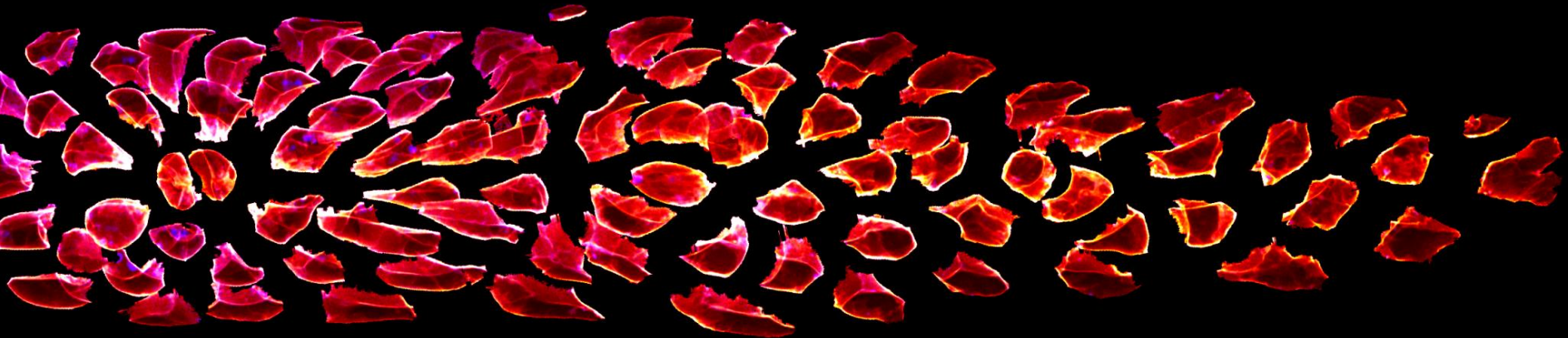


# Bonus Lecture! Yay!

EMBL Bio-IT/ALMF Course

*Image Analysis with Python 2018*

Sessions 3 – 5



Jonas Hartmann

Gilmour group, EMBL Heidelberg

# Agenda

- ▶ **Parameter optimization**
- ▶ **Smart microscopy**
- ▶ **Object tracking**
- ▶ **Machine learning for segmentation**
- ▶ **More machine learning**

# Parameter Optimization

# Parameter Optimization

## ► Selecting parameters is important but difficult!

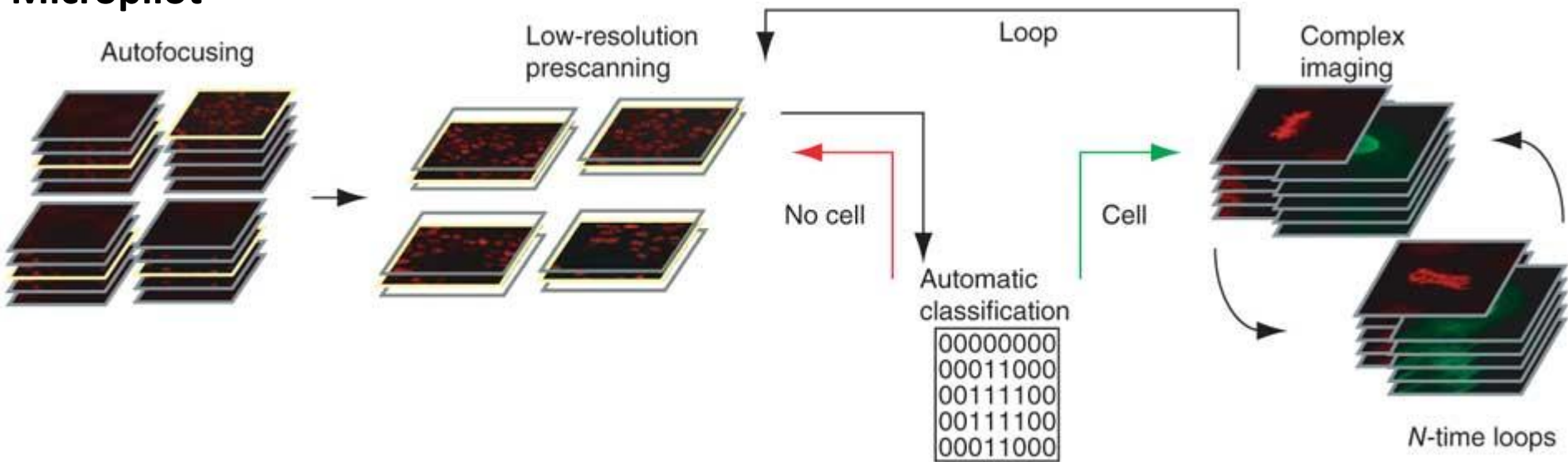
- Examples: *sigma*, *SE size*, *morphology params*, *DT sigma*, *min\_distance*, ...
- Option 1: Use reasoning based on 'limits'
- Option 2: Trial and error
  - This can be automated!
    1. Create some manual gold standard segmentations
    2. Automatically screen through 100s of parameter combinations
    3. This becomes feasible by using the compute cluster!

# Smart Microscopy

# Smart Microscopy

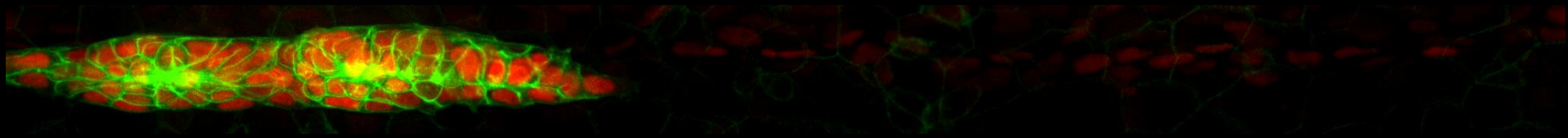
- ▶ Direct coupling of microscopy and image analysis
- ▶ Example 1: Region Of Interest (ROI) detection
  - Identify ROI in low-res high-FOV image
  - Instruct microscope to acquire ROI in high-res

## Micropilot

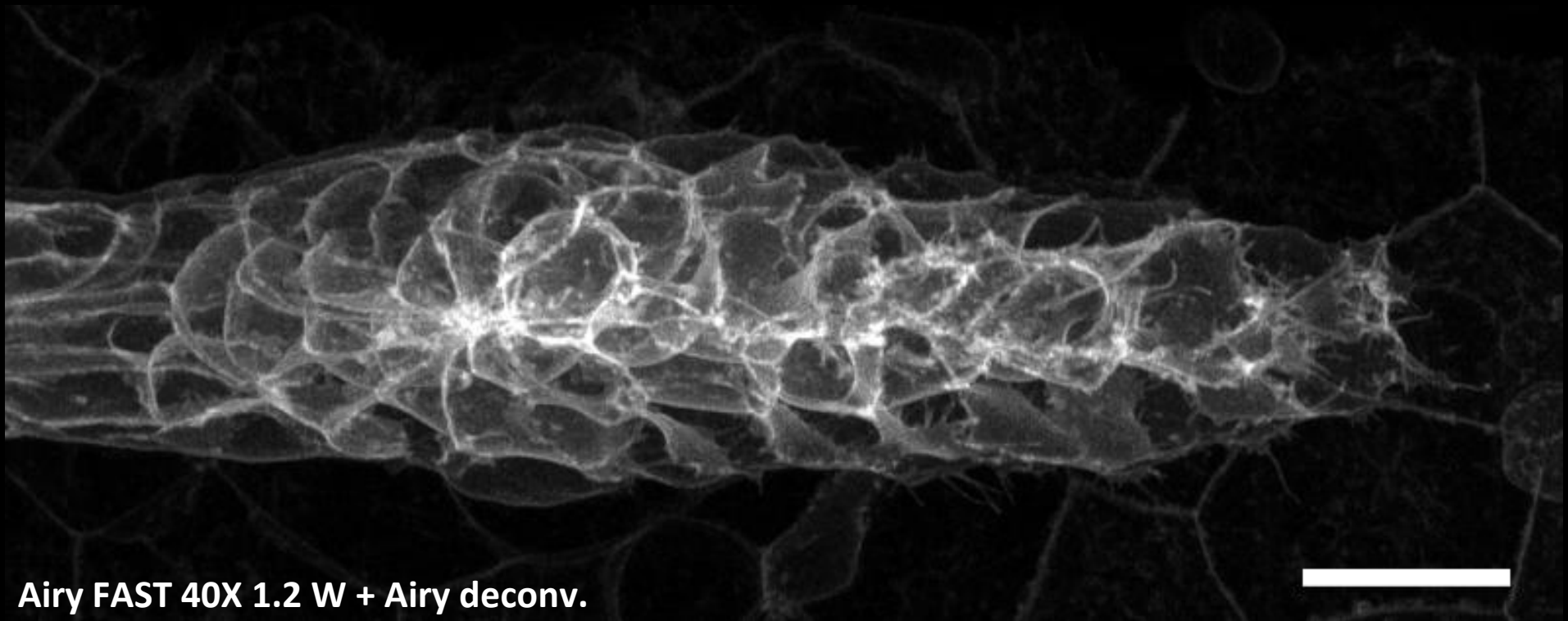


Published 2011 in Nature Methods by Ellenberg & Pepperkok groups

# Smart Microscopy



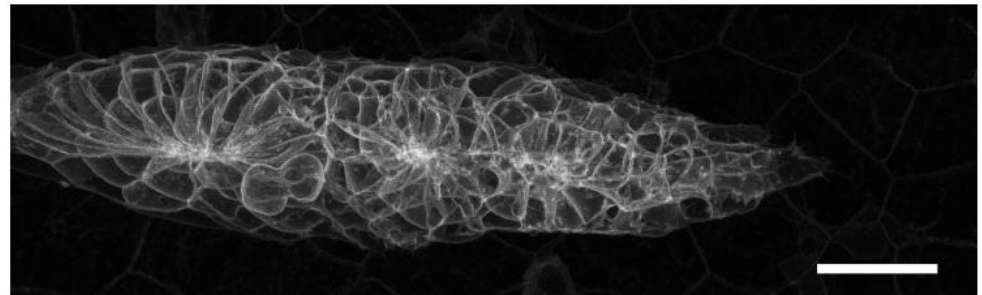
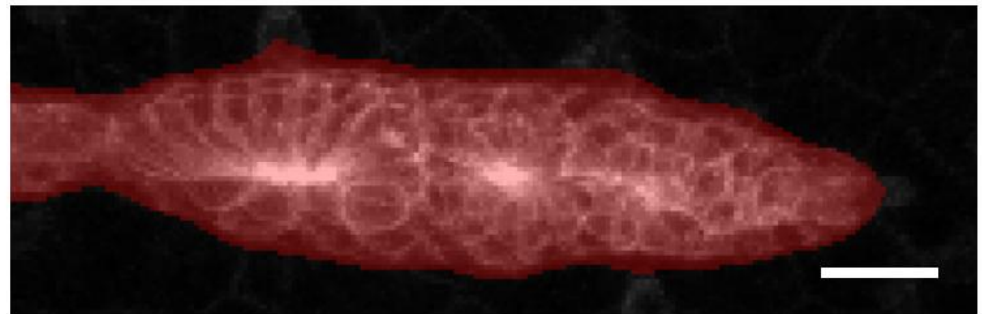
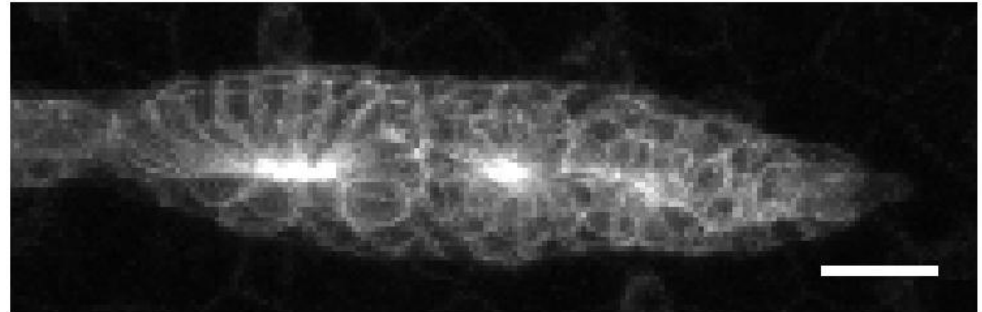
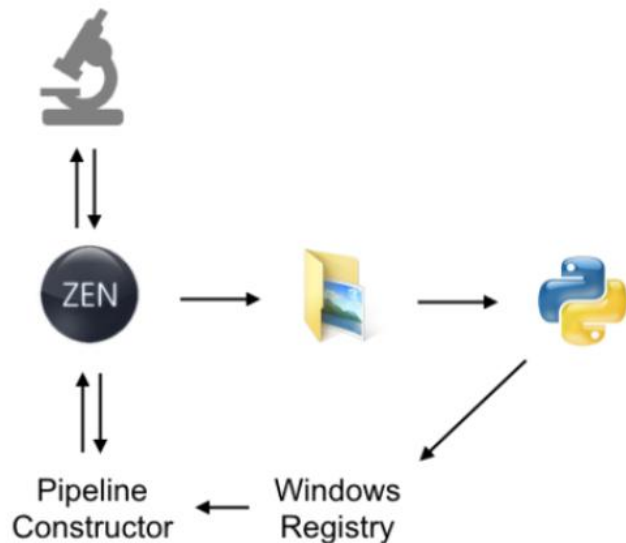
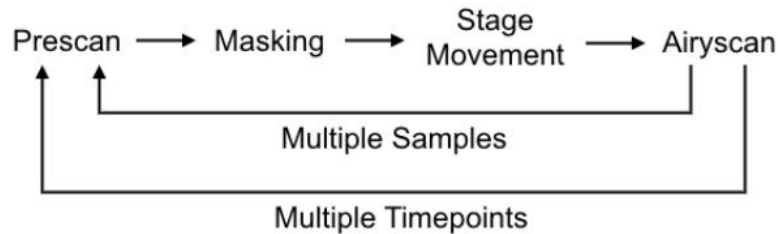
VoX Spinning Disk 40X 1.2 W



Airy FAST 40X 1.2 W + Airy deconv.

# Smart Microscopy

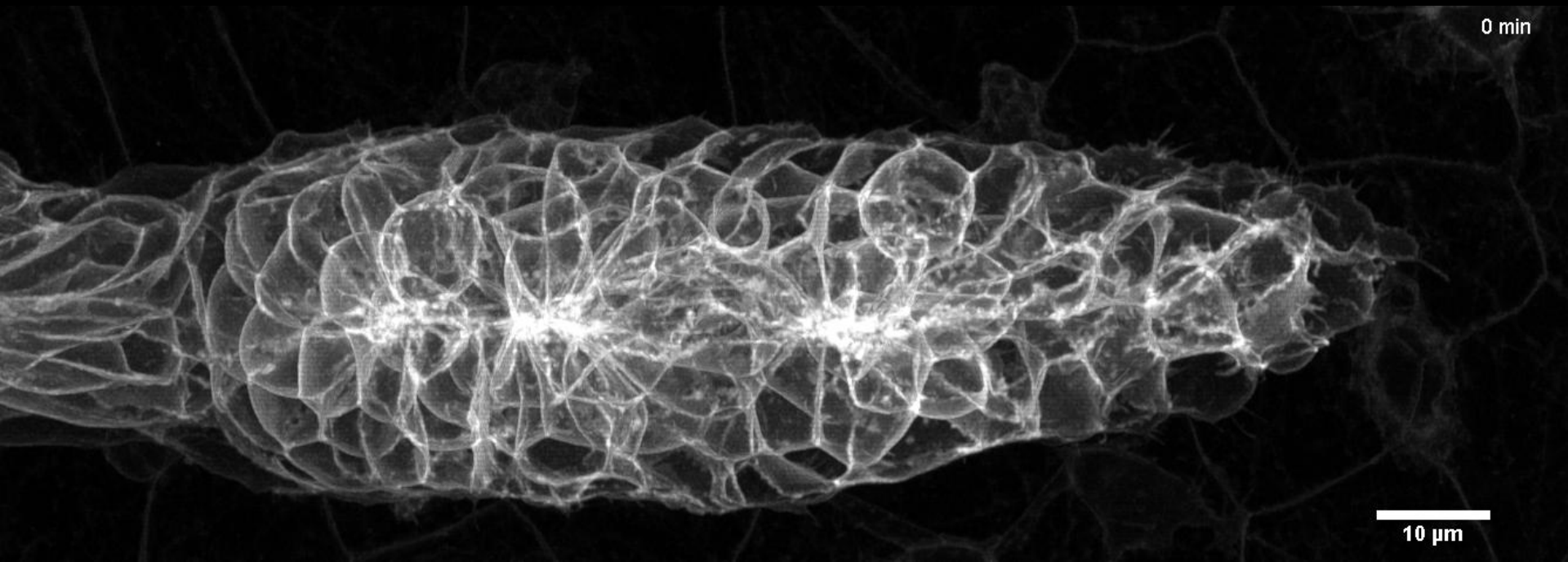
- ▶ Direct coupling of microscopy and image analysis
- ▶ Example 2: Live object tracking



Scale bars: 15 $\mu$ m



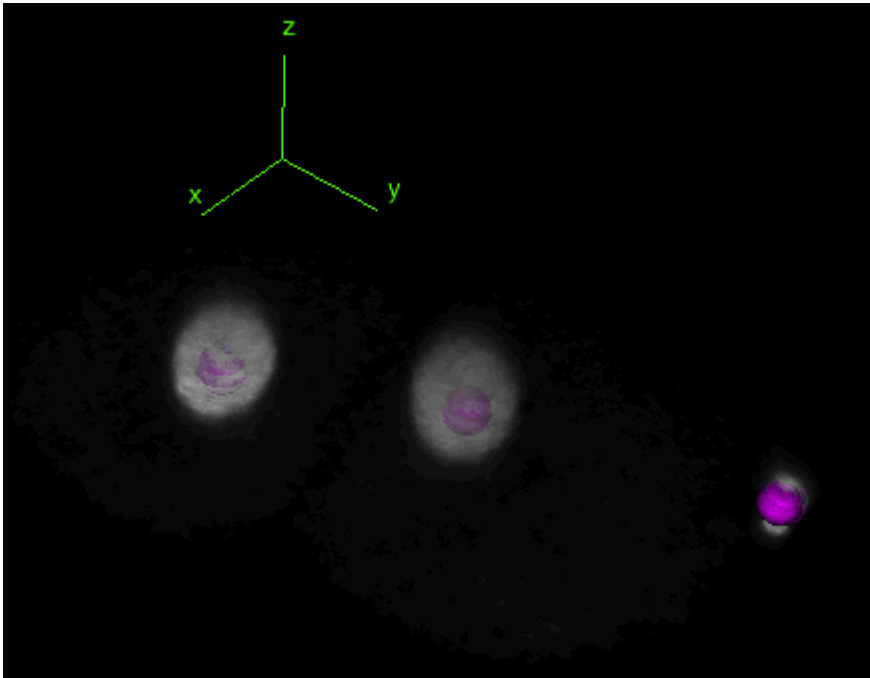
# Smart Microscopy



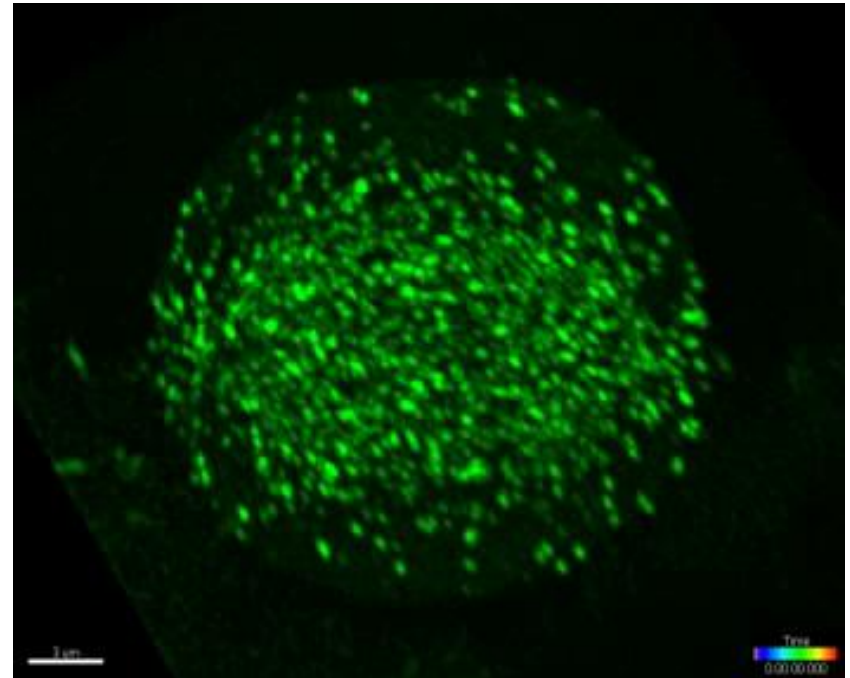
# Object Tracking

# Object Tracking

- Assign each labeled object at time  $t_0$  a corresponding object at  $t_1$ 
  - Approach: segment independently, then link objects (by optimization)
  - Linking based on: space, movement, object properties
  - Easier for small  $\delta t$
  - Challenges: poor segmentation, overlapping objects, dividing/merging objects



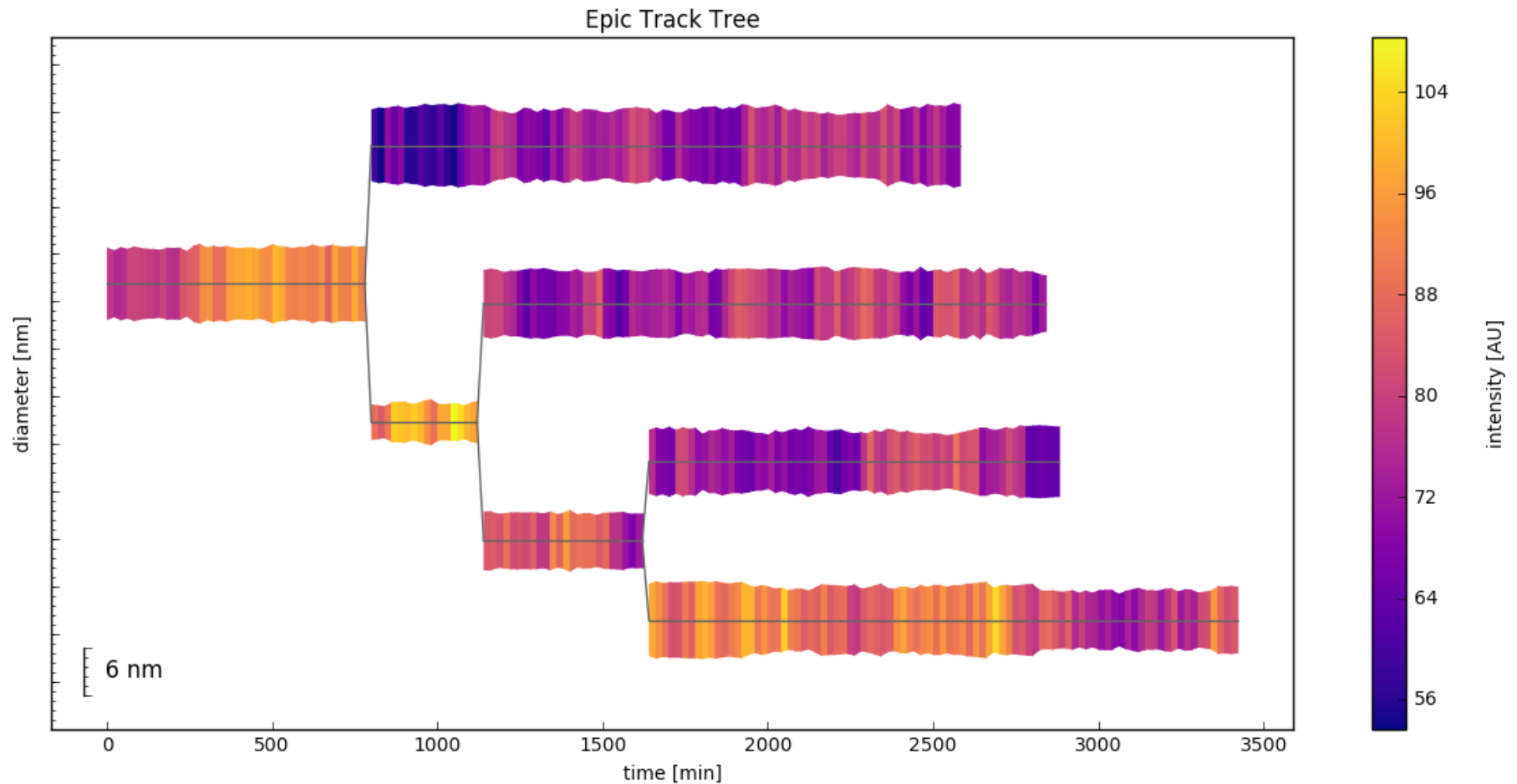
From Fiji's TrackMate plugin



From Eric Betzig's group

# Object Tracking

- Assign each labeled object at time  $t_0$  a corresponding object at  $t_1$



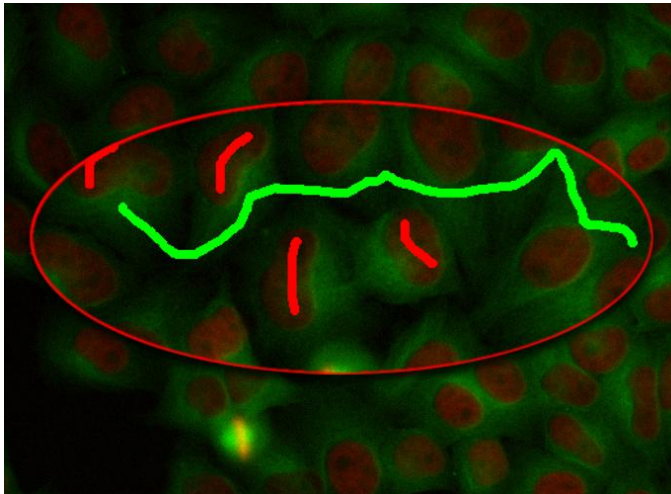
# Machine Learning for Segmentation

# Machine Learning for Segmentation

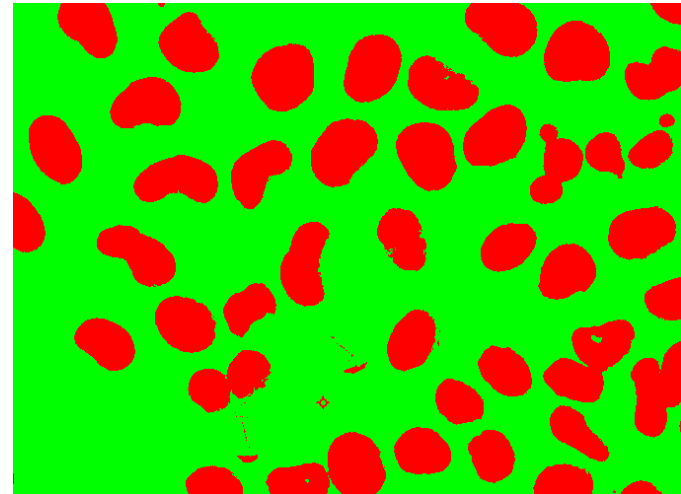
- ▶ **Goal:** foreground-background detection by machine learning
  - This is a classification task: classify pixels into groups
  - Approach: supervised learning
    - Manually label example pixels
    - Extract features (intensity, neighborhood, ...)
    - Train classifier, e.g. random forest
    - Make prediction for all other pixels



Ilastik



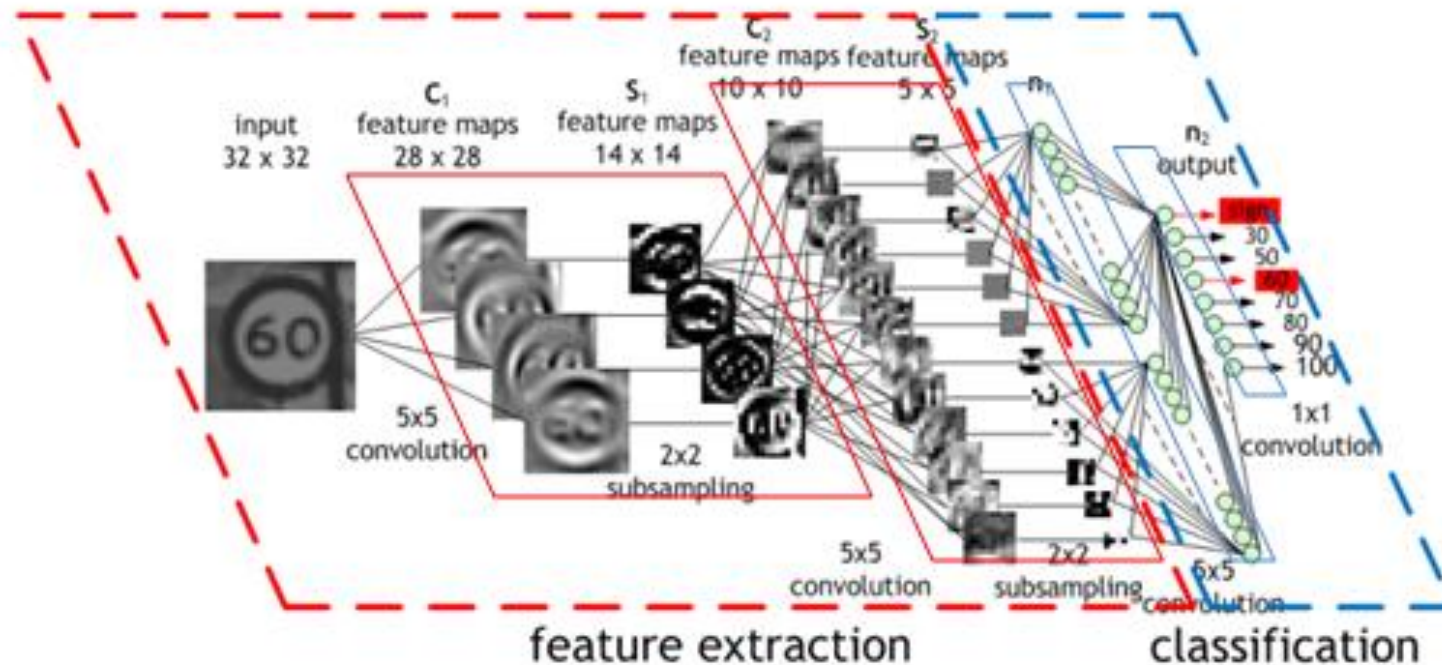
From the Ilastik website



Also useful as `preprocessing` (use probabilities)!

# Machine Learning for Segmentation

- ▶ Machine learning in python
  - scikit-learn (sklearn)
- ▶ Deep Learning
  - Keras, Tensorflow, PyTorch



# Machine Learning for Segmentation

## Machine Learning for Image Analysis

**Date:** Monday 29 - Wednesday 31 October 2018

**Venue:** EMBL - EMBL- Heidelberg, Meyerhofstraße, 69117, Heidelberg, Germany

**Application opens:** Monday May 07 2018

**Application deadline:** Friday June 15 2018

**Participation:** Open application with selection

This is a blended learning course on Machine Learning for Image Analysis, consisting of three online sessions with associated hands-on exercises prior to the workshop, a three day face-to-face workshop at EMBL Heidelberg and two optional online sessions with associated hands-on exercises after the workshop.

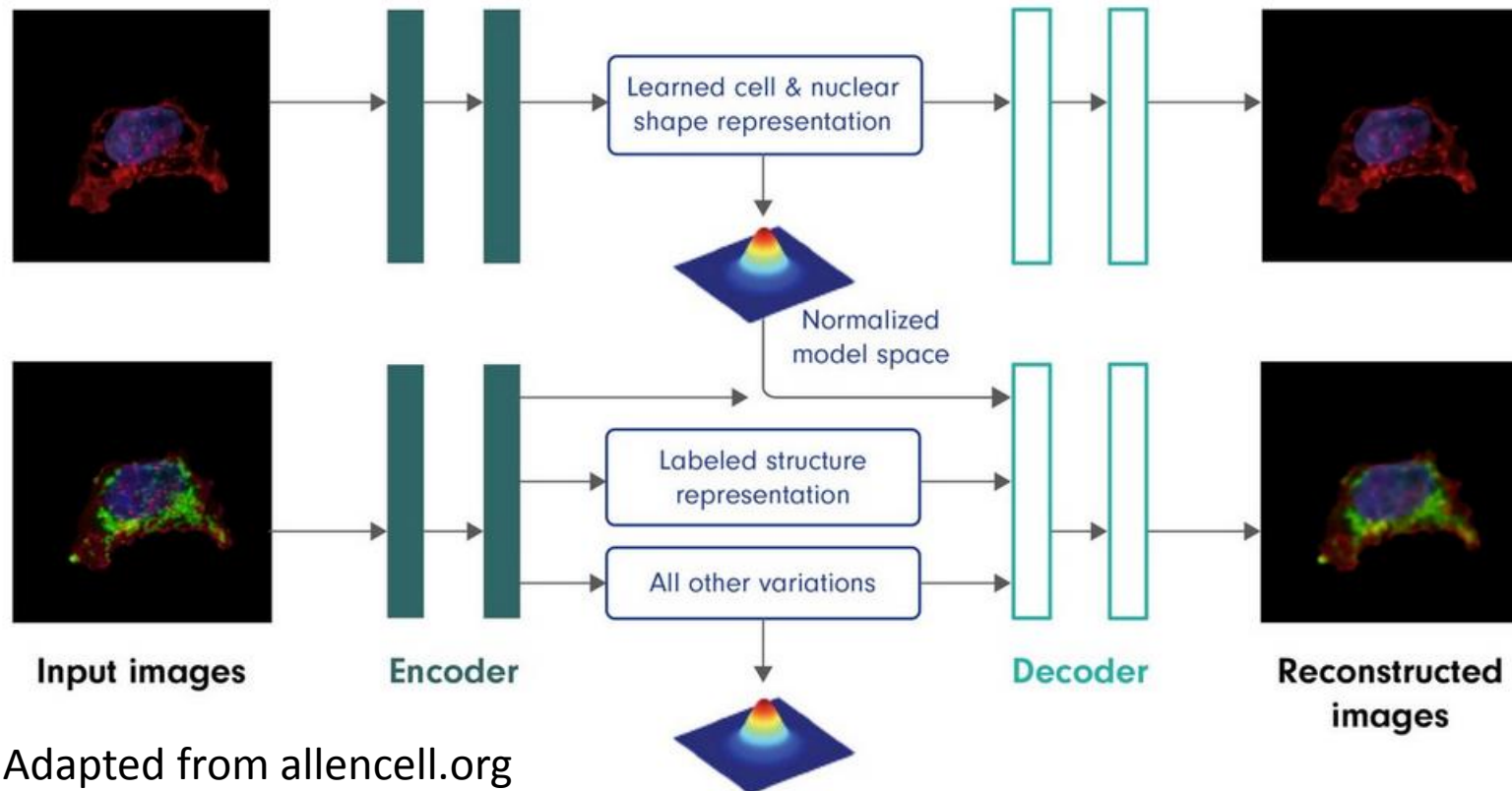


# More Machine Learning

# More Machine Learning

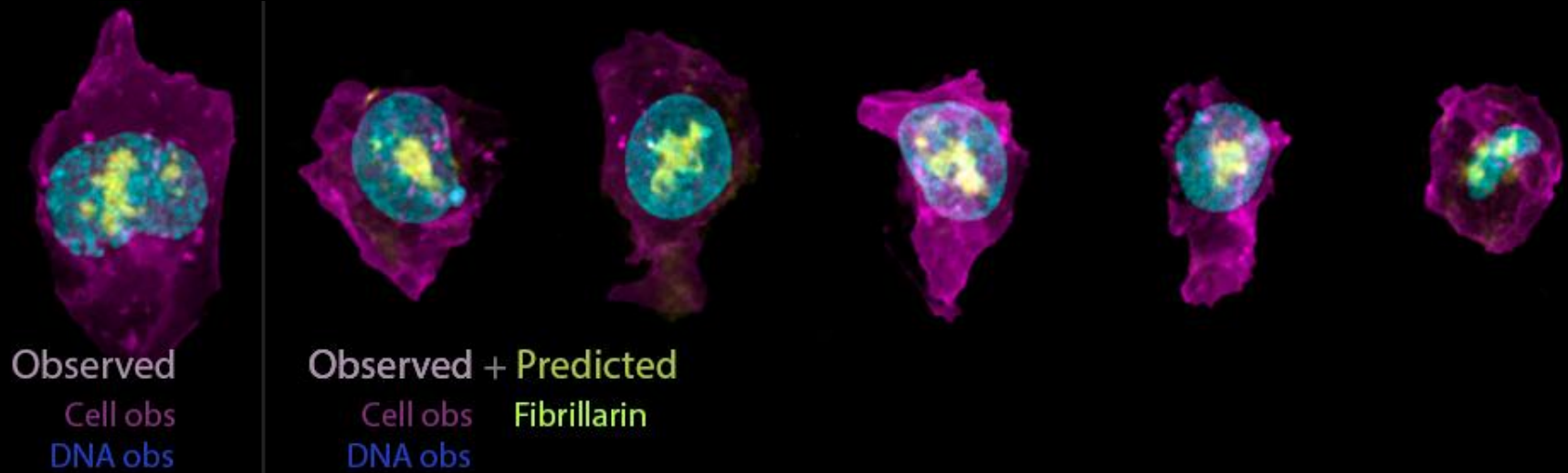
## ► Deep neural network autoencoder

- First Learn to reconstruct mem+nuc image from more sparse representation
- Next learn to reconstruct other channels based on mem+nuc



# More Machine Learning

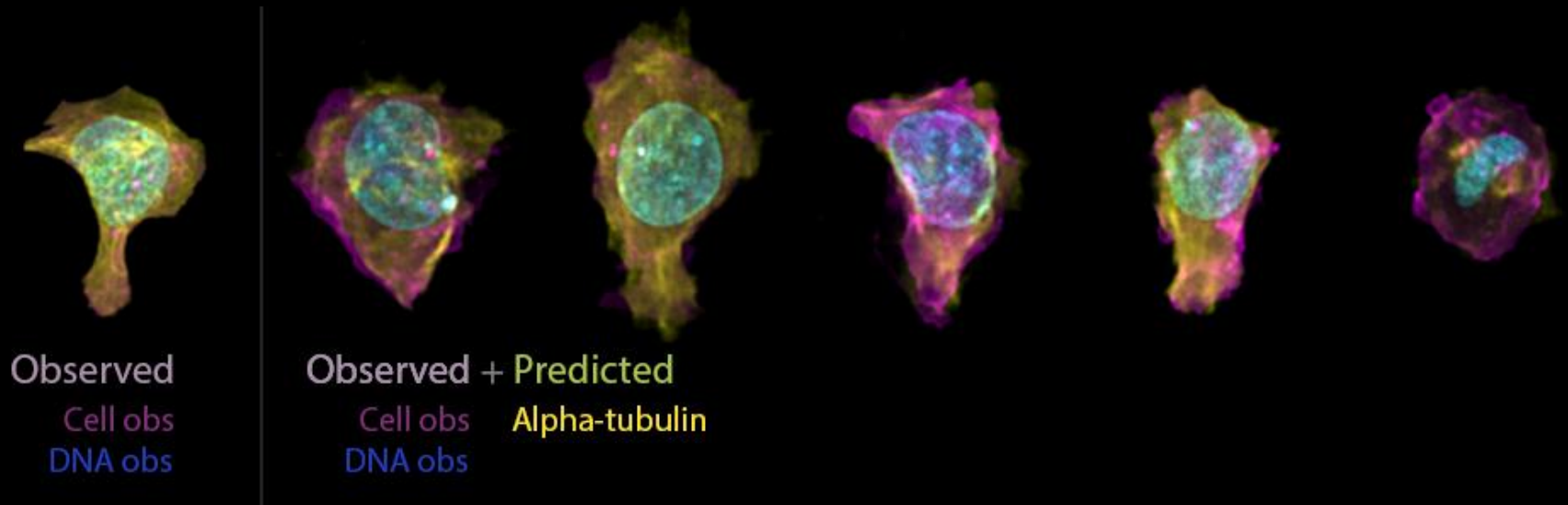
## ► Deep neural network autoencoder



Adapted from allencell.org

# More Machine Learning

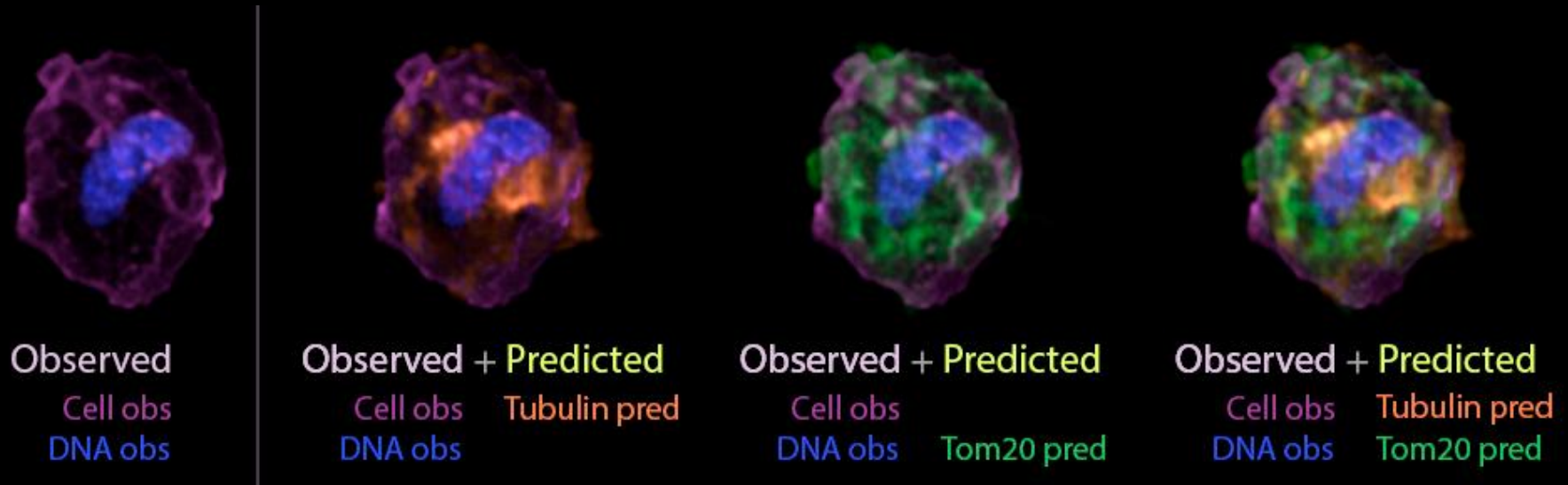
## ► Deep neural network autoencoder



Adapted from allencell.org

# More Machine Learning

## ► Deep neural network autoencoder



Adapted from allencell.org

- **Advantages:** construction of ‘atlases’ without feature engineering!
- **Disadvantages:** encoded model is a ‘black box’

**That's all!**

**Thanks!**

**:)**