

# Image analysis for microscopy with machine learning

BIO-221

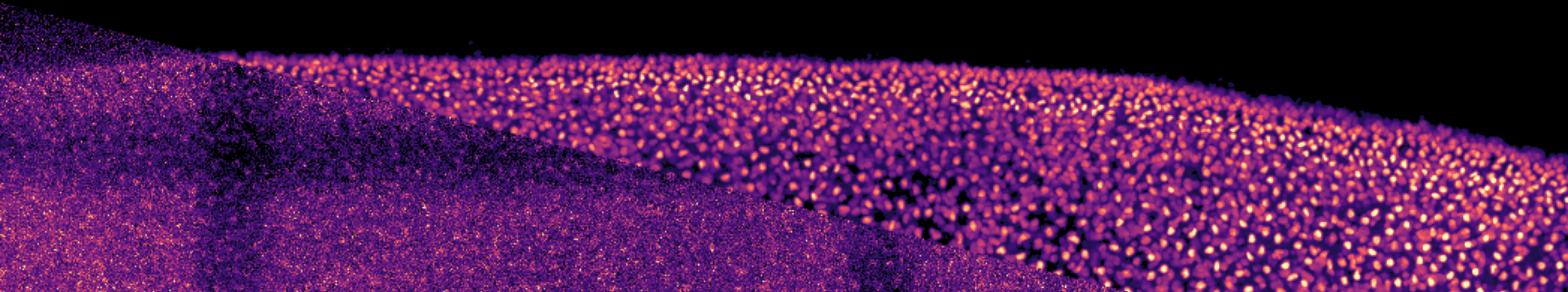
October 28th, 2025

Albert Dominguez Mantes

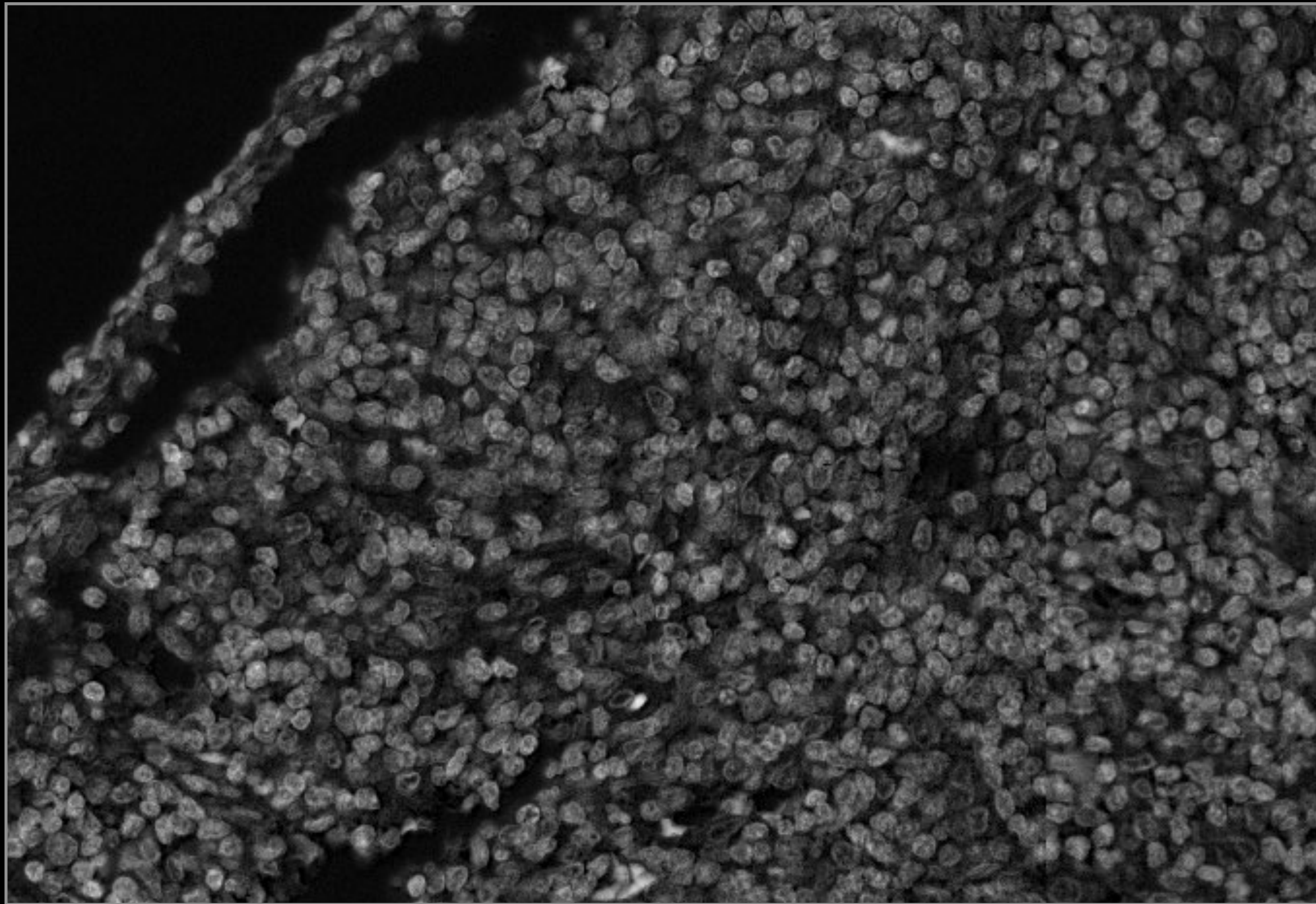
La Manno Group, EPFL  / Weigert group, TU Dresden 

<https://www.epfl.ch/labs/nsbl/>

<https://weigertlab.org/>

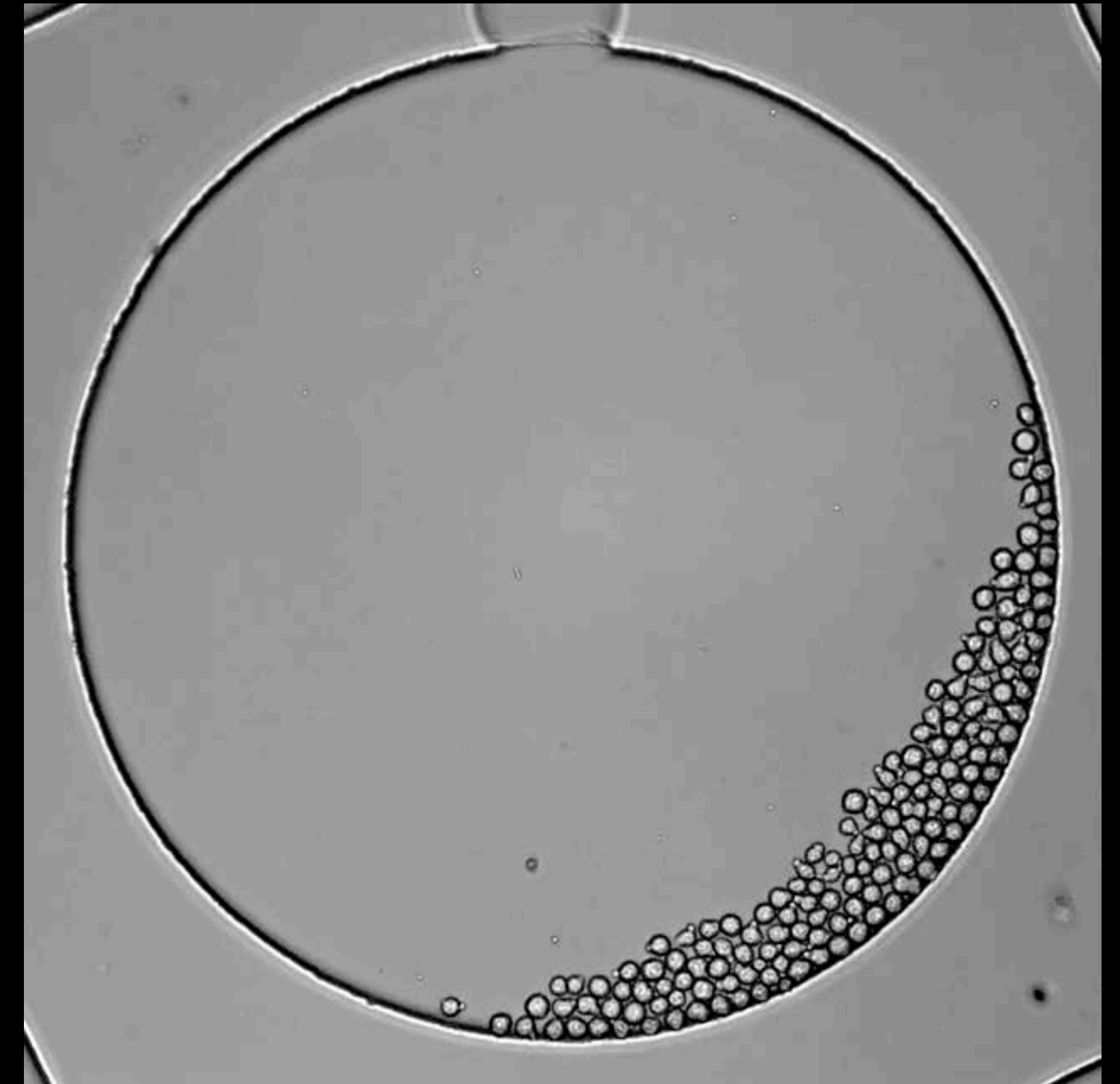


2D (fluorescence)



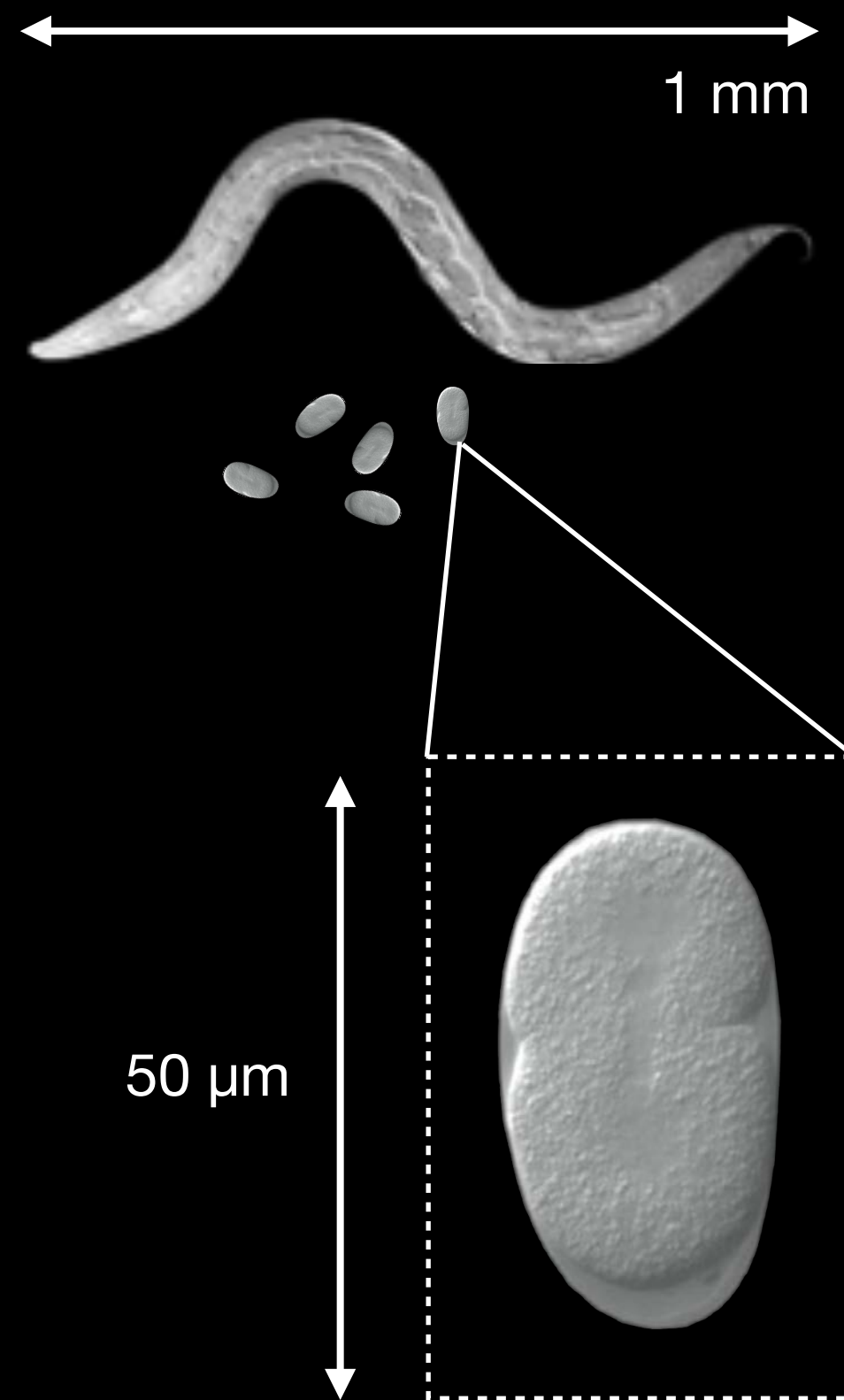
*Data from Anna Maria Tsakiroglou (Manchester)*

2D + time (brightfield)

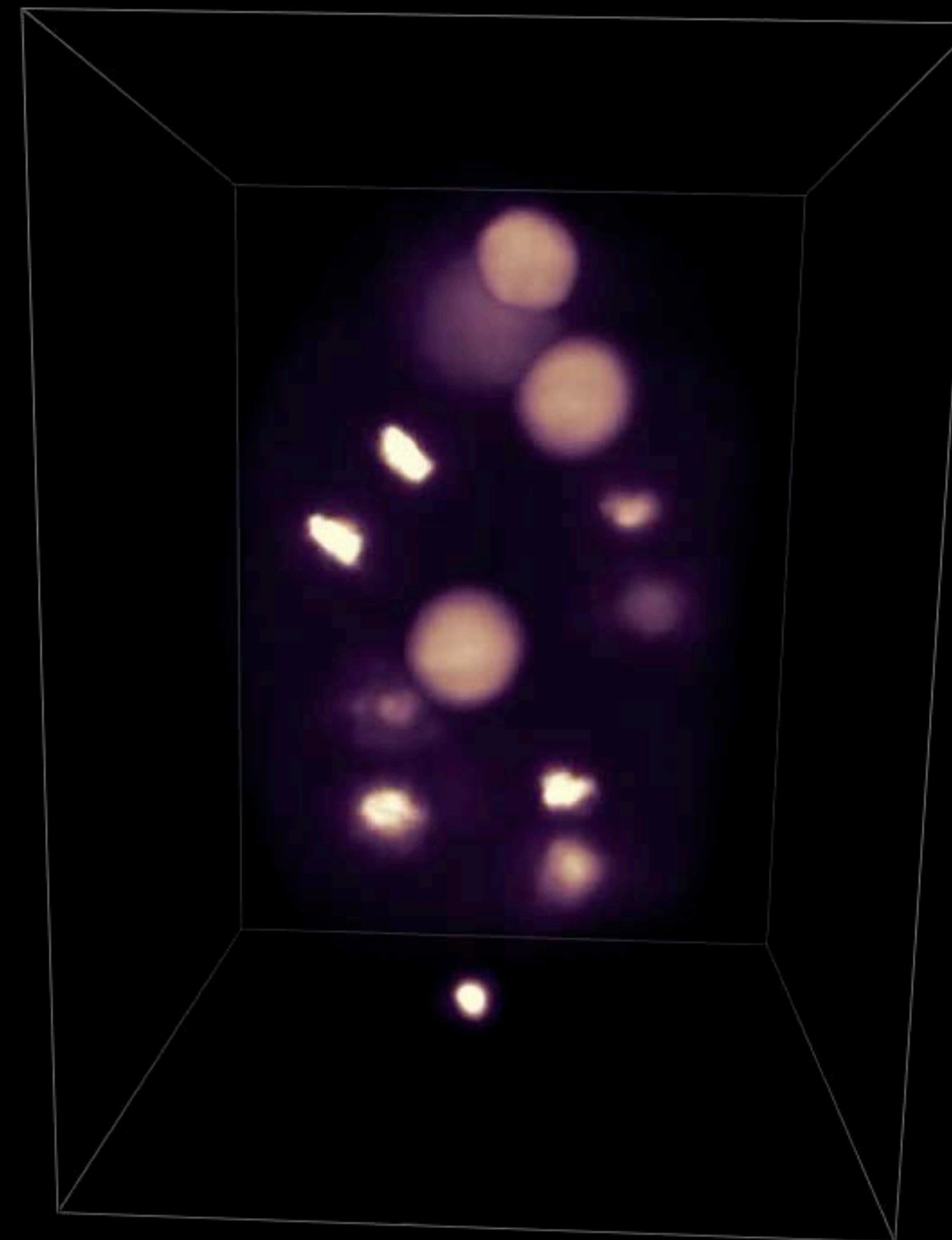


*Mouse stem cells, data from cell tracking challenge*

3D + time - fluorescence microscopy of developing organisms



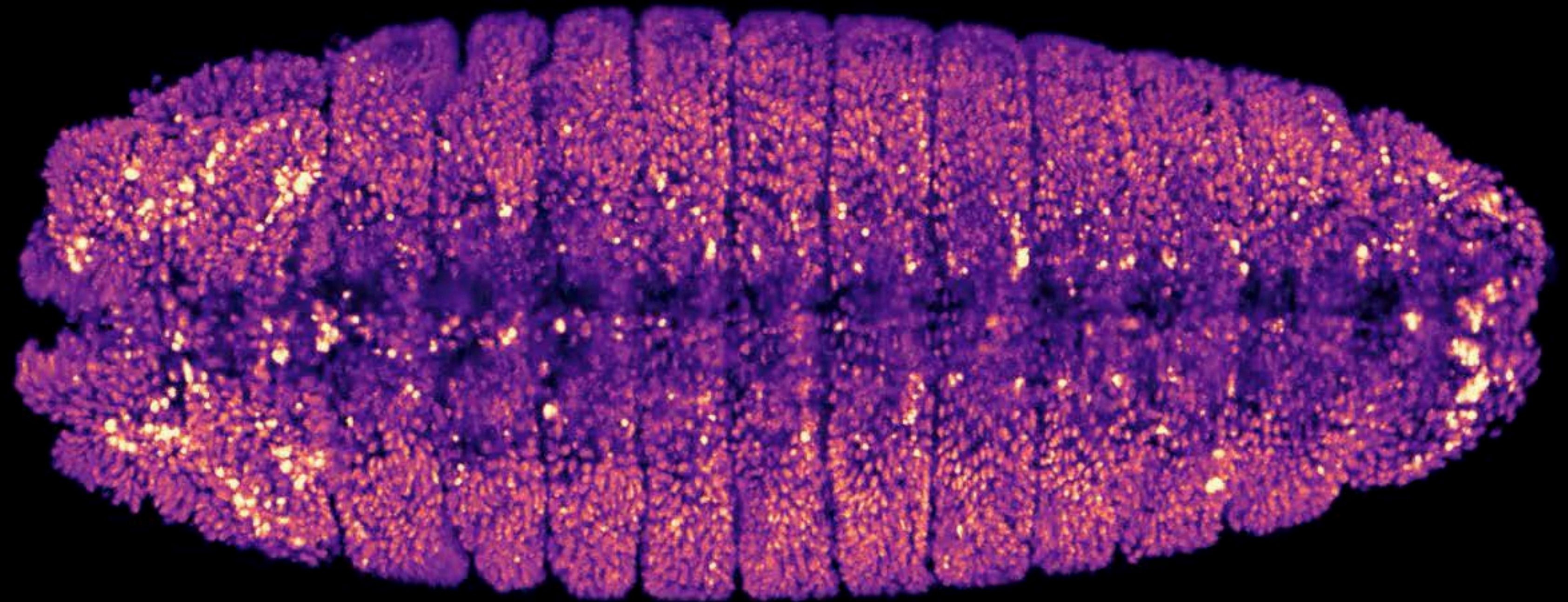
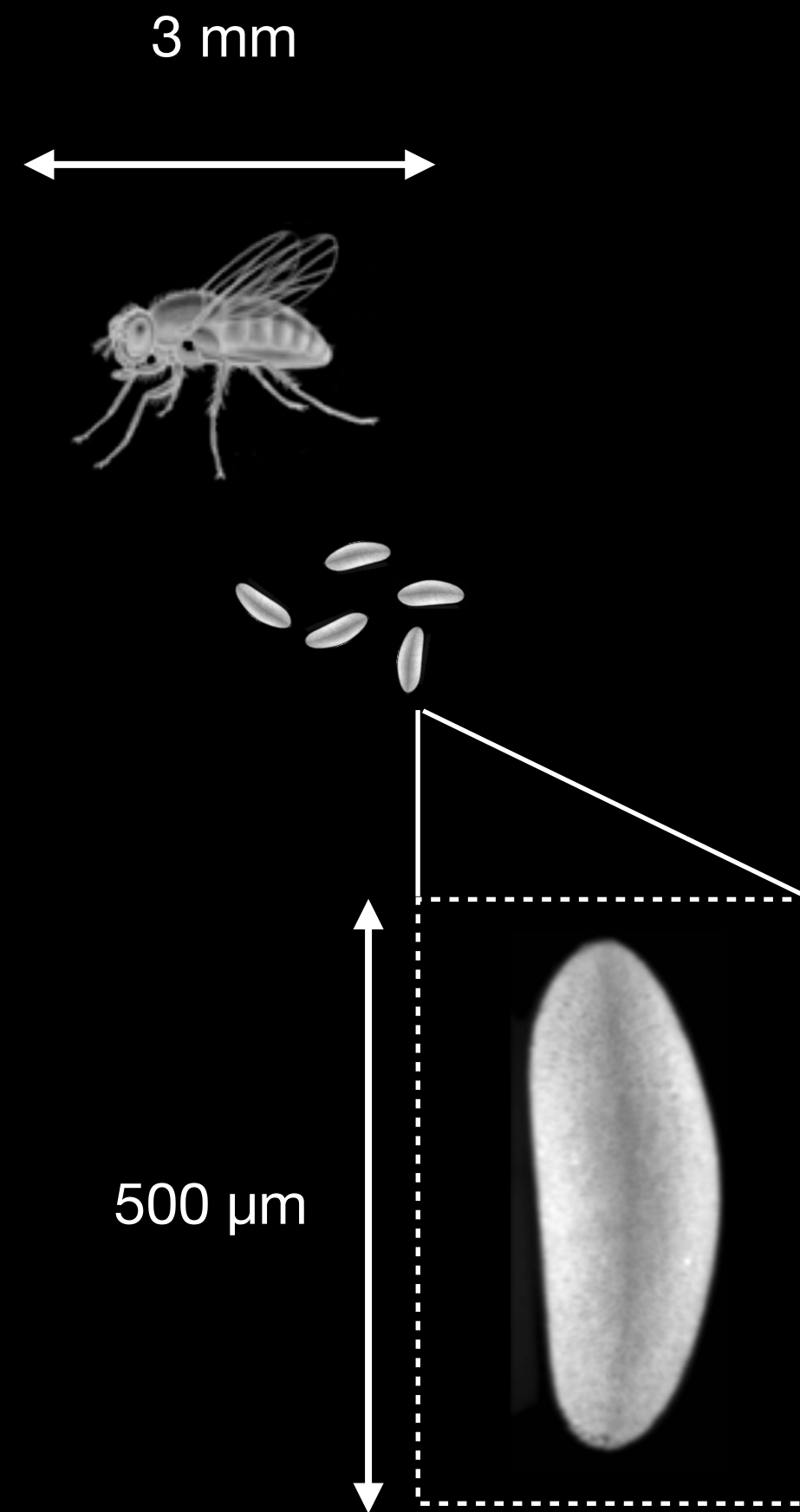
*C. elegans*



1:22:04

Data: Gene Myers Lab (Dresden)

3D + time - fluorescence microscopy of developing organisms



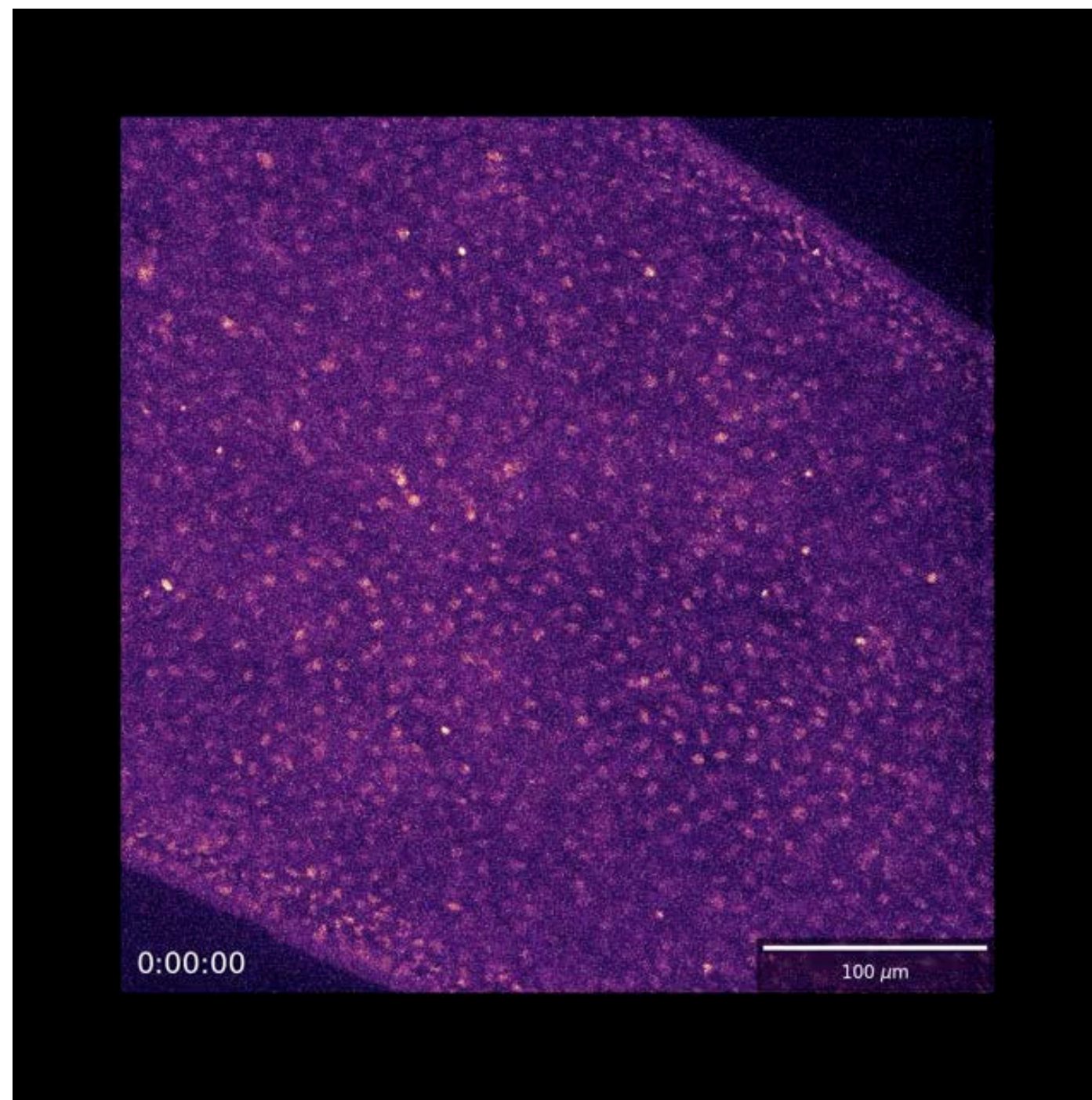
*Drosophila melanogaster*

Royer et al. (2016)

Extraction of quantitative biological information from digitised camera images

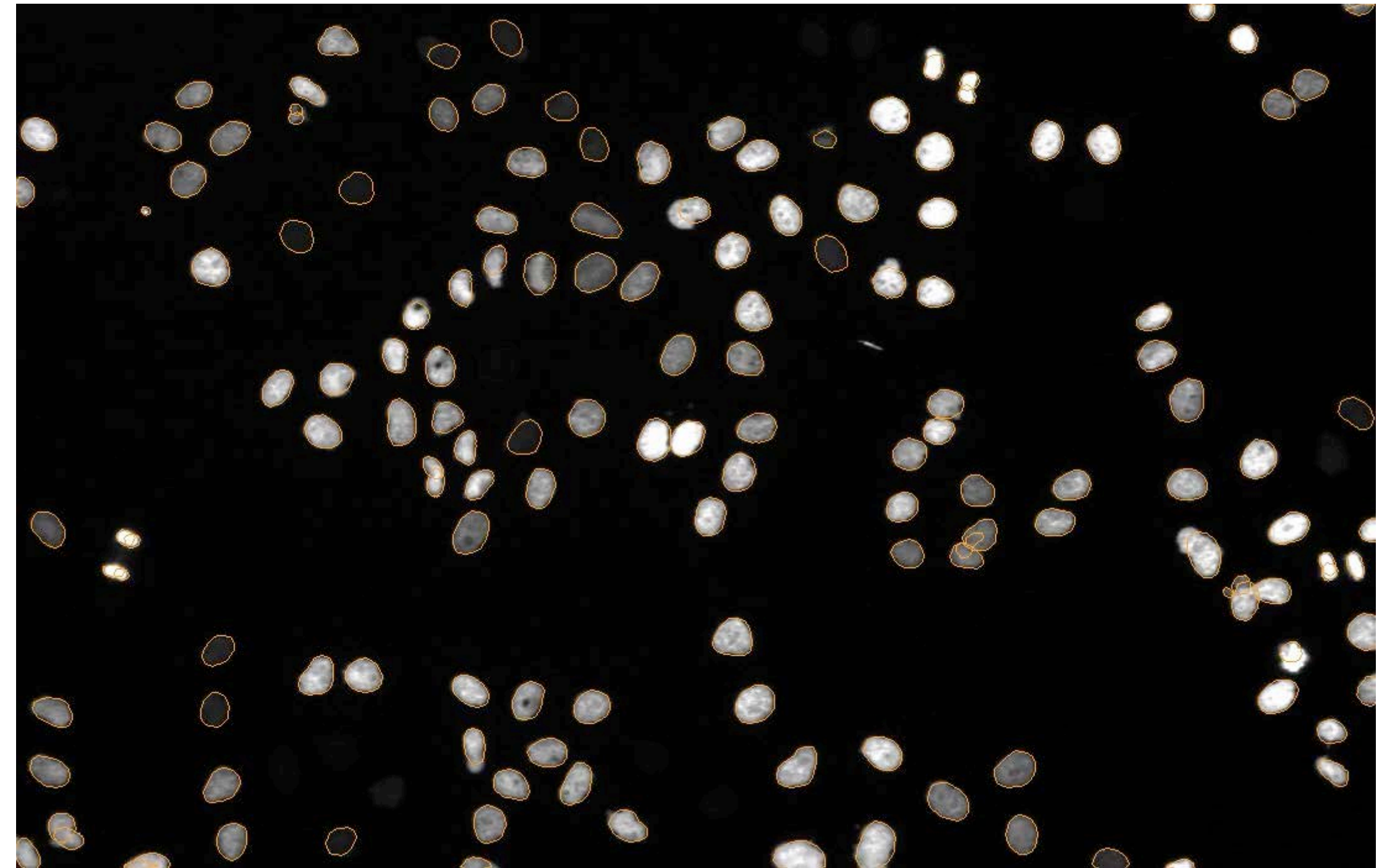
## Image denoising/restoration

Remove camera noise, remove blur...

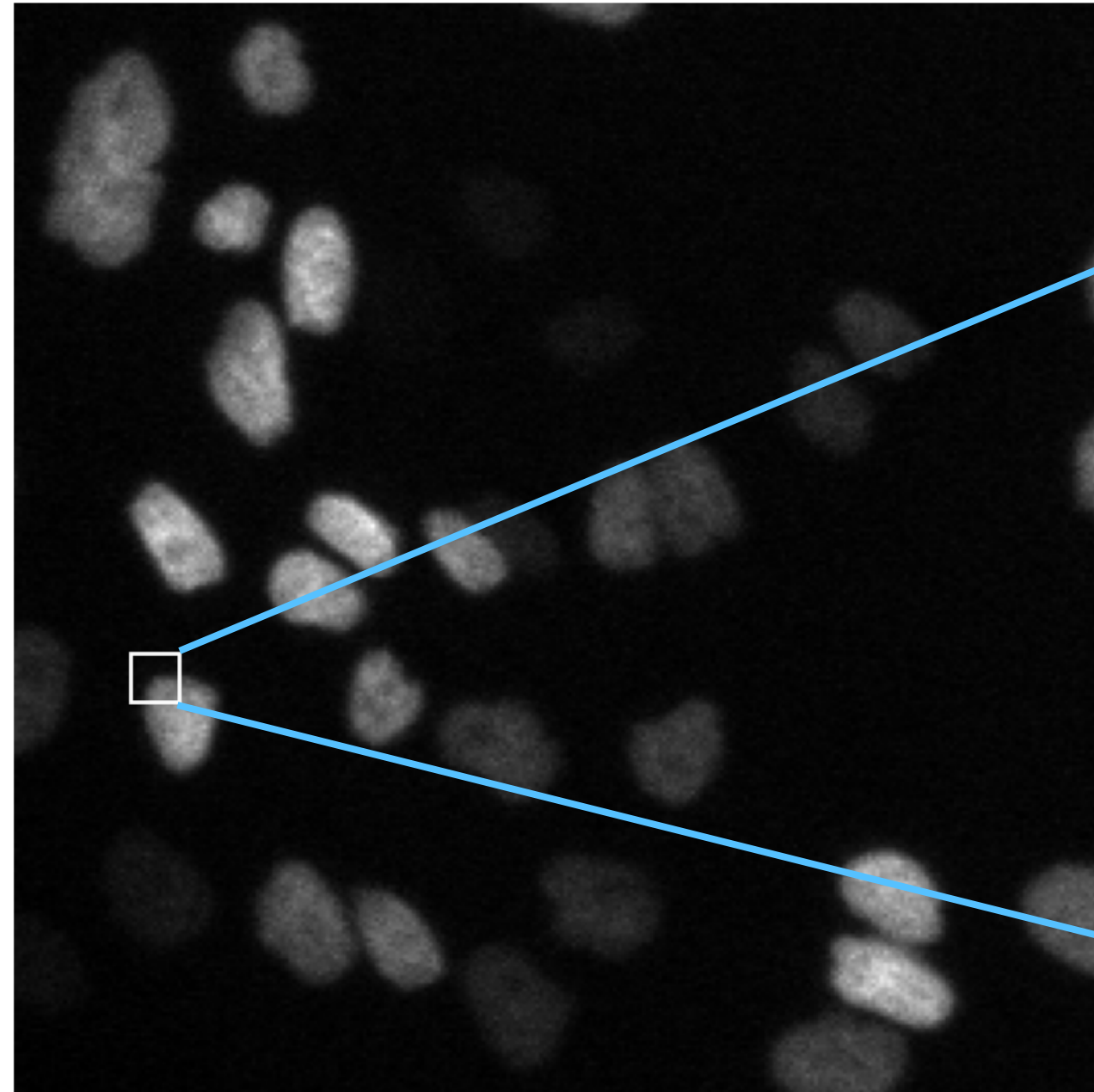


## Object Detection and Segmentation

Find contours of cells, detect spots, track cells over time, ...



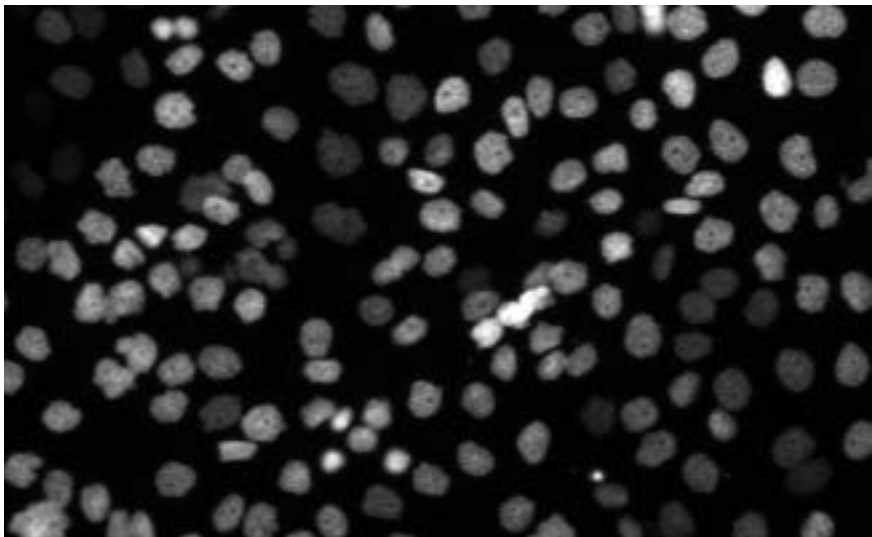
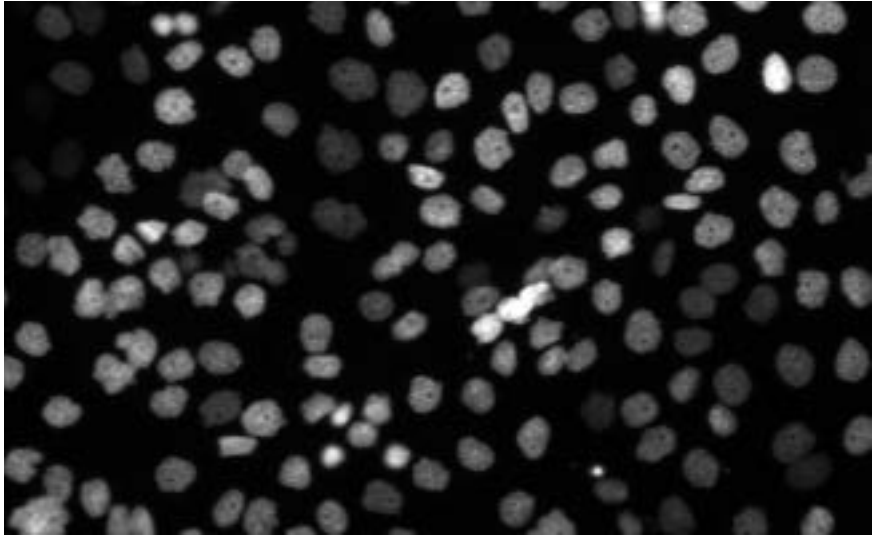
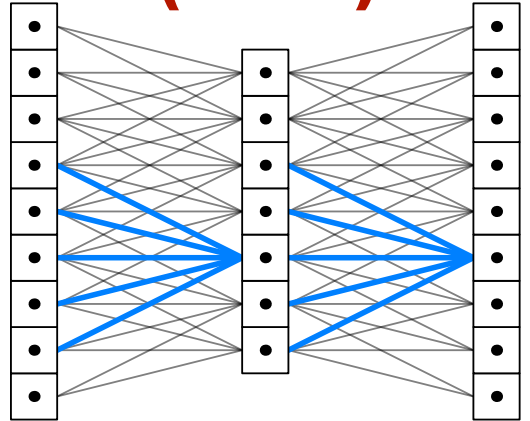
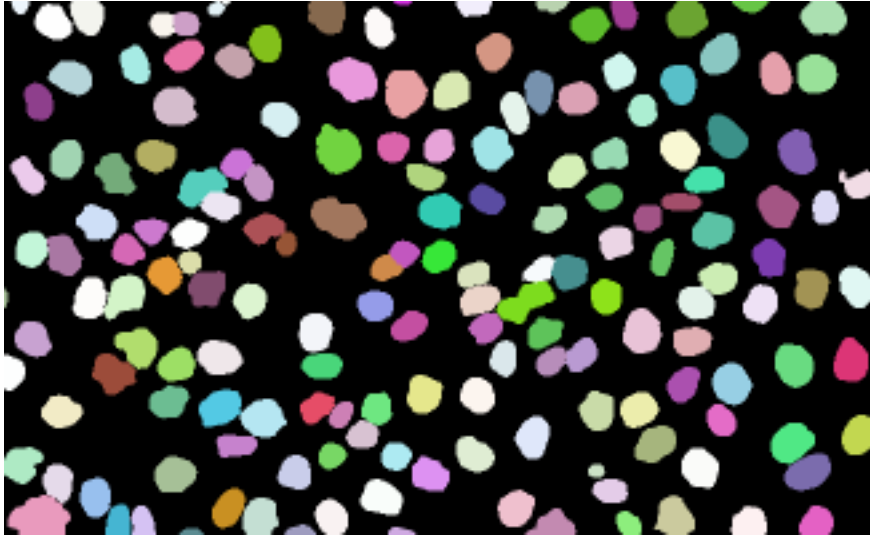
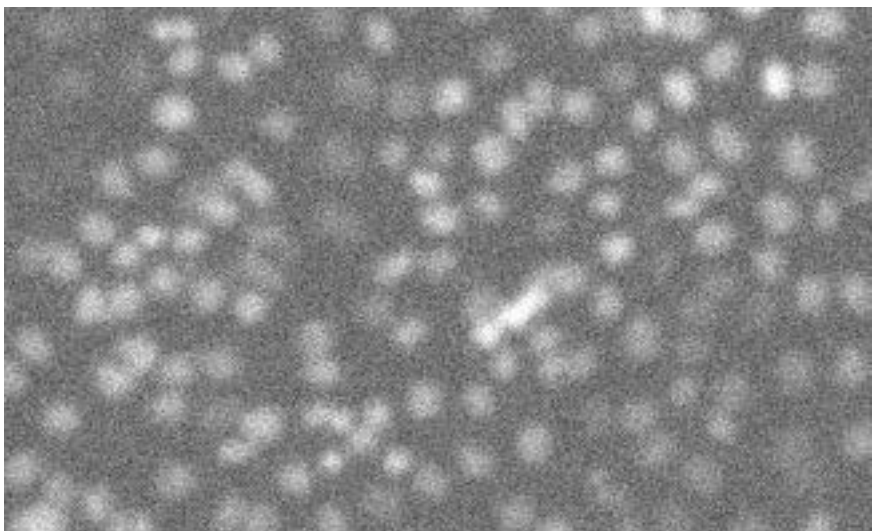
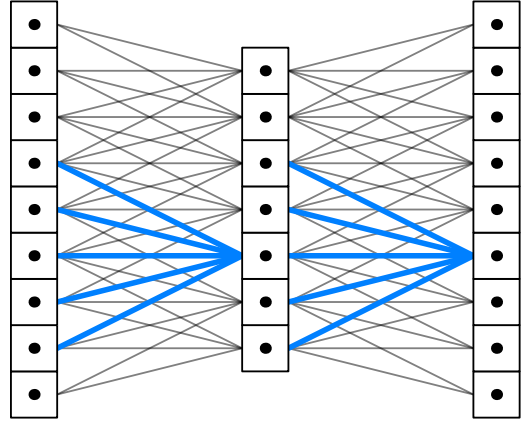
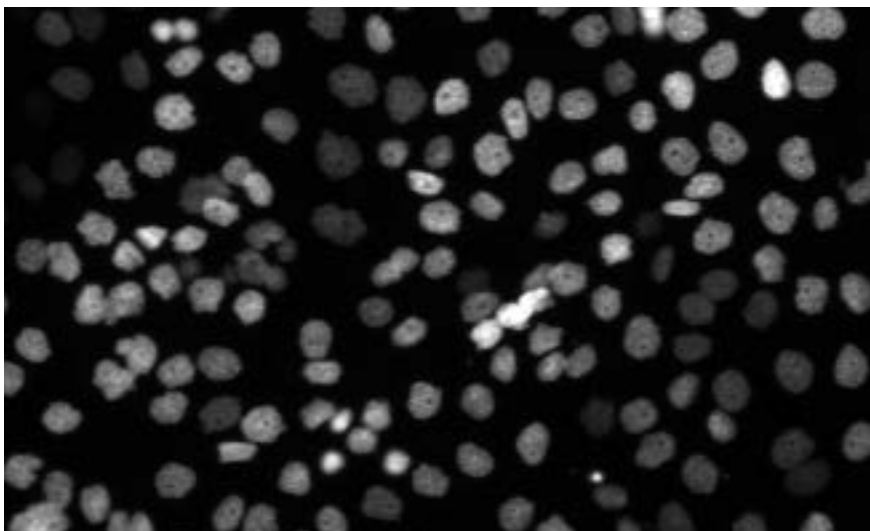
Acquired Image



Pixel level

13	7	10	11	18	17	19	6	11	5	12	13
12	11	6	7	12	13	13	8	11	7	10	13
10	9	17	11	17	17	13	19	14	14	16	31
9	6	14	21	20	16	21	17	30	24	18	14
19	14	23	28	18	23	17	29	24	16	31	26
14	19	17	15	24	21	26	41	48	48	48	56
11	11	14	23	33	46	106	111	112	116	108	103
16	16	17	26	41	80	153	188	169	199	203	182
16	12	40	35	77	123	158	223	229	207	240	209
13	25	16	50	112	175	203	227	234	252	222	224
18	18	16	54	131	198	221	233	247	199	219	232
21	18	36	73	126	187	234	256	240	225	236	258

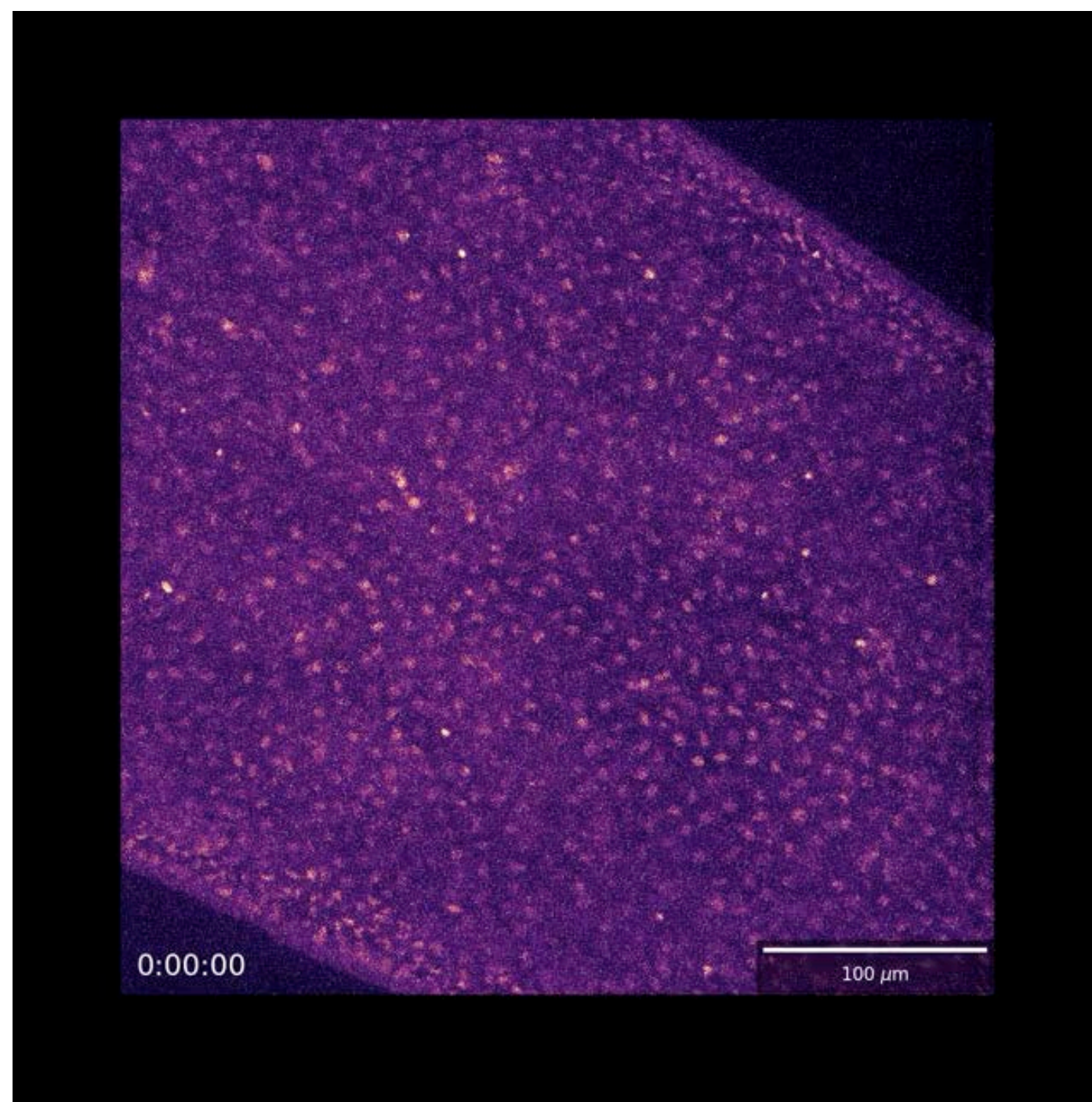
# Artificial neural networks are powerful for many bioimage tasks

Input	Output	Task
<i>Image</i> 	<i>Discrete-valued class</i>  "wildtype" or "mutant"	<b>Classification</b>
<i>Image</i> 	<b>Convolutional Neural Networks (CNN)</b> 	<i>Label Mask (class per pixel)</i> 
<i>Image</i> 		<i>Image</i> 
		<b>(Dense) Regression</b> <b>Image2Image</b> <b>Image Restoration</b>

# Extraction of quantitative biological information from digitised camera images (Computer Vision)

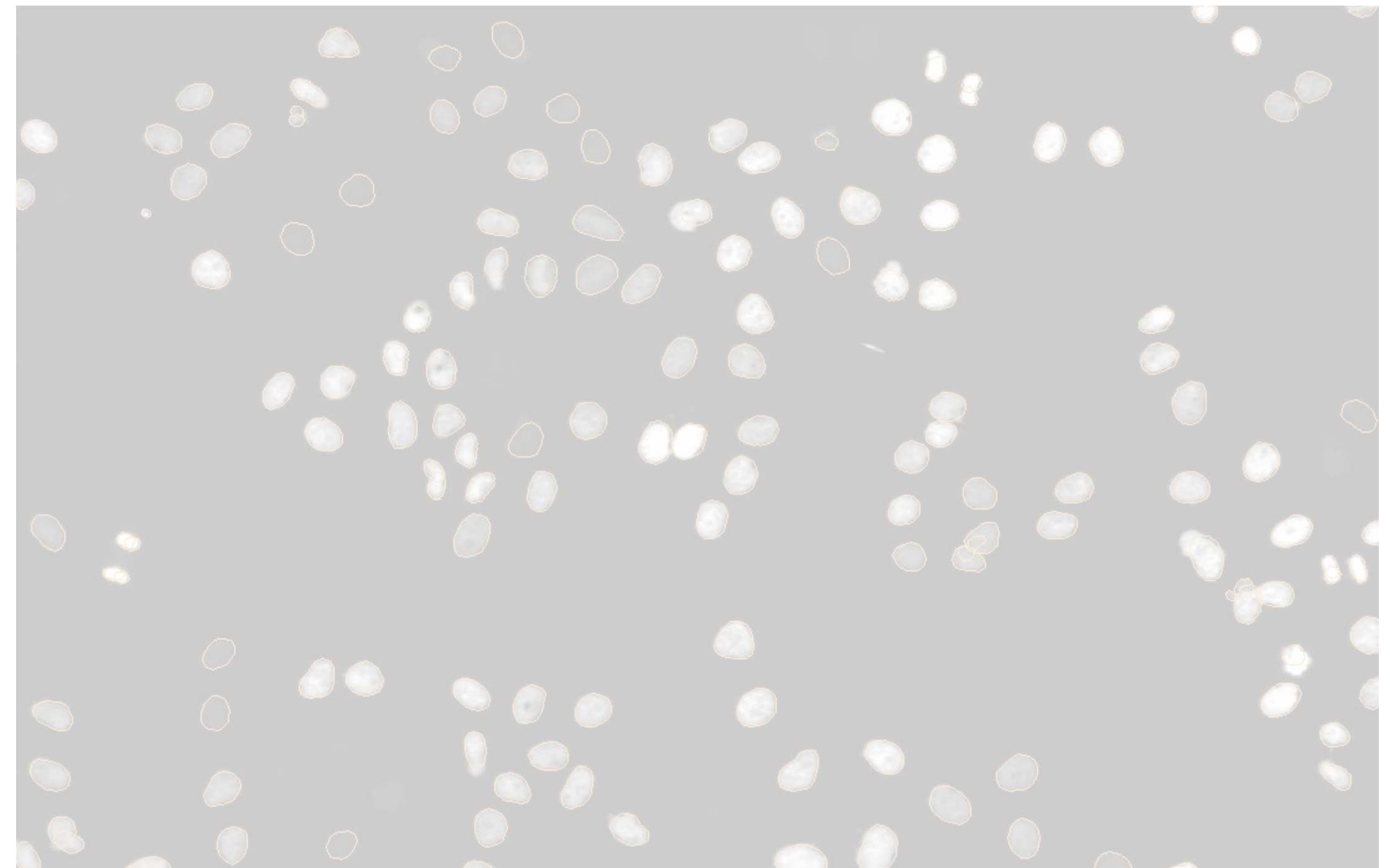
## Image Processing/Restoration

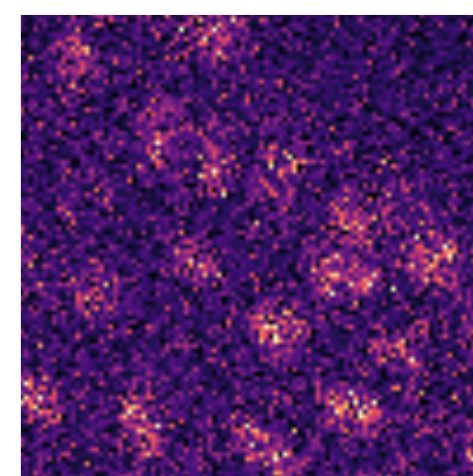
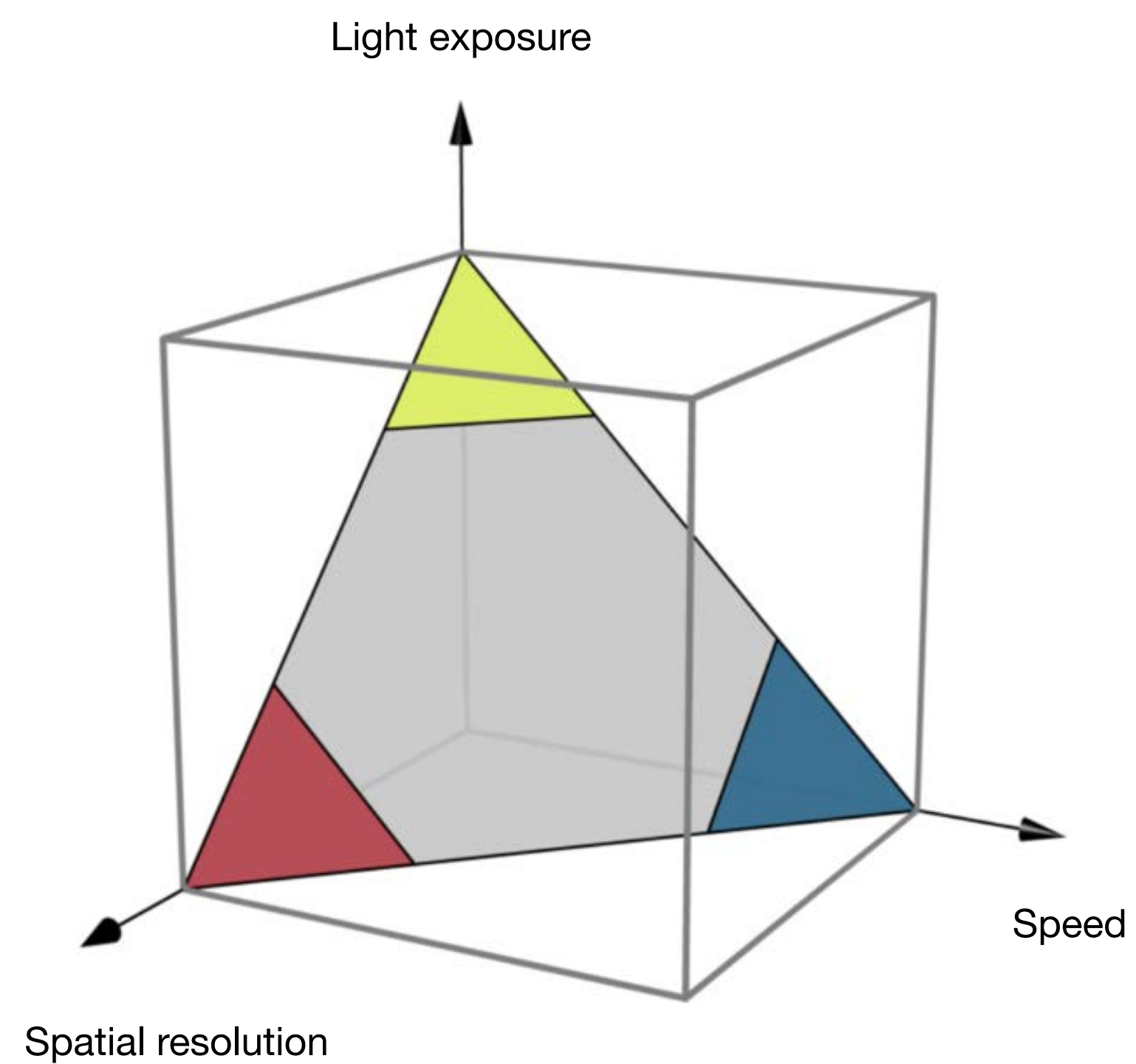
Remove camera noise, remove blur...



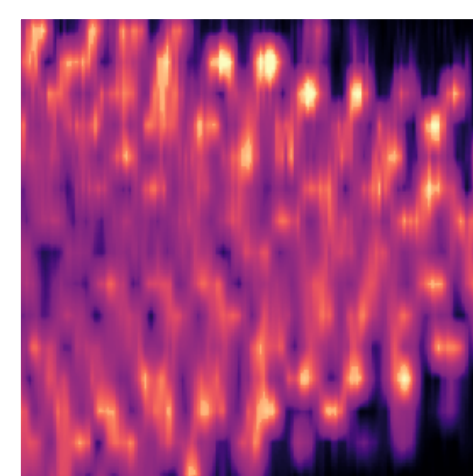
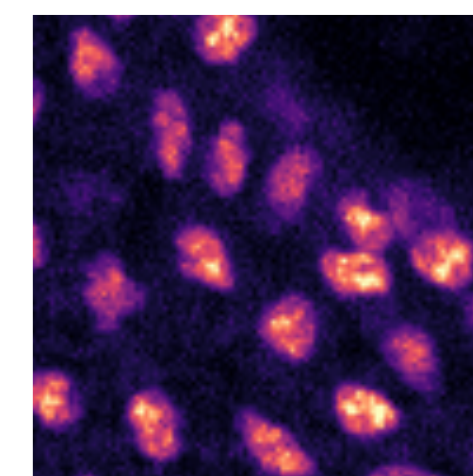
## Object Detection and Segmentation

Find contours of cells, track cells over time, ...

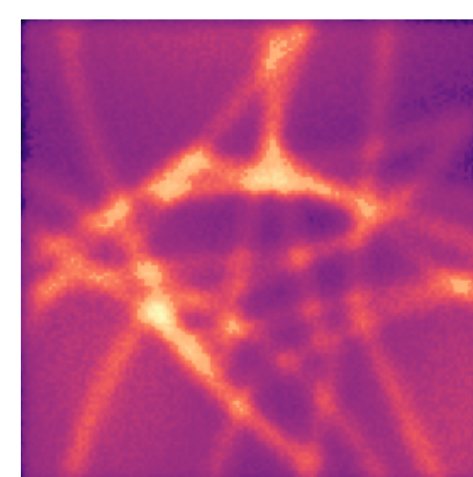
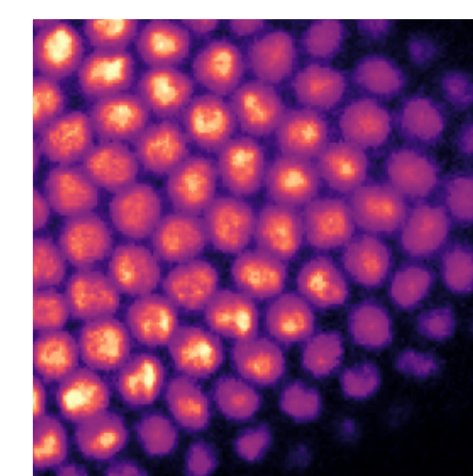




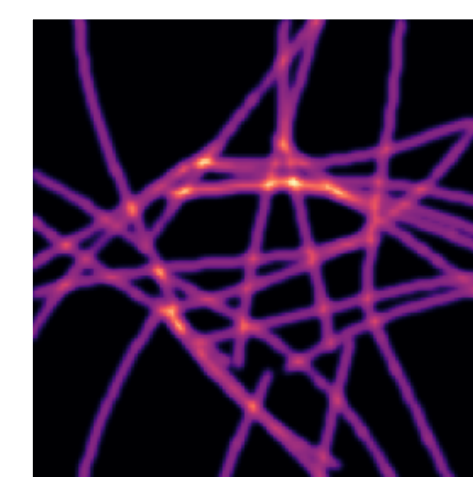
Light  
→  
*Denoising*



Speed  
→  
*Upsampling*

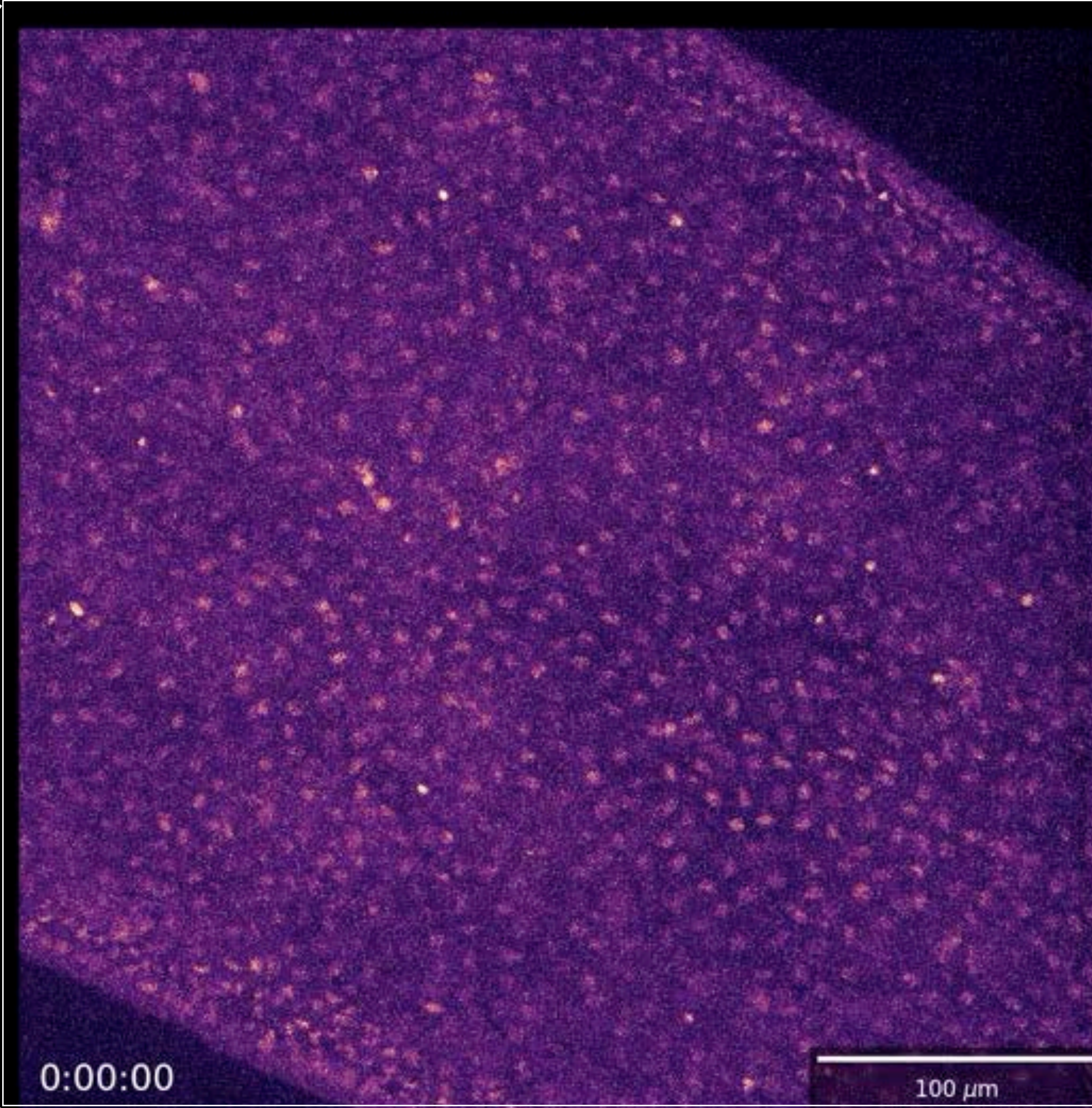
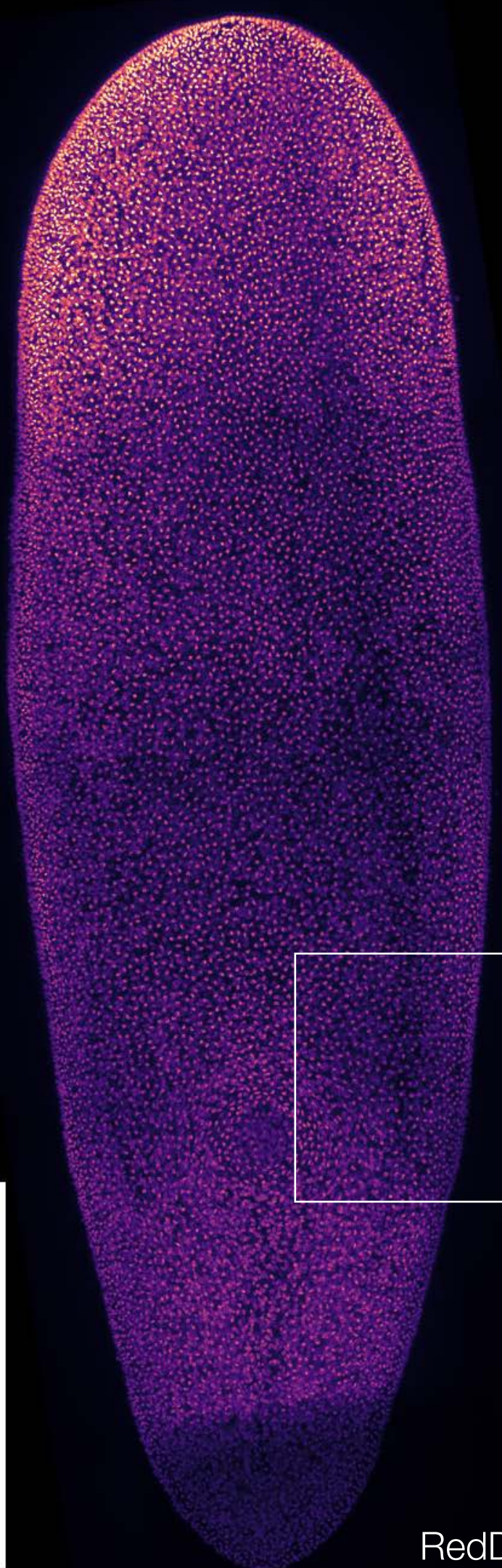


Spatial  
→  
*Deconvolution*



Planaria (Flatworm)

Low light dosage / low SNR  
(laser/exposure: 0.3%/10ms)



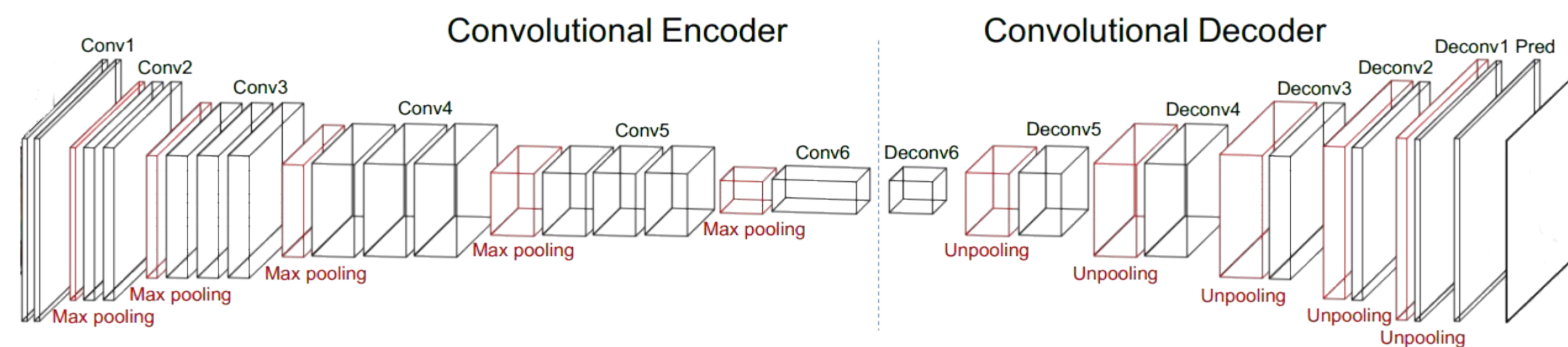
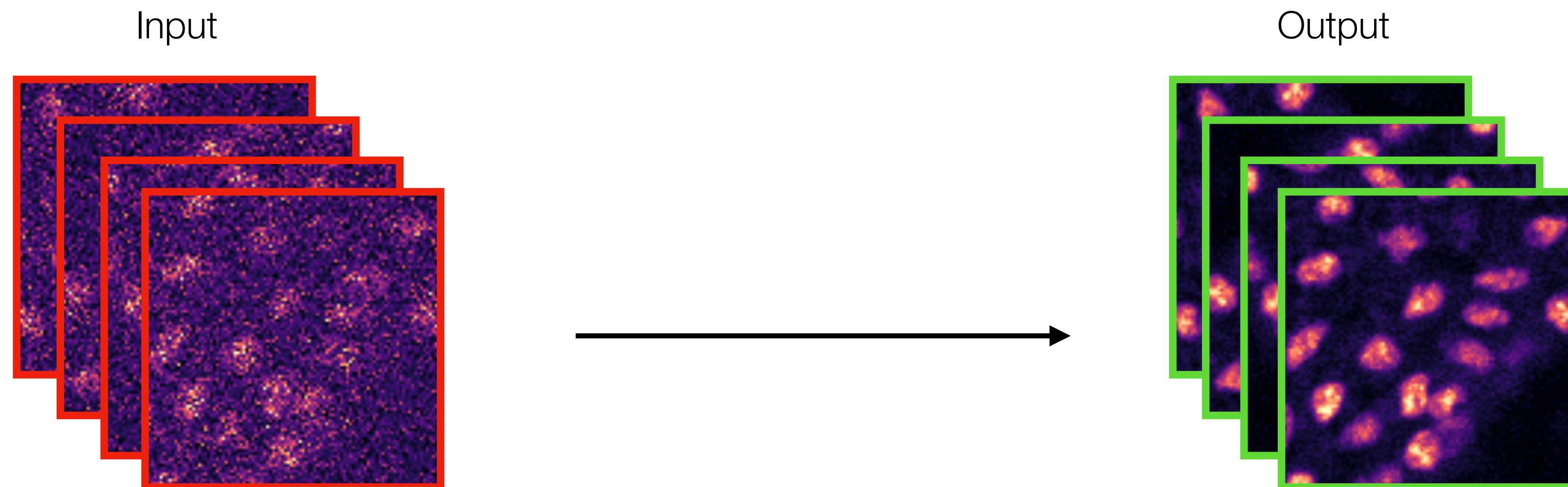
400µm

RedDot1 (Nuclei)

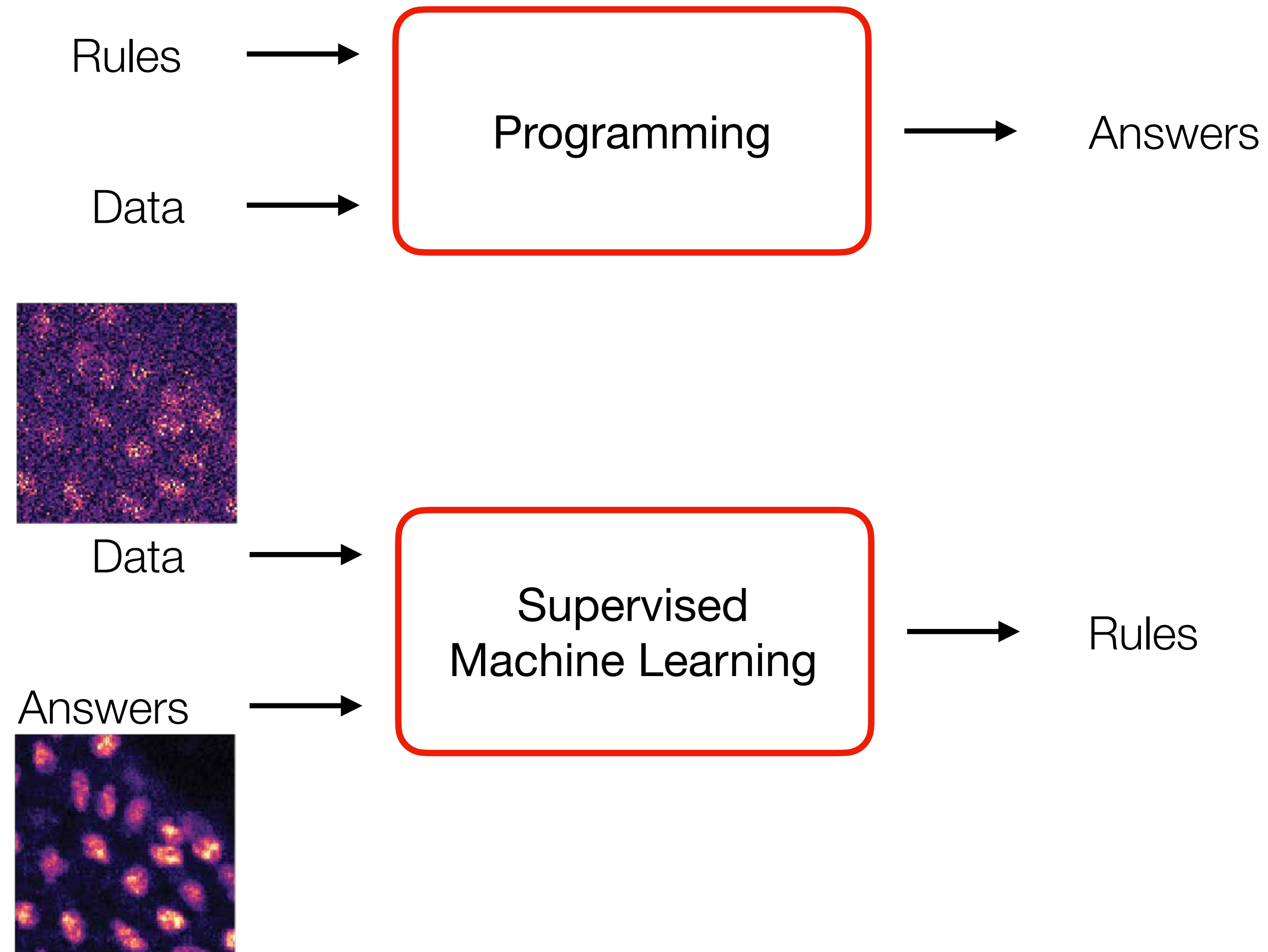
100 µm

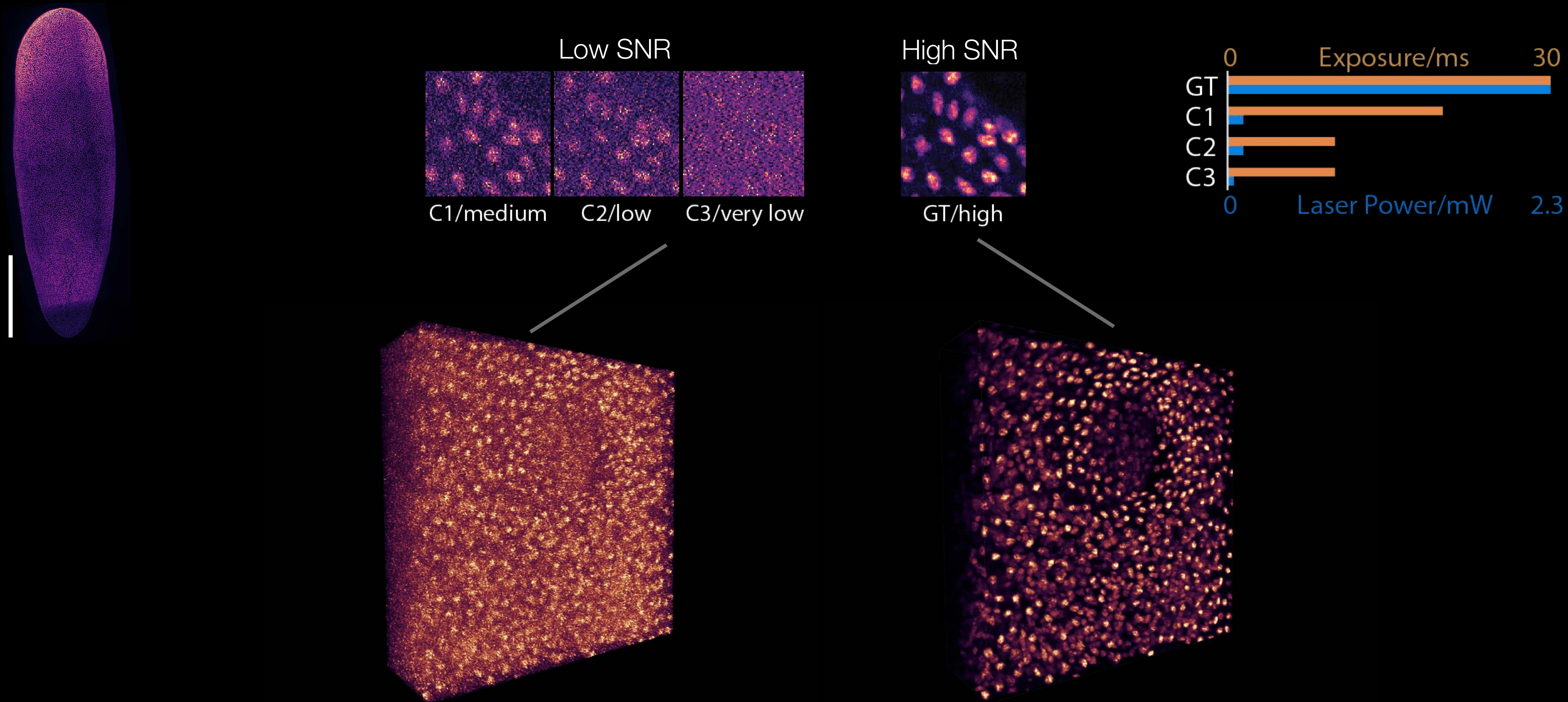
Data from Tobias Boothe

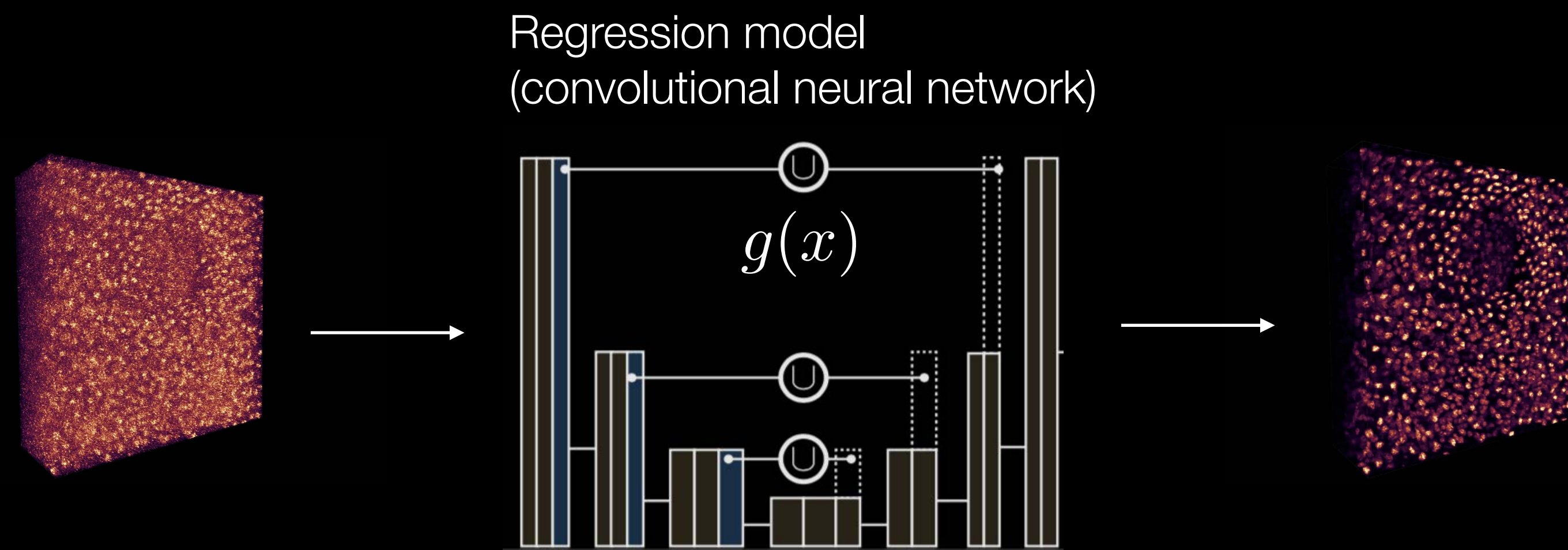
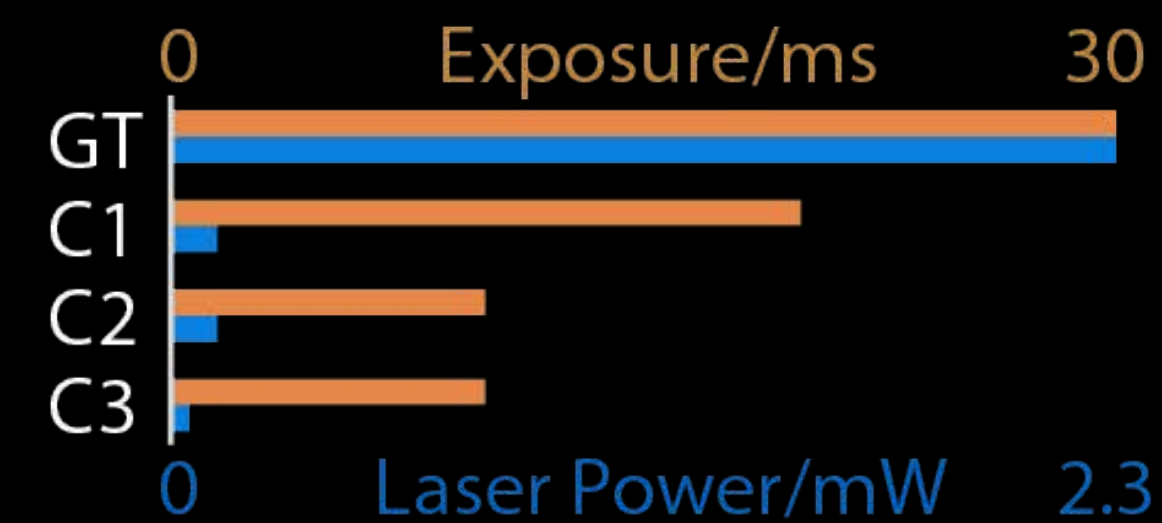
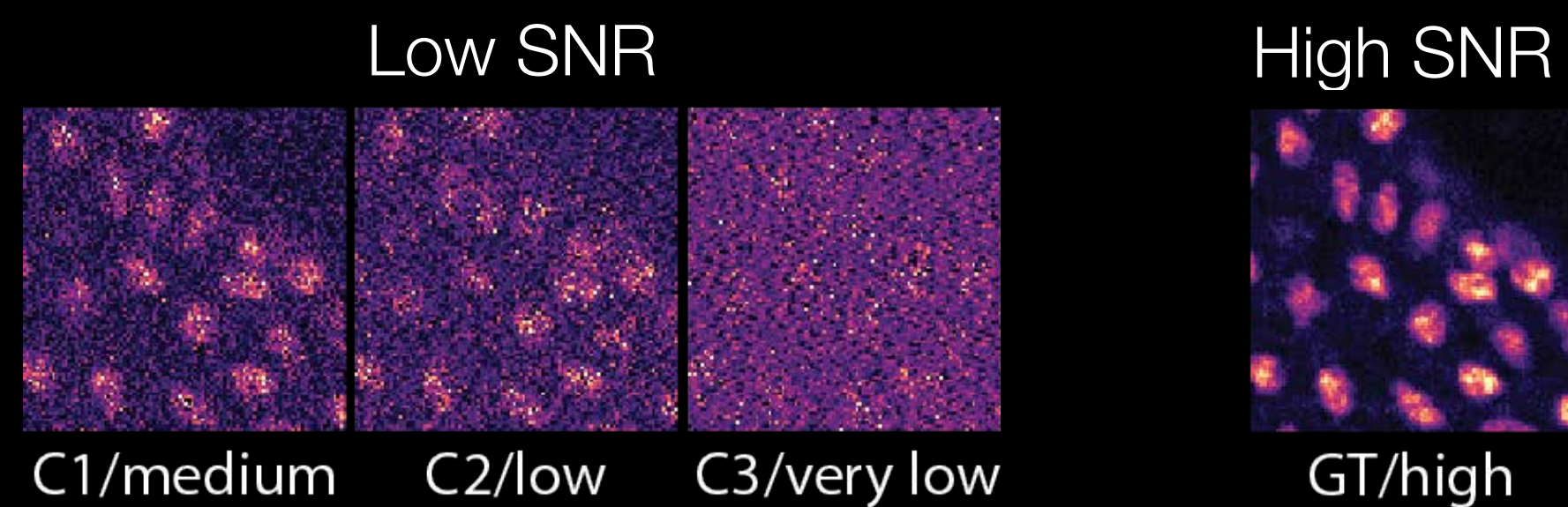
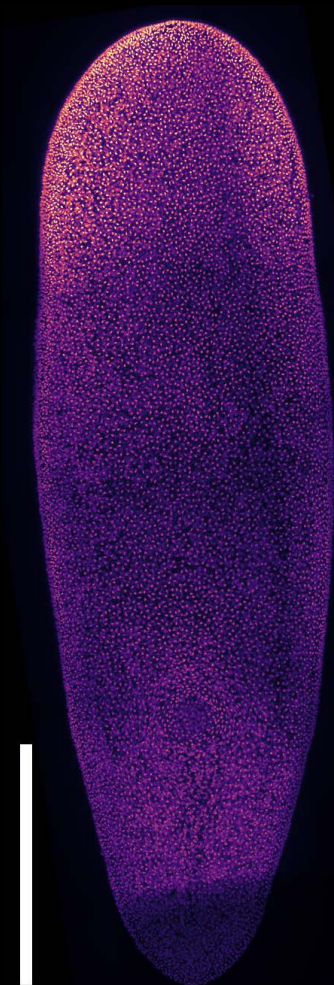
Pairwise training data of input/output pairs  
Learn a function that implements the mapping noisy  $\rightarrow$  clean



Artificial Neural Network



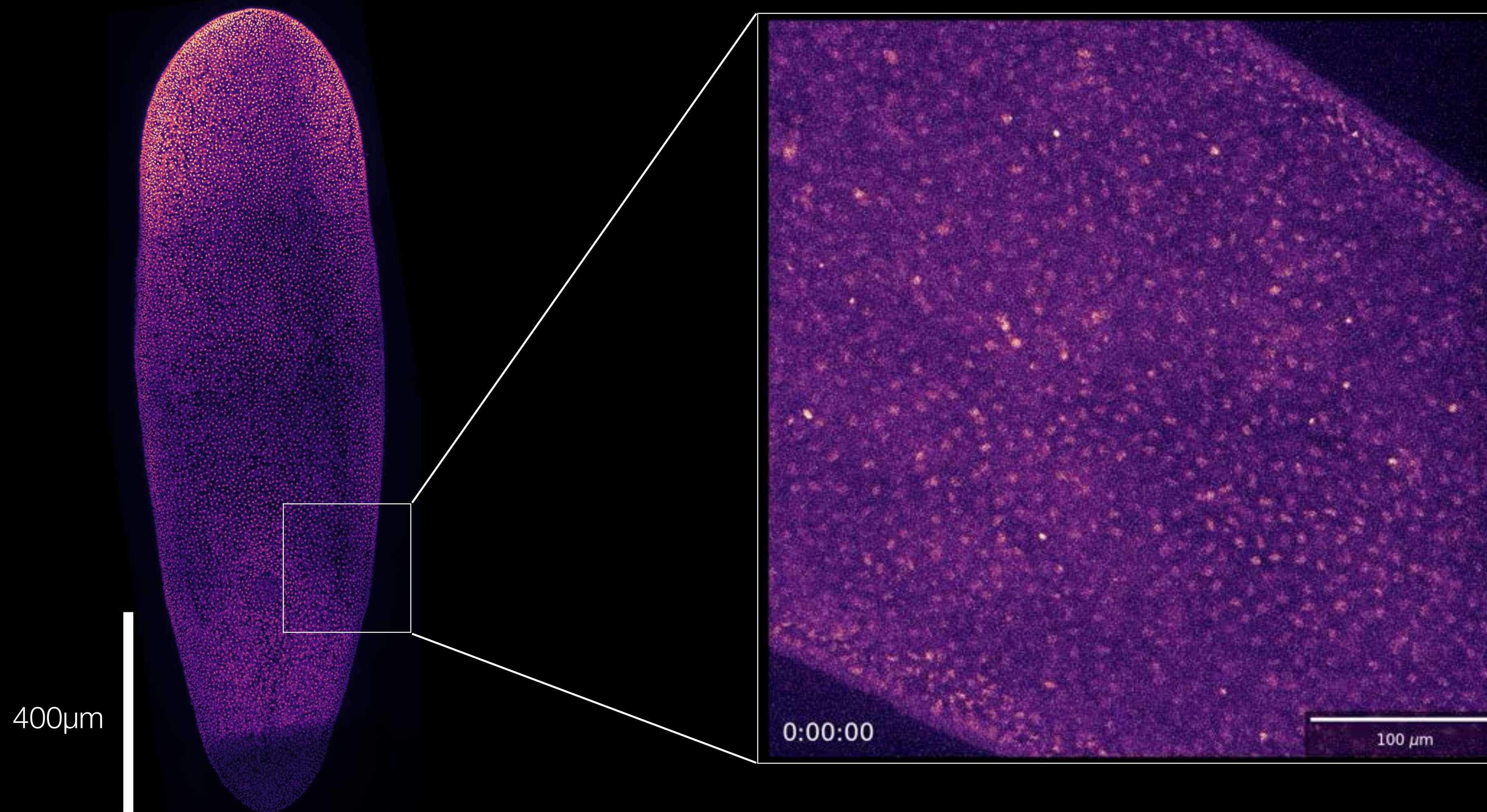


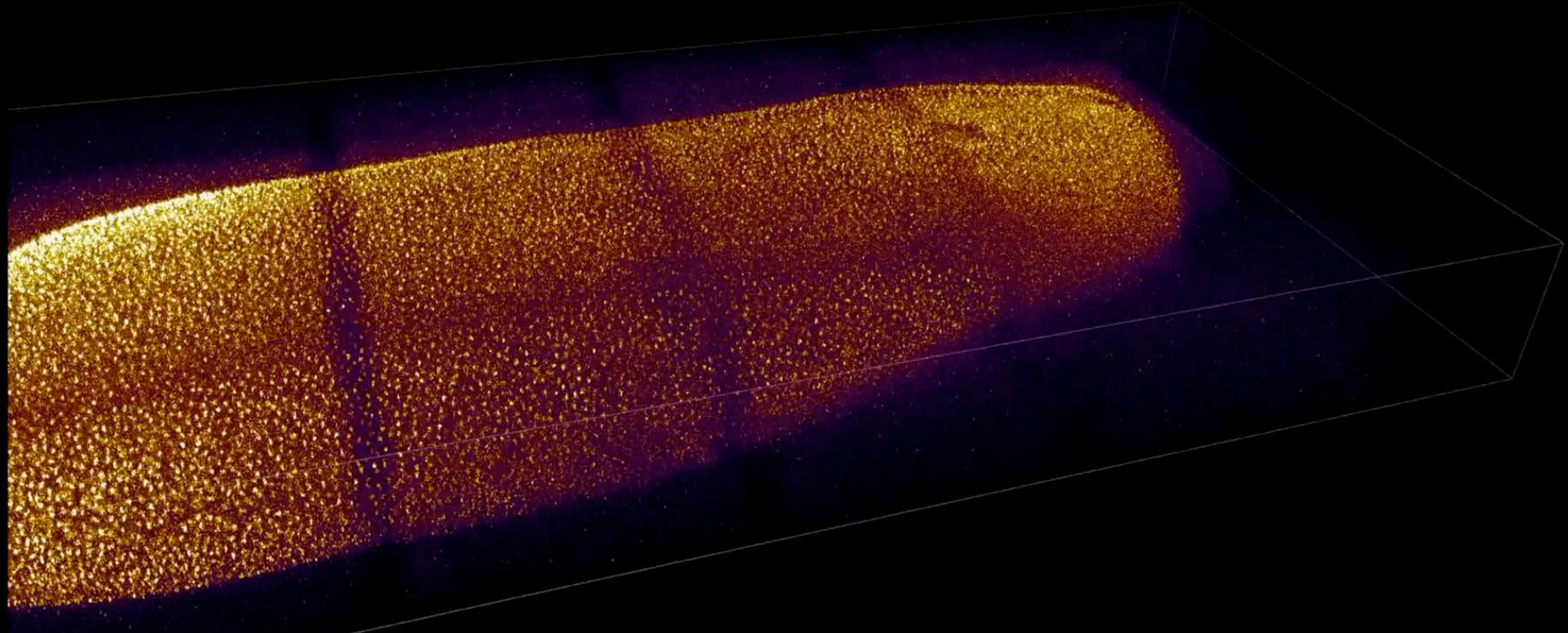


- Trained on 96 pairs (160GB)
- Training time ~ 8hrs

Planaria (Flatworm)

Restoration

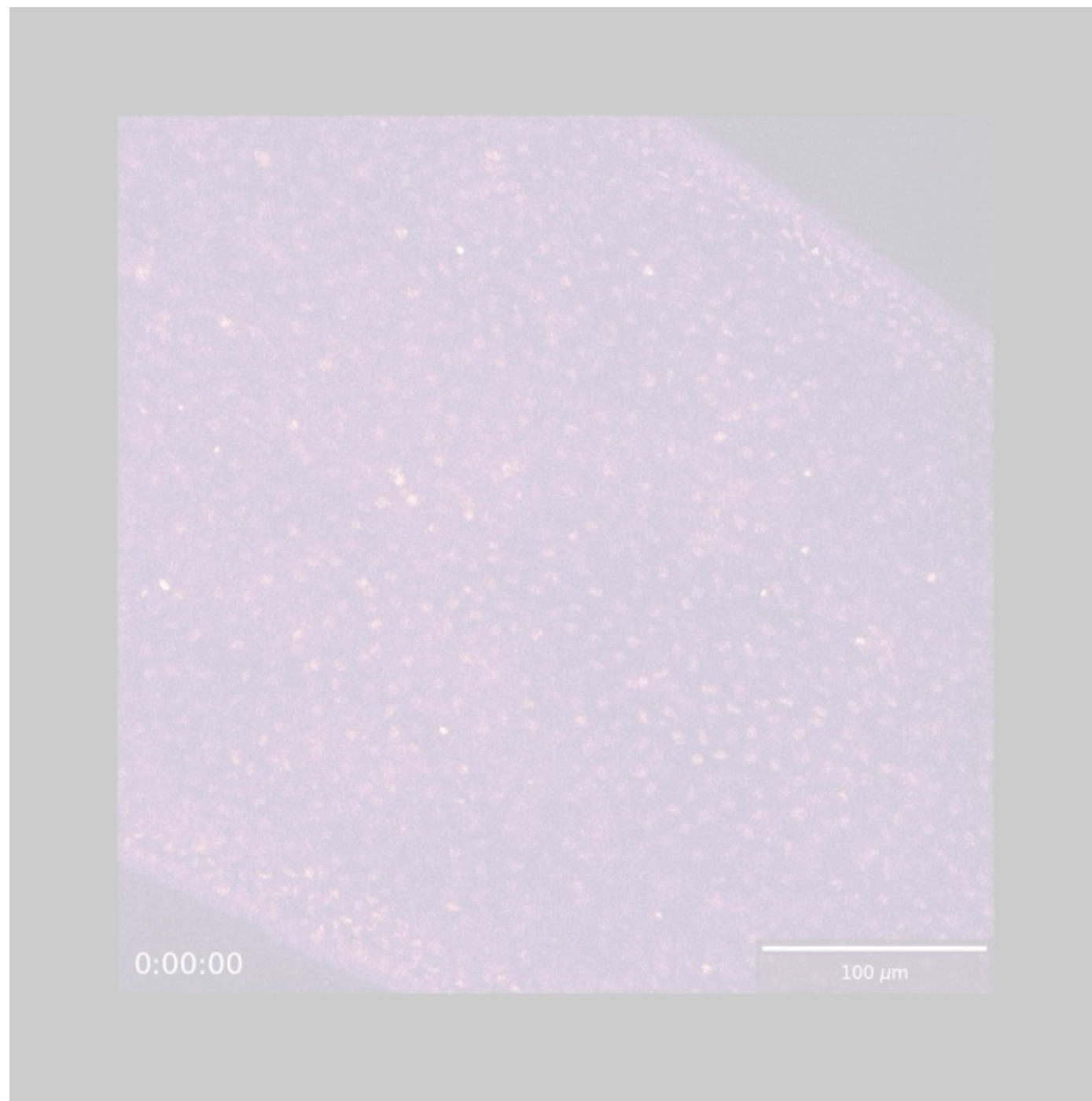




# Extraction of quantitative biological information from digitised camera images (Computer Vision)

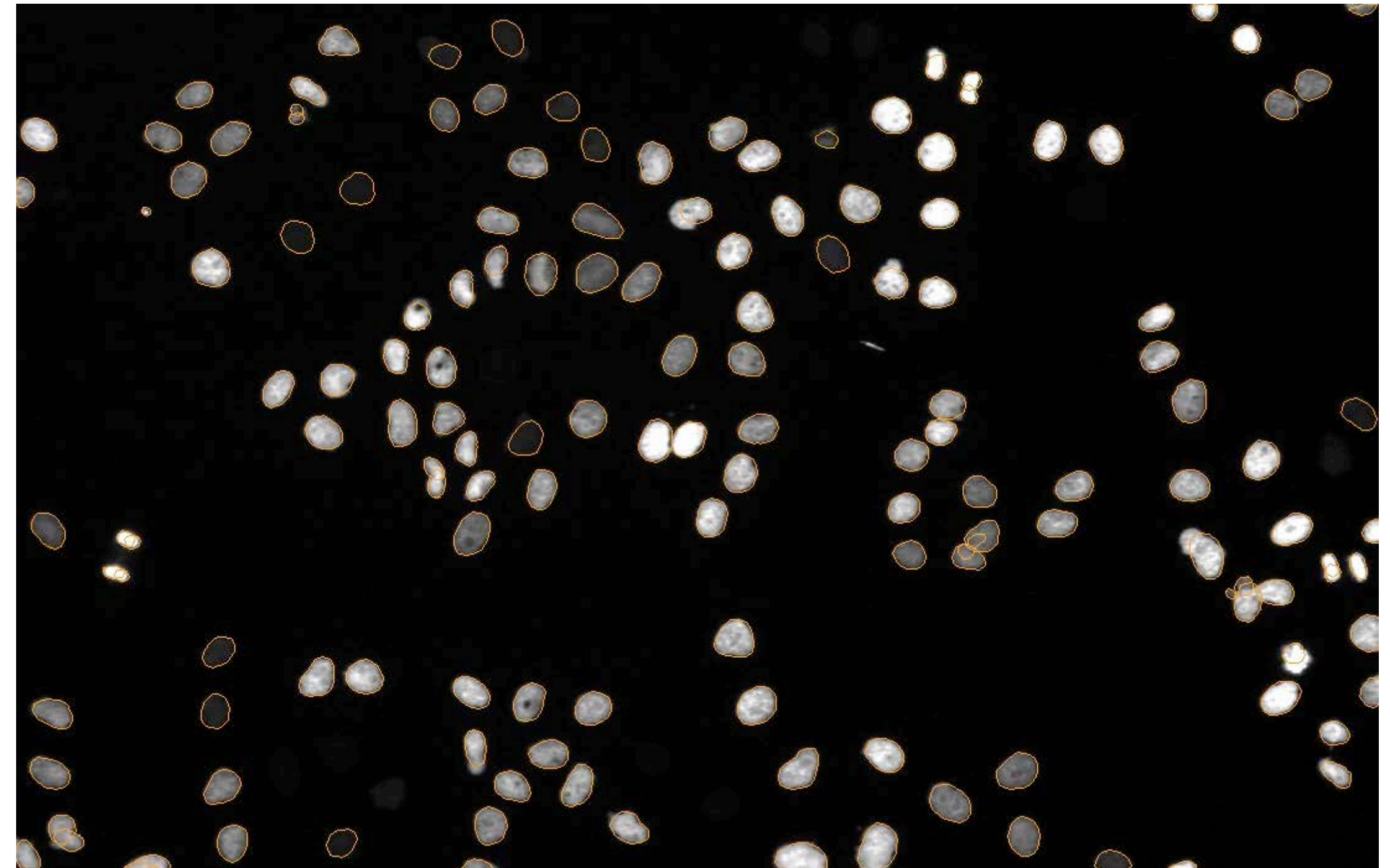
## Image Denoising/Restoration

Remove camera noise, remove blur...



## Object Detection and Segmentation

Find contours of cells, detect spots, track cells over time, ...



## Single Object

## Multiple Objects

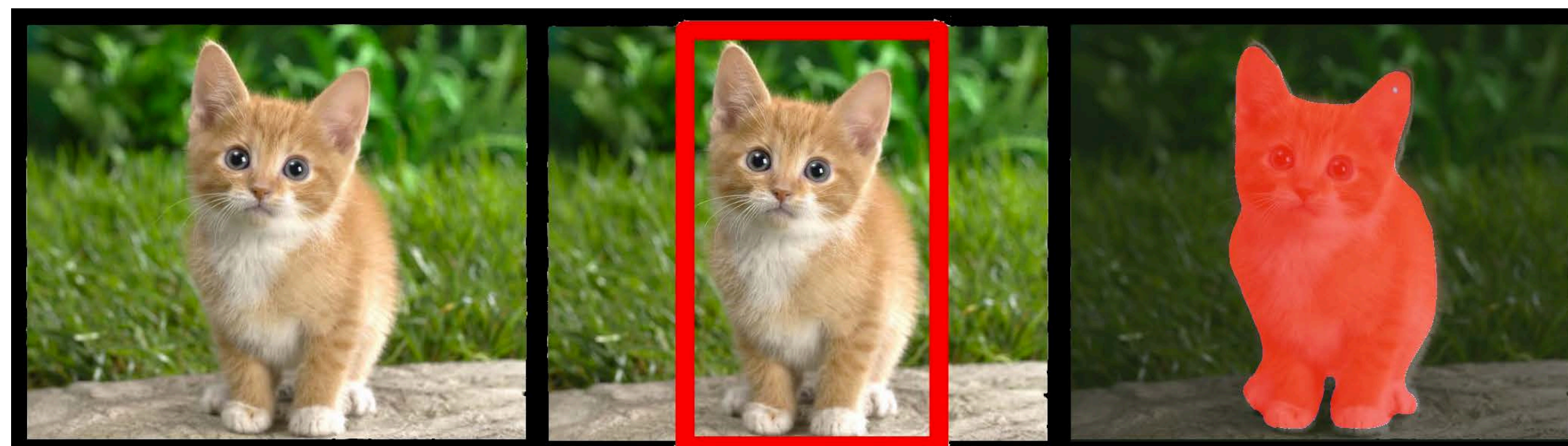
Image Classification

Object Detection (Localization)

Dense/Semantic Segmentation

Object Detection

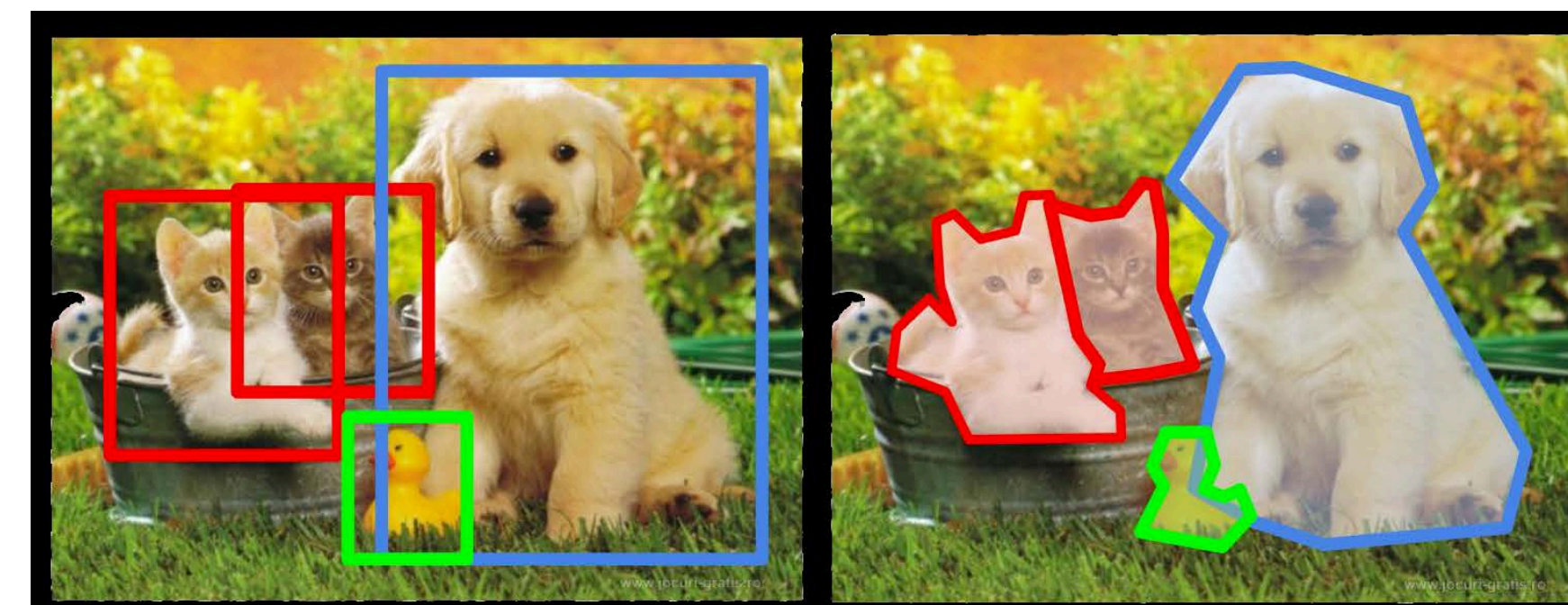
Instance Segmentation



Cat

Cat

Cat



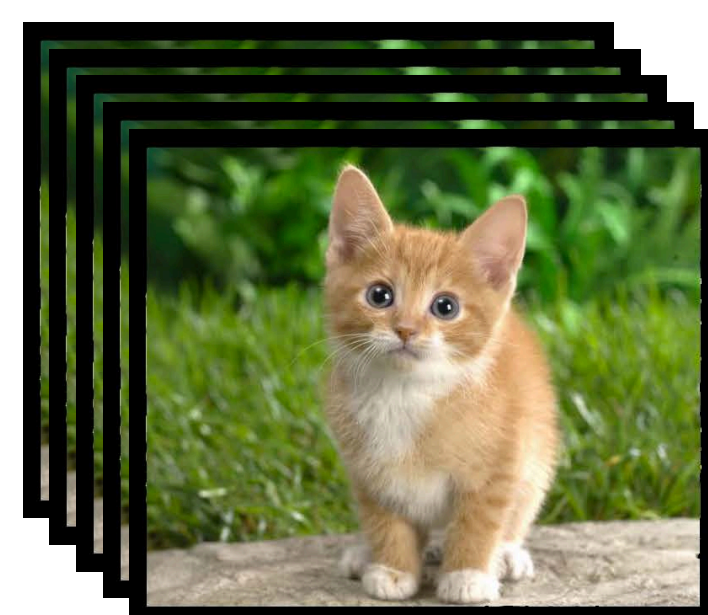
Cat, Dog, Duck

Cat, Dog, Duck

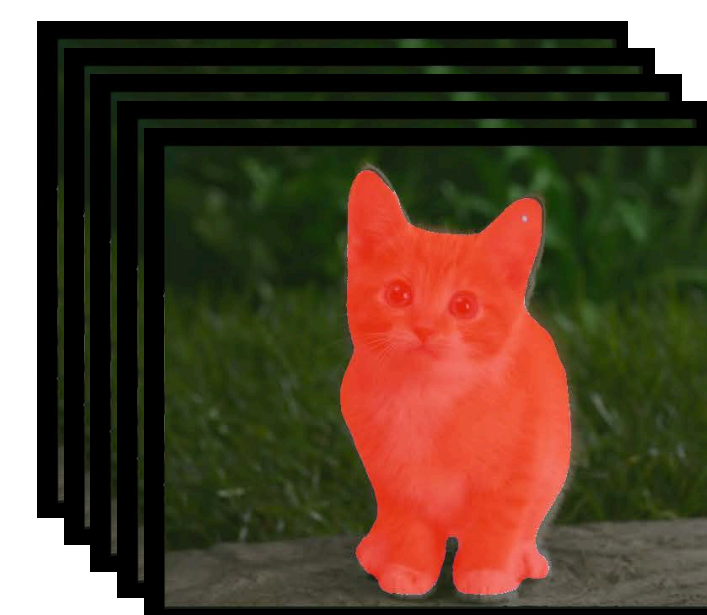
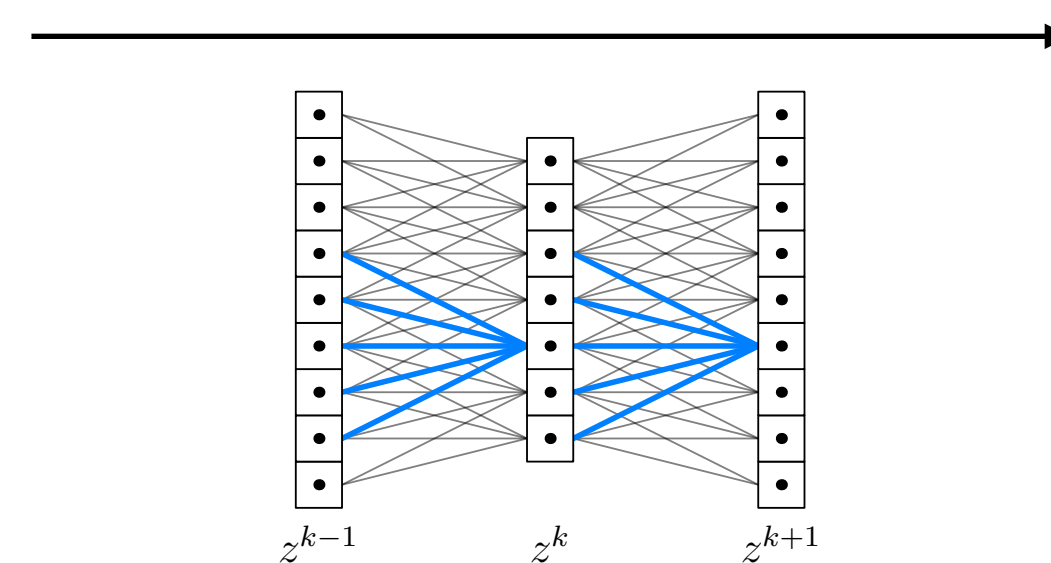
Currently the most successful paradigm: Supervised deep learning

Input

Output

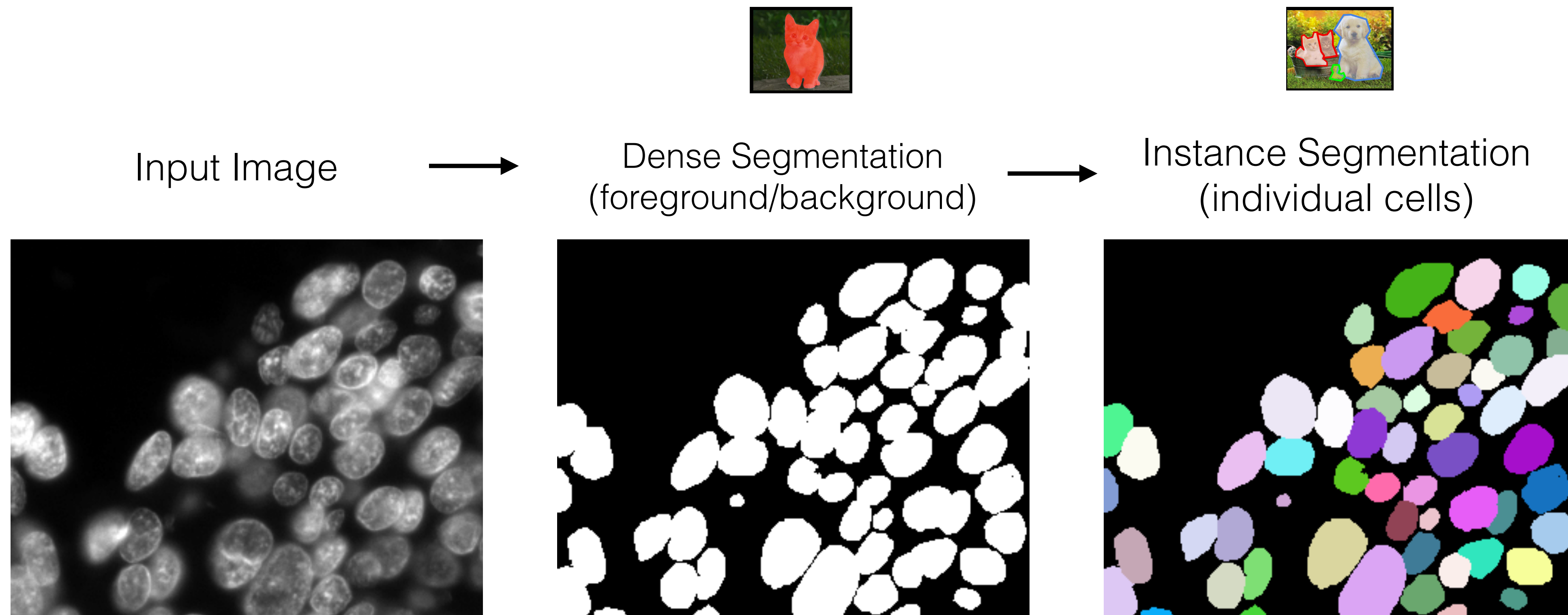


Deep Neural Network

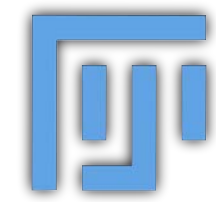


- U-Net Ronneberger et al (2015)
- YOLO Redmond et al (2016)
- Mask-RCNN He et al (2017)

Deep Neural Networks trained on annotated training data (ground truth, GT)

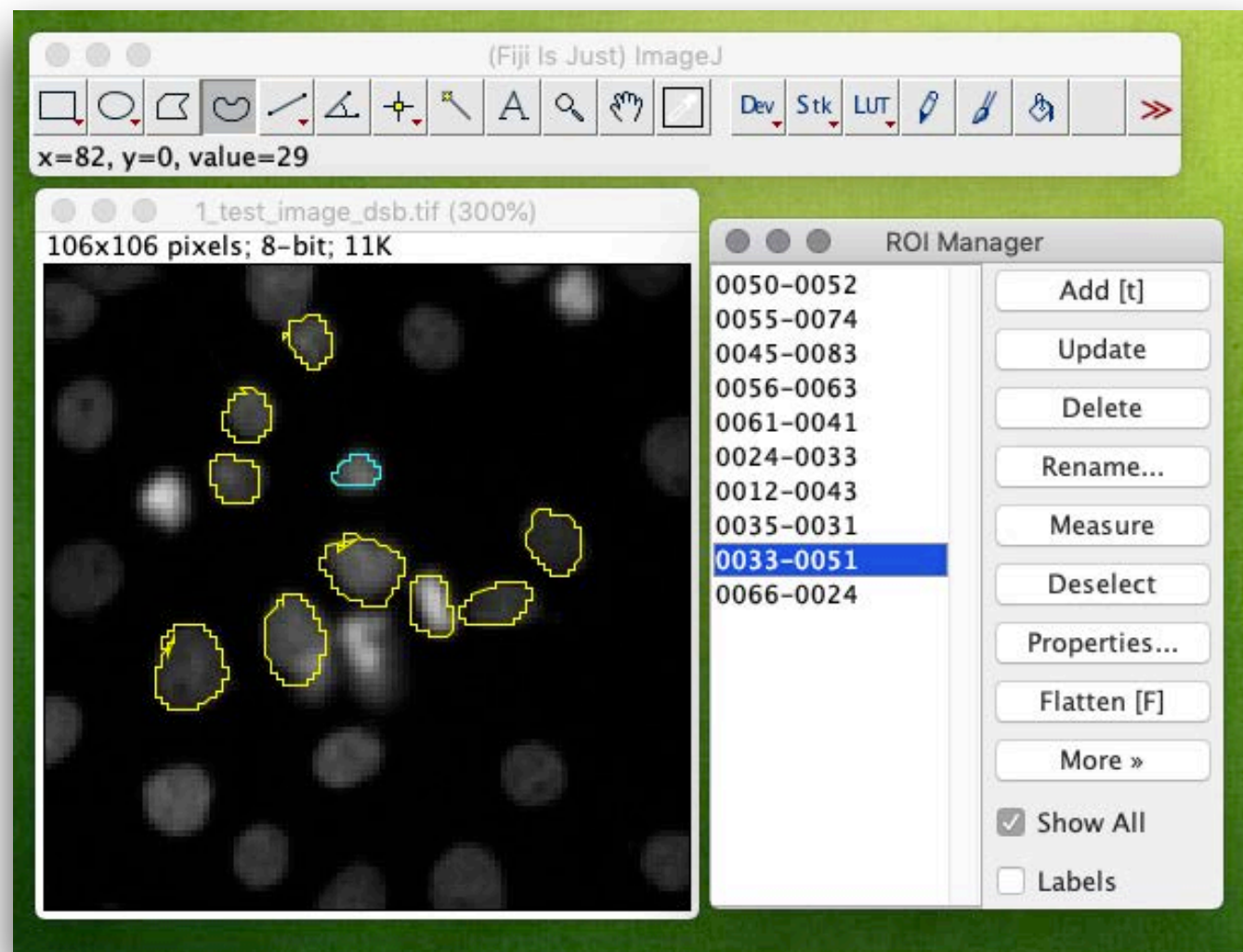


Challenges: many crowded objects, noisy images,  
different from natural images (so knowledge cannot be directly transferred)



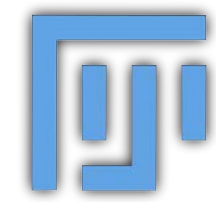
## Fiji/ImageJ

- Draw Roi per object (Roi-Manager)
- Convert to label mask (e.g. see [here](#))



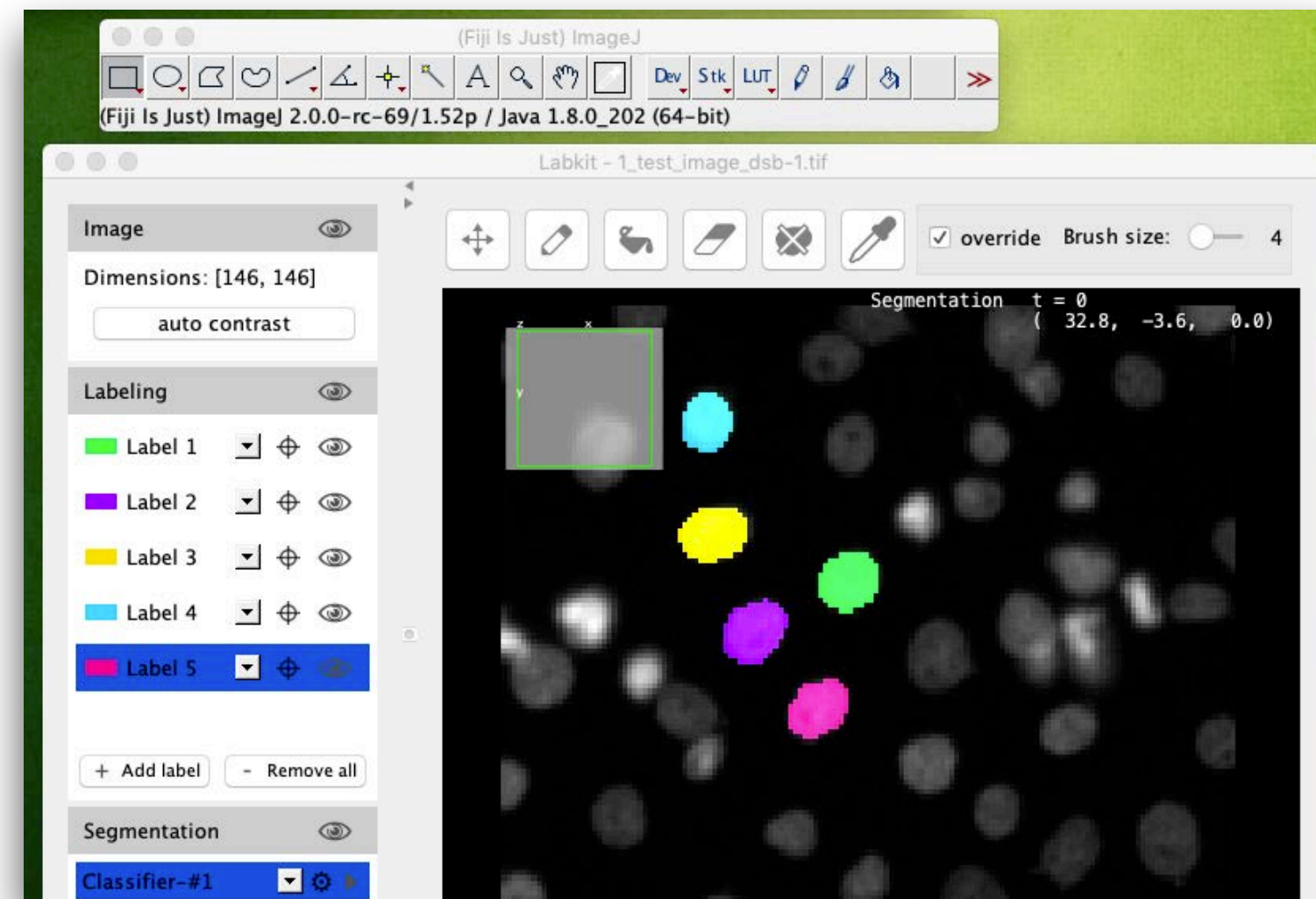
<http://fiji.sc/>

Schindelin et al. (2012)  
Schneider, Rasband et al (2012)



## Fiji + LabKit

- Directly draw label mask



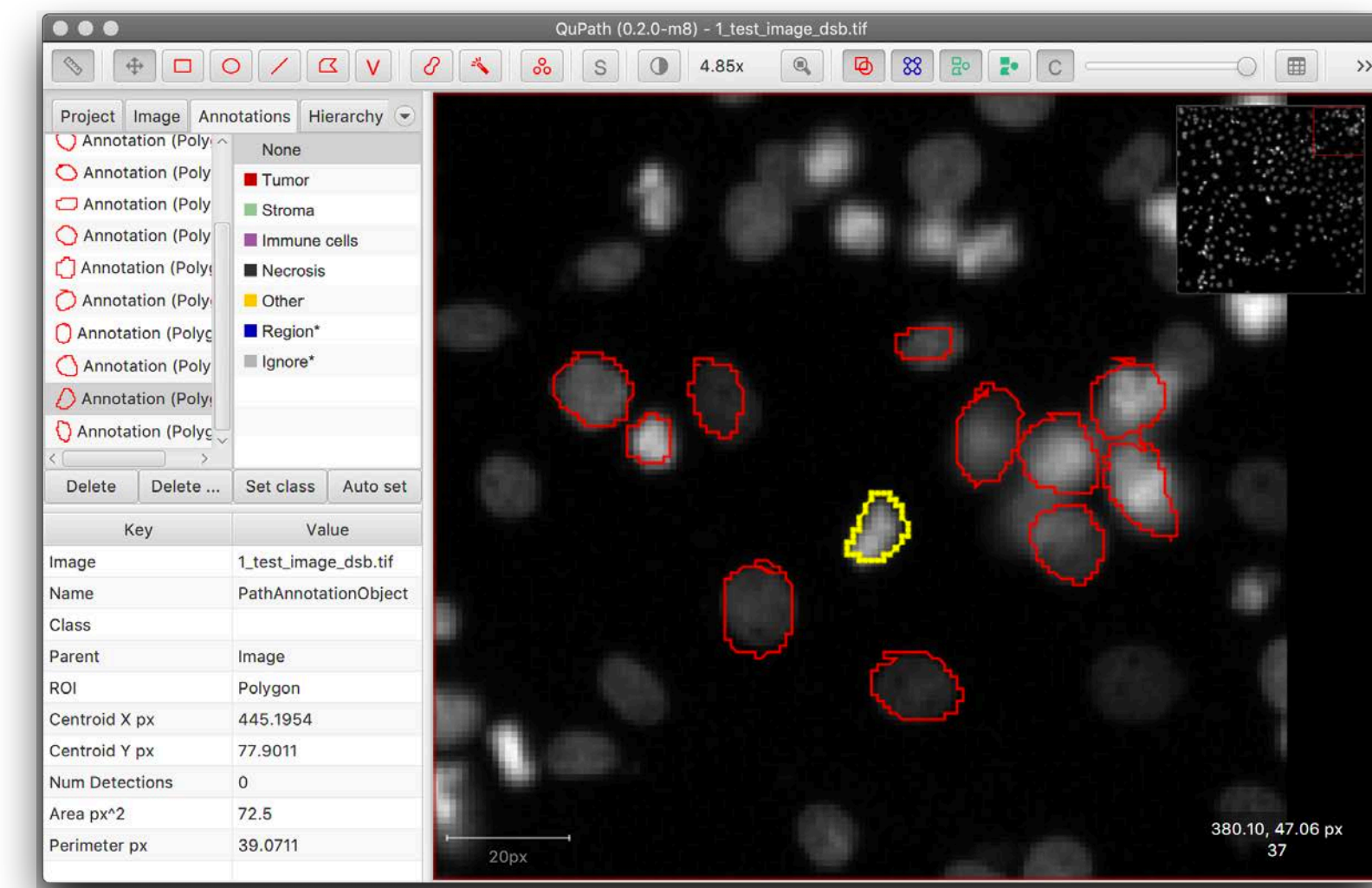
<https://imagej.net/Labkit>

M. Arzt, MPI-CBG



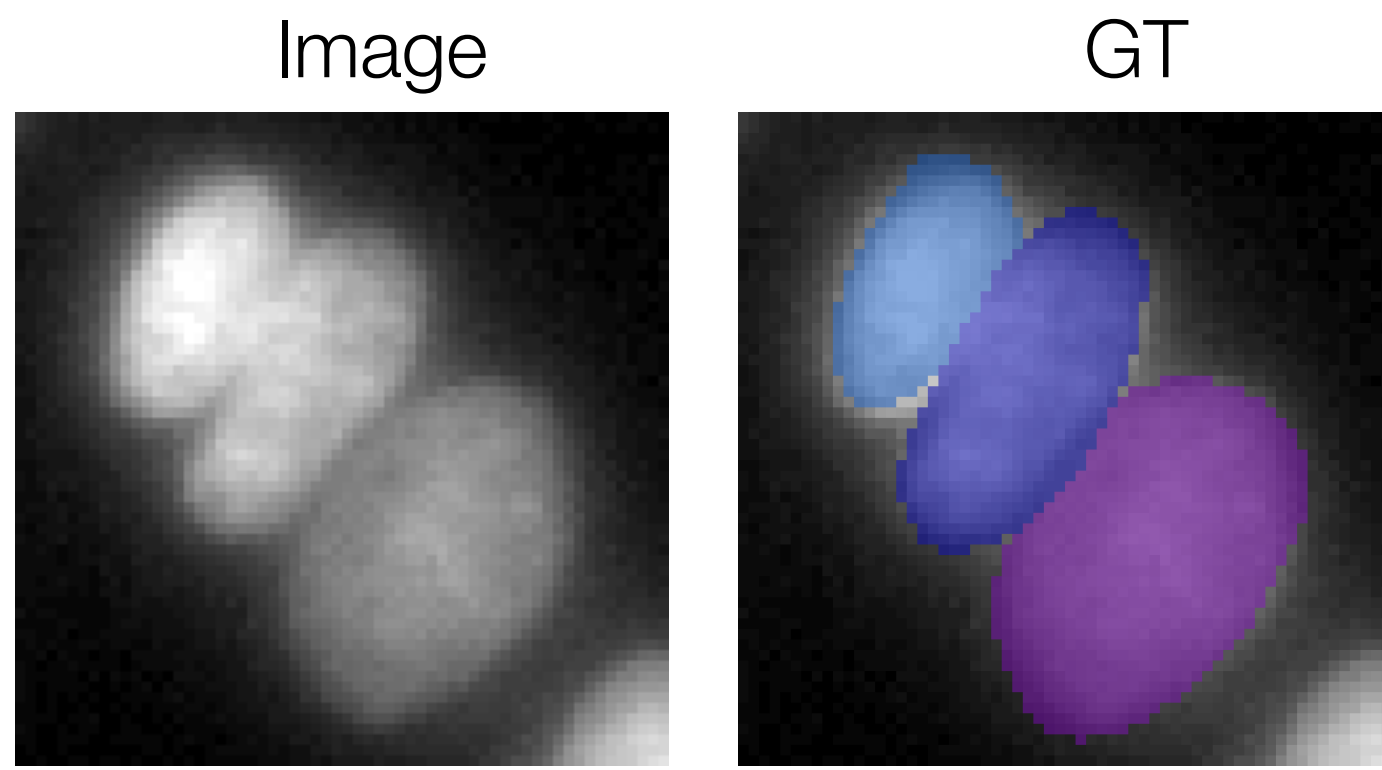
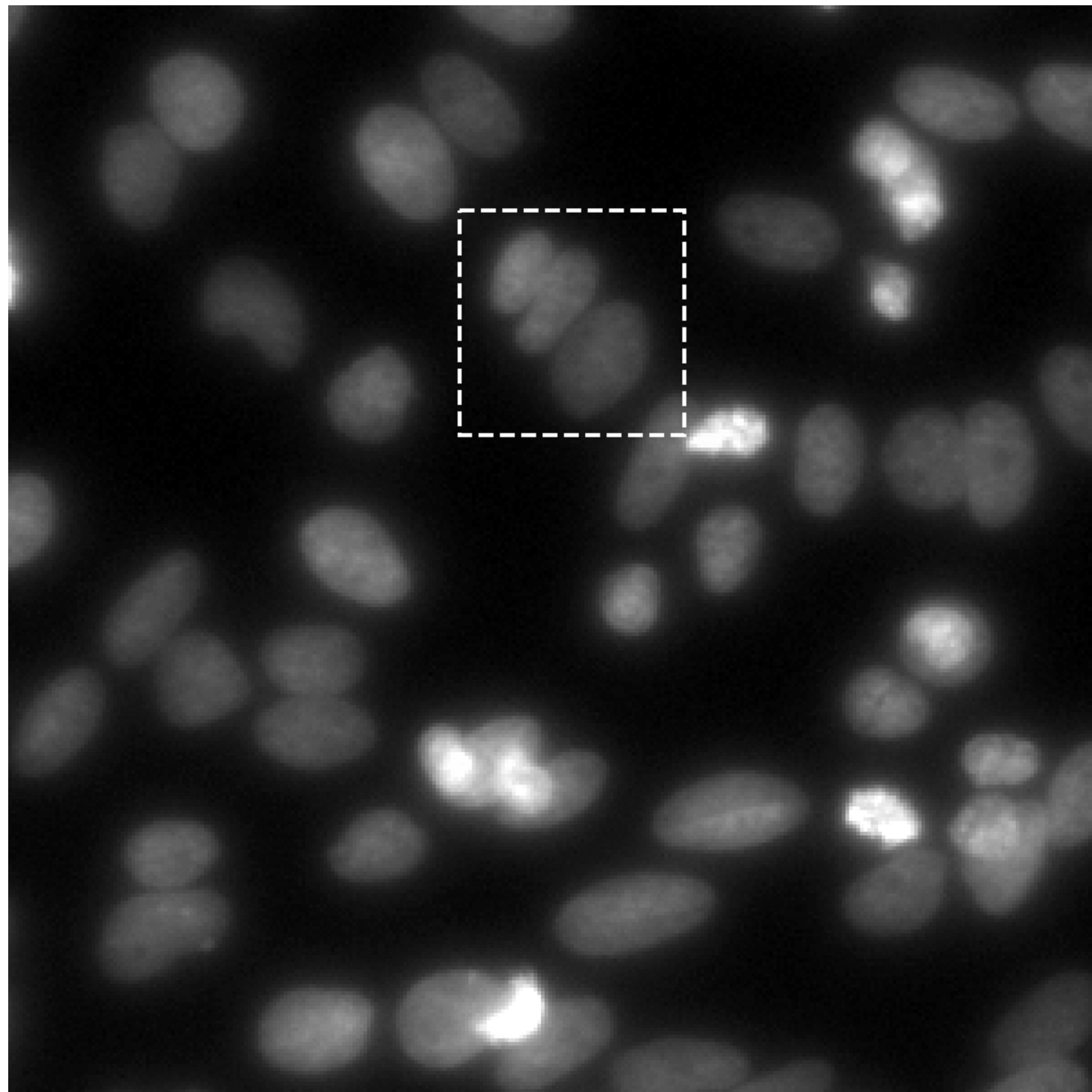
## QuPath

- Draw ROI per object
- Convert to label mask



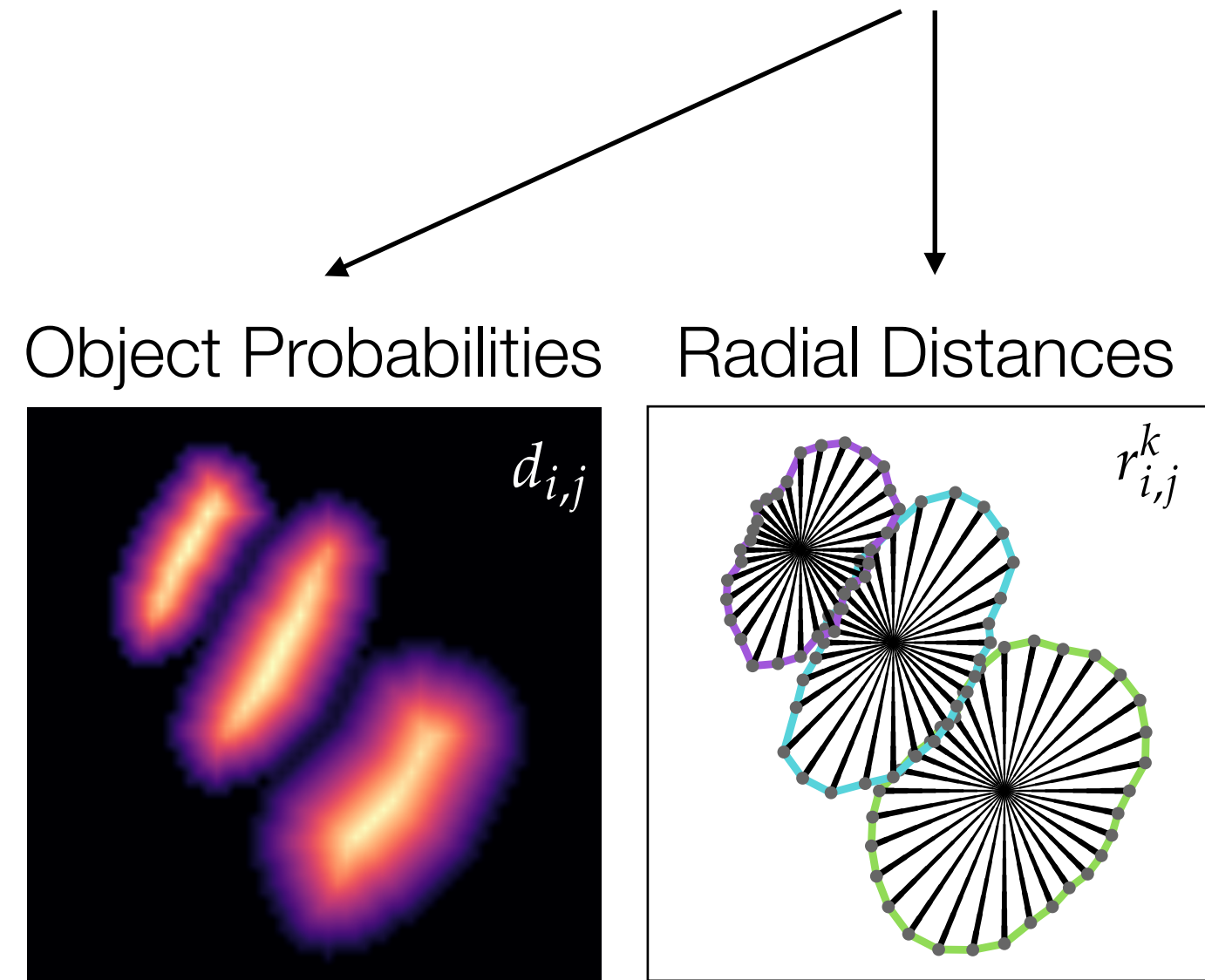
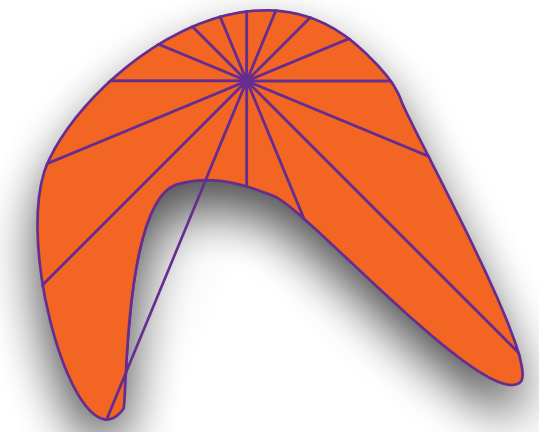
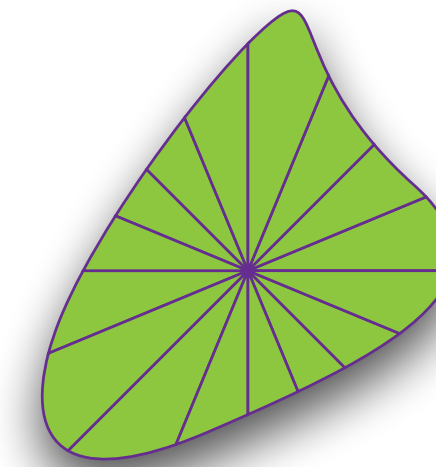
<https://qupath.github.io>

Bankhead et al. (2017)

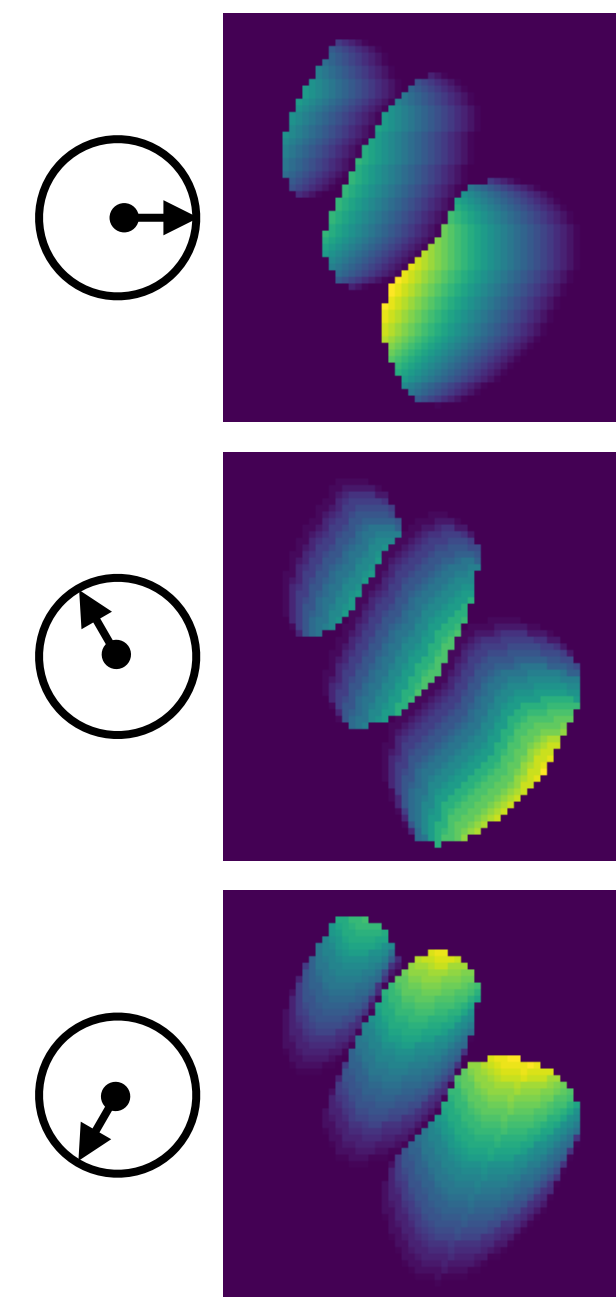


Star-Convex

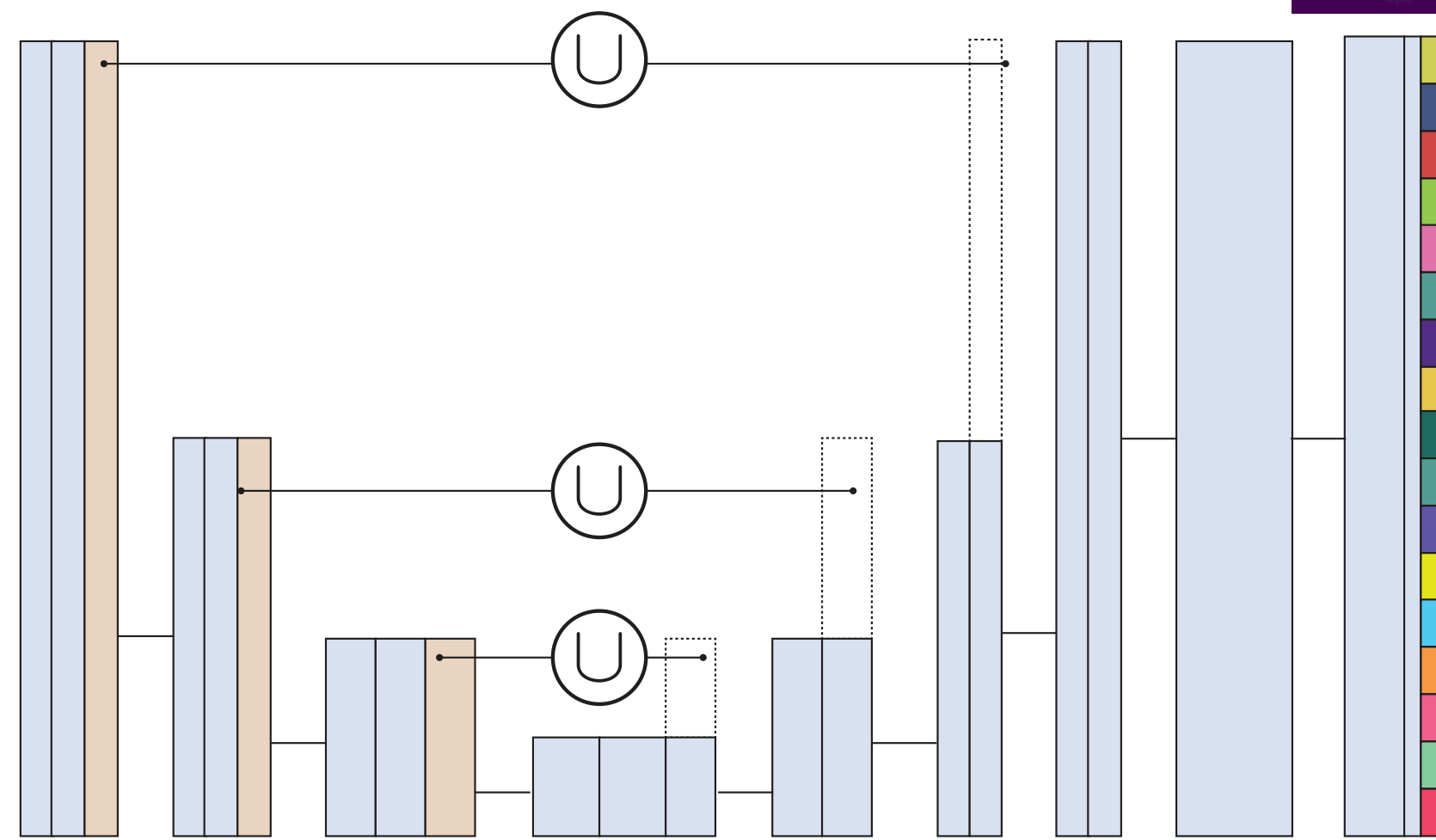
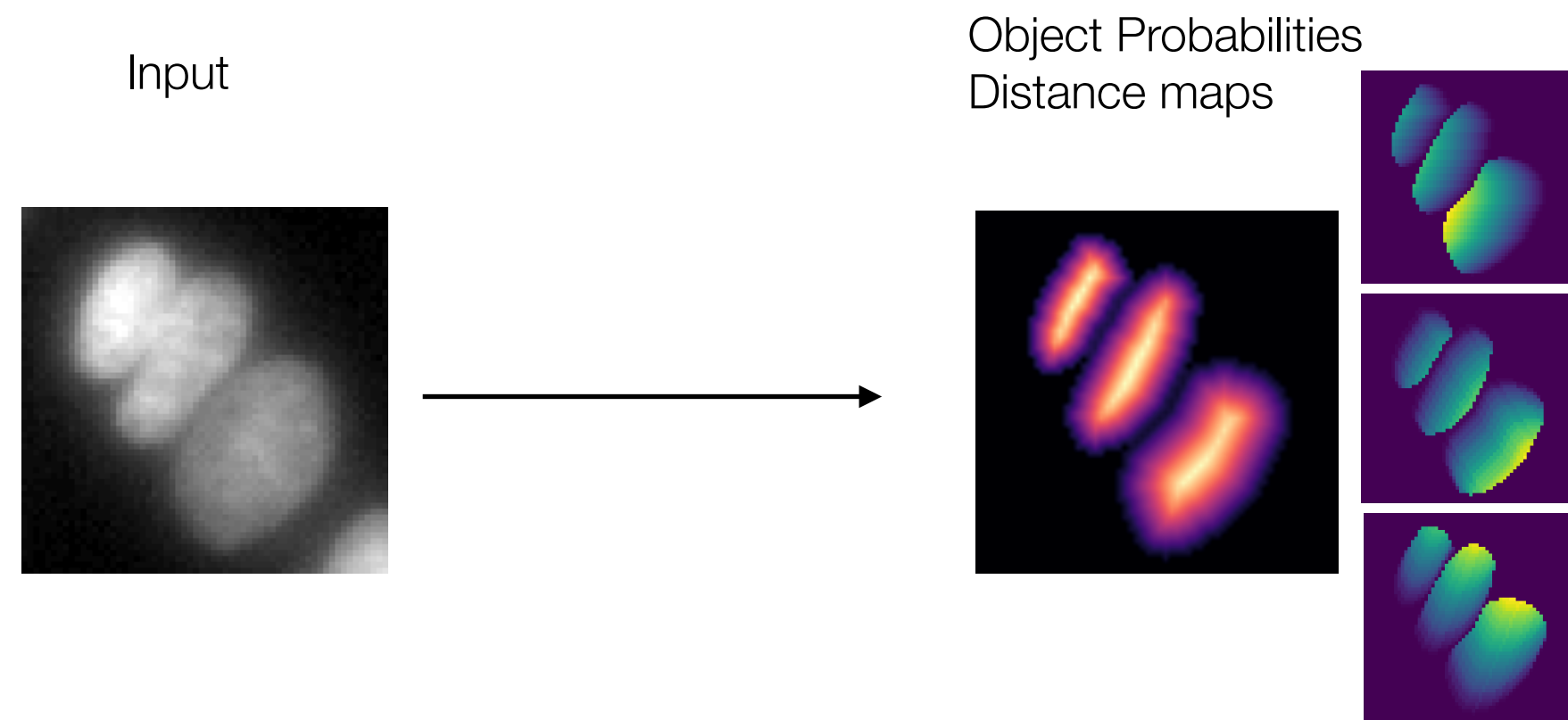
Not Star-Convex



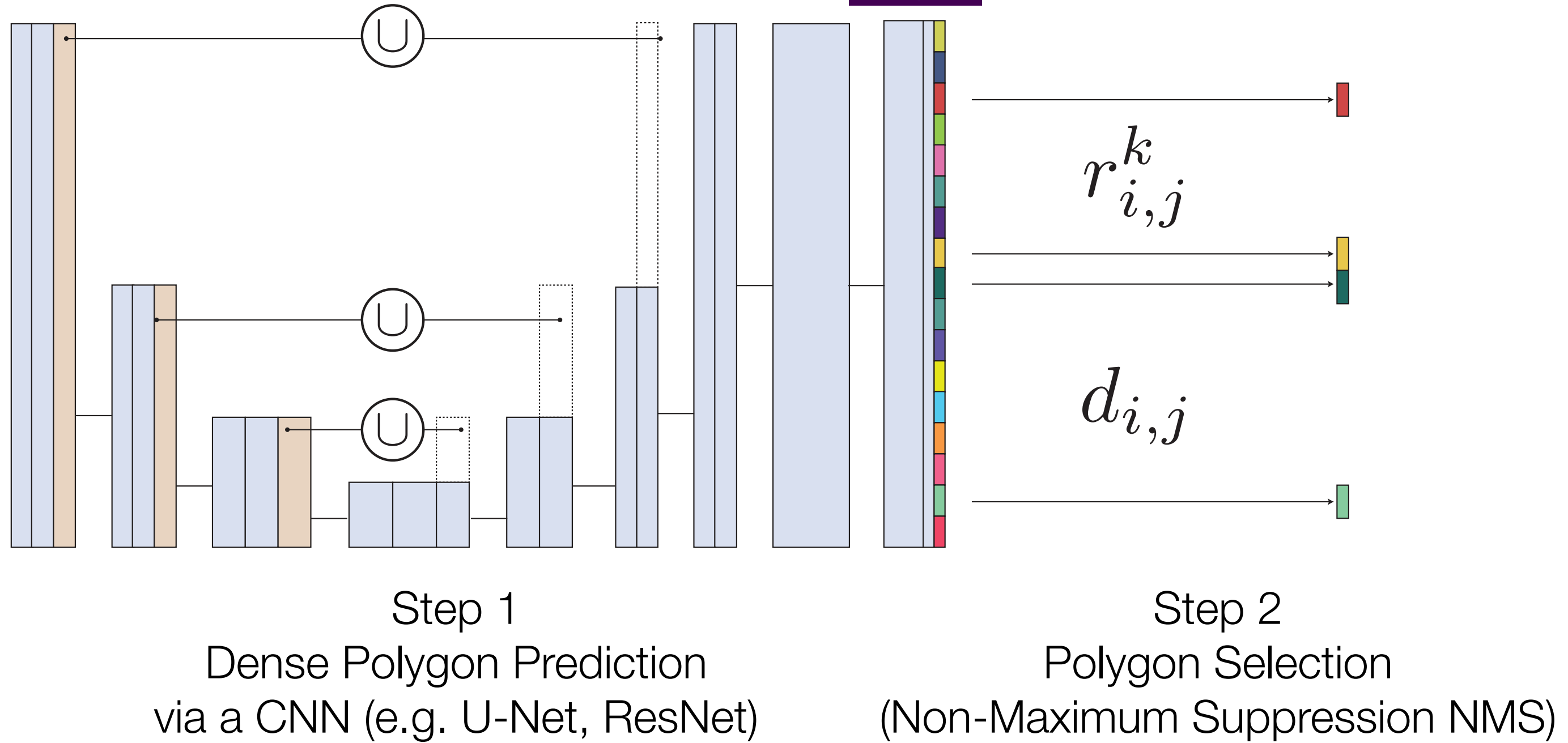
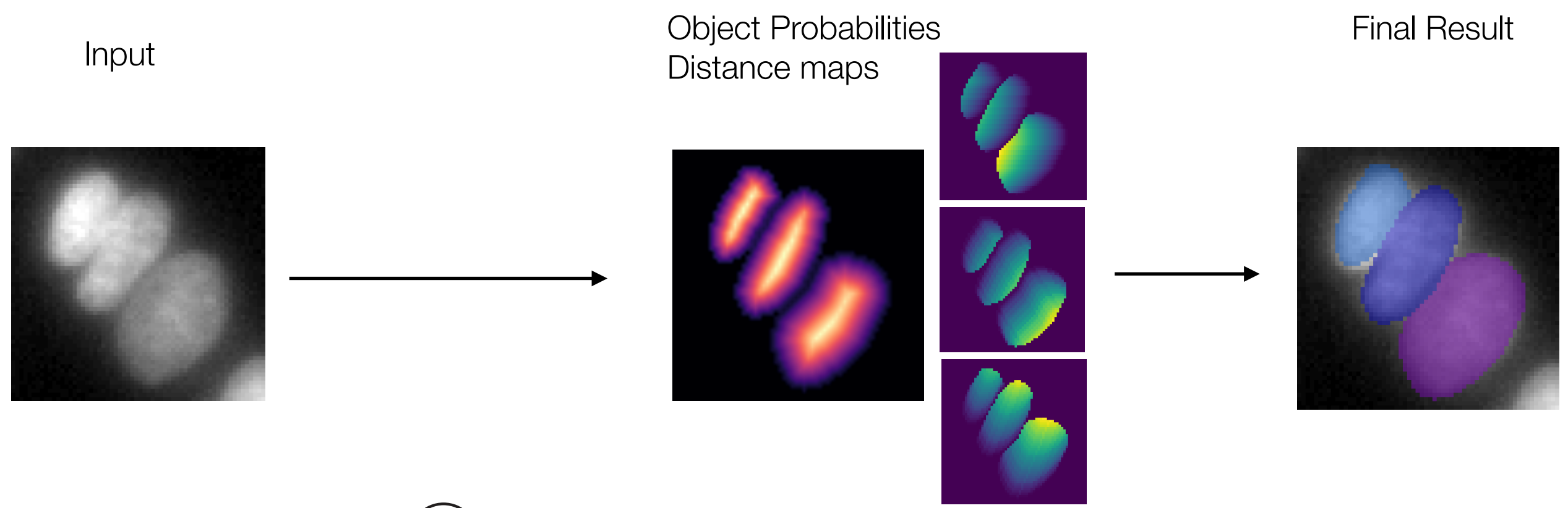
Distance maps (~32)



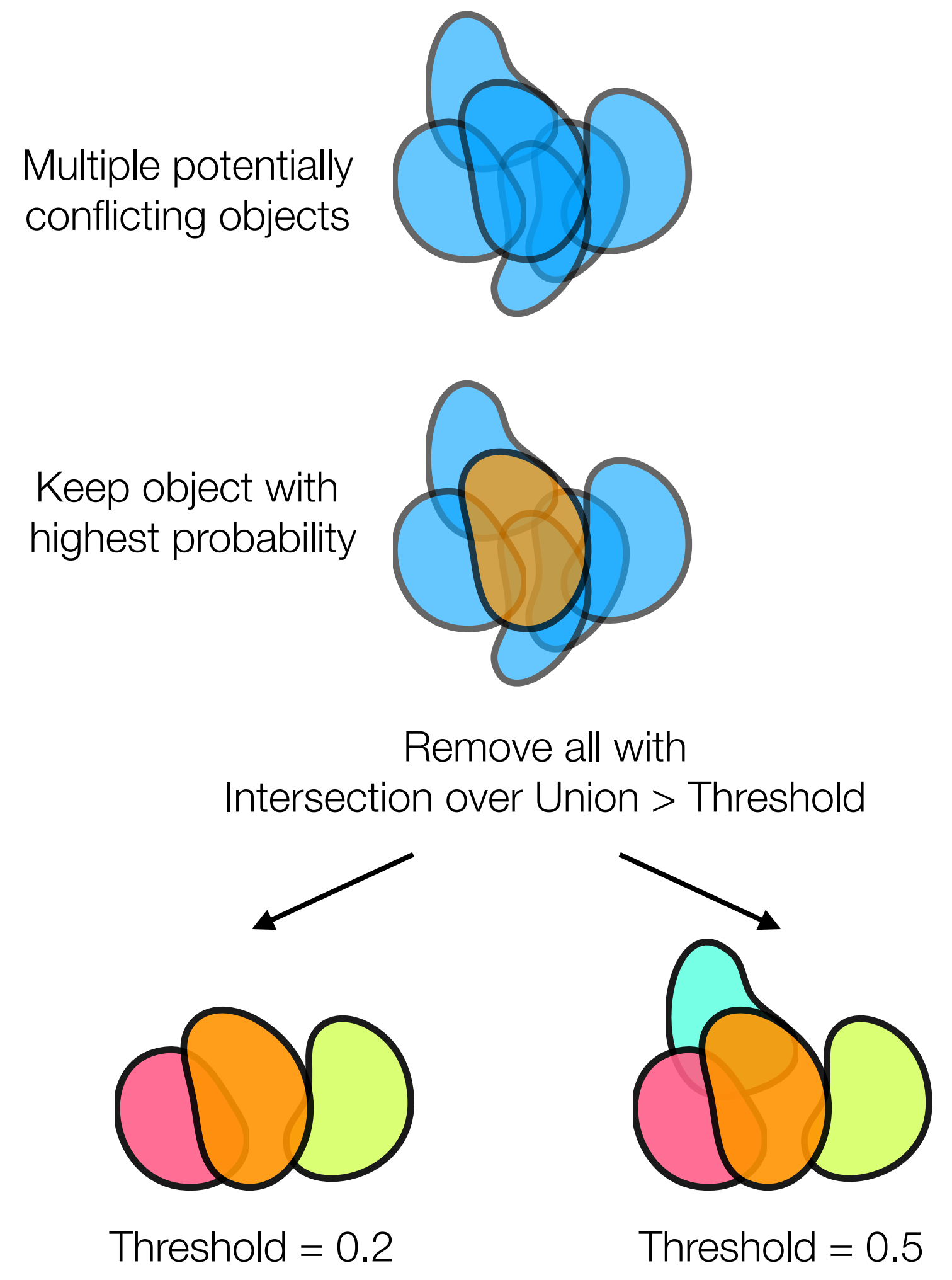
Approximate cells/nuclei with star-convex bounding polygon



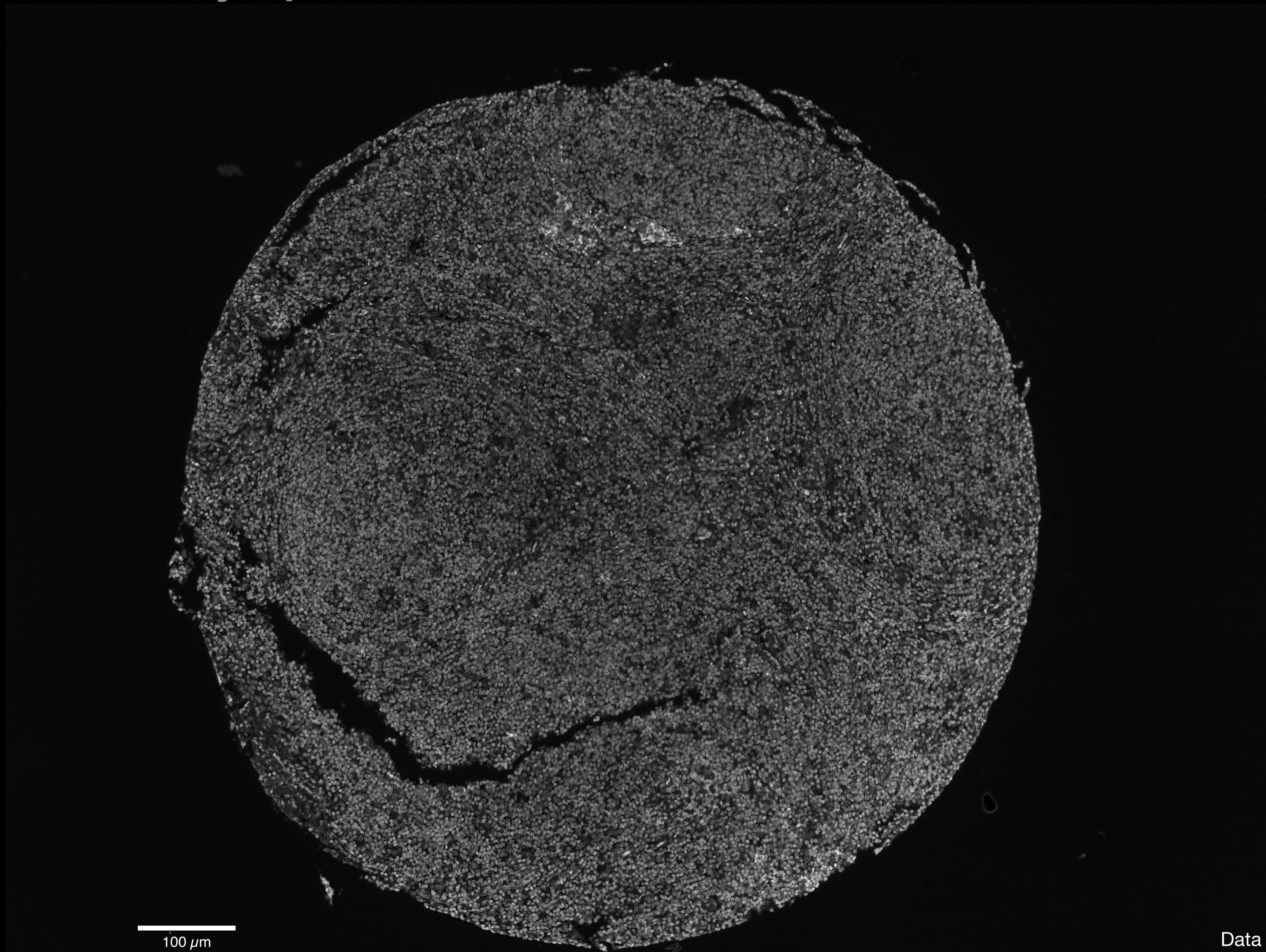
Step 1  
Dense Polygon Prediction  
via a CNN (e.g. U-Net, ResNet)

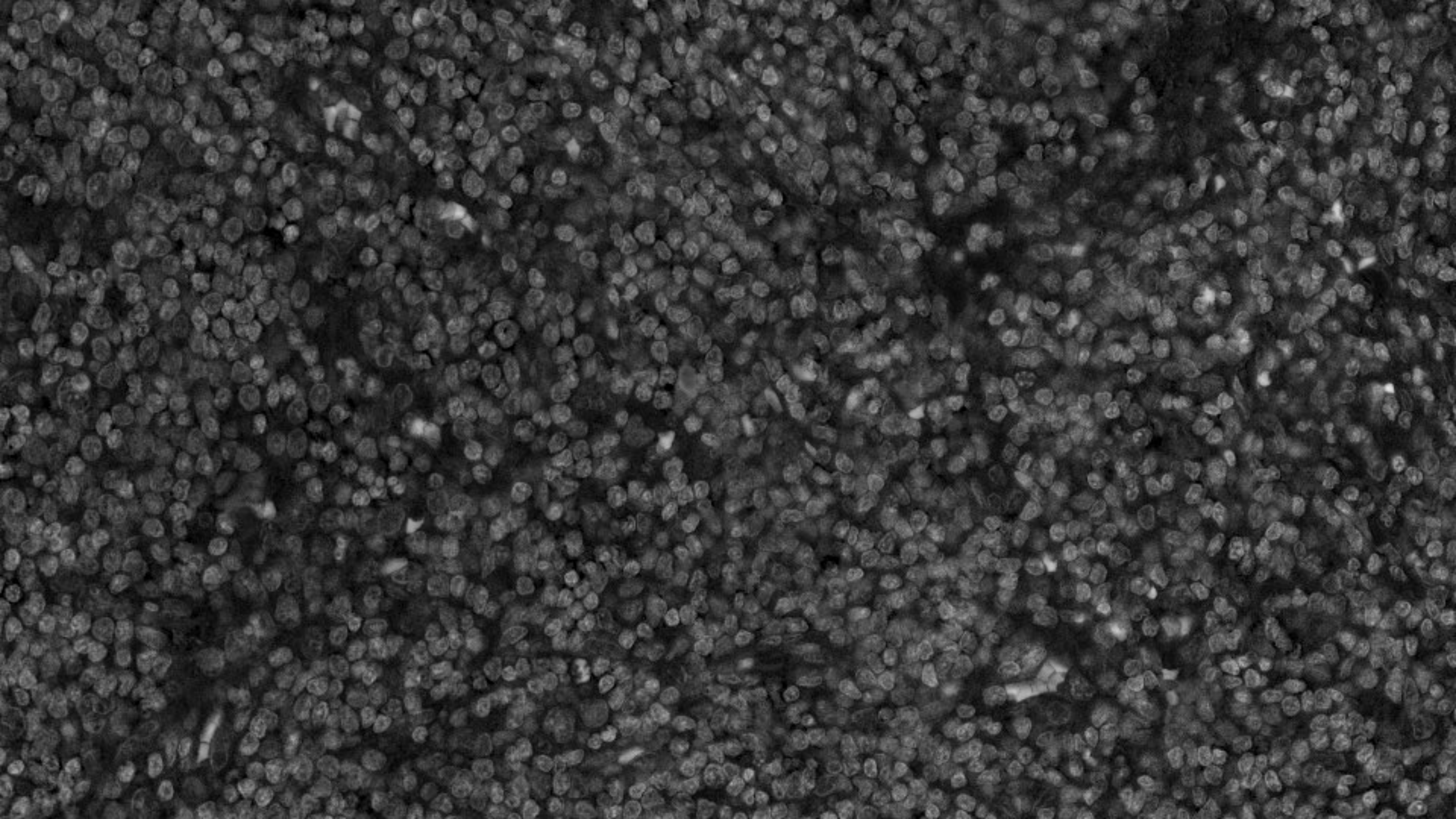


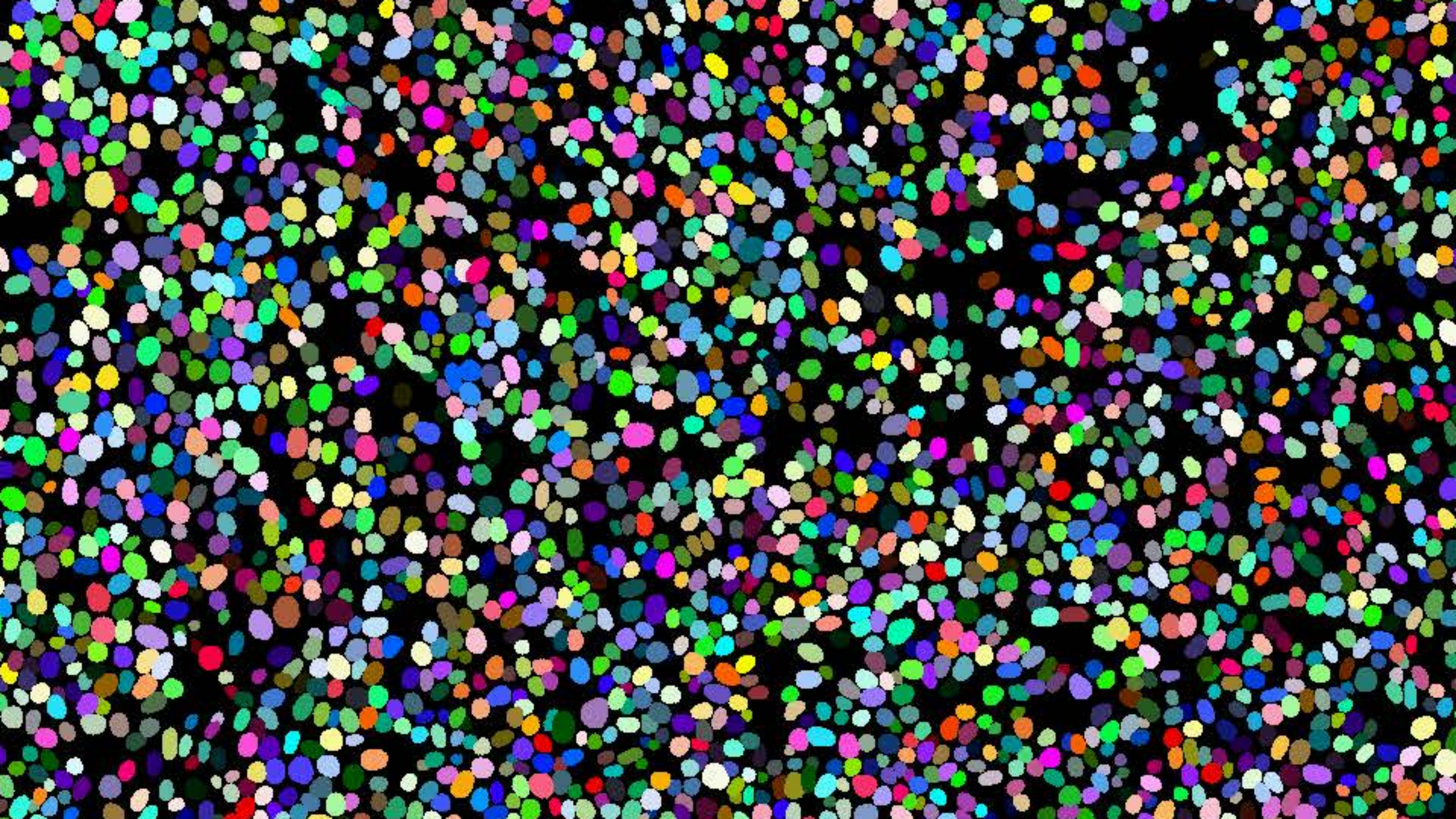
**Non-Maximum-Suppression (NMS)**



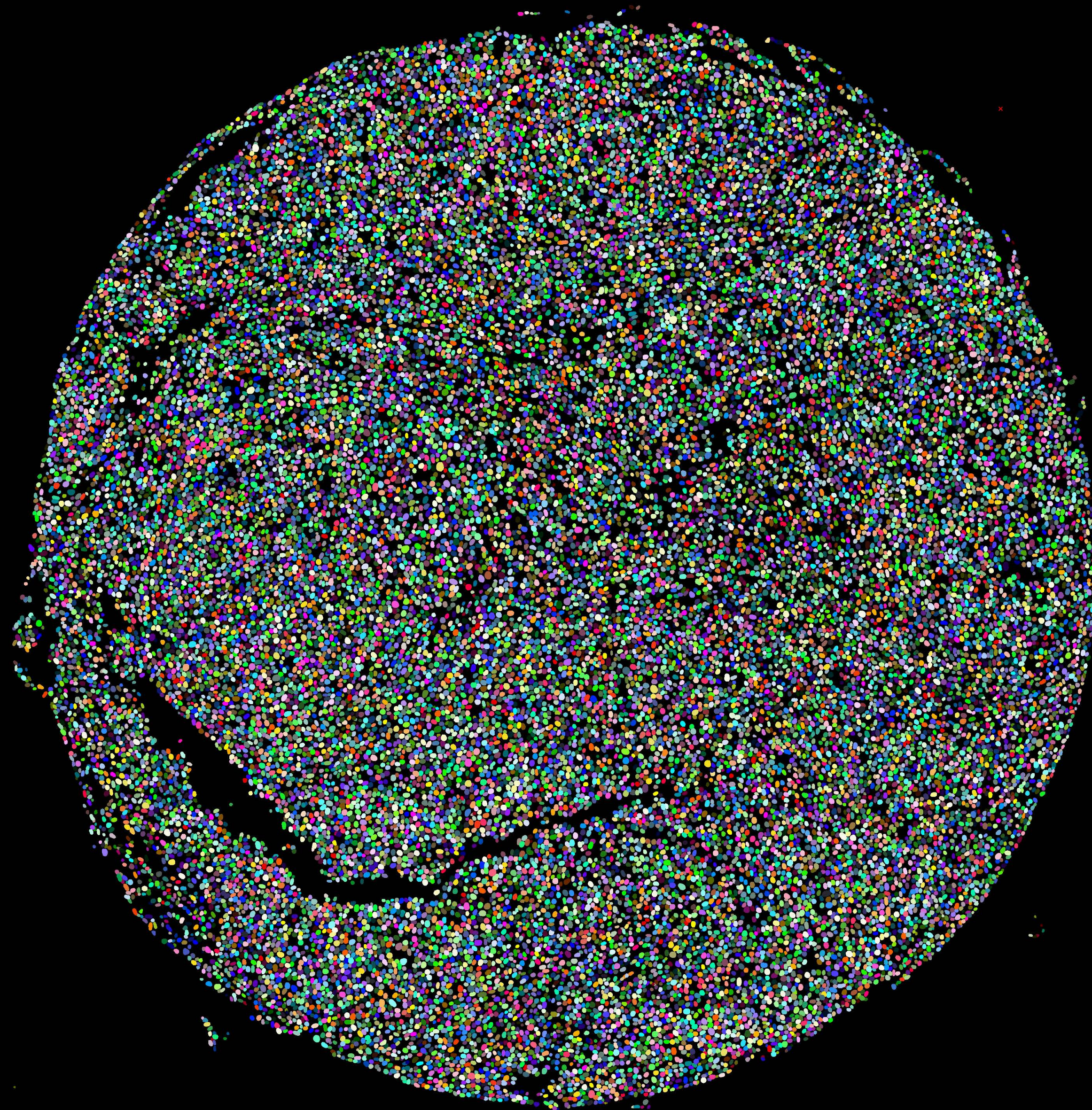
# Example: crowded lymphoma cells

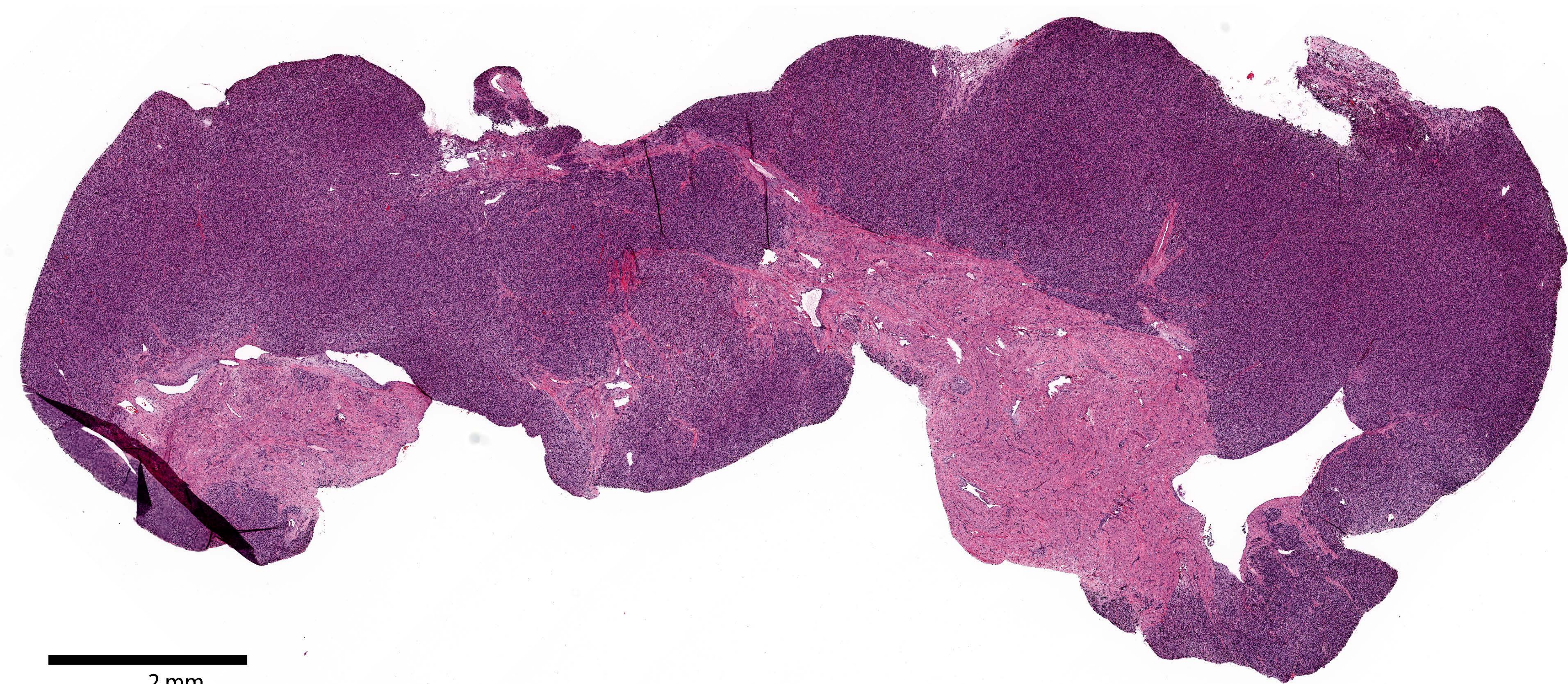




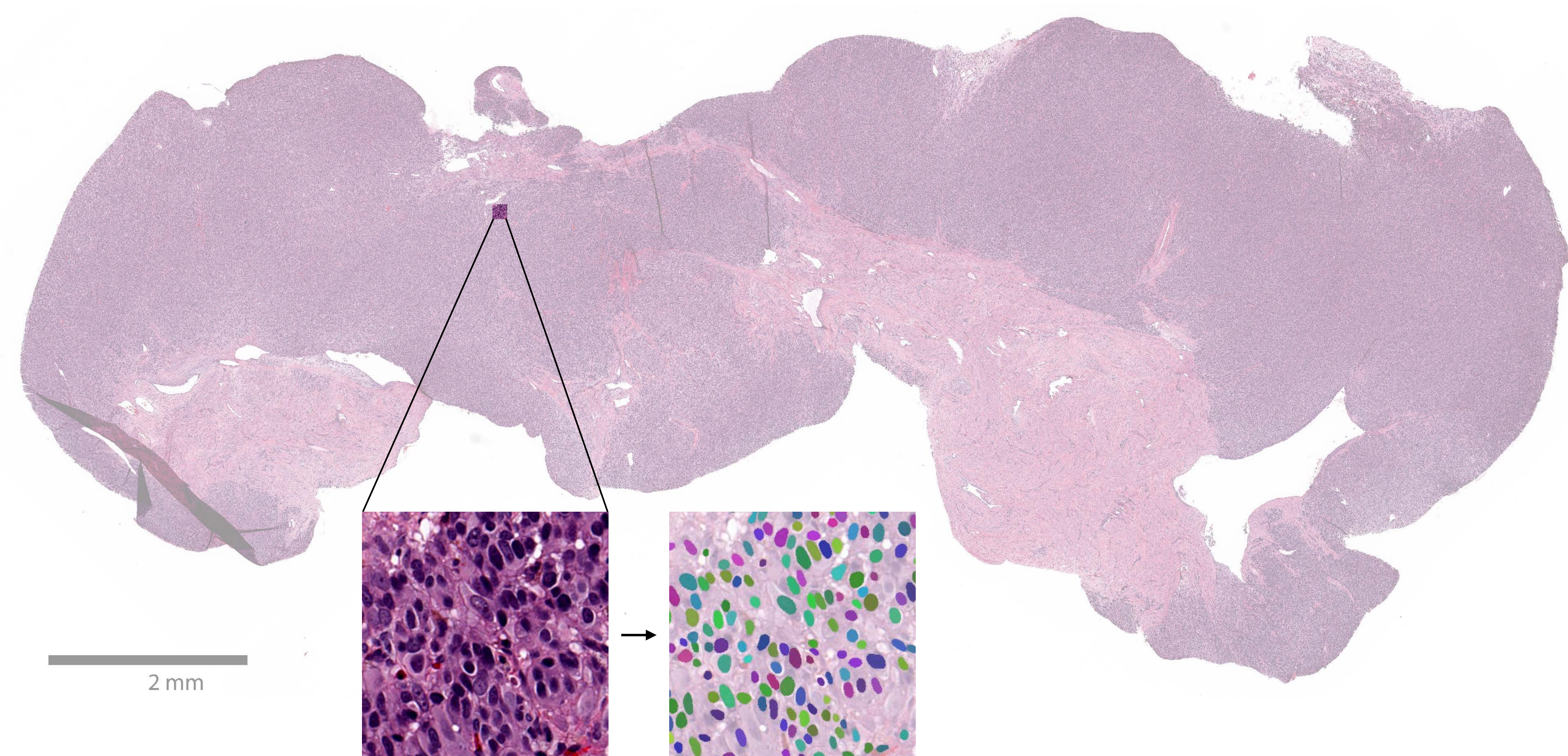


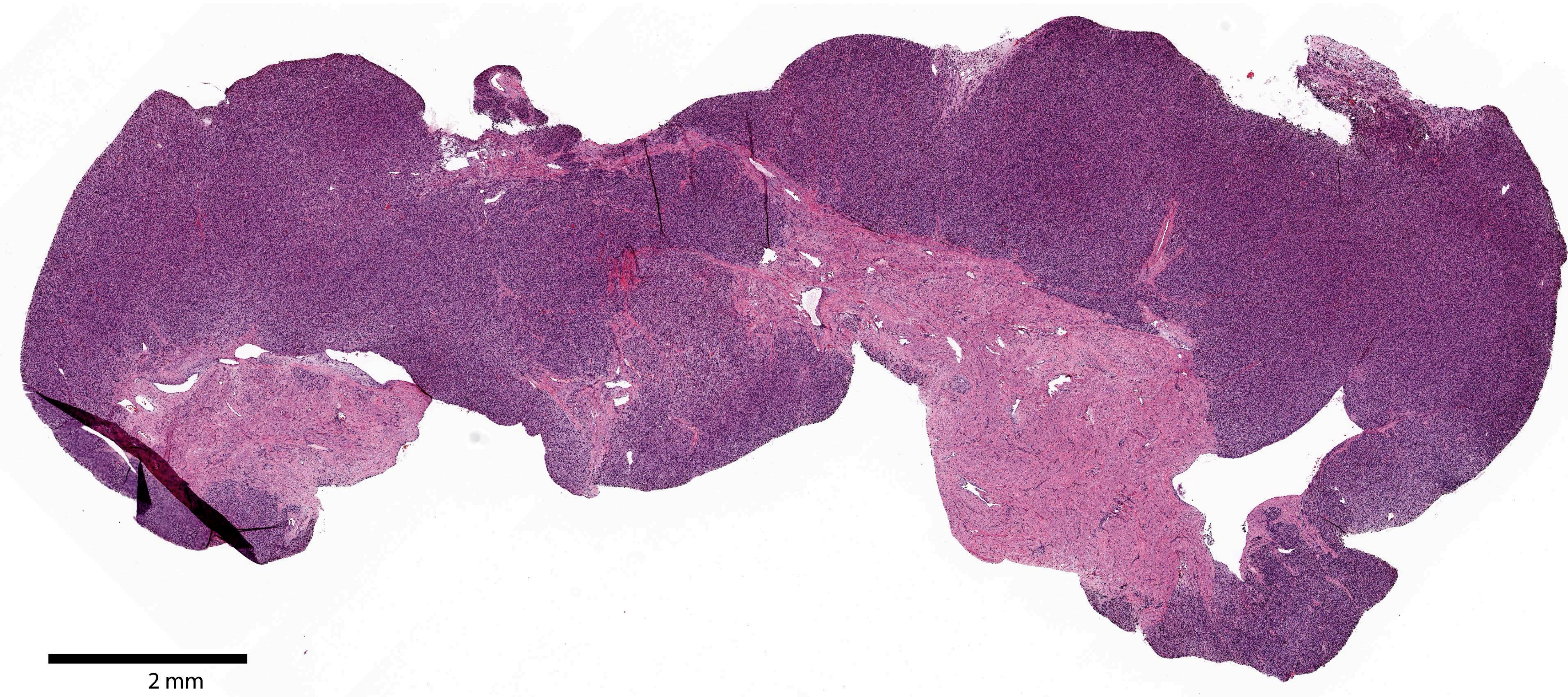
Example: crowded lymphoma cells



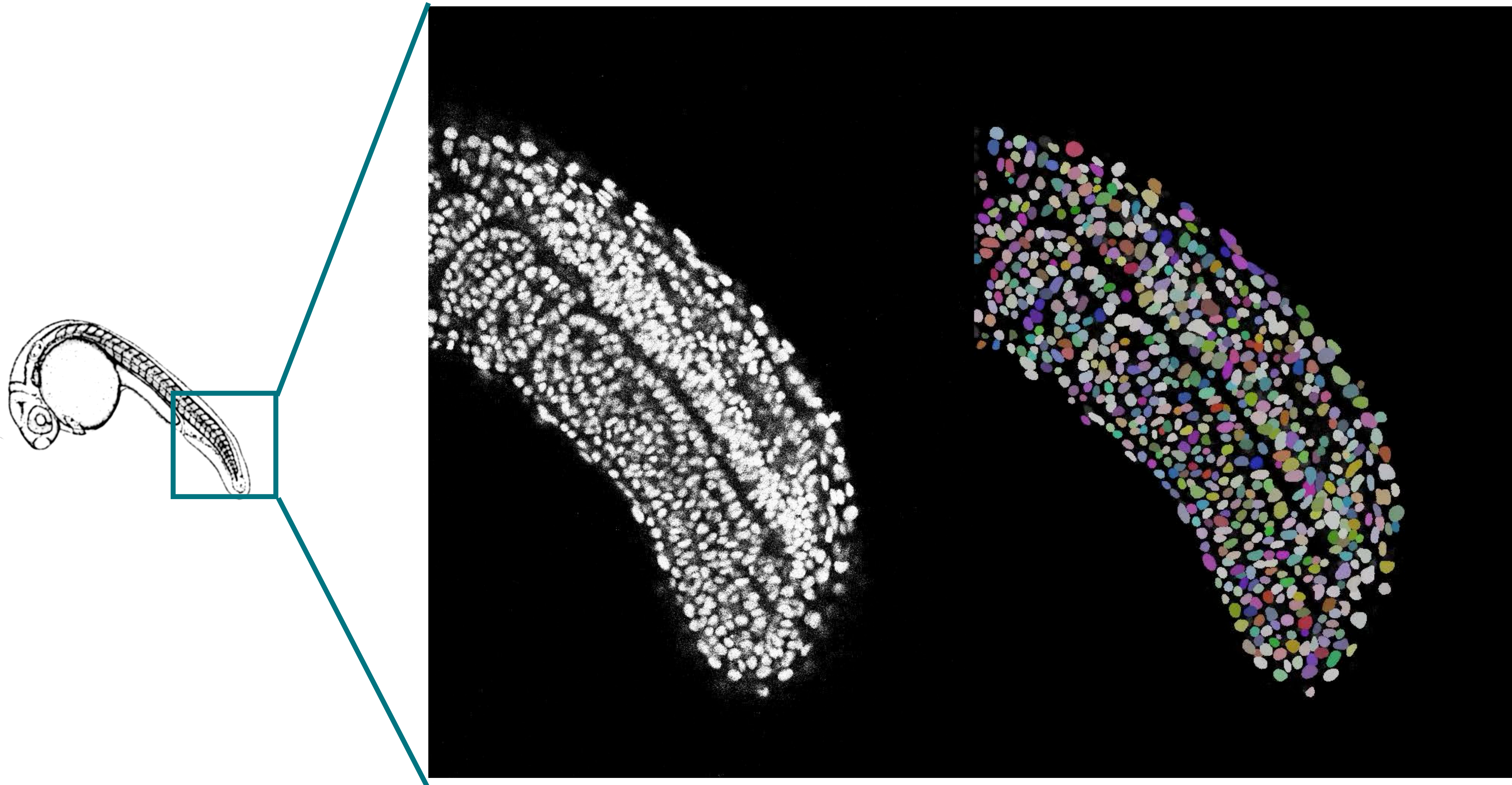


Whole Slide H&amp;E (Sarcoma), (15mm x 6mm, 32k x 14k pixels)

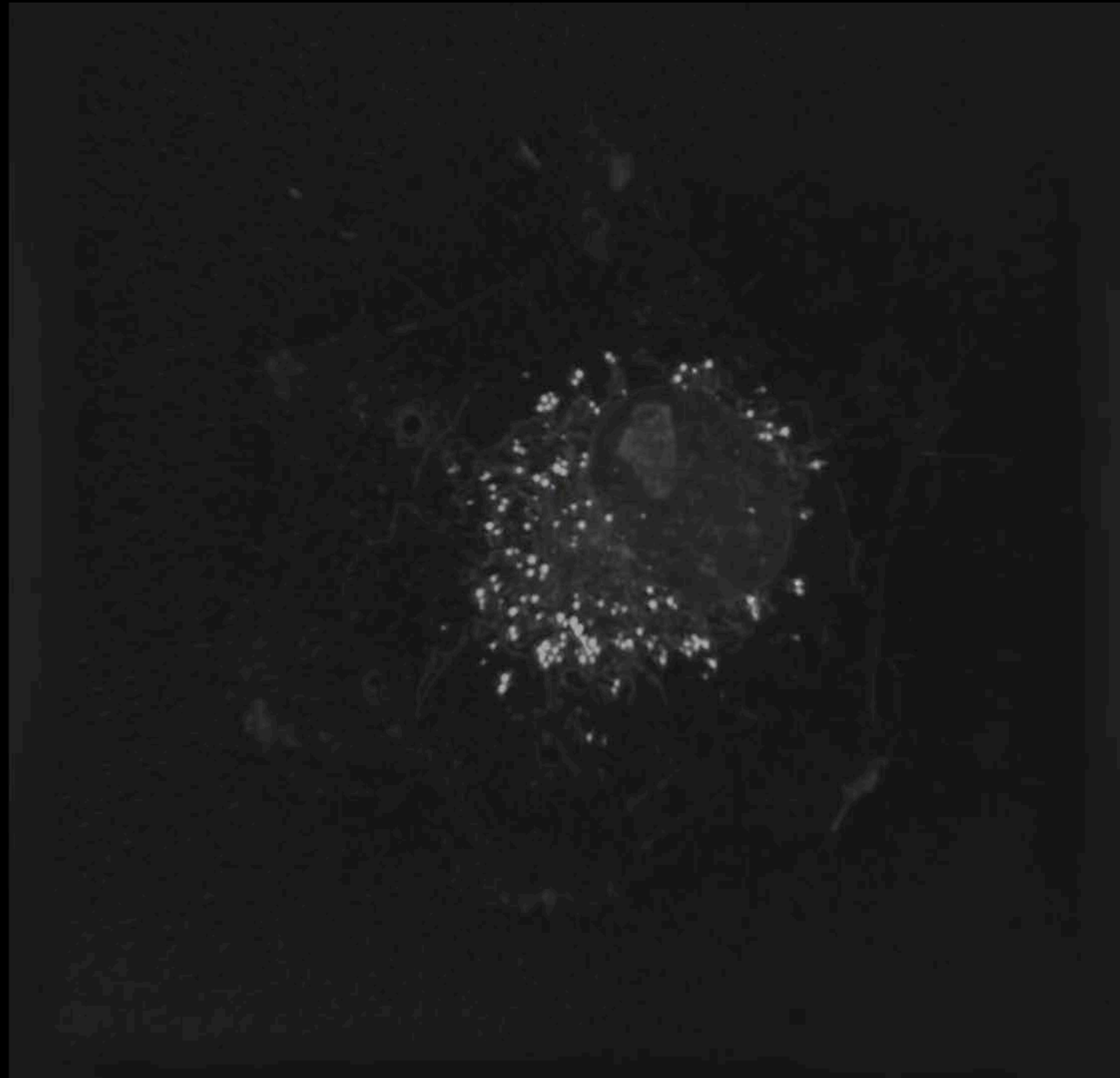




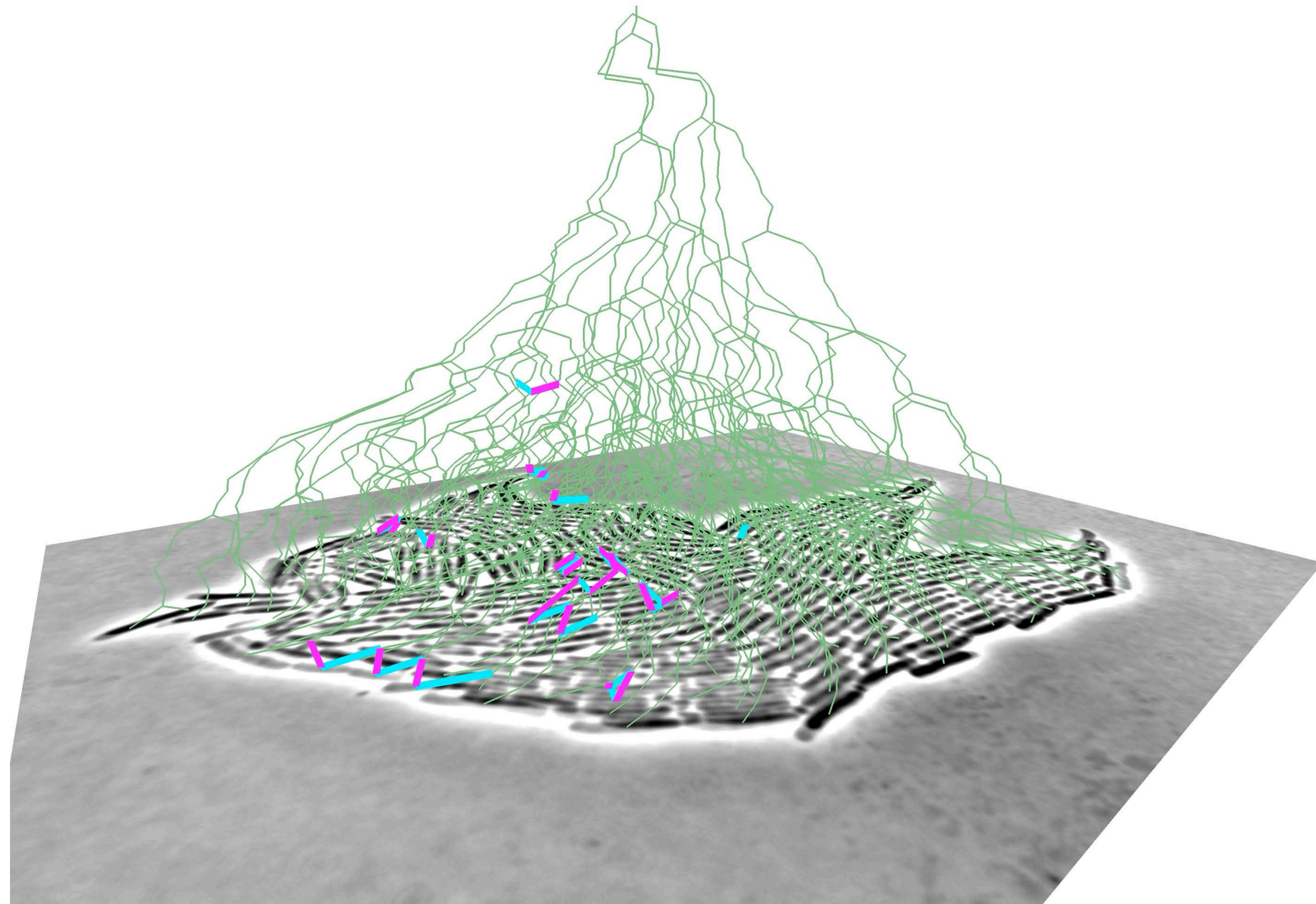
Whole Slide H&amp;E (Sarcoma), (15mm x 6mm, 32k x 14k pixels)



Zebrafish tail development, Data from Oates lab, EPFL

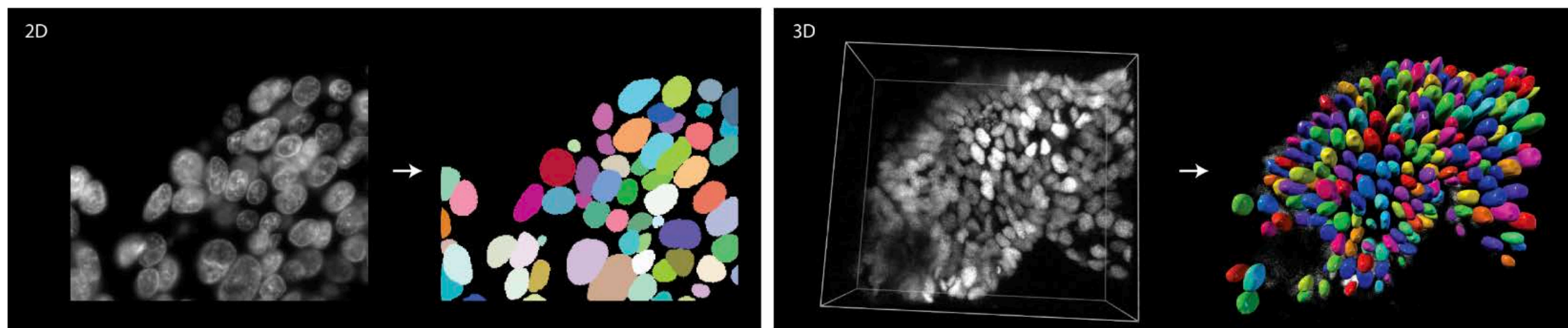


Method	Linking	TRA $\uparrow$	AOGM $\downarrow$
Distance	greedy	0.944	1511
Distance	ILP	0.962	1065
TrackMate (overlap)	LAP	0.957	872
DELTA 2.0	greedy	0.996	118
TRACKASTRA (points only)	ILP	0.995	136
TRACKASTRA	greedy	<b>0.999</b>	36
TRACKASTRA	ILP	<b>0.999</b>	23



[pypi package 0.9.1](#)
[Anaconda.org 0.9.1](#)
[Test passing](#)
[Test \(PyPI\) failing](#)
[forum 459 topics](#)
[downloads 22k/month](#)

## StarDist - Object Detection with Star-convex Shapes



Spotiflow Spotiflow 0.5.0 documentation

Spotiflow 0.5.0 57 7

**Spotiflow 0.5.0 documentation**

- Predicting spots using the napari plugin
- Inference via CLI
- Available pre-trained models
- Training a Spotiflow model on a custom dataset
- Fine-tuning a Spotiflow model on a custom dataset
- API Reference

# Spotiflow

Spotiflow is a learning-based subpixel-accurate spot detection method for 2D and 3D fluorescence microscopy. It is primarily developed for spatial transcriptomics workflows that require transcript detection in large, multiplexed FISH-images, although it can also be used to detect spot-like structures in general fluorescence microscopy images and volumes. For more information, please refer to our [paper](#).

**Spotiflow**

- Getting Started
- Installation
- Predicting spots in an image
- Python API
- Command Line Interface (CLI)
- Napari plugin
- Contents

