



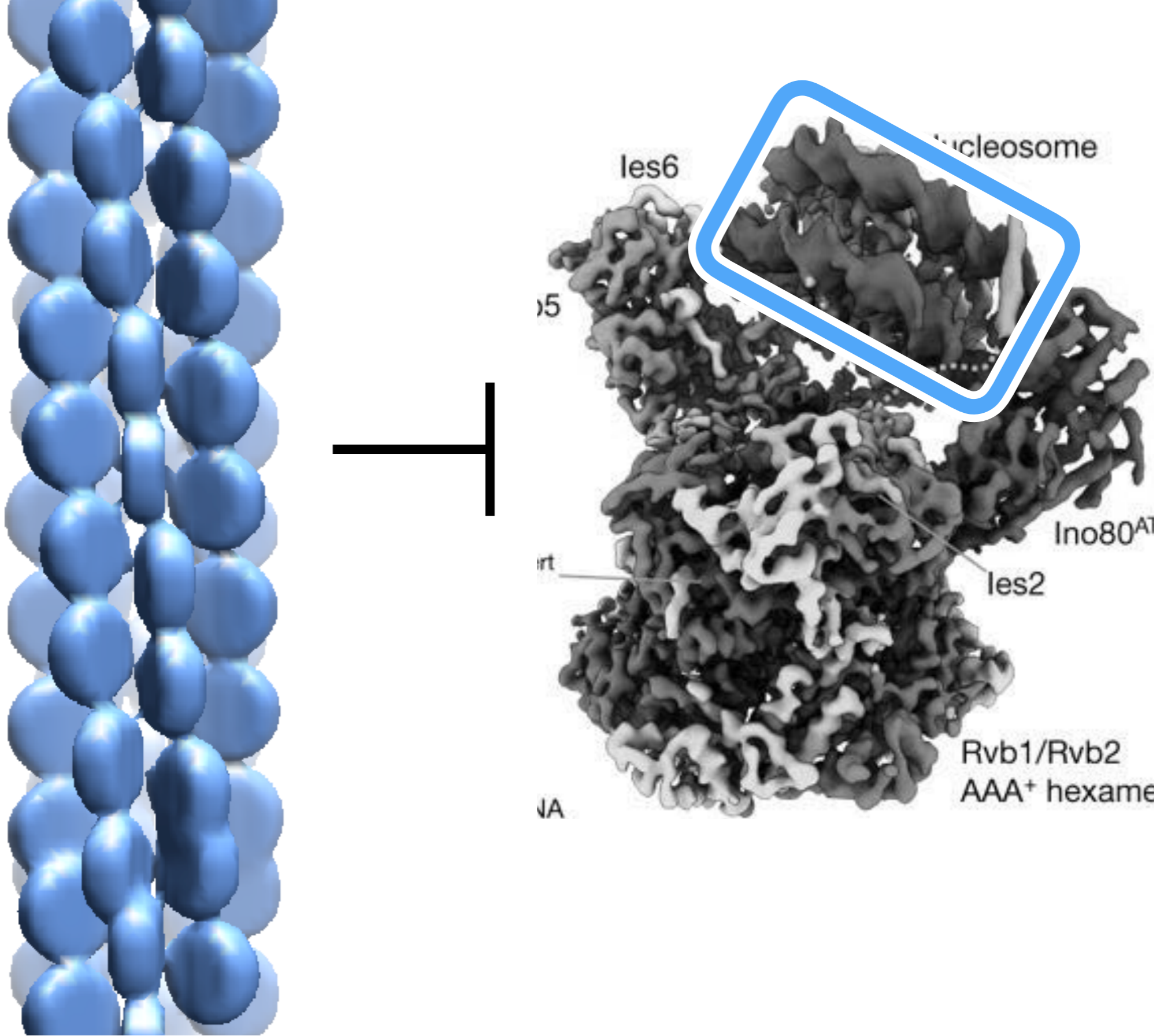
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Collaborators:



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Funding: NUS, LHK fund, CBIS, YIA, MOE



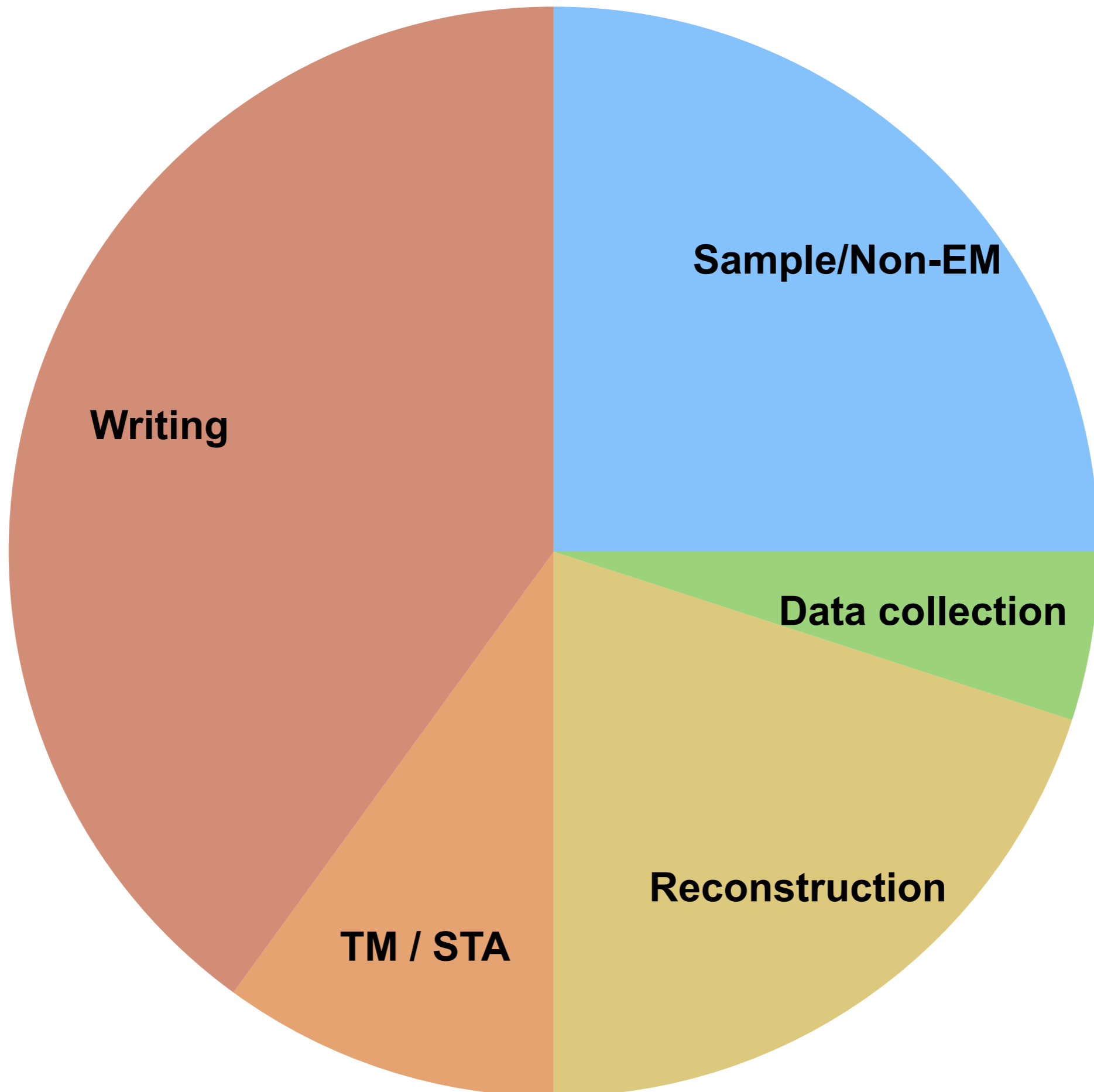
# A small structural-cell bio groups' perspective

- A pragmatic case for H-T tomo
- A scientific case for H-T tomo

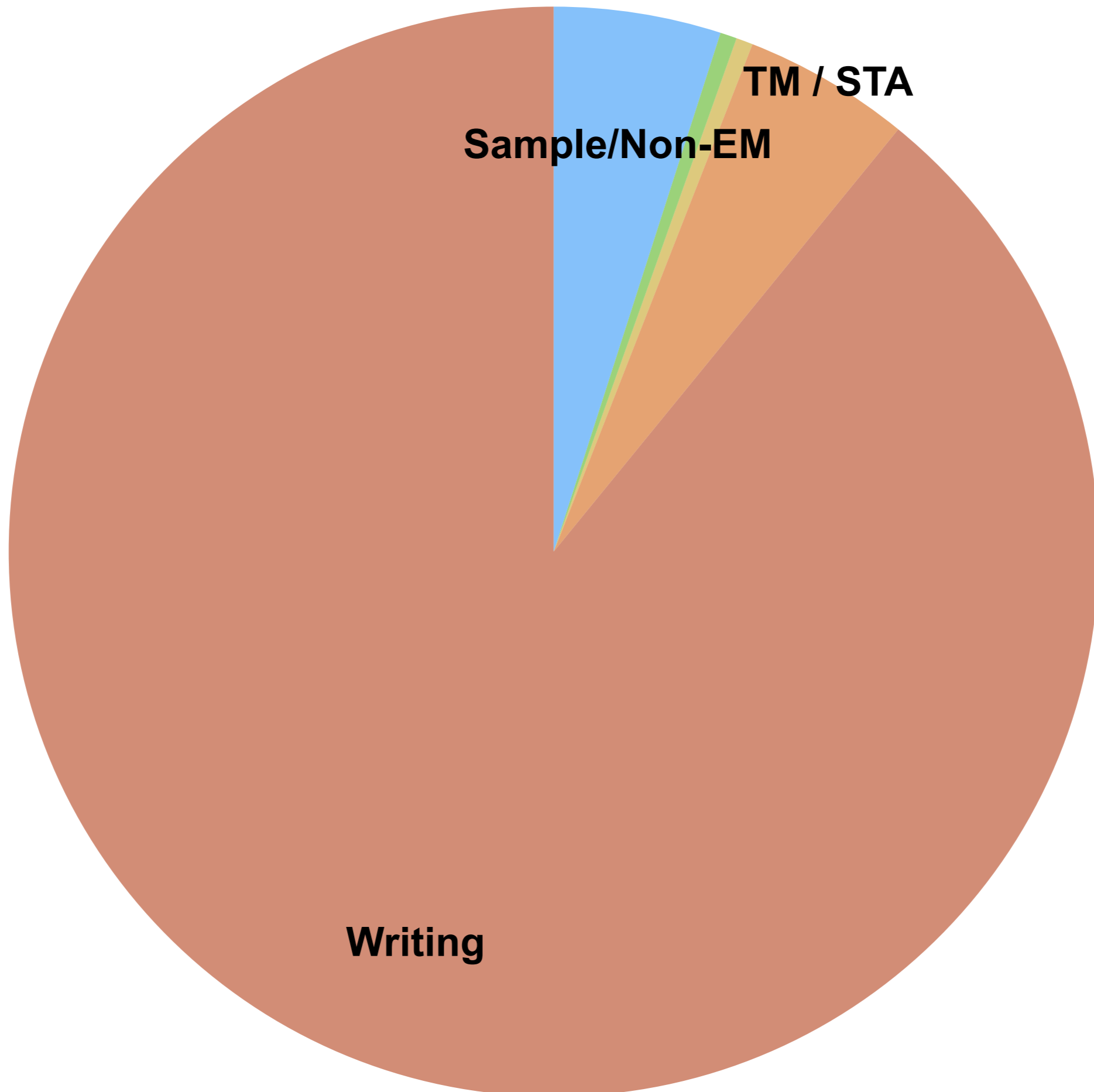
# Timeline (PhD student-driven lab)

- First 2 years for cryomicrotomy skills to start plateauing (inclusive of cryo-EM and image-processing training).
- Next 3+ years to do vast majority of experiments for thesis / paper. How is this time spent?

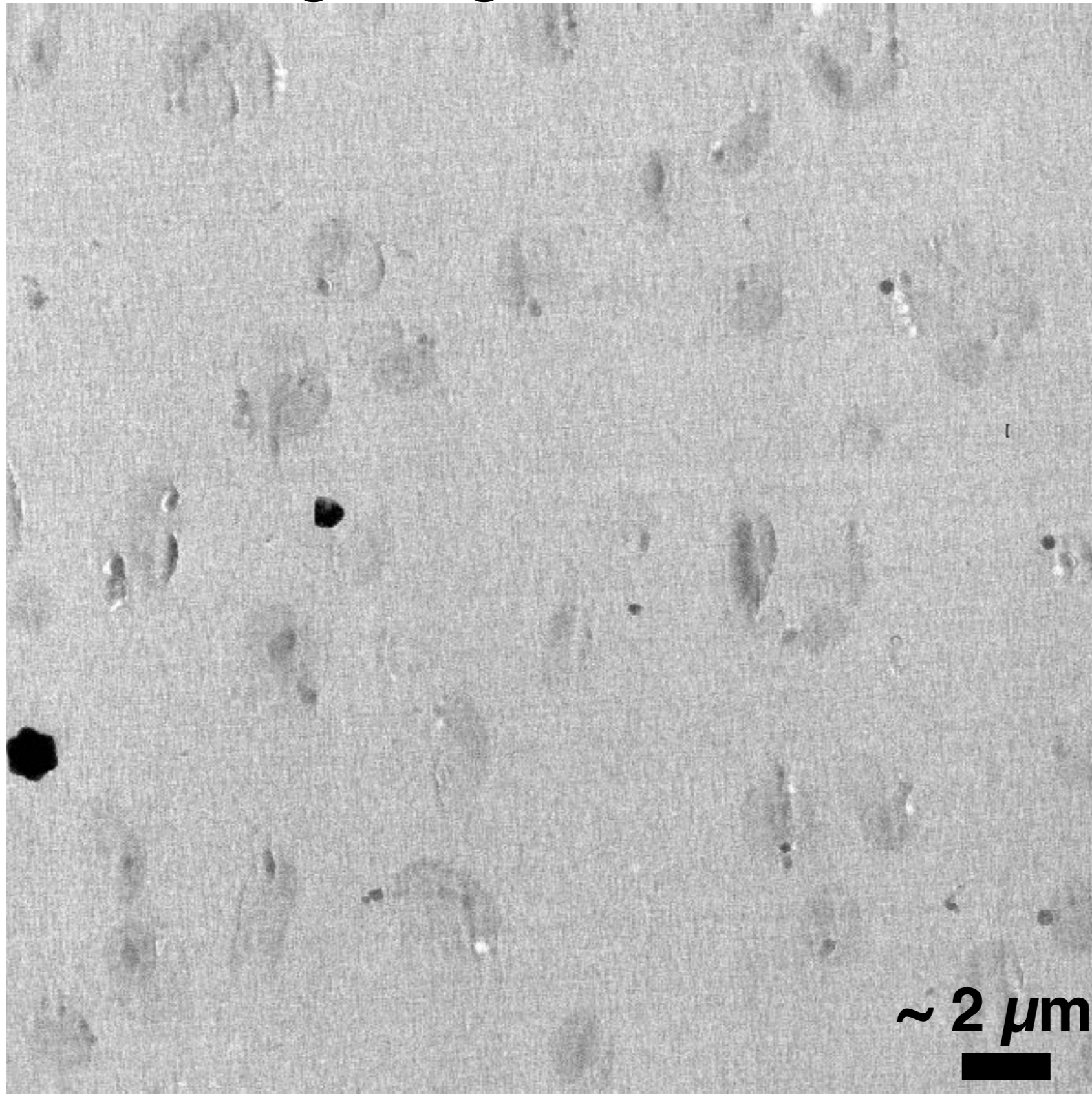
# Time commitment



# Ideal time commitment



# Targeting is not trivial



- A pragmatic case for H-T tomo
- **A scientific case for H-T tomo**



Shujun CAI



Désirée Böck  
(ETHZ)



Martin Pilhofer  
(ETHZ)

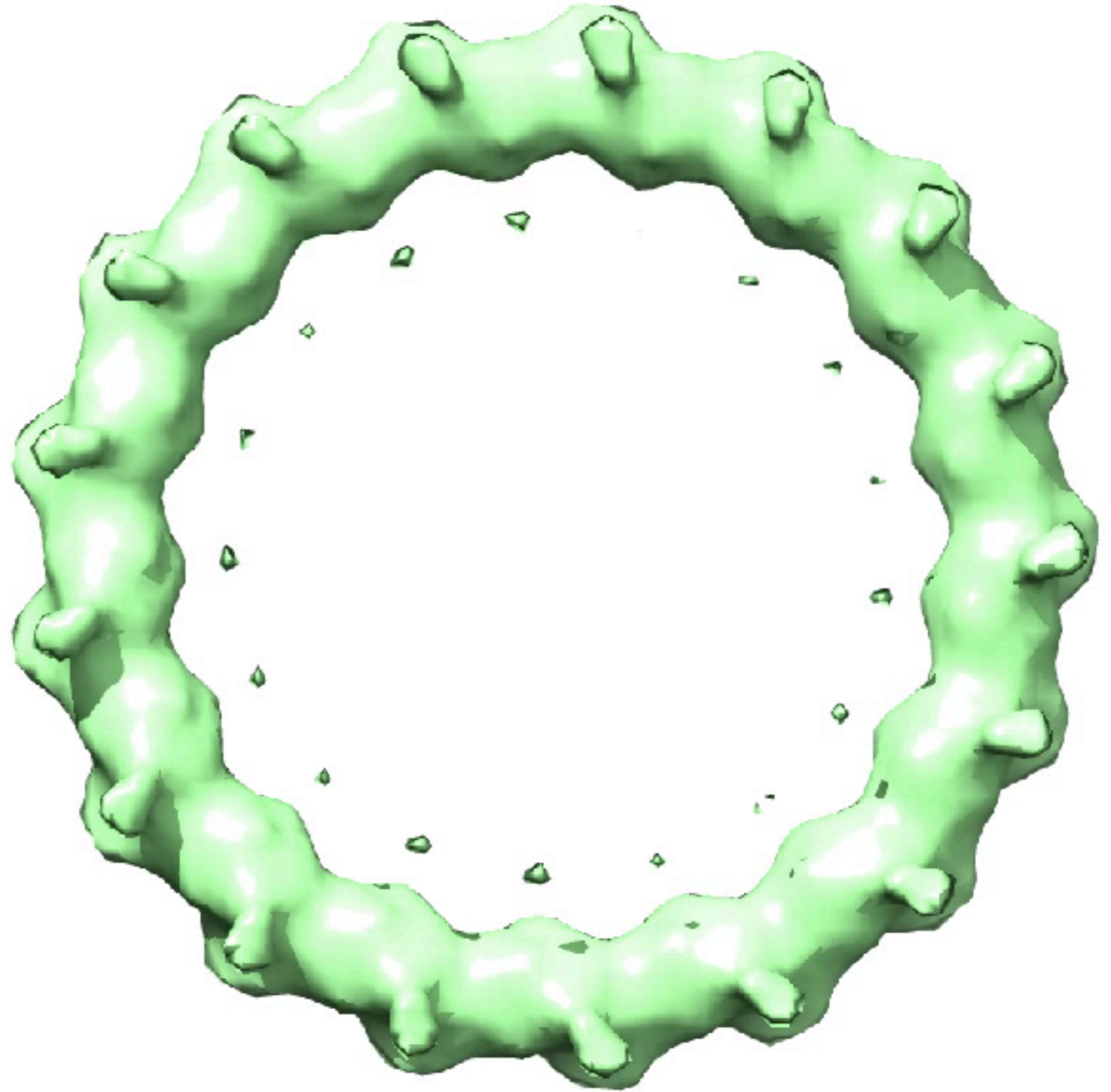
Nucleosomes:  $> 1,000$  per tomogram



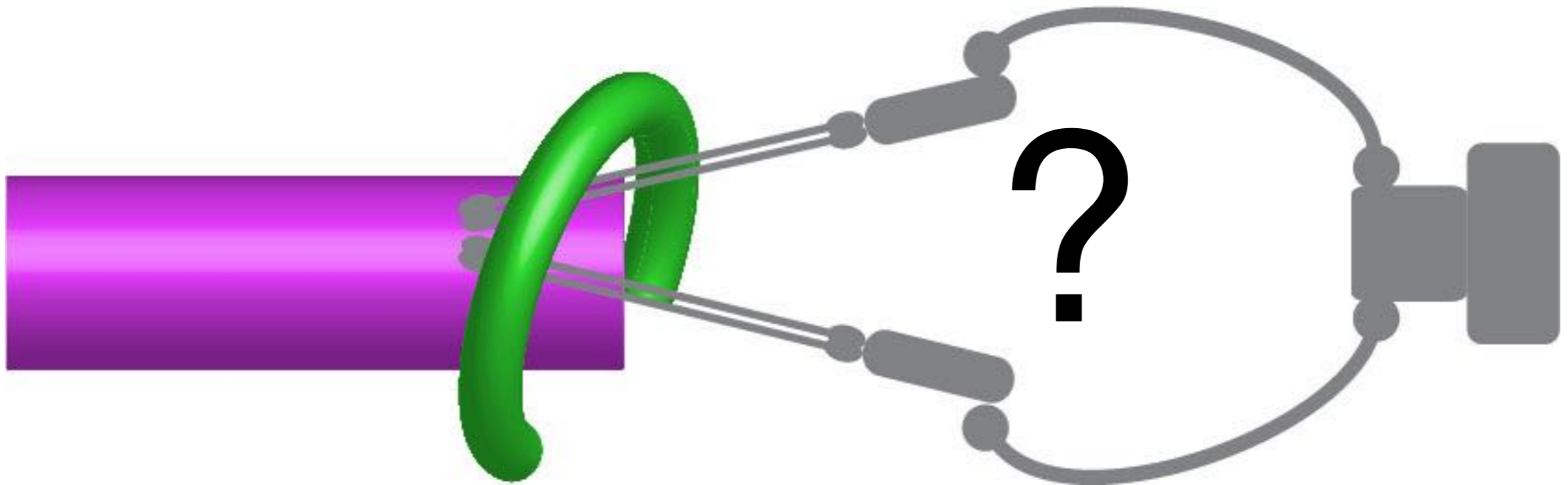
# Kinetochores (subcomplex): $< 10$ per tomogram



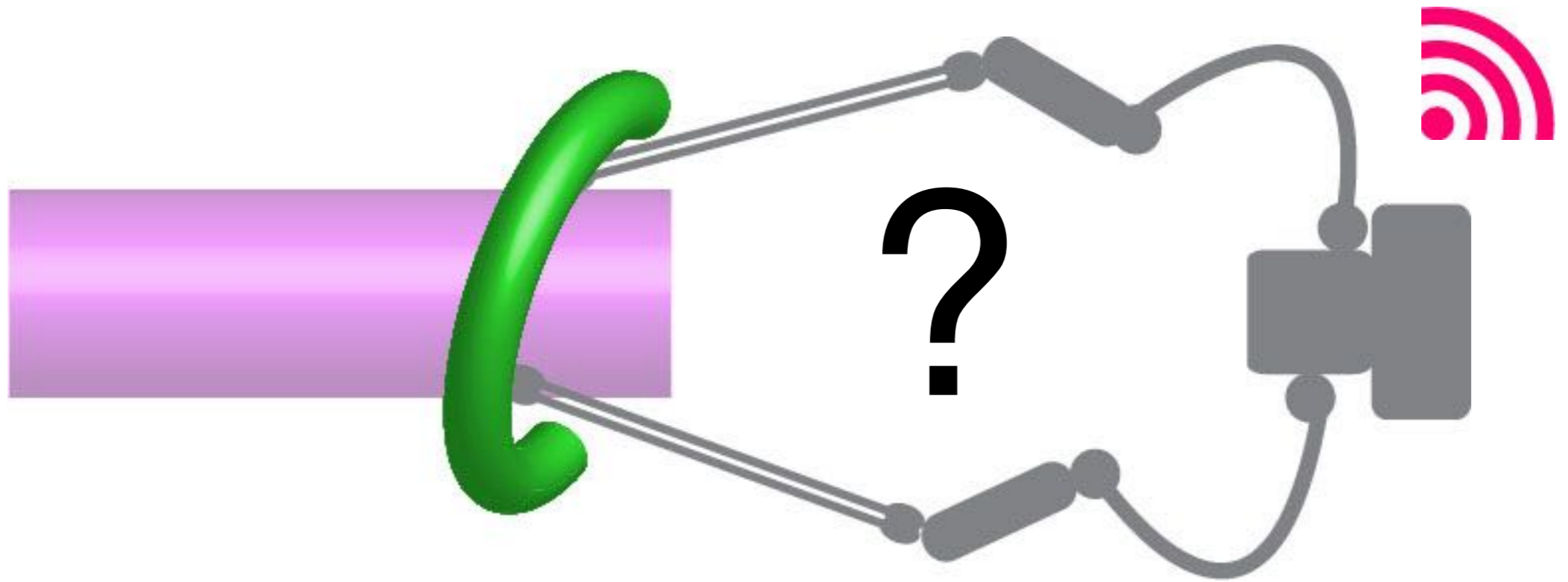
Cai Tong NG

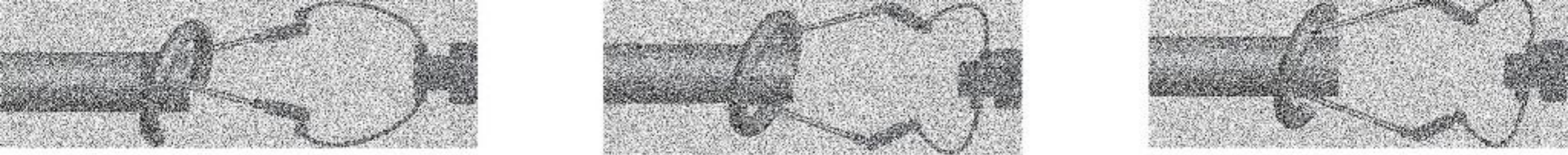


Lots of interesting biology is 'heterogeneous'



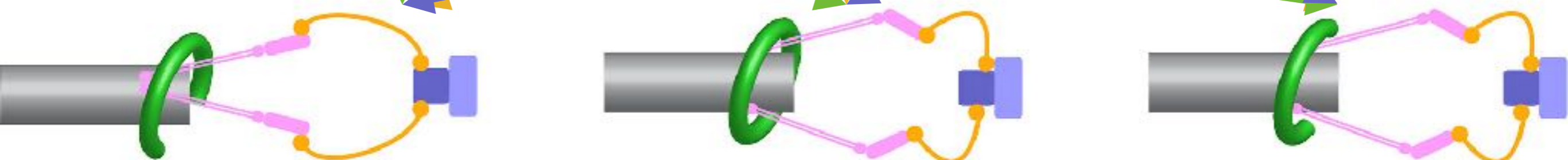
Lots of interesting biology is 'heterogeneous'





Align +  
Classify

Remap



New mechanisms & hypotheses

# Wishlist

- Auto cryo-CLEM & target selection
- High-precision/speed stage
- Fiducialless alignment\*
- Instant binning/low-pass filters

\* “one-click” and indistinguishable from “manual”  
reconstructions

New Results

## ETDB-Caltech: a blockchain-based distributed public database for electron tomography

Davi R Ortega, Catherine M. Oikonomou, H. Jane Ding, Prudence Rees-Lee, Alexandria, Grant J Jensen

doi: <https://doi.org/10.1101/453662>

This article is a preprint and has not been peer-reviewed [what does this mean?].

Abstract

Info/History

Metrics




 Preview PDF

### Abstract

Three-dimensional electron microscopy techniques like electron tomography provide valuable insights into cellular structures, and present significant challenges for data storage and dissemination. Here we explored a novel method to publicly release more than 11,000 such datasets, more than 30 TB in total, collected by our group. Our

# EMPIAR-10227

## A collection of yeast cell cryo-ET data

**Publication:** A collection of yeast cell cryo-ET data  
[Gan L](#) , [Cai S](#) , [Ng C](#) , Chen C

**Related EMDB entry:** [EMD-8157](#)

**Deposited:** 2018-10-03

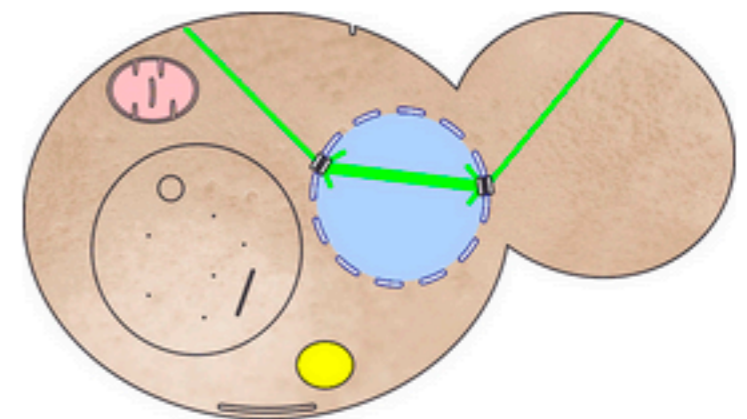
**Released:** 2018-11-16

**Last modified:** 2018-11-21

**Dataset size:** 1.8 TB

**Dataset DOI:** [10.6019/EMPIAR-10227](#)

### Contains:

 tilt series

### Image set

—A collection of yeast cell cryo-ET data

**Category:** tilt series (.mrc files) and tomograms (.rec files)

**Image format:** MRC

**No. of images or tilt series:** 1203



# Current practice

Typical cryosection ribbons:

- Gold beads spread uniformly on carbon
- 100 - 150 nm thick

One session 48 hours

- 3 hours to load grids & locate target positions

- 45 hours data collection

- $\pm 60^\circ$ ,  $2^\circ$  incr (60 min / tilt series)

- 40 - 45 tilt series / session

Reconstruction

- 20 - 40 minutes, w/ Etomo batch + manual adjustment

# Centre for Biolmaging Sciences (CBIS)

Vitrobot

Leica UC7/FC7 (> 90% of our samples)

T12 (screening & training)

CCD camera

Krios

Falcon 2 & Greg McMullan's hack

Volta phase plate

Leginon & Tomo 4

