

Automatic segmentation of punctate 3D super-resolution microscopy data

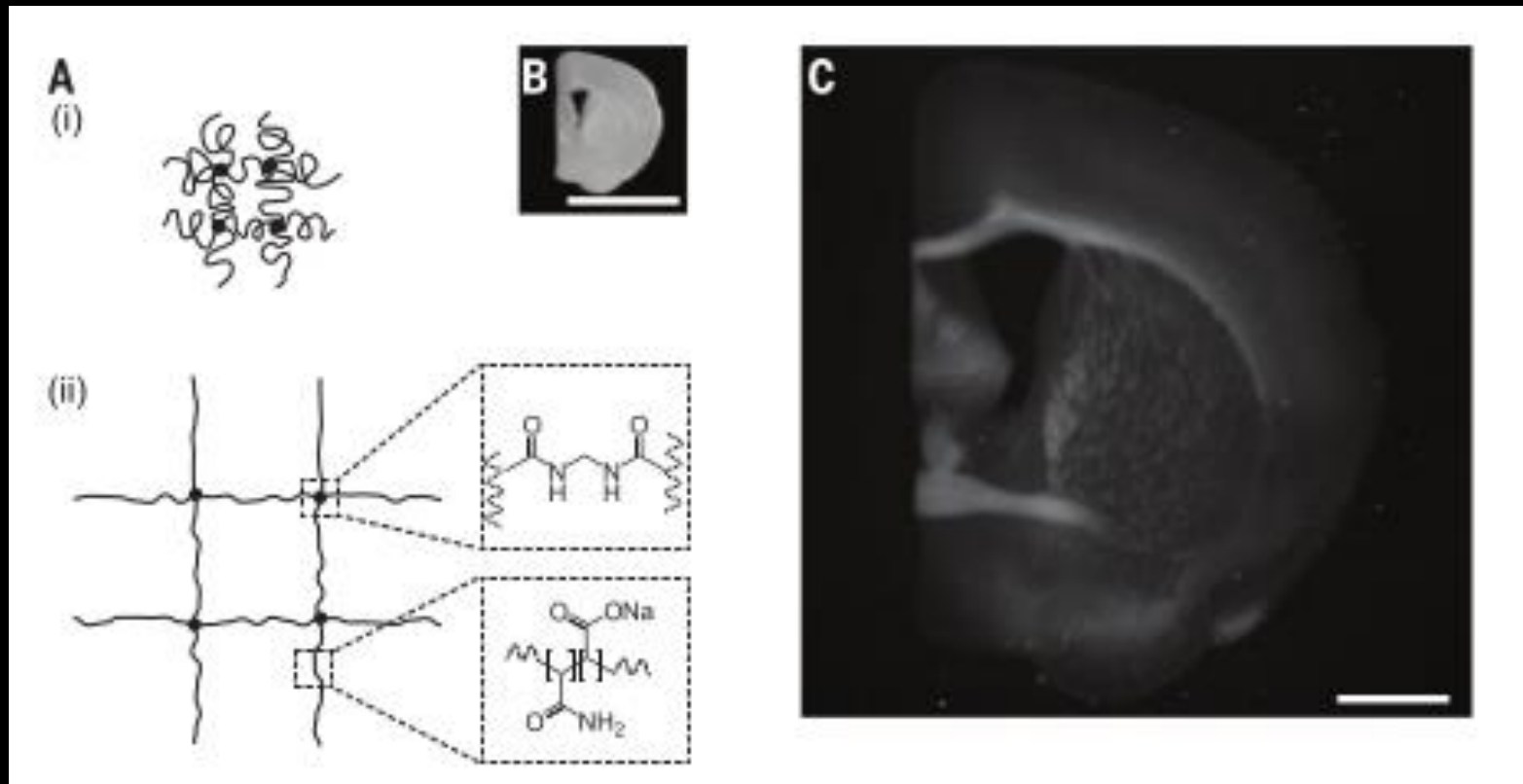
Zachary Steinberg

MIT PRIMES

May 23, 2016

Expansion Microscopy

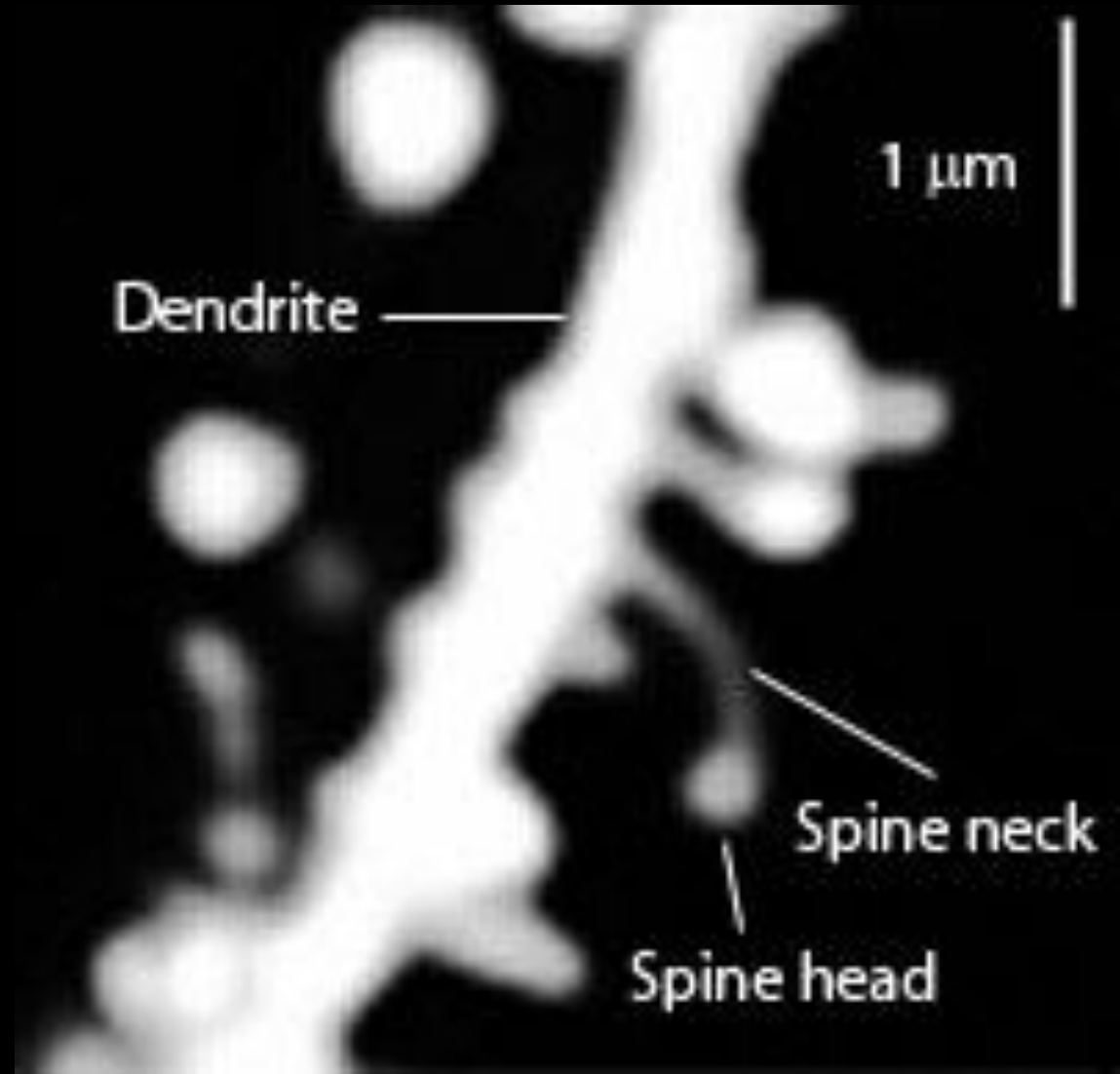
- Small structures with light-based microscopes



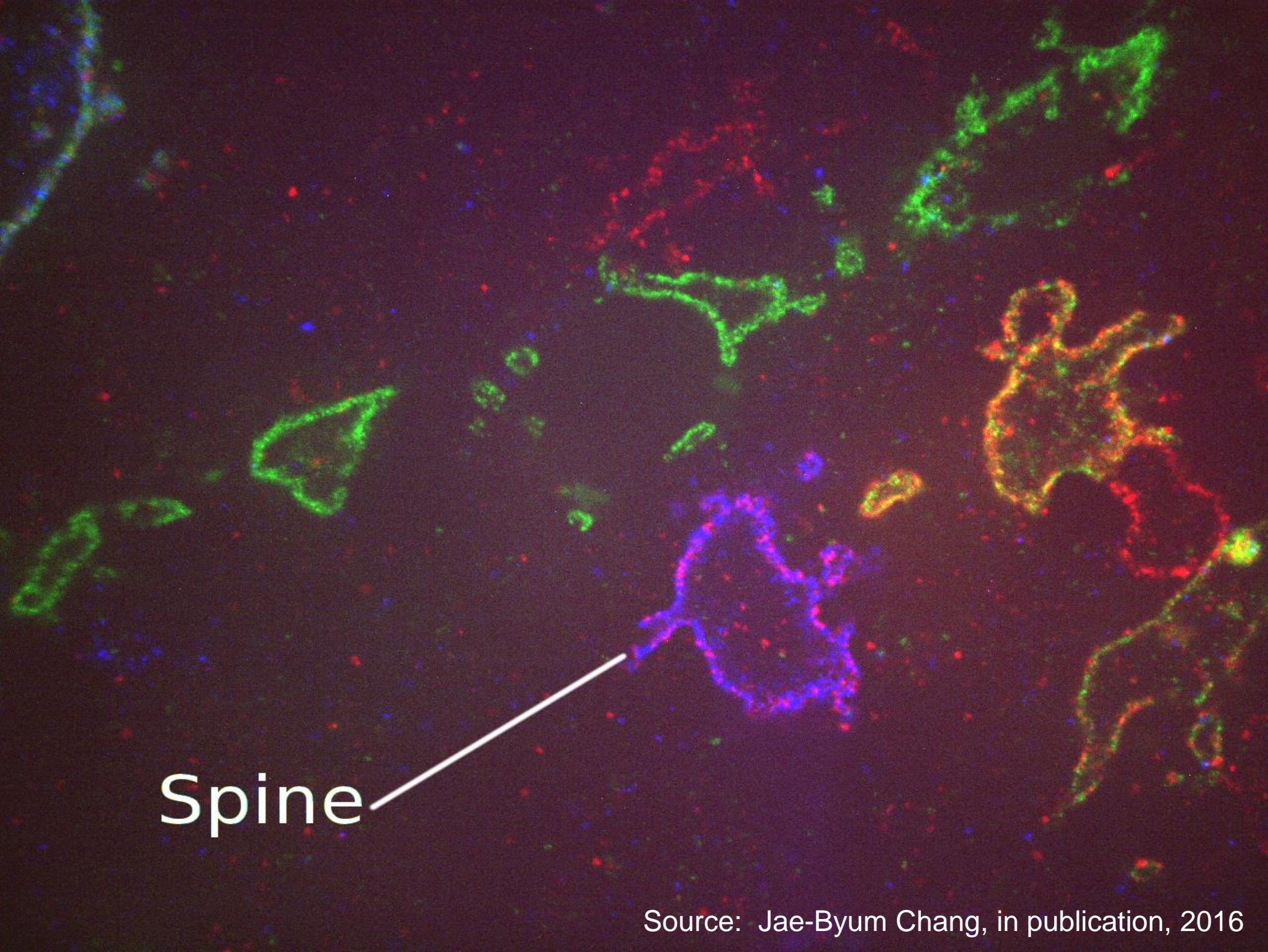
Source:
Chen et al, Science 2015

Dendritic spines

- Where synapses occur, connecting neurons
- Dendritic spines are too small to see with light, so usually imaged with electron microscopy (EM)



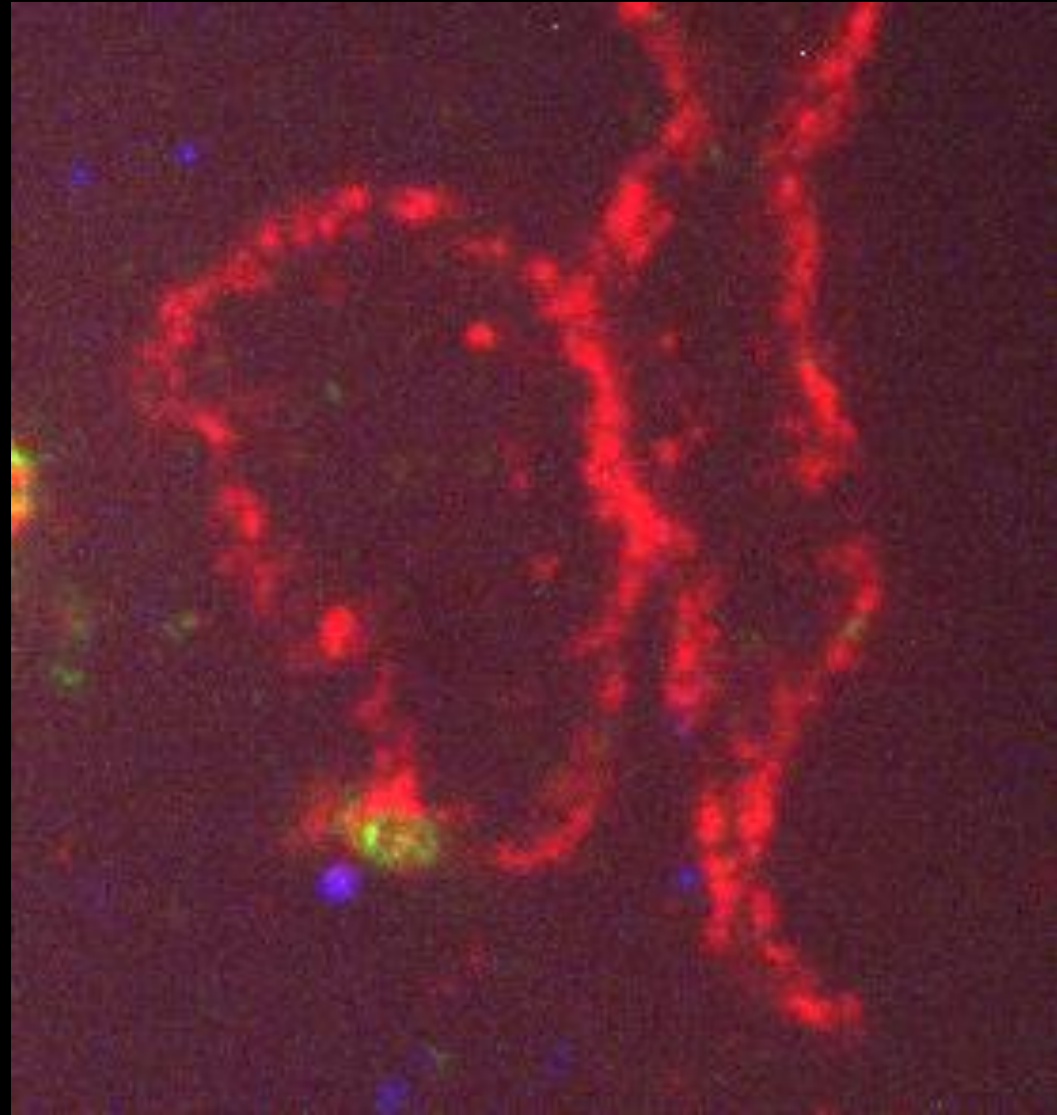
Source: public domain by user CopperKettle



Spine

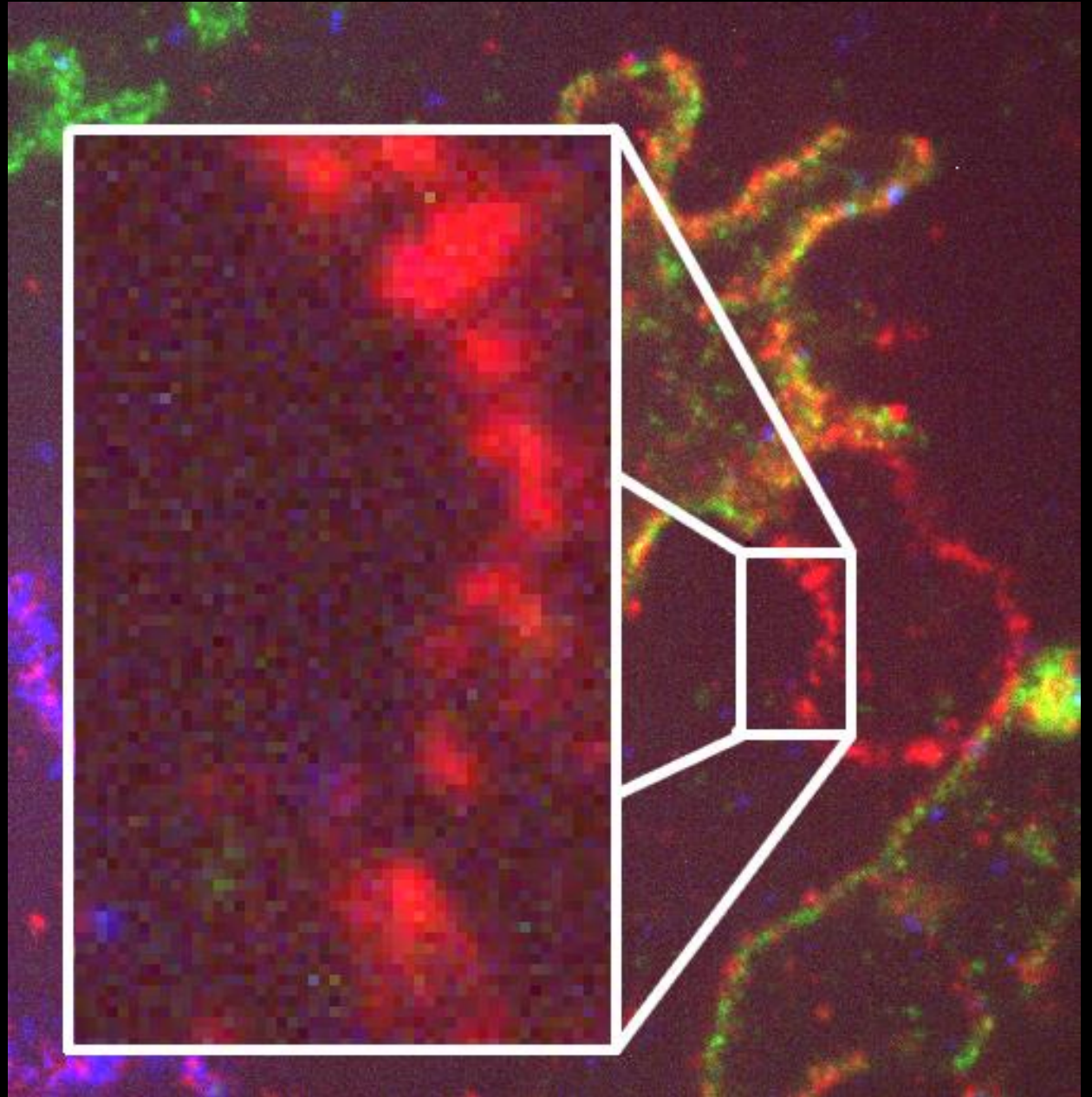
Connectomics

- Mapping connections in the brain
- The challenge: telling neurons apart in an automated way
- Methods exist for low resolution, but not high



Challenges

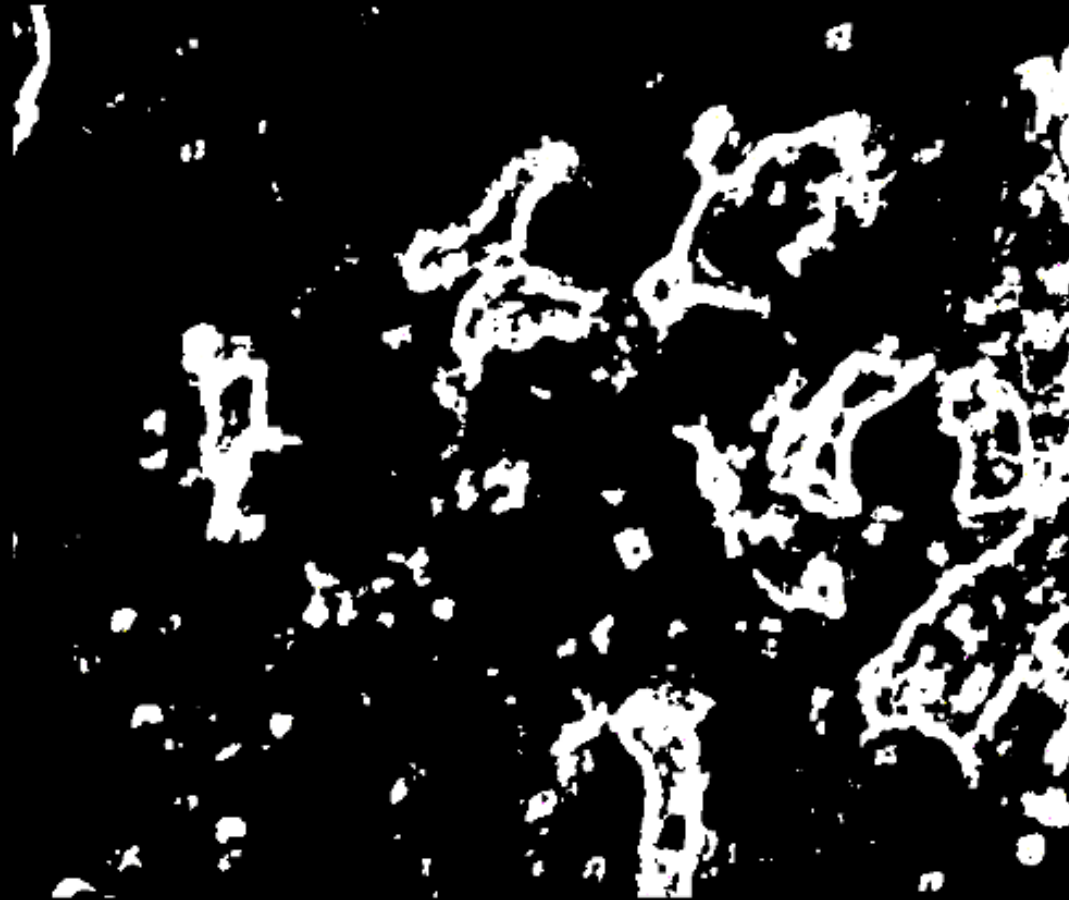
- 1080 x 1200 x ~200 – almost 1GB!
- Punctate



Our Solution

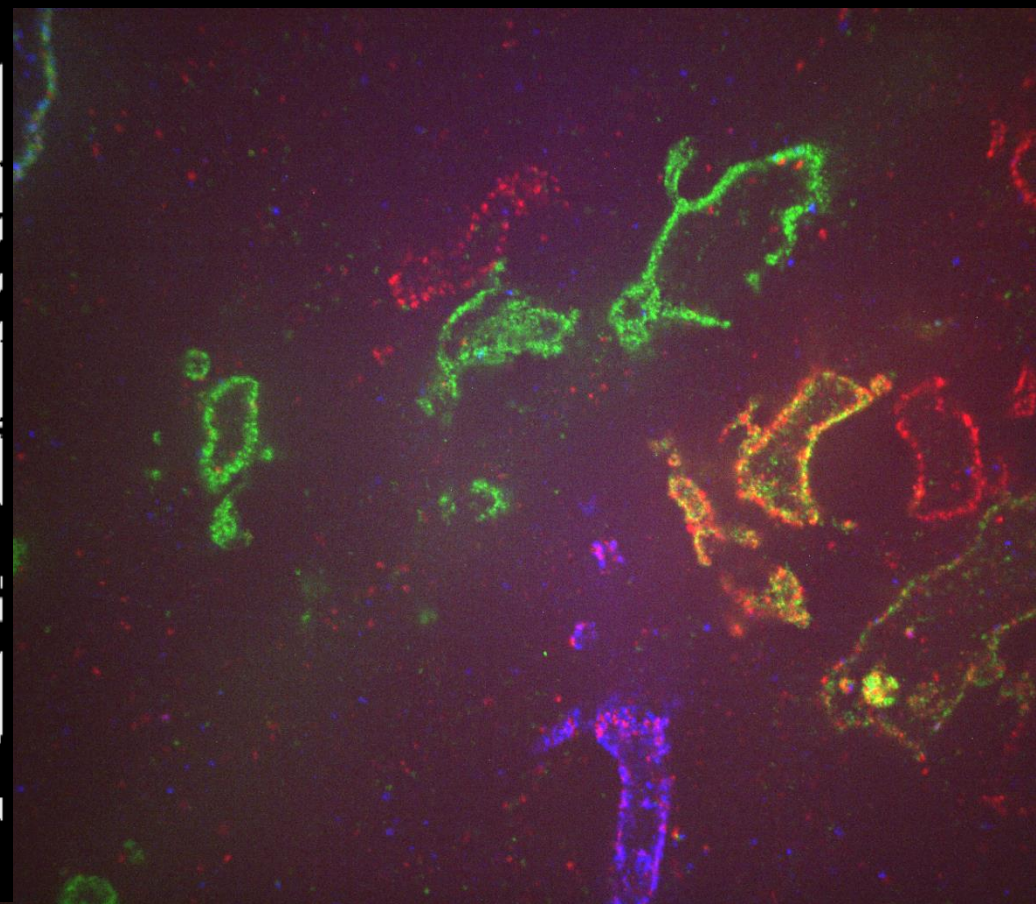
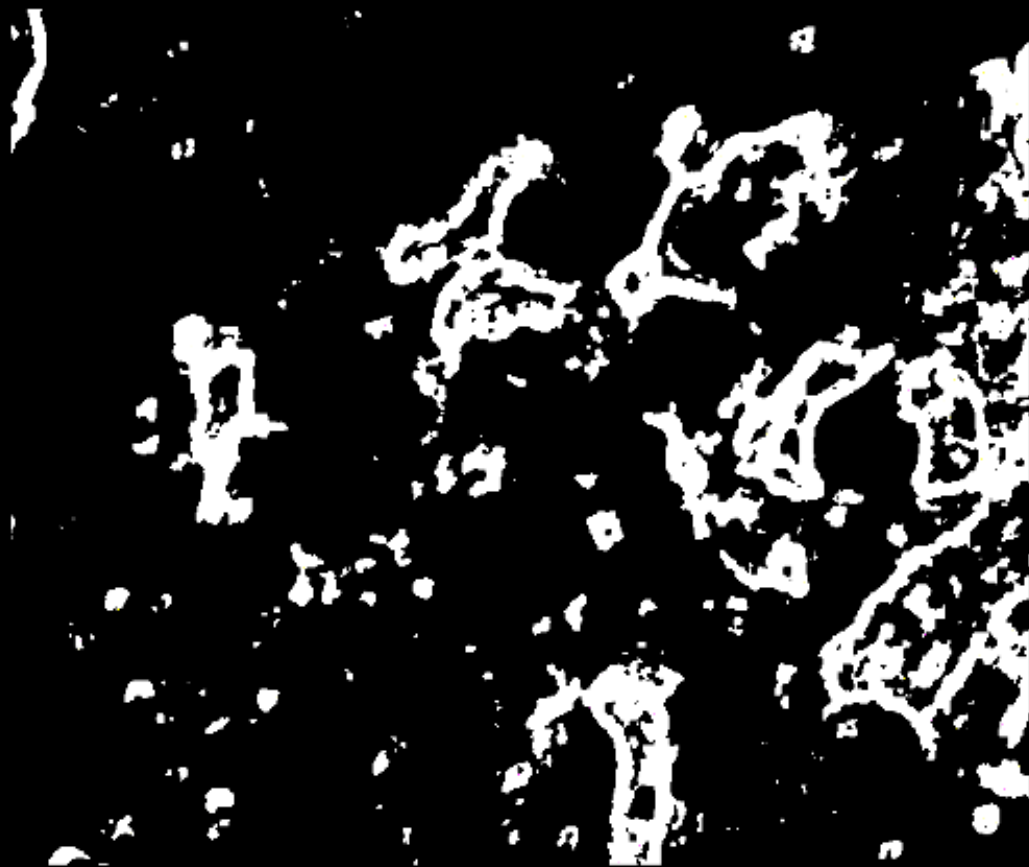
- Mask
- Clustering
- Connect components

Step 1: Mask

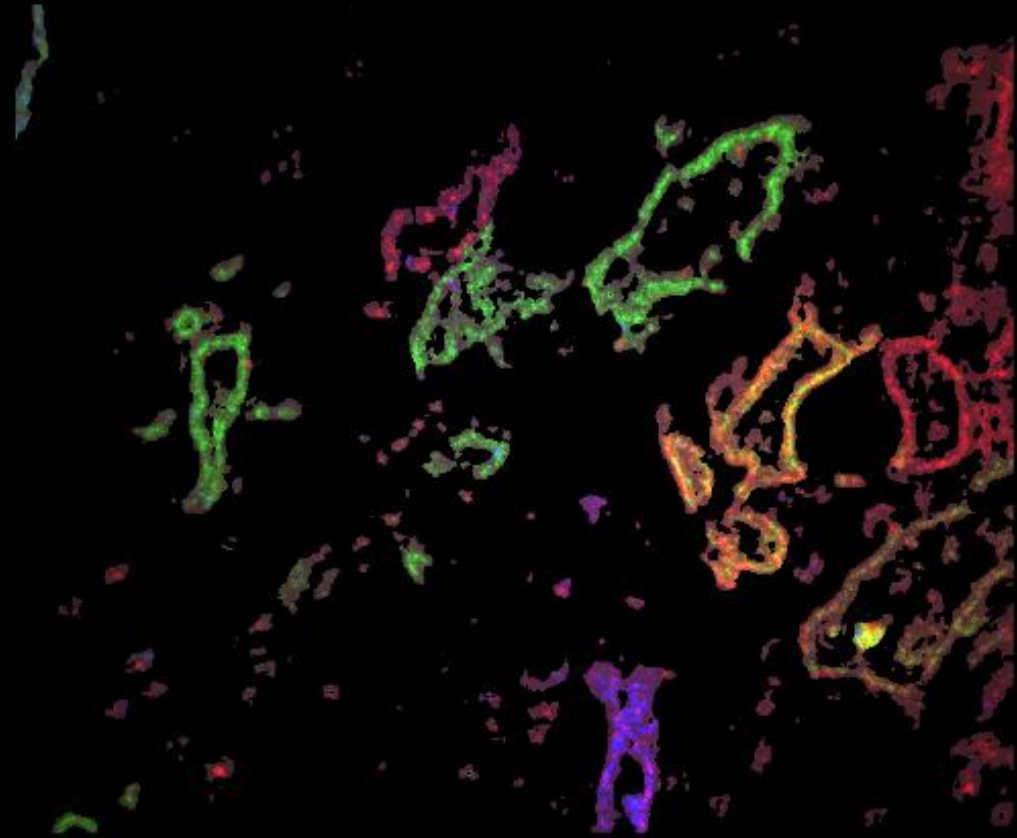
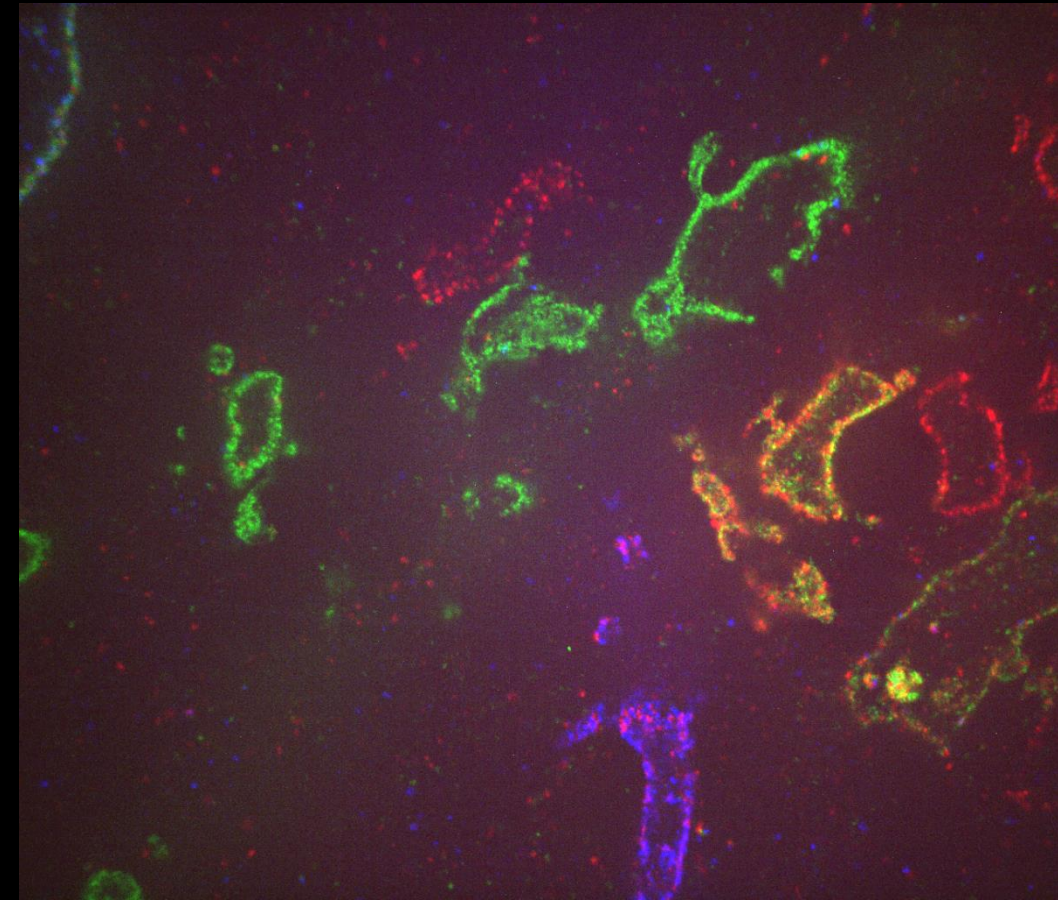


- Provided as an “oracle” to discard pixels
- Convolutional neural network

Mask vs base image



With mask applied

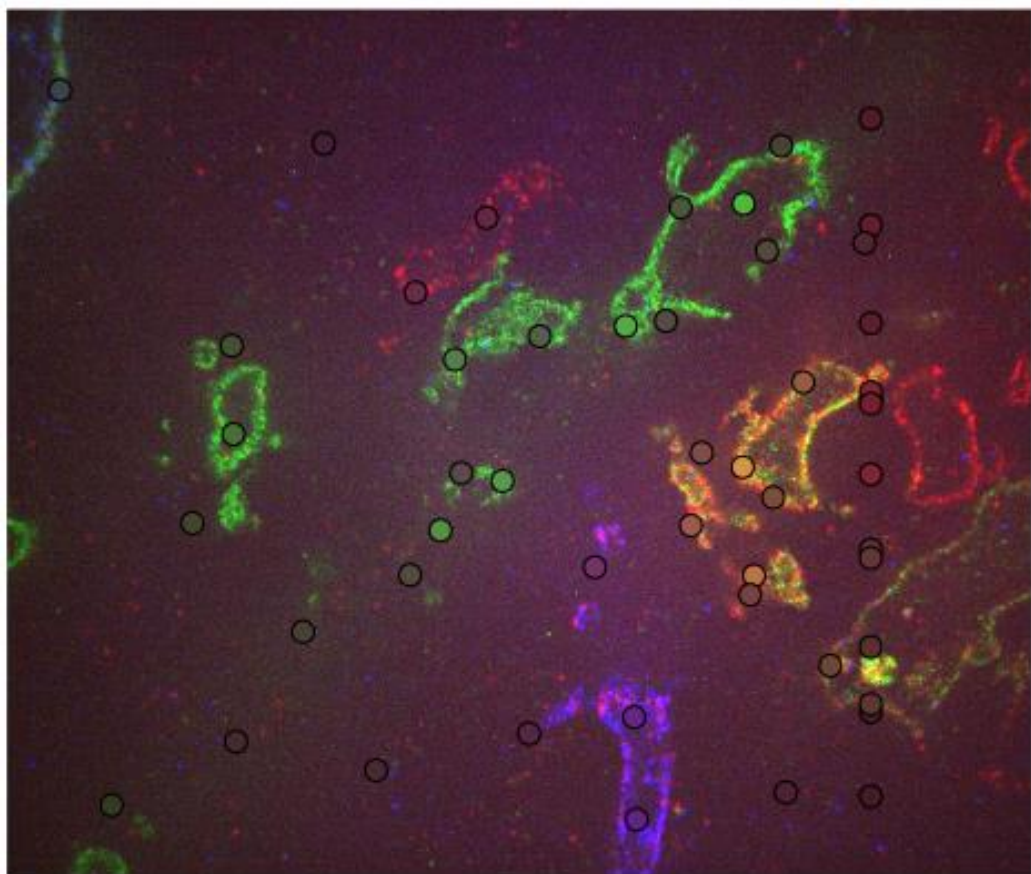


Step 2: Clustering

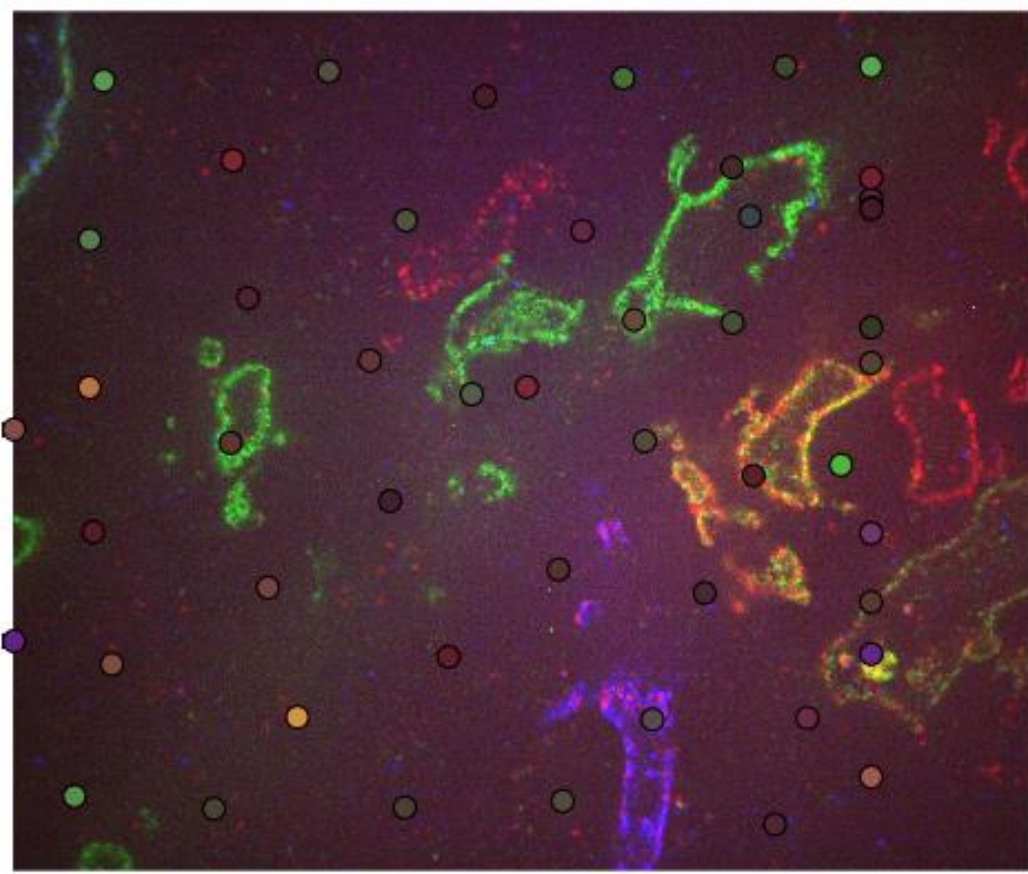
- K-means:
Classify similar points together, forming k groups
- RGB vs RGBXYZ
- Choice of k



The effects of masking on clustering



Mask

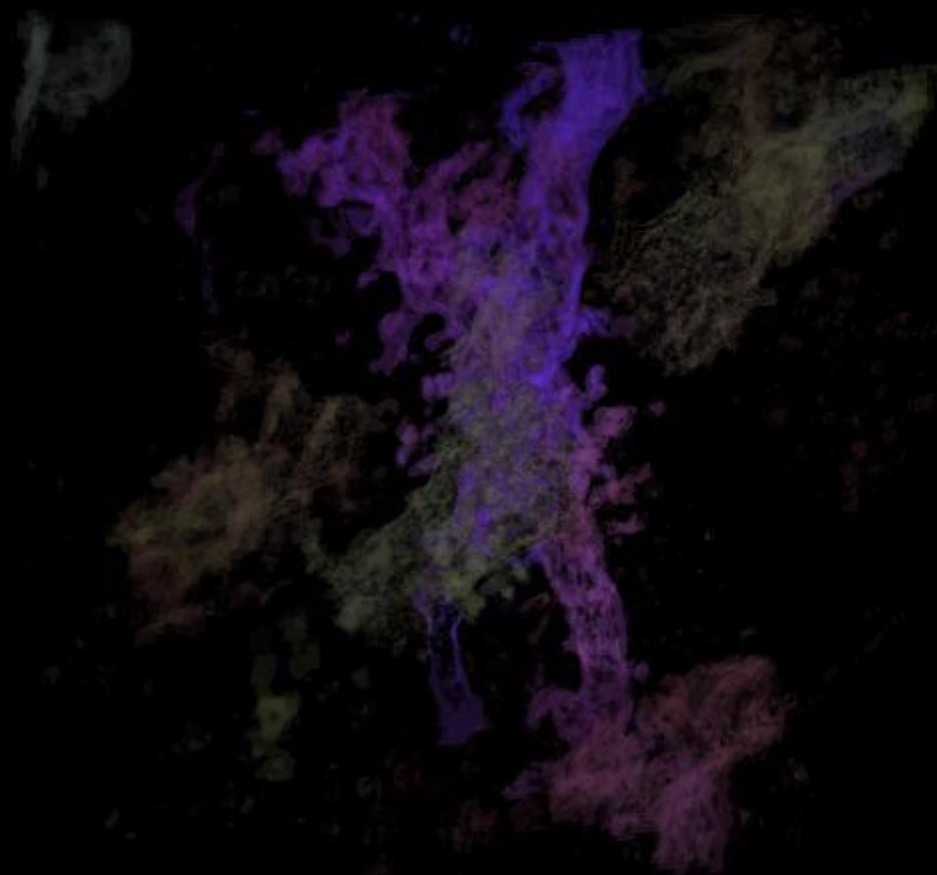


No Mask

Step 3: Connected components

- Each “neuron” is actually made up of multiple, slightly differing centroids
- Merge together regions that touch
- After, combine individual slices to create the final product

Results



Future work

- Differentiating within a region
- Memory usage
- Further merging
- Using more of the z axis for further benefits

Acknowledgements

- My mentors
- MIT PRIMES
- The Boyden lab
- My Family
- The brain, both for being amazing to study and amazing to study with

Questions?