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PLANT GENETICS

Spatial transcriptomics in plants

> high-throughput and spatially resolved transcriptomics in plant tissues

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Spatial transcriptomics combines histological imaging and RNA sequencing to simultaneously quantify and localize gene expression. Giacomello *et al.* have now optimized this technique for plant tissues, which pose specific challenges compared with mammalian tissues, such as the presence of cell walls, vacuoles, chloroplasts and secondary metabolites.

Spatial transcriptomics is performed by placing cryosections on arrays coated with reverse transcription oligo(dT) primers, which harbour unique positional barcodes. After fixation, staining and permeabilization of the section, mRNA is captured locally and reverse transcribed, tissue is removed, mRNA–cDNA hybrids are released and, finally, tissue section morphology is combined with the barcoded gene expression data.

To enable the use of this technique in plant tissues, the authors modified this protocol. Technical adjustments included using less intense fixation to prevent irreversible attachment; the use of Toluidine blue staining to visualize plant morphology; enzymatic permeabilization optimized for each tissue type; the capture of secondary metabolites by additional steps during permeabilization and cDNA synthesis; as well as the development of an enzyme cocktail to degrade the tissue sections without interfering with the captured gene expression information. The method was validated for a wide range of herbaceous and woody plant model systems, which included the inflorescence meristem of *Arabidopsis thaliana*, developing and dormant leaf buds of *Populus tremula* and the female cones of *Picea abies*.

The proposed method showed high biological and technical reproducibility. Validation of the spatial gene expression data by comparison with the AtGenExpress Development dataset showed high specificity (93%), accuracy (71%) and a low false-positive rate (6.5%). Furthermore, localization of known genes and gene clusters obtained via hierarchical clustering confirmed the gene expression pattern that was derived using the optimized spatial transcriptomics protocol. Indeed, genes encoding proteins that are involved in flowering localized in the flowers at the expected developmental stage, and the identified gene clusters corresponded to the main organ types that were present in the tissue sample. Finally, the quality of the data combined with analytic approaches allowed for quantification of differences in gene expression and functional analysis (pathway scores) between different regions of the same tissue section and among replicates.

In conclusion, the method developed by Giacomello *et al.* enables high-throughput and spatially resolved transcriptomics in plant tissues and has clear advantages over techniques that are currently available in plant research (such as fluorescence-activated cell sorting or laser capture microdissection), which require the creation of transgenic plant lines or cannot be scaled up.

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ORIGINAL ARTICLE Giacomello, S. *et al.* Spatially resolved transcriptome profiling in model plant species. *Nature Plants* http://dx.doi.org/10.1038/nplants.2017.61 (2017)
FURTHER READING Crosetto, N., Bienko, M. & van Oudenaarden, A. Spatially resolved transcriptomics and beyond. *Nat. Rev. Genet.* **16**, 57–66 (2015)