

The kinetics of Interferon- γ induced RNA flow

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Summary

Attempting to cover everything from imaging strategy to analysis method

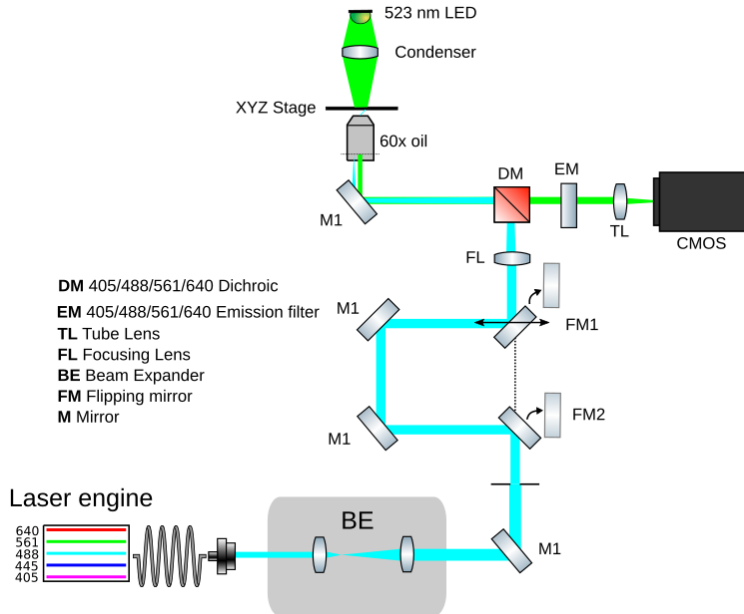
Part 1

- ▶ Imaging strategy and BASIC processing
- ▶ Image stitching, registration, segmentation with MCMICRO

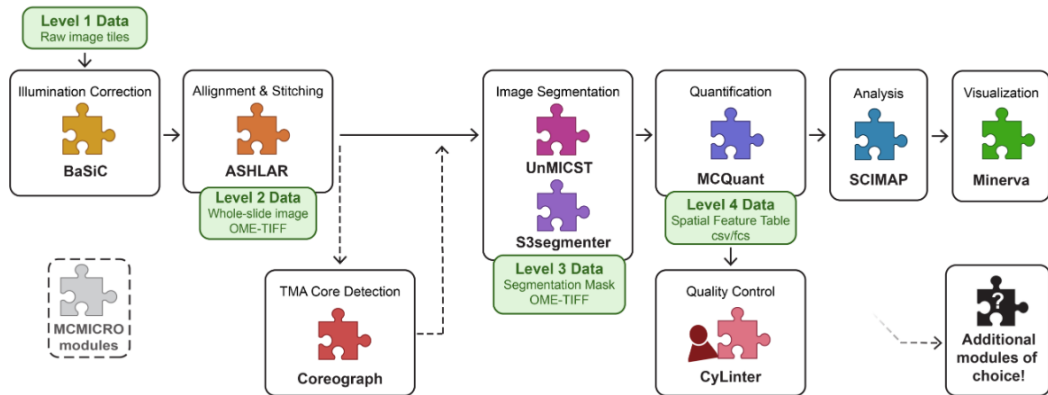
Part 2

- ▶ Recap of the compartment model for RNA flow
- ▶ Gene selection based on RNA flow model
- ▶ Brief comments on probe design

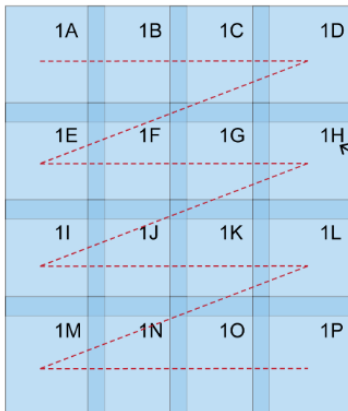
Imaging setup



The MCMICRO pipeline



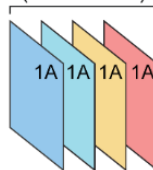
4-Channel acquisition



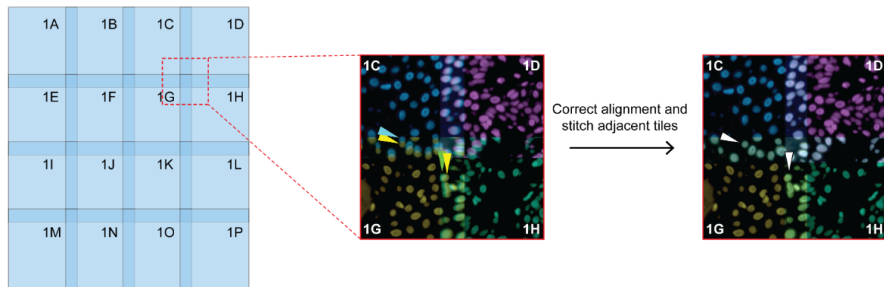
---- Microscope Path

Image identifier:
Cycle 1
Location H

Each image tile may encompass
4-6 channels of data
(Level 1 Data)

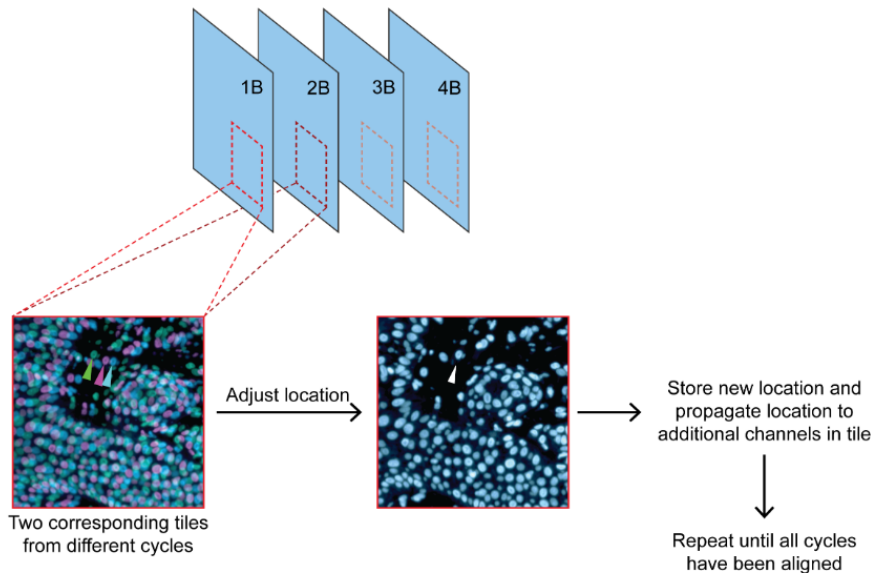


Stitching a single channel with ASHLAR

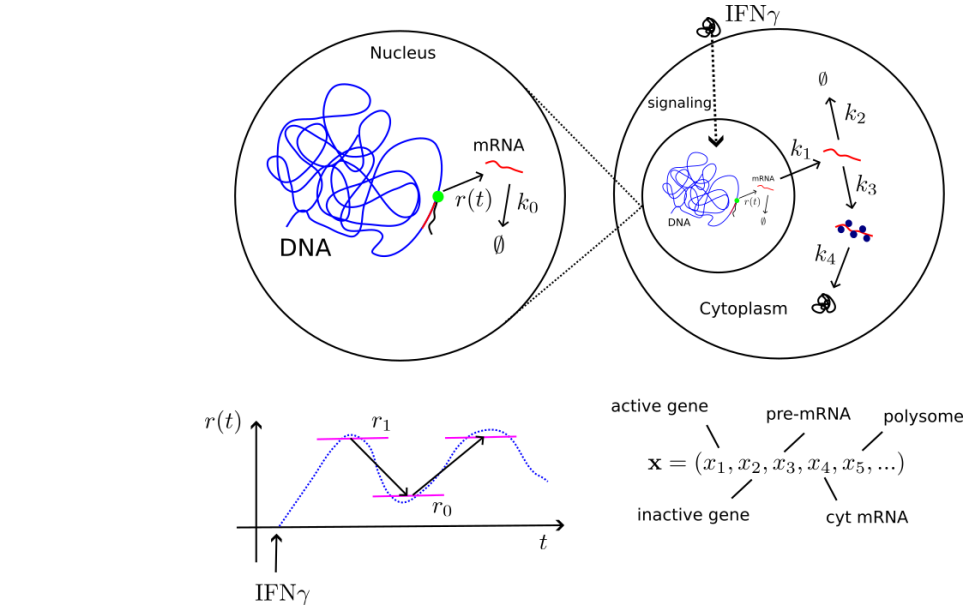


Courtesy of the Laboratory of Systems Pharmacology at Harvard

Registering images between cycles with ASHLAR



A model for RNA Flow



The principle of Interferon- γ induced transcriptional memory

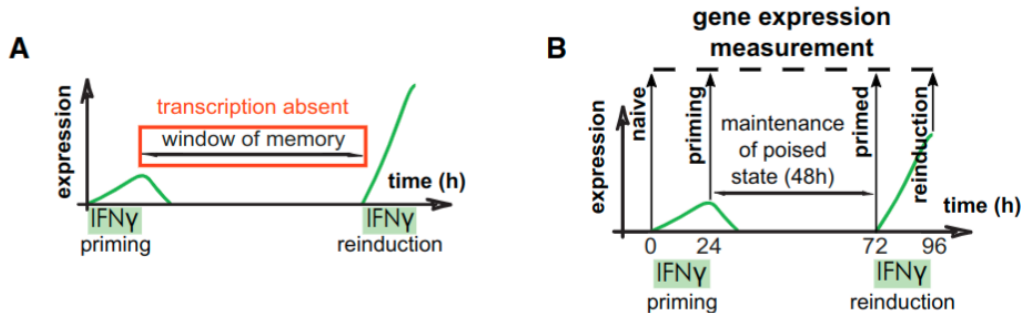
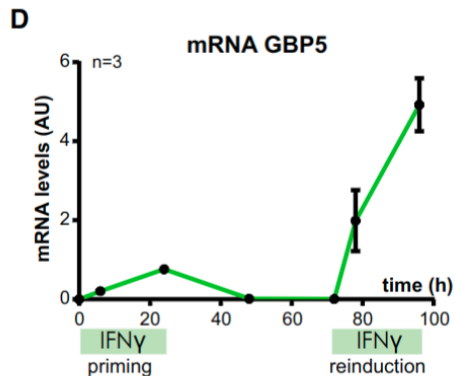
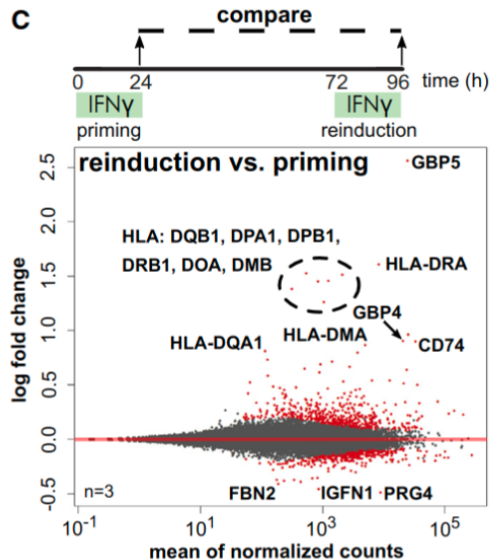


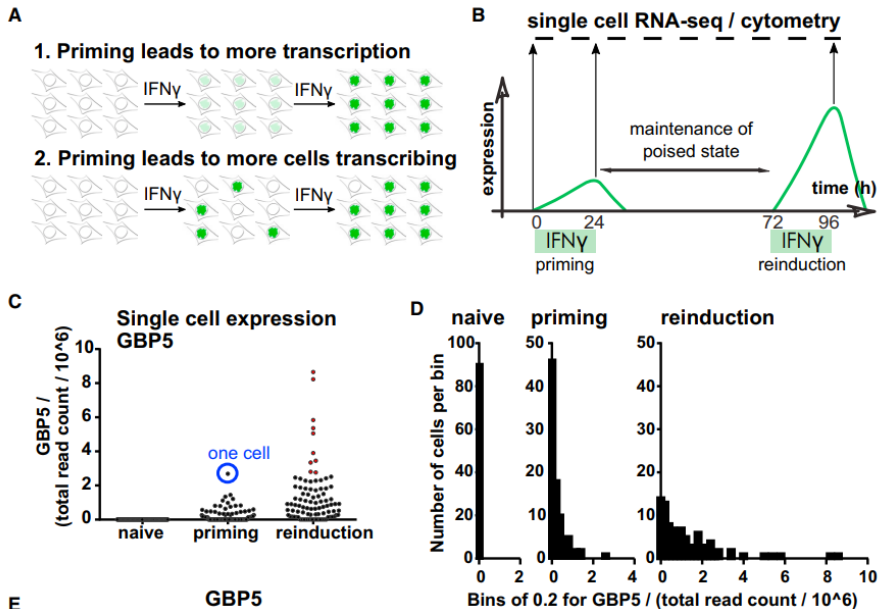
Figure 1

Siwek et al. *Activation of Clustered IFN γ Target Genes Drives Cohesin-Controlled Transcriptional Memory*. Molecular Cell 2020

Total RNA-seq identifies GBP5 as a memorized gene



Single-cell RNA-seq identifies GBP5 as a memorized gene



BLASTing probes for GPB5 and CD274

