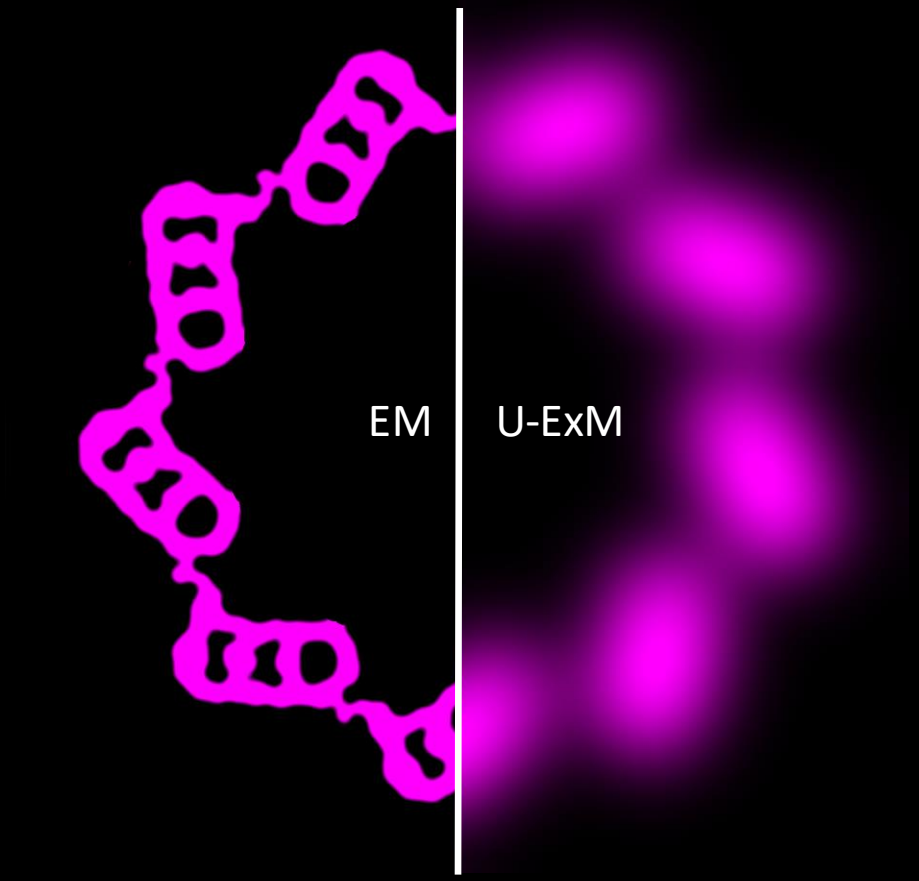
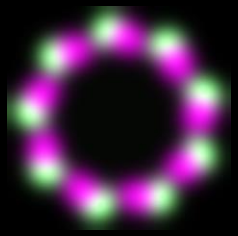
EM to U-ExM resolution



Centriole Lab

data

simulation



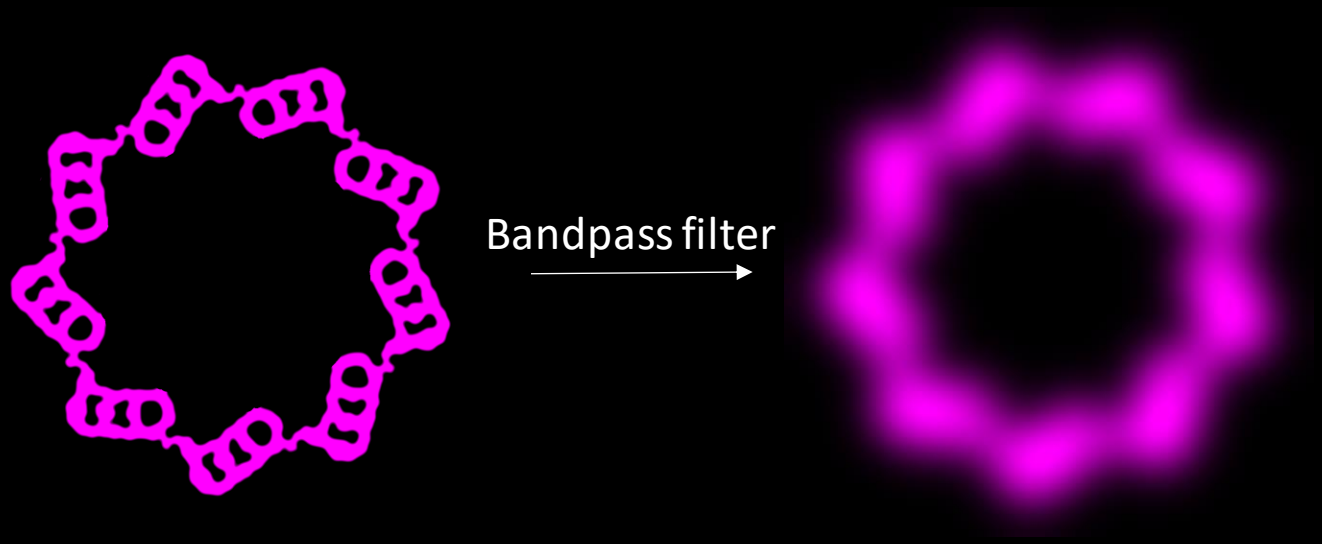
Gambarotto et al.
Nature Methods
2019

data

simulation

Le Guennec et al.
Science Advances
2020

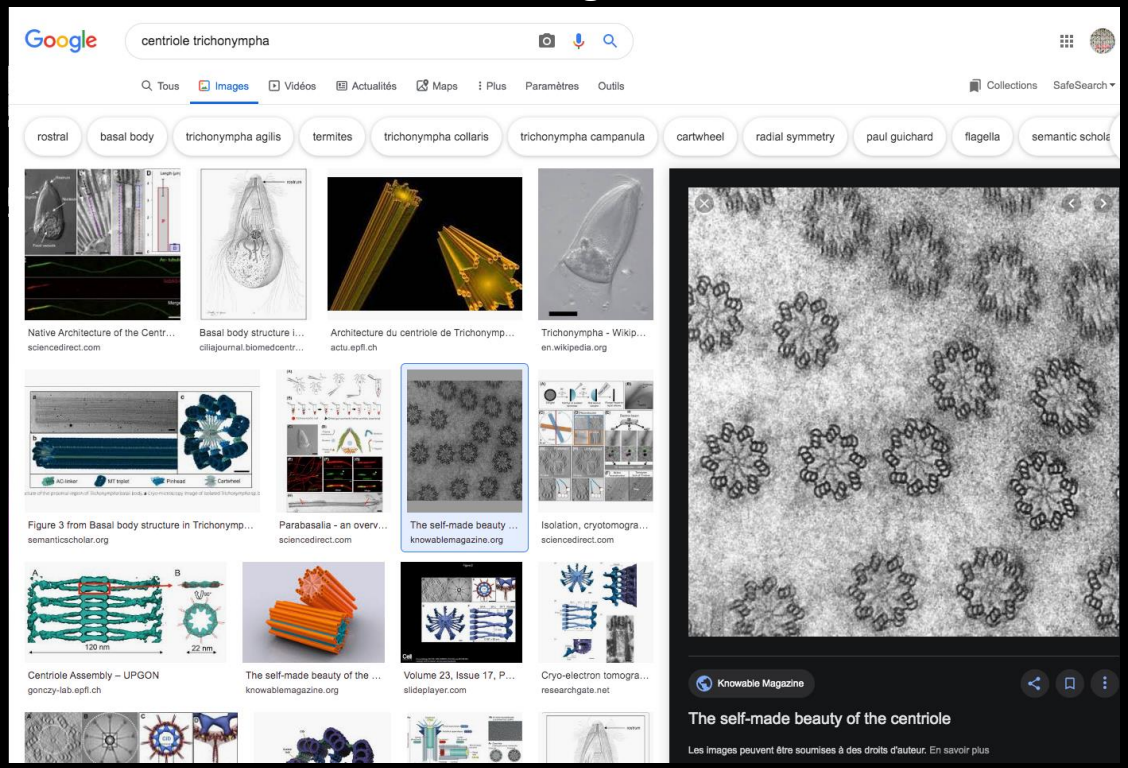
Simulation of ExM data from drawing of cryoEM images using imageJ



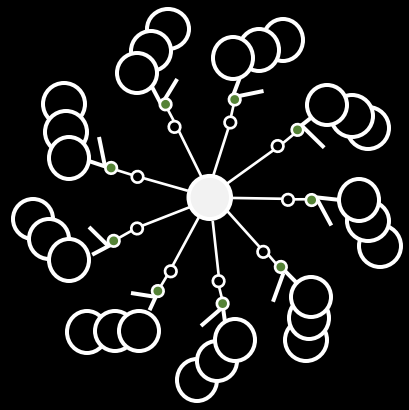
Let's start with the initial image

Find a nice image of your object of interest

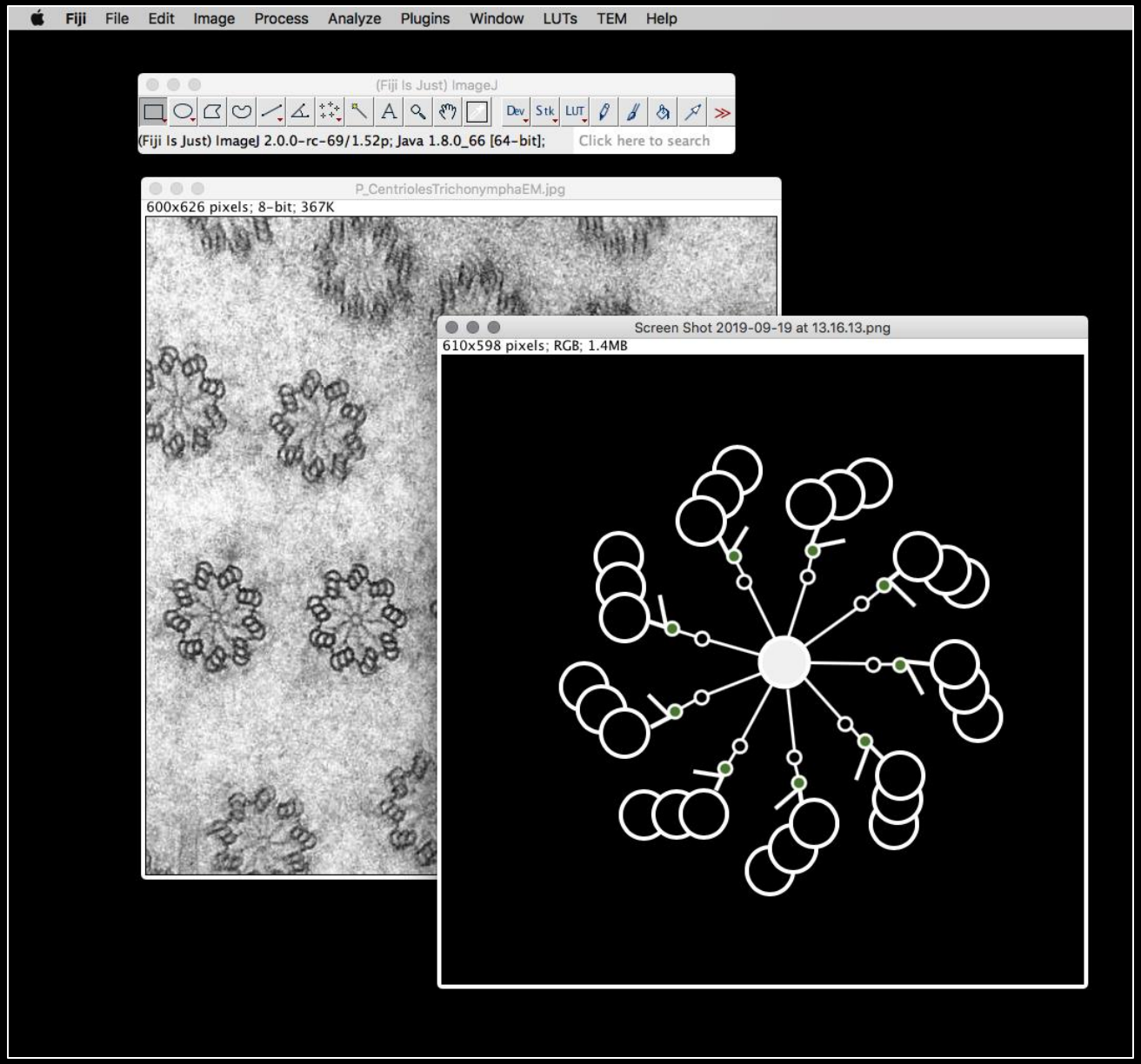
Google



Drawing



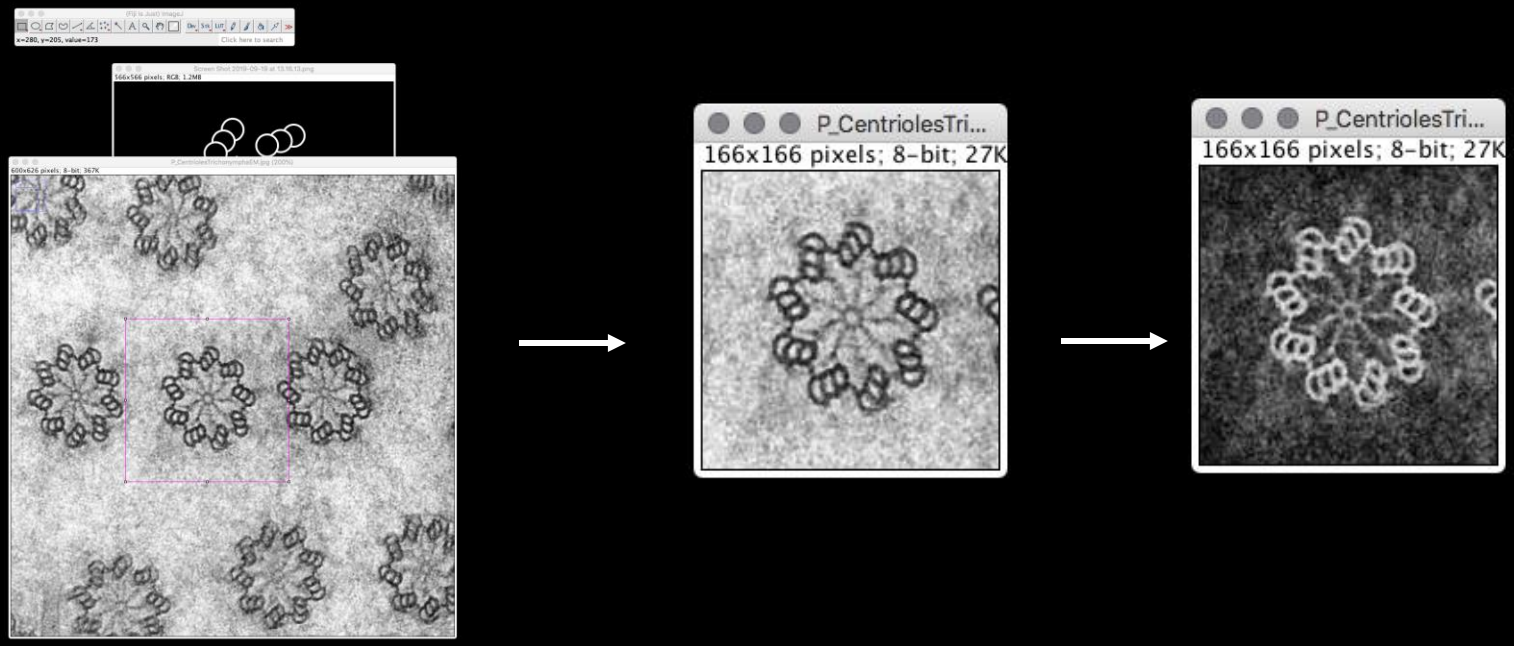
Open them with ImageJ



Prepare your image – EM image

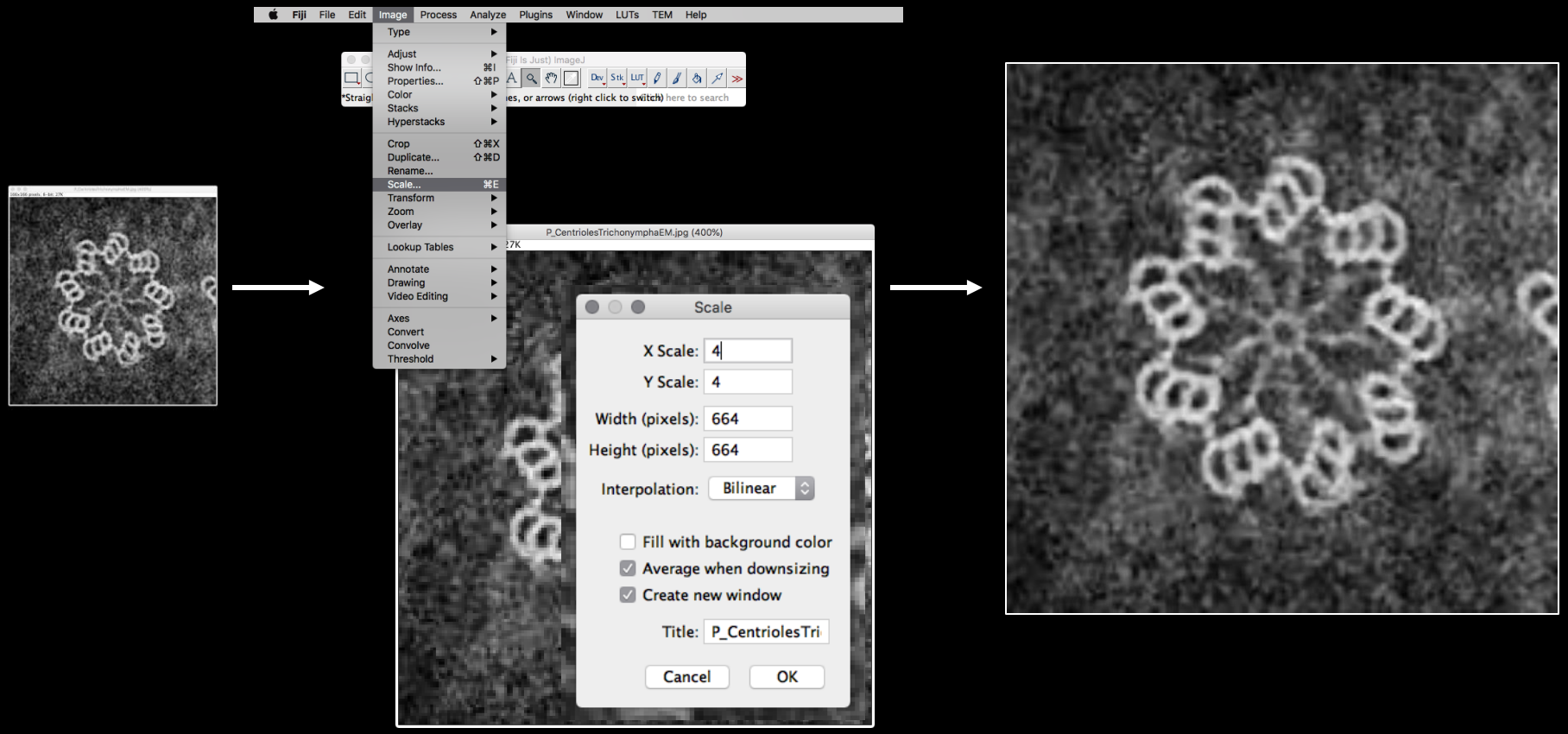
Crop the region of interest

Contrast – white on black – similar to fluorescent microscopy data



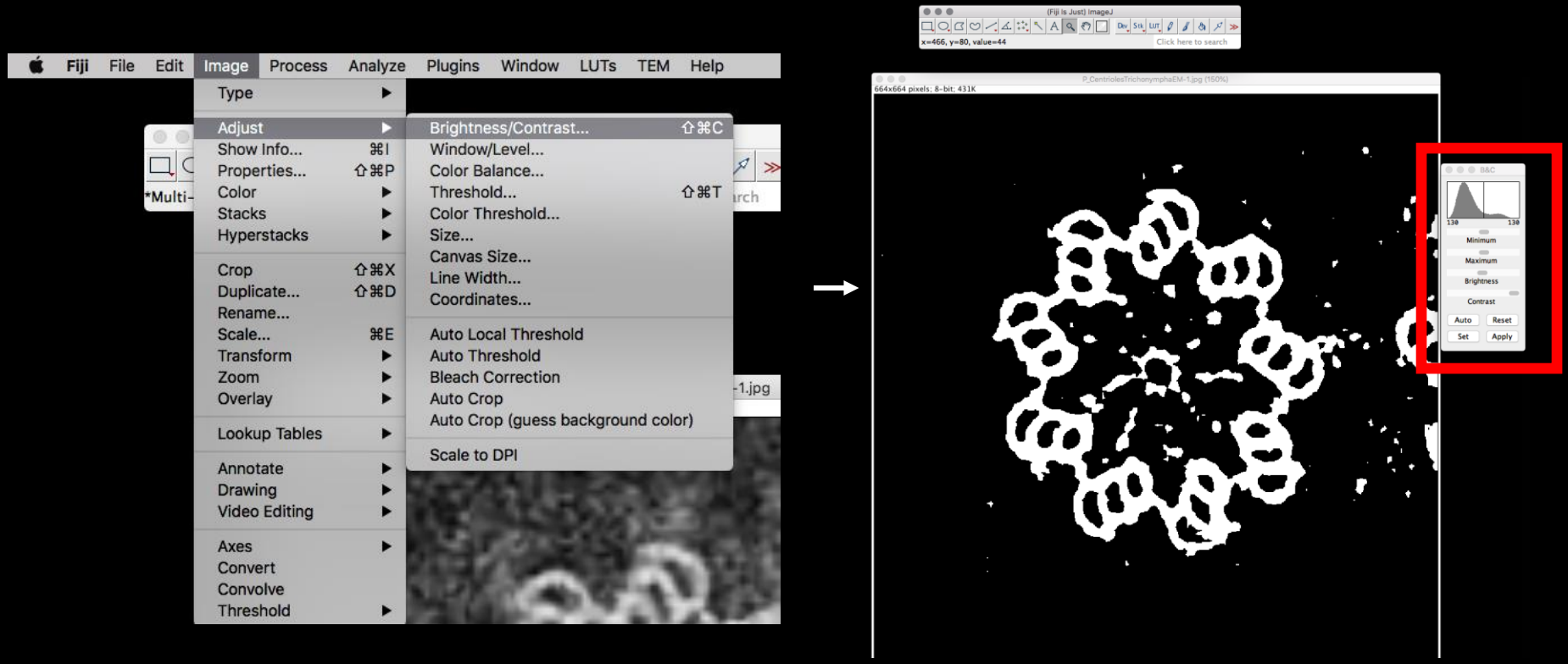
Prepare your image – EM image

Rescale your image



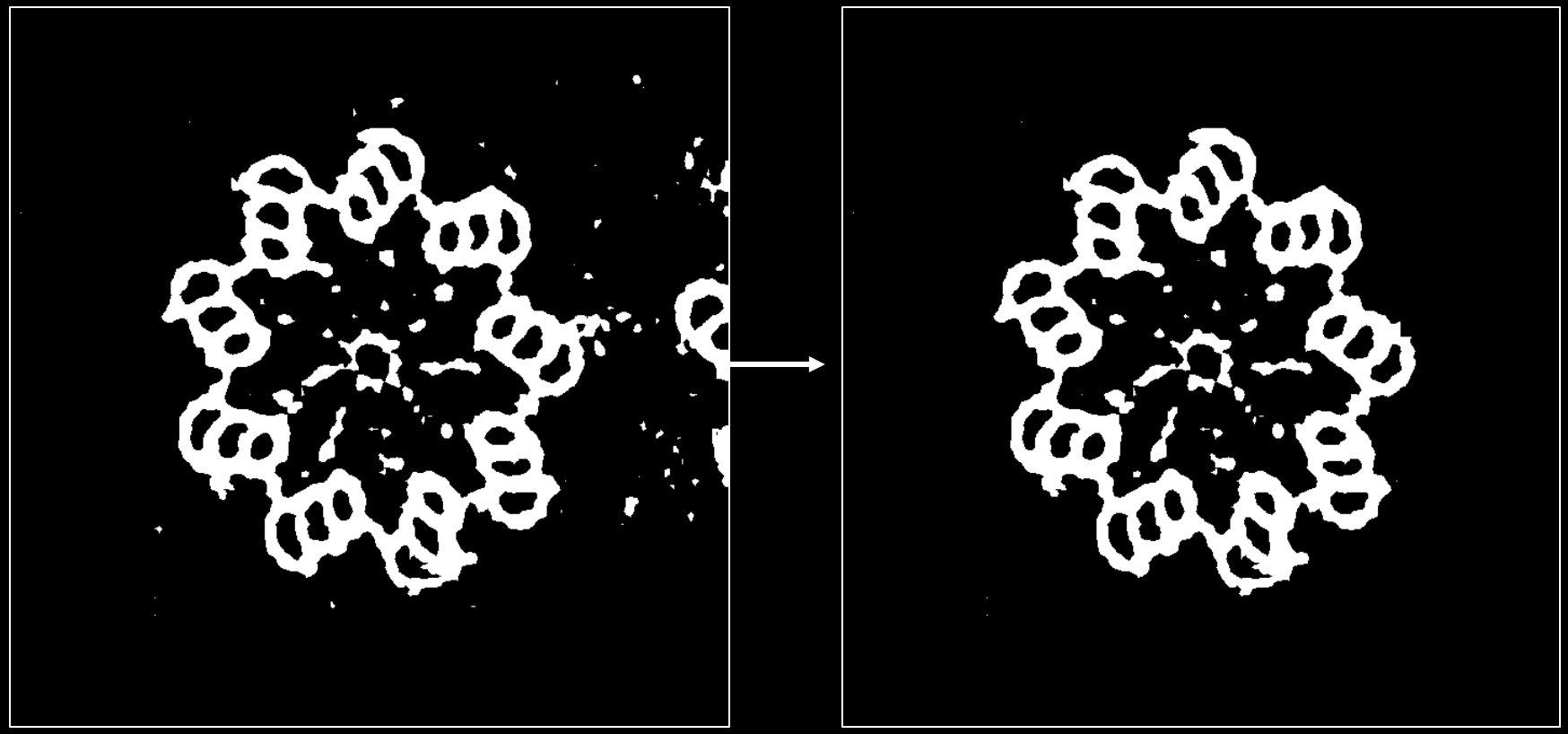
Prepare your image – EM image

Adjust the contrast

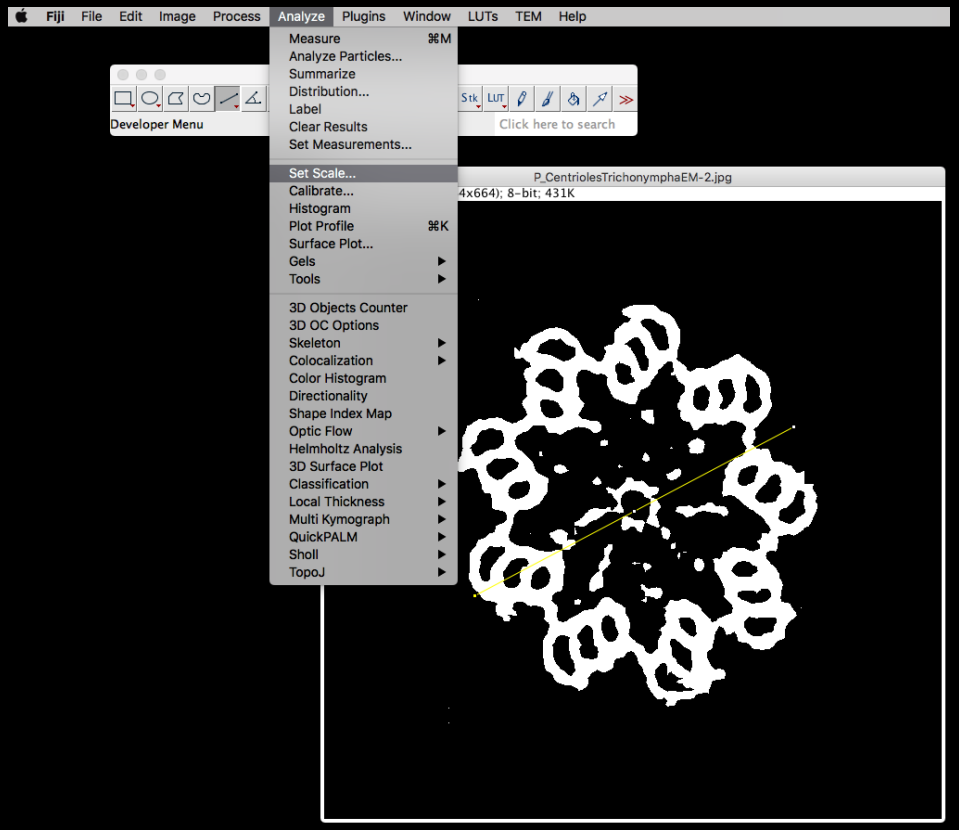


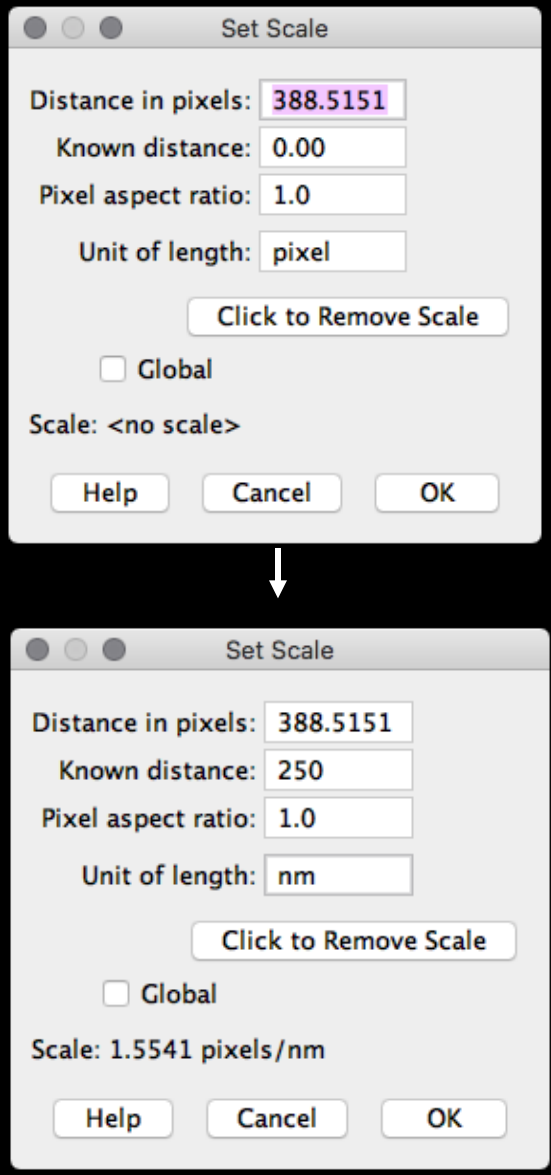
Let's start with the drawing

Clean your image



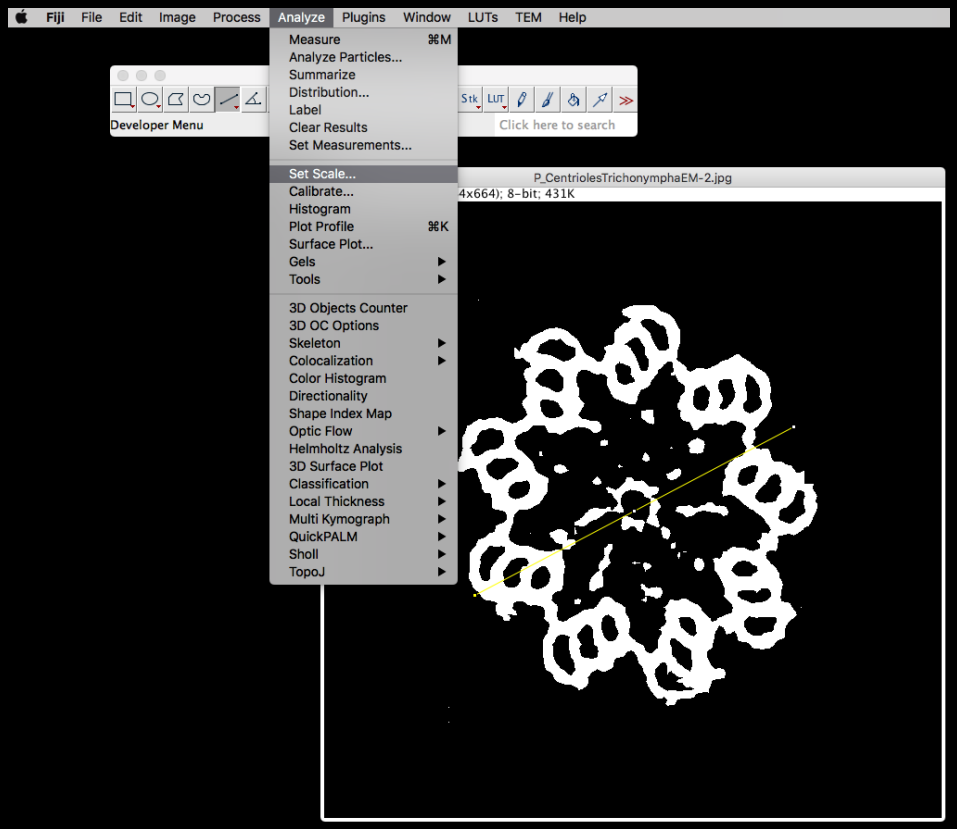
Scale you image – real size

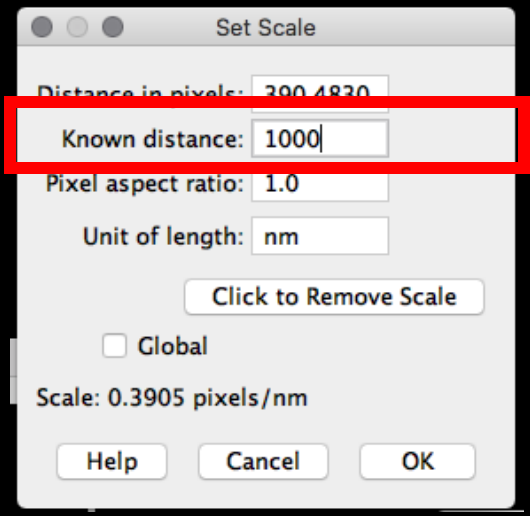




Save the image - B-ExM!!!

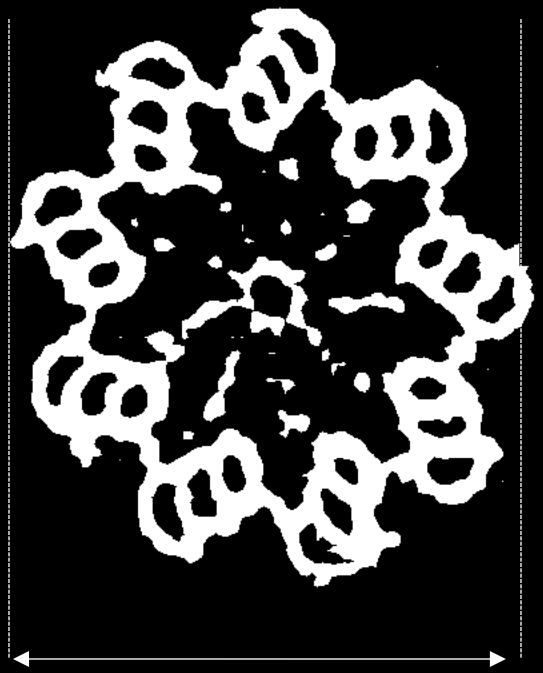
Scale you image – After expansion (4X)



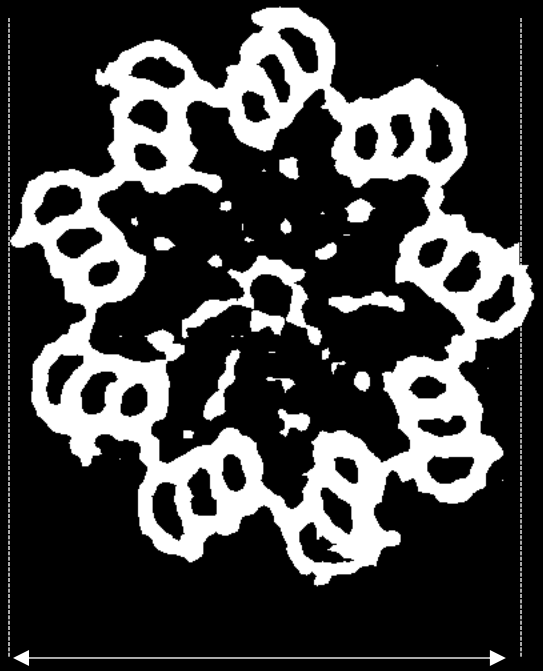


Save the image - after ExM!!!

Microtubule triplet in ExM

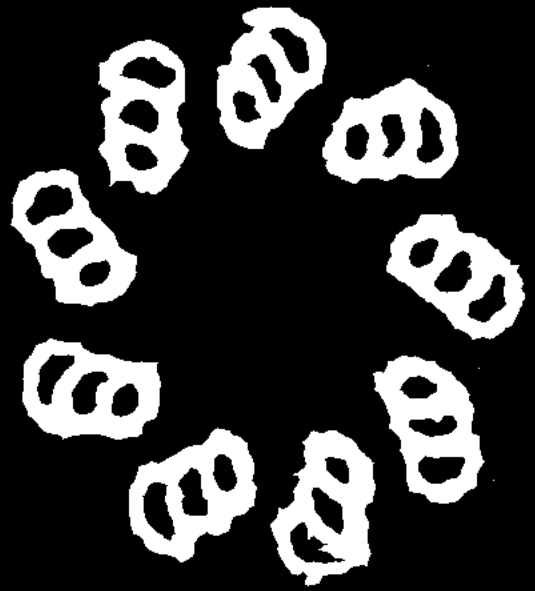


Microtubule triplet in ExM



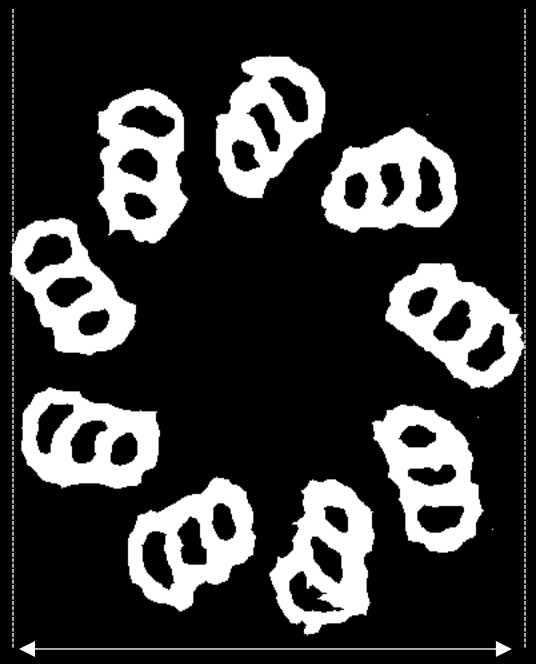
1000nm

Clean

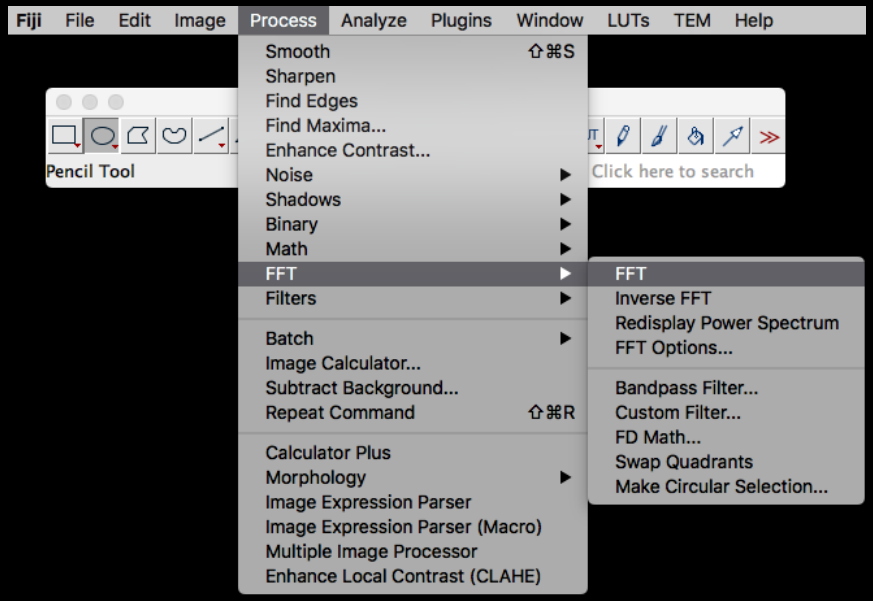


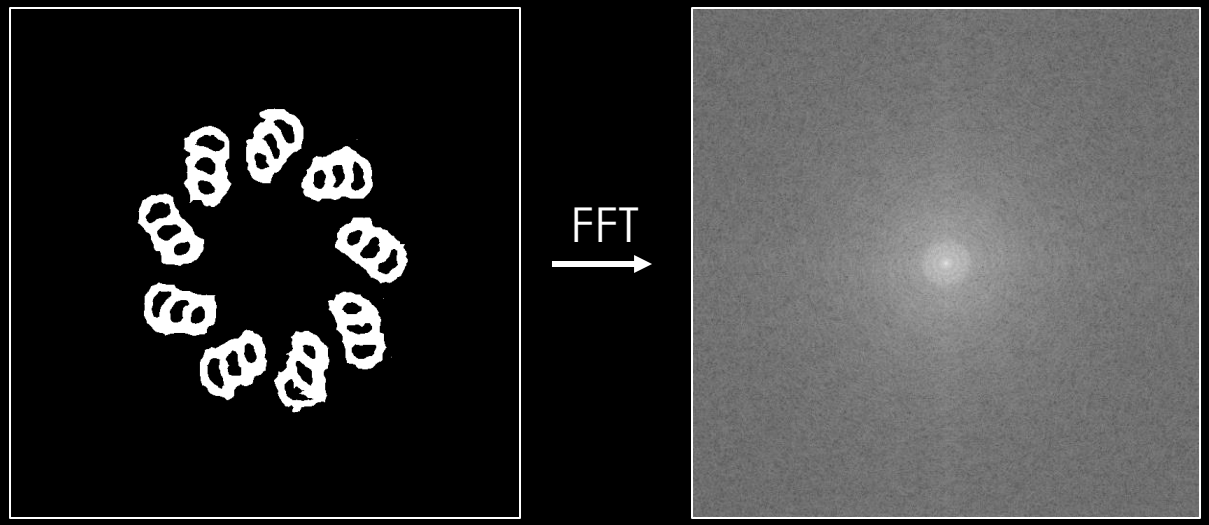
Save the image - triplet

Filtering at the resolution of ExM

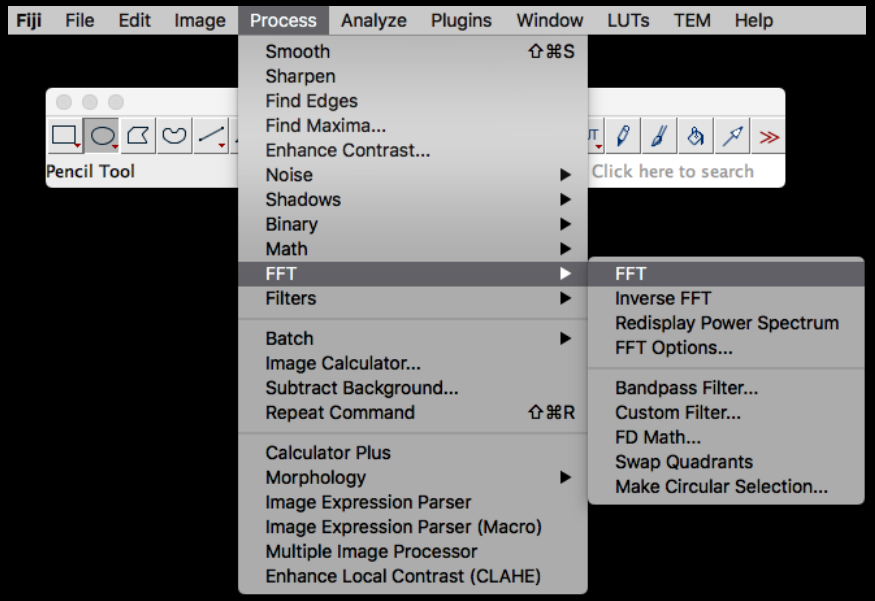


Bandpass filter in the fourier space

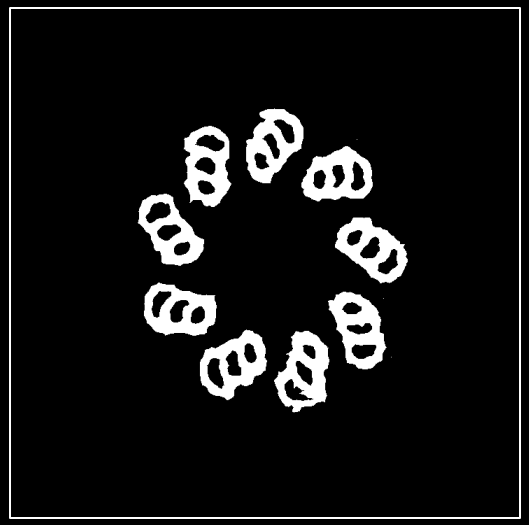


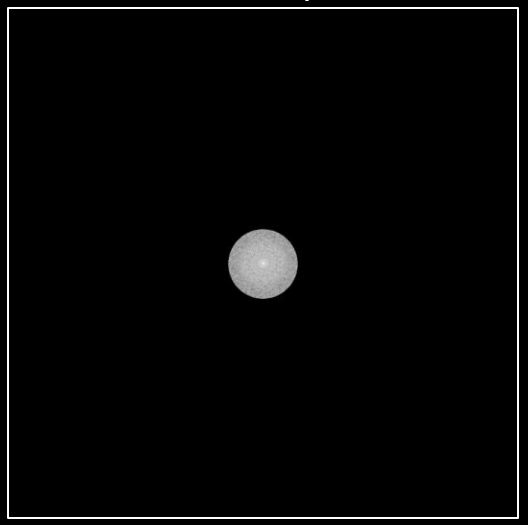


Bandpass filter in the fourier space

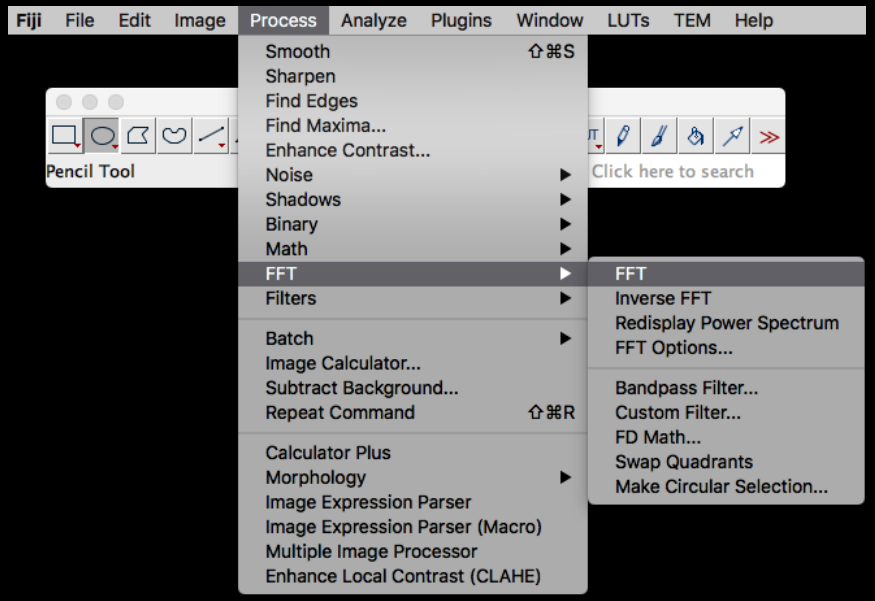


Bandpass filter

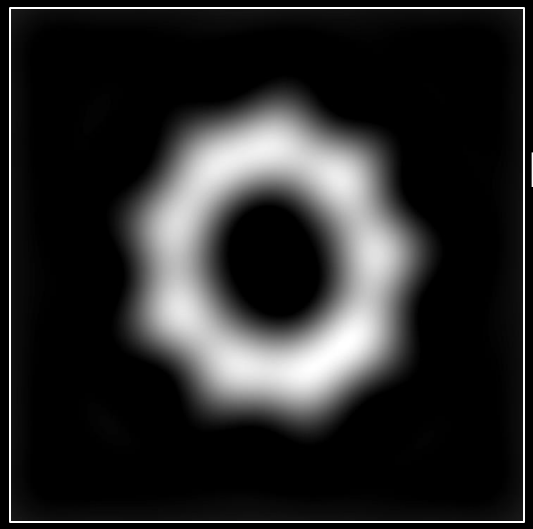




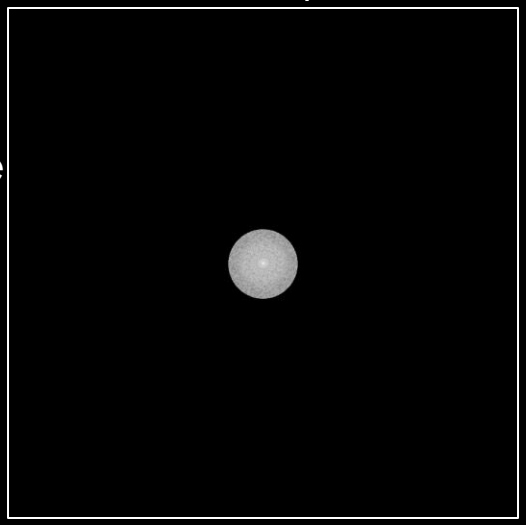
Bandpass filter in the fourier space



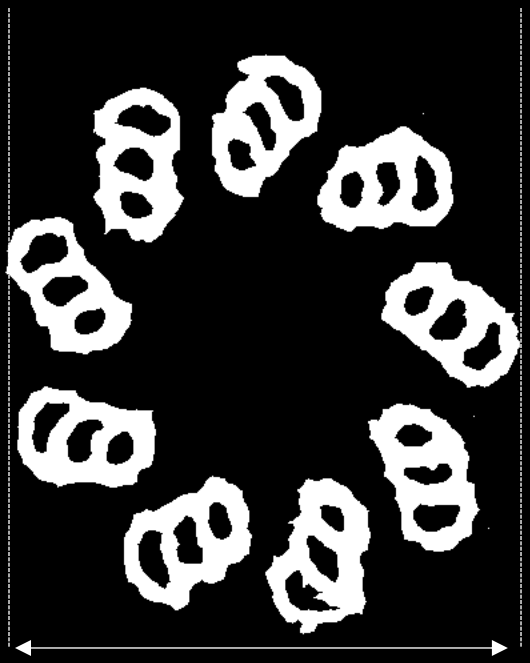
Bandpass filter



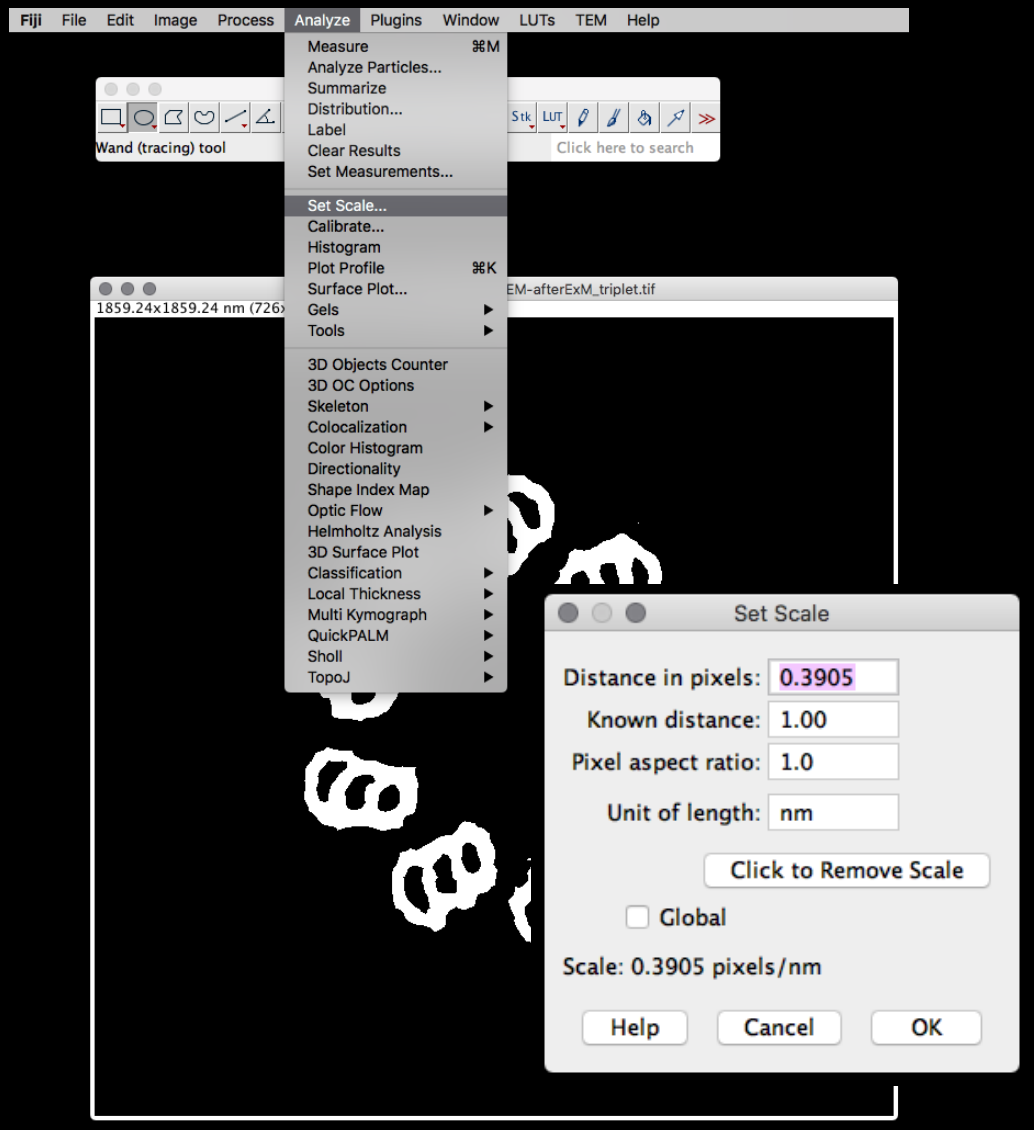
Inverse FFT



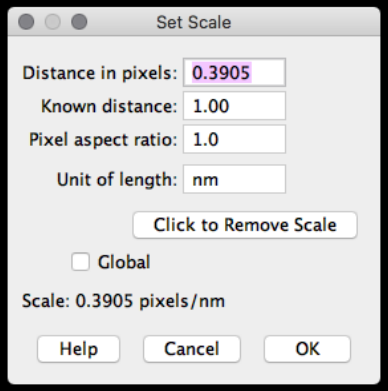
Filtering at the resolution of ExM



1000nm



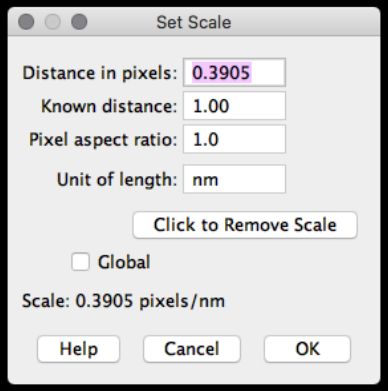
Filtering at the resolution of ExM



1nm = 0.3905 pixel

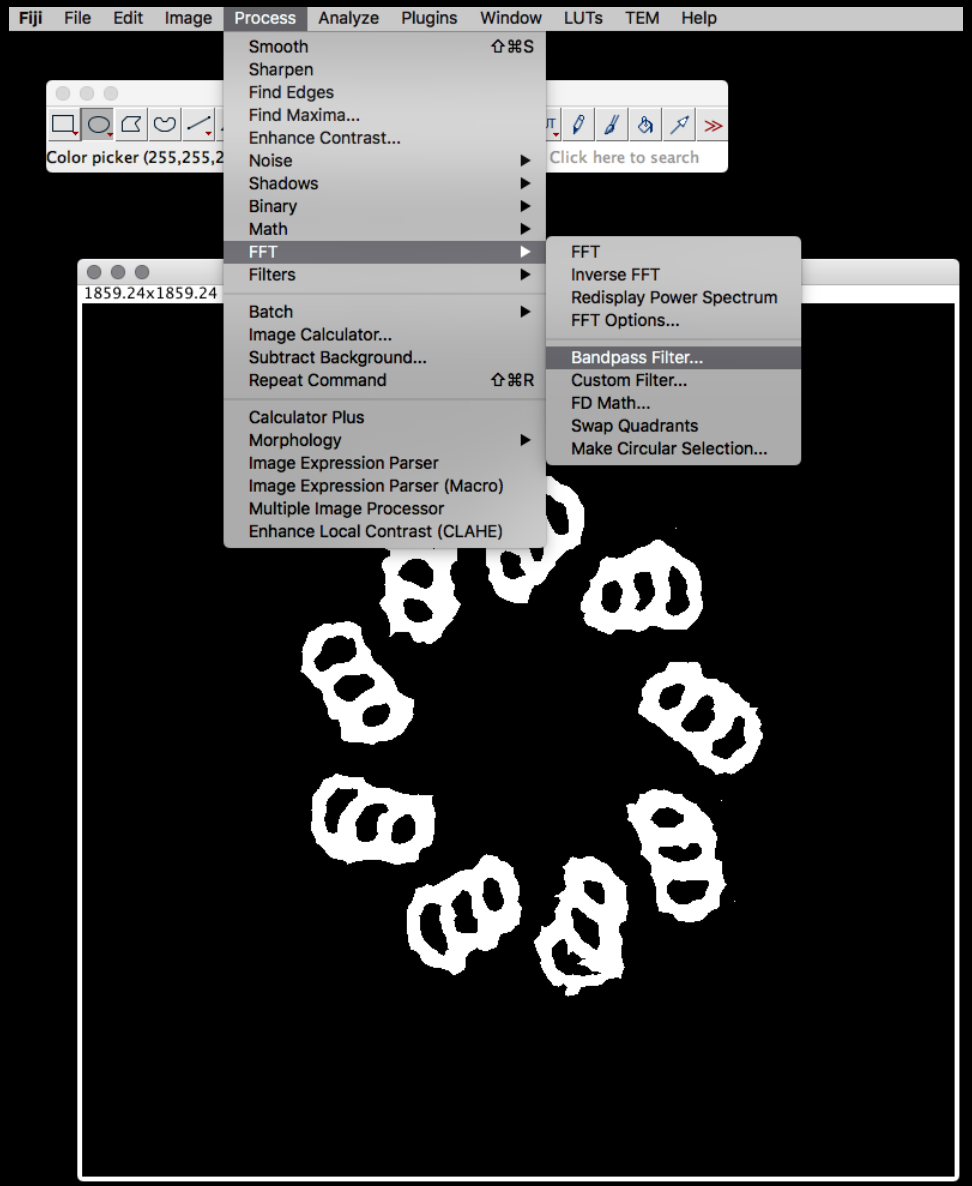
250nm = 98 pixels

Filtering at the resolution of ExM

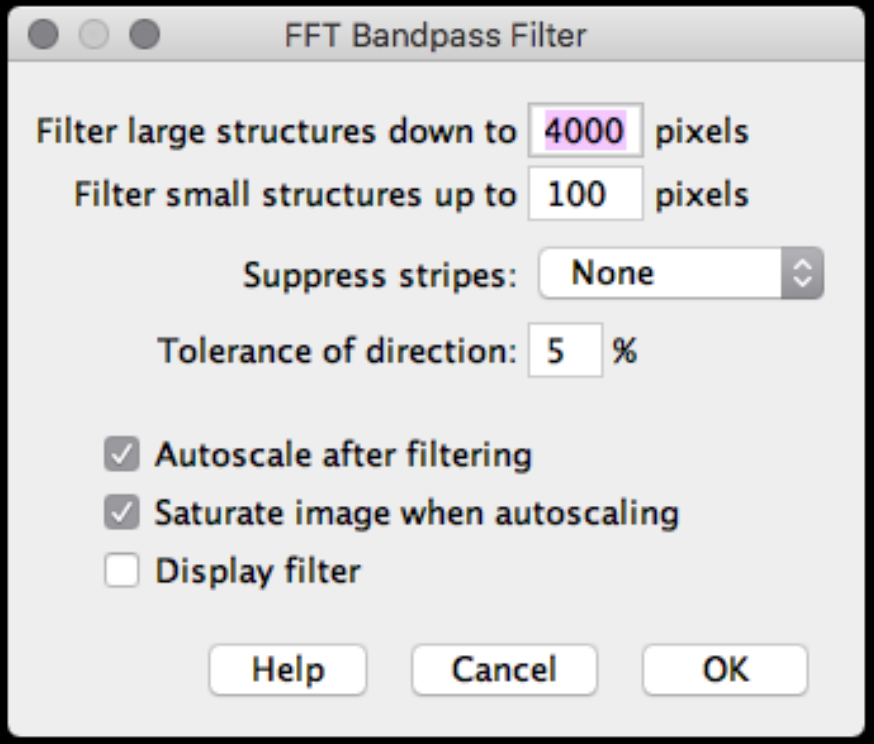


1nm = 0.3905 pixel

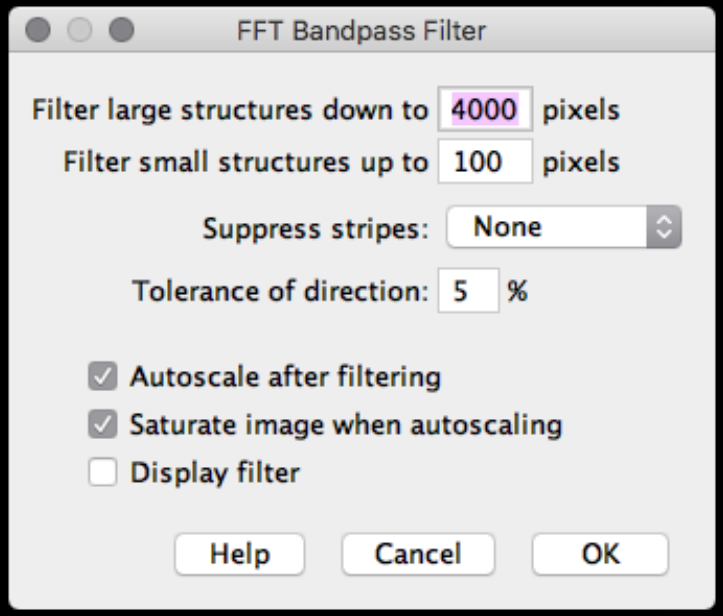
250nm = 98 pixels



FFT Bandpass filter



Filtering at the resolution of ExM

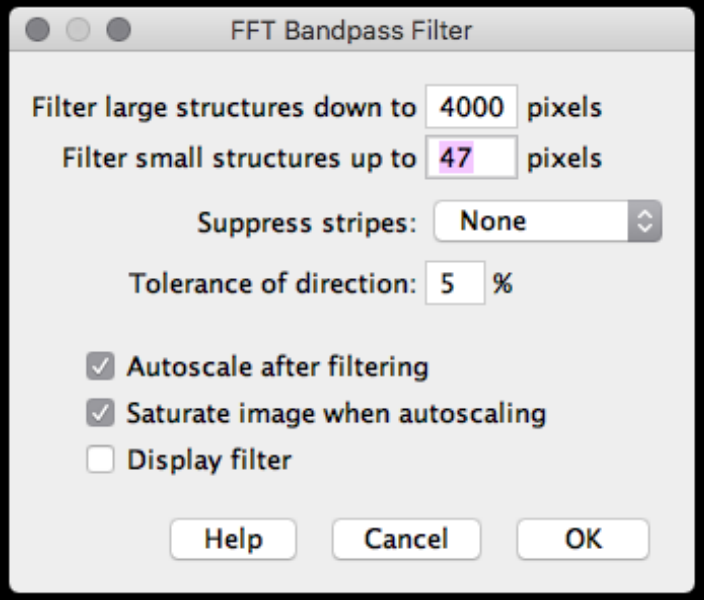


1nm = 0.3905 pixel

250nm = 98 pixels

REAL SIMULATION

Filtering at the resolution of ExM

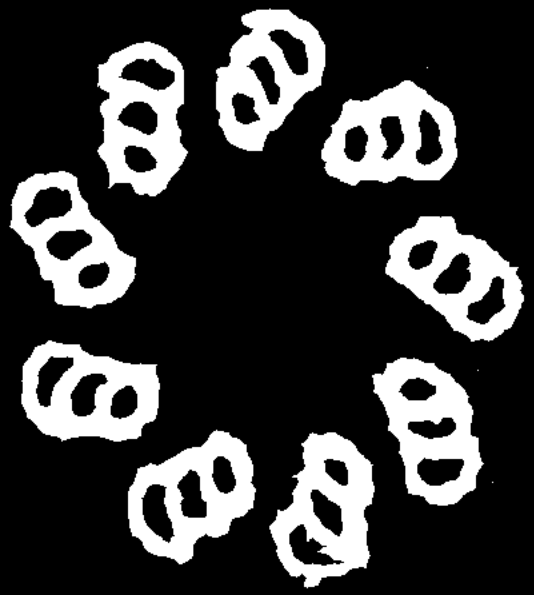


1nm = 0.3905 pixel

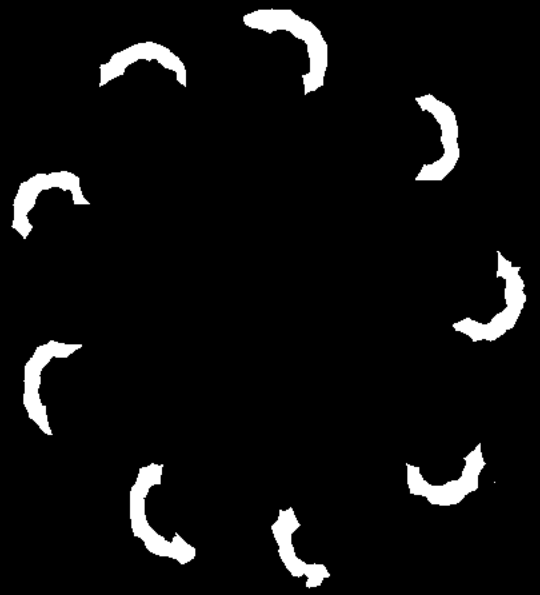
120nm = 47 pixels

DUAL COLOR SIMULATION

Full triplet

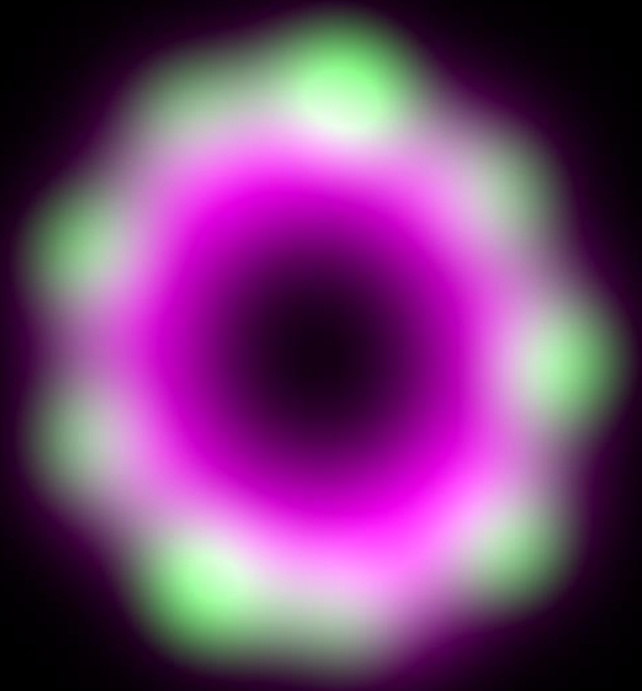


Only C-microtubules

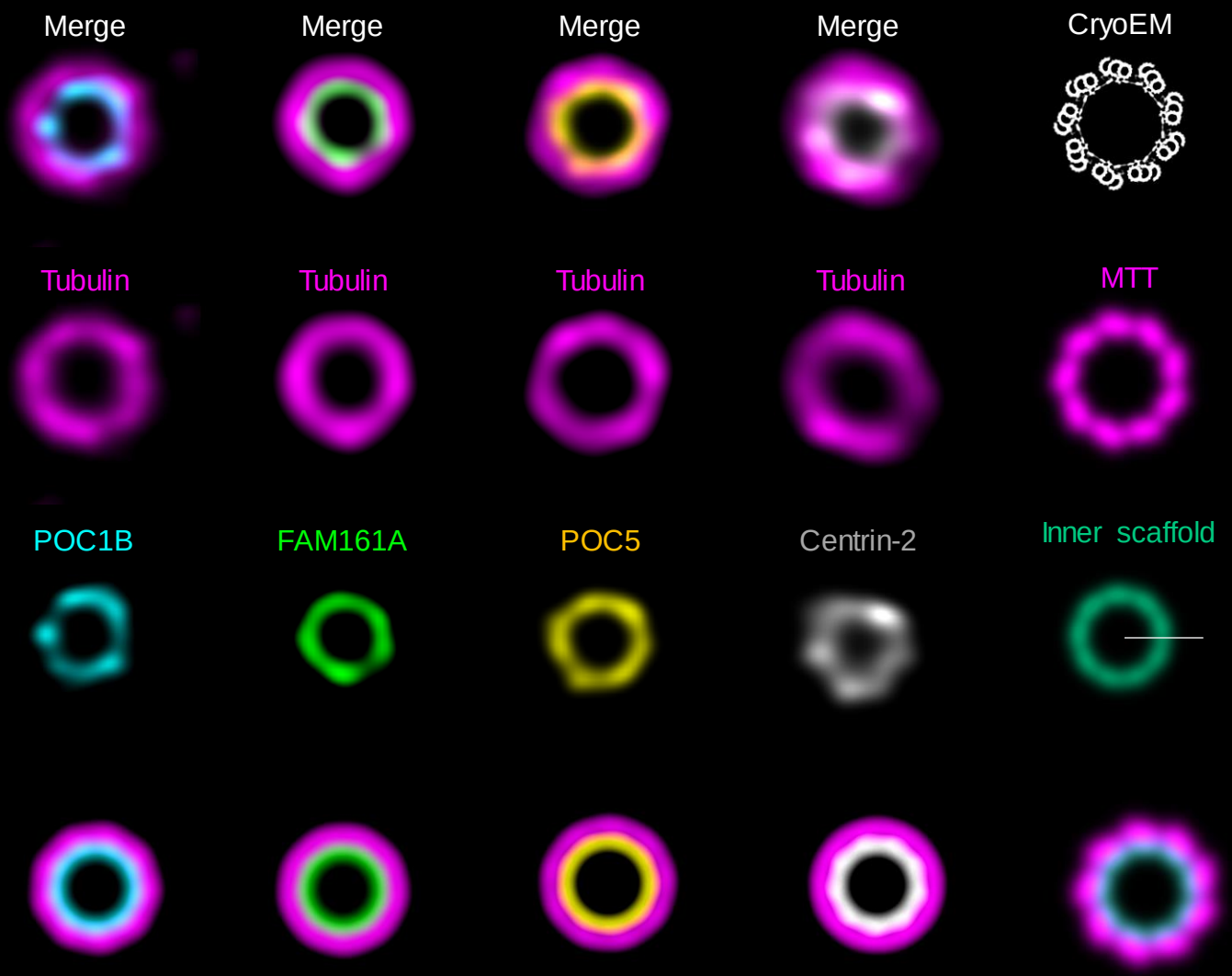


DUAL COLOR SIMULATION

DUAL COLOR SIMULATION - MERGE



Examples

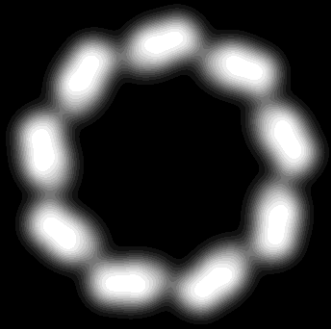

Merge
Merge
Merge
Merge
CryoEM
Tubulin
Tubulin
Tubulin
Tubulin
MTT
POC1B
FAM161A
POC5
Centrin-2
Inner scaffold

Experiments versus simulation

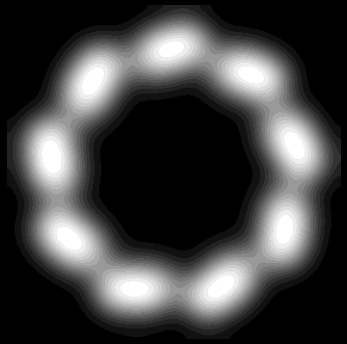
Centriole model

FFT Bandpass filter = 10nm

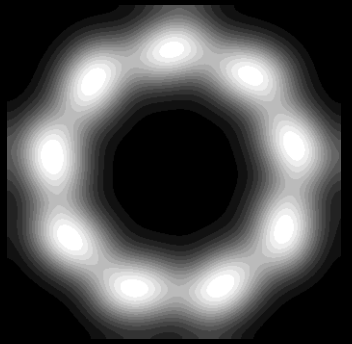
FFT Bandpass filter = 20nm



FFT Bandpass filter =30nm



FFT Bandpass filter = 40nm



UltraExM + DyMIN