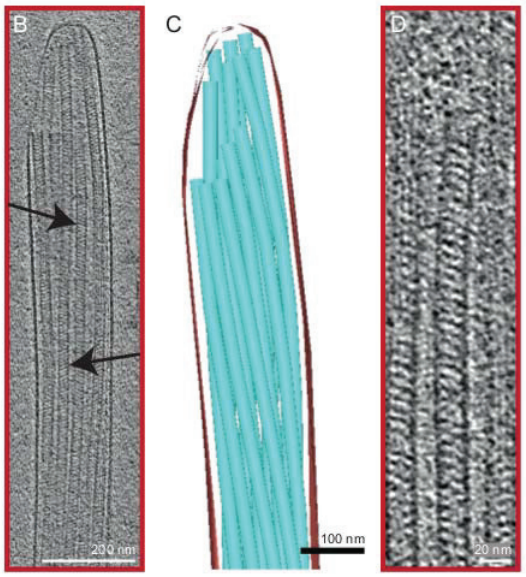
Verifying Results

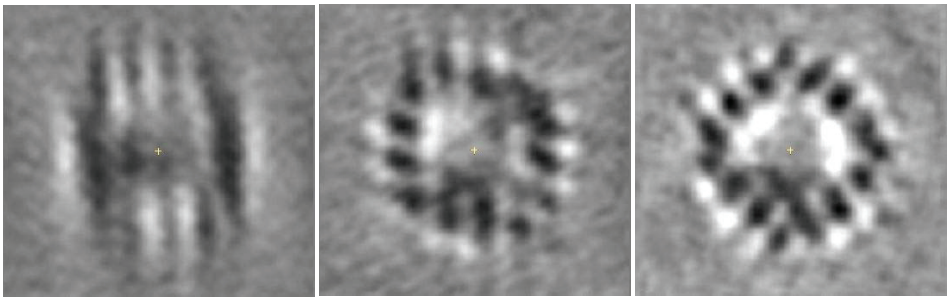
General Precautions

- Always double-check your settings
- Are results
  - Consistent with other information?
    - Includes raw tomogram(s)!
  - Highly sensitive to settings?
    - Initial reference
    - Mask size
    - Sensitivity is sometimes unavoidable
- Be especially skeptical of your own results!
- Are scores and alignments reasonable?
- Check for heterogeneity
- Estimates of resolution / frequency response

A Sensitive Example: Sperm TAILS

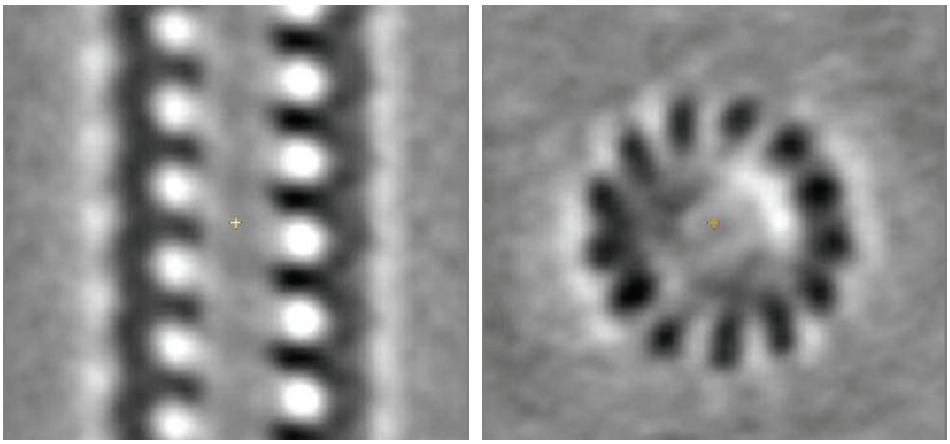


Reference / Mask Sensitivity Is Sometimes Unavoidable

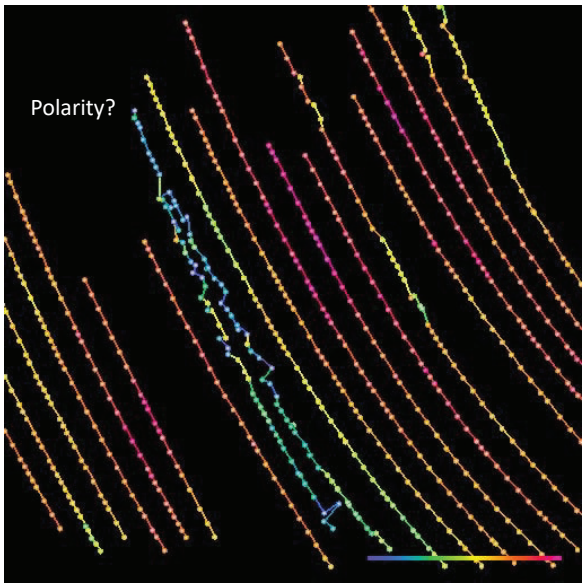


In this case, masking and choice of reference strongly affect the tradeoff between aligning real features and missing wedge artifacts!

TAILS Final Average



Checking Alignment and Scores



Score Visualization

- createAlignedModel assigns pseudo-colors based cross-correlation range within a volume
  - Virtual volumes treated as separate volumes
- To use the same mapping across multiple volumes, need to specify min and max values:

  ```
  echo >dummy.txt                [create an empty file]
  imodsetvalues –values dummy.txt –minMax map.txt
  <inputModel> <outputModel>
  rm dummy.txt
  ```

  where map.txt contain lines like:

  ```
  <object number> <minimum value> <maximum value>
  ```


Checks for Heterogeneity

- Averaging assumes a single class
- Colors assigned by createAlignedModel
- Histogram of cross-correlation scores
- Missing-wedge compensated principal components analysis (PCA) followed by k-means (or other) clustering
  - Separate and realign discrete subclasses if found
  - Will discuss in separate lecture / lab

Resolution and Frequency Response

- Spectral Signal-to-noise Ratio (SSNR)
- Fourier Shell Correlation (FSC)
  - Odd / even (generalizes to random splitting)
  - Gold standard
  - Comparison with Model or other solution
  - FSC will also be covered in it's own lecture / lab
- External Programs… *e.g.* ResMap

Questions?