

# The kinetics of Interferon- $\gamma$ induced RNA flow

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# Summary

Attempting to cover everything from imaging strategy to analysis method

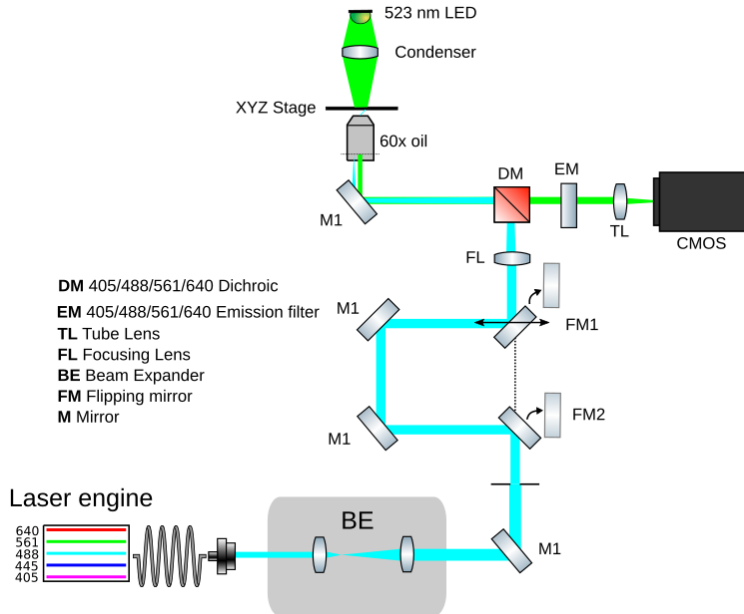
## Part 1

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

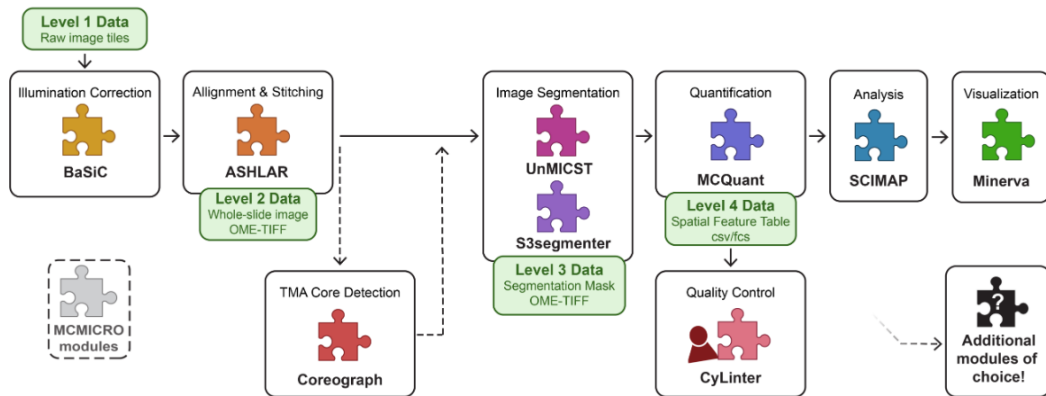
## Part 2

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

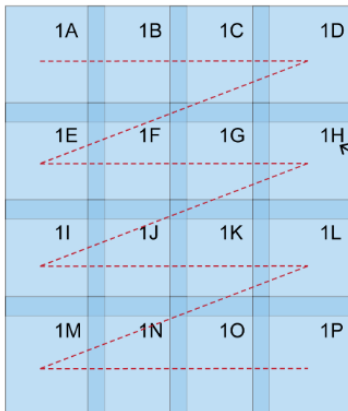
# Imaging setup



# The MCMICRO pipeline



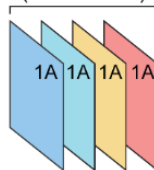
## 4-Channel acquisition



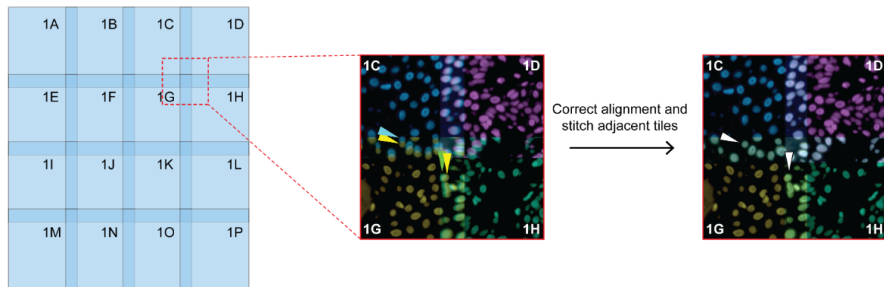
---- Microscope Path

Image identifier:  
Cycle 1  
Location H

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

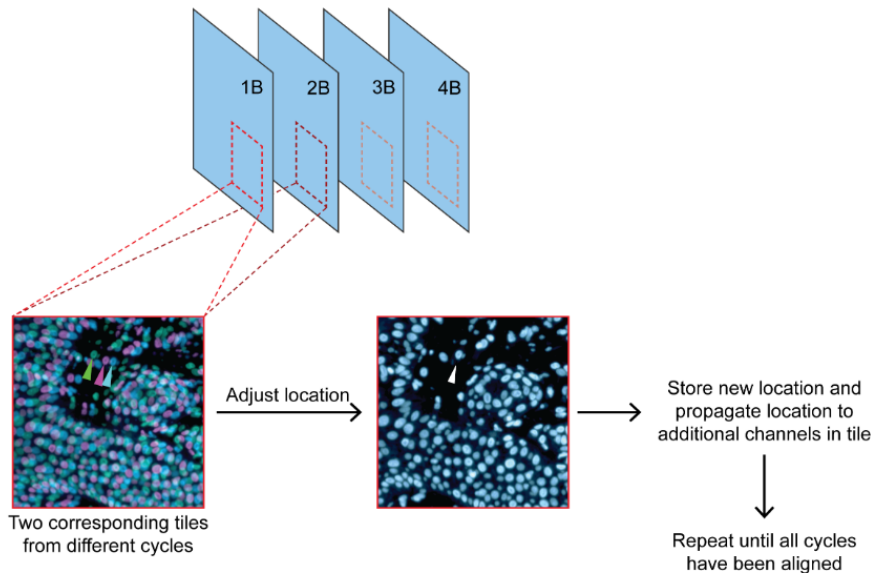


# Stitching a single channel with ASHLAR

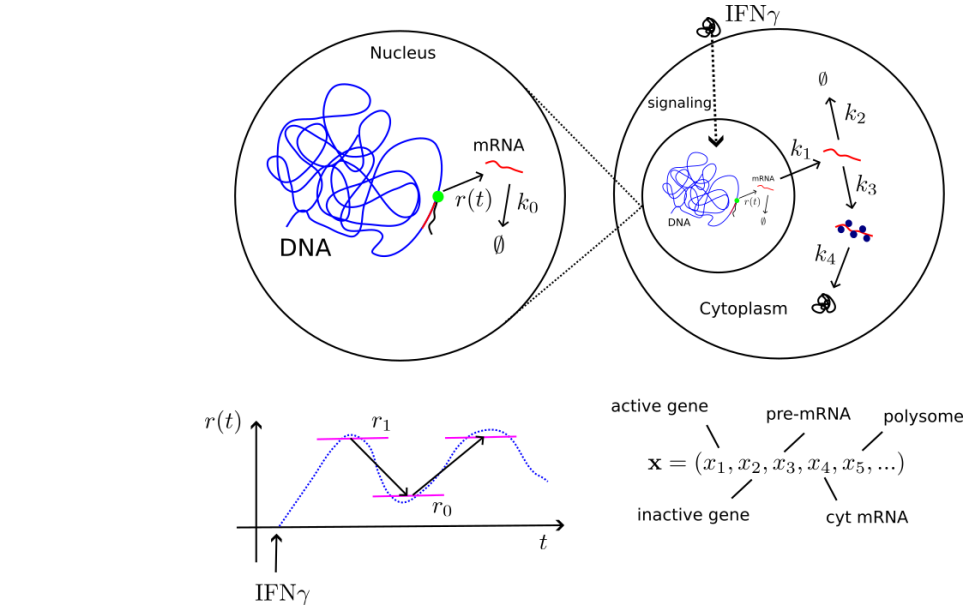


*Courtesy of the Laboratory of Systems Pharmacology at Harvard*

# Registering images between cycles with ASHLAR



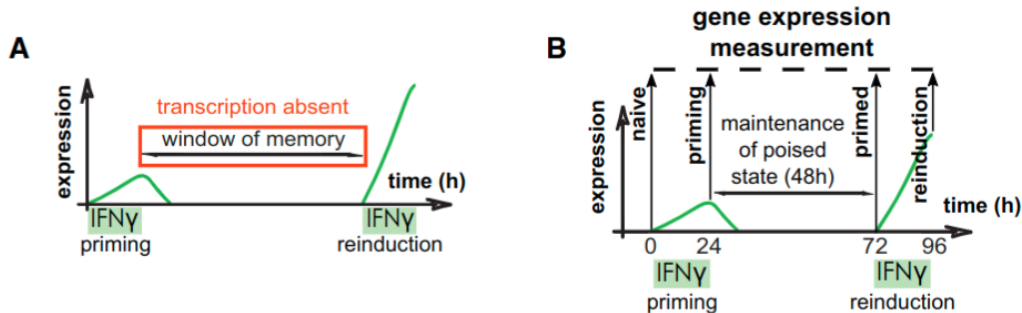
# A model for RNA Flow





# Bayesian inference of RNA flow parameters

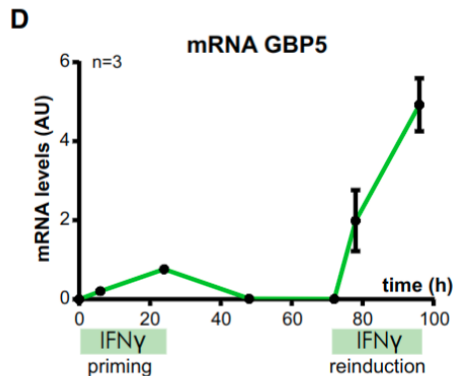
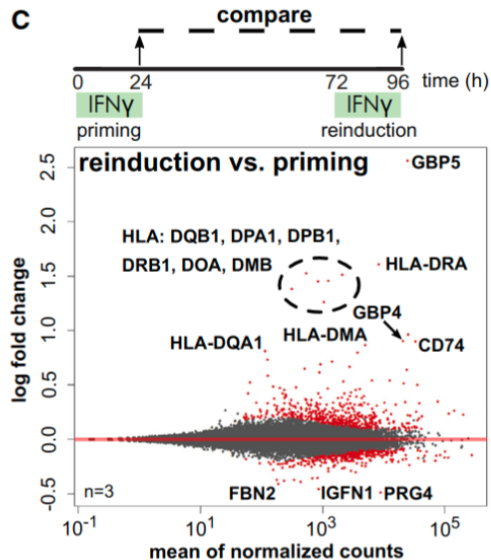
# The principle of Interferon- $\gamma$ induced transcriptional memory



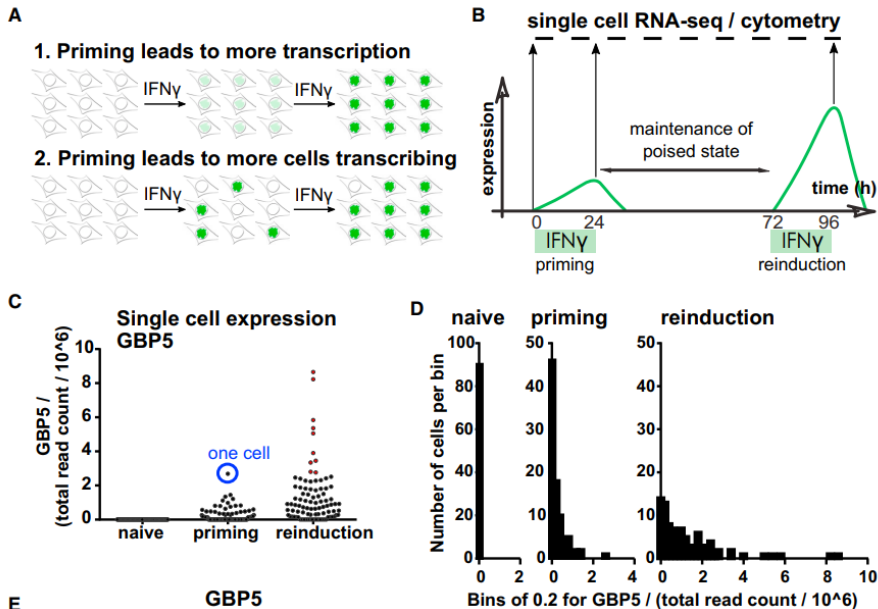
**Figure 1**

Siwek et al. *Activation of Clustered IFN $\gamma$  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

