

Defining gene regulation and co-regulation at single cell resolution in grapevine

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Abstract

Conventional molecular analyses provide bulk genomic/transcriptomic data that are unable to reveal the cellular heterogeneity and to precisely define how gene networks orchestrate organ development. We will profile gene expression and identify open chromatin regions at the individual cells level, allowing to define cell-type specific regulatory elements, developmental trajectories and transcriptional networks orchestrating organ development and function. We will perform scRNA-seq and snATAC-seq on leaf/berry protoplasts and nuclei and combine them with the leaf/berry bulk tissues obtained results, where the analysis of transcripts, chromatin accessibility, histone modification and transcription factor binding sites showed that a large fraction of phenotypic variation appears to be determined by regulatory rather than coding variation and that many variants have an organ-specific effect. By bioinformatics approaches we will identify cell and gene clusters, interpreting the heterogeneity from single-cell transcriptomes; subsequently, we will perform in situ hybridizations to corroborate already predicted cell-type annotations and to identify new cell-type marker genes, required for the cell identity definition, and for the experimental validations of scRNAseq data. The realization of a single cell resolution spatiotemporal transcriptomic and chromatin accessibility map of grapevine berry will allow to link gene expression profiles to cellular and developmental processes, uncovering part of the molecular mechanisms of ripening and slowly providing the key in maintaining high quality grapes and wine. Building organ-scale gene expression maps is essential to drive technological innovation such as reprogramming cell identity and inducing phenotypic changes via cell-type-specific gene editing.

Keywords: Single-cell RNA-seq, single nucleus ATAC-seq, gene expression regulation, gene network, developmental trajectories.

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