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Setting up high-content applications for detection of mRNAs and miRNAs in plants

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Plant functional biology studies rely largely on the characterization of differential gene expression in different experimental conditions. Gene expression studies at the RNA level have undergone tremendous progress, reaching a high level of throughput and enabling the simultaneous analysis of entire transcriptomes through microarrays or deep sequencing RNA-Seq approaches. These powerful approaches however do not provide the high spatial resolution given by techniques detecting mRNA or miRNA expression in their tissue context by *in situ* hybridization (ISH). The adoption of ISH has been hampered by its low throughput, sensitivity and reproducibility. We describe the optimization of automated liquid handling procedures for the high-throughput reliable application of ISH for the detection of relatively large numbers of mRNAs and miRNAs on sections and on whole mount samples for the two model plants *Zea mais* and *Arabidopsis thaliana*. We show that by adopting optimized conditions ISH can be brought to high level of processivity and reproducibility. Case studies of applications of the automated protocols developed are described for their potential to enable obtainment of tissue arrays, including the evaluation of currently used modified oligonucleotides for the detection of miRNAs on plant tissues.