological changes throughout development. Given the importance of grape composition in wine quality, we aimed at measuring developmental changes in the metabolome of Syrah single berries from anthesis to over-ripening, without a priori preconceived. Non-targeted UHPLC-HRMS analyses of single berries yielded 9,335 compounds with specific mass and retention time. This dataset was submitted to an analysis workflow, combining classification and dimension reduction tools, to reveal the dynamics of metabolite composition. The outcomes of this workflow highlight an innovative redefinition of developmental stages, through the clustering of metabolites into 11 specific kinetic patterns. More precisely, the usual double sigmoidal growth pattern could be split into more transient stages characterized by the accumulation of specific metabolites. For instance, we identified a cluster of metabolites annunciative of the onset of ripening at the end of the herbaceous plateau which witnesses transient lipidic changes. We also found a cluster composed of stilbenes that accumulate during berry shriveling, following sugar loading. This non-targeted approach enables a more precise and unbiased characterization of grapevine berry development through the metabolomic clock paradigm, paving the way for a better assessment of berry physiological stage in genetic studies and ultimately for varietal selection and adaptation to climate change.

**Keywords:** Vitis vinifera L., untargeted metabolites, single berry, phenology