Electron Tomography: Processing Pipeline

Contents

1. Choose dataset for reconstruction

Expected time: ~5-10 minutes

Before processing our data, we need to look at our available datasets and decide which is worth processing. The easiest way to do this is to open each dataset sequentially in imageJ/Fiji and checking to see that as far as possible:

- The images are not saturated (*very important!)*
  - You should auto adjust the brightness contrast, then hover the cursor over the centre of the particle and scroll through the full tilt series, checking that the intensity value doesn't cap out
- High angle projections are not obscured
- Images are all in focus
- There is minimal diffraction contrast

When you have chosen which dataset to reconstruct, export it as a .tif file.

2. Set up python environment

Expected time: ~30 minutes for initial setup

Note that for optimal performance you will need a computer with an Nvidia GPU that has CUDA installed.

The processing pipeline is all done in python, so we need to set this up with the correct packages installed. There are three options for doing this, ranging here from easiest to hardest and correspondingly least to most flexibility:

2.1 Use the electron microscopy workstation

The EM workstation Orodruin is already set up for tomography recosntructions

1. Reserve a session using the booking form
2. Access using Anydesk (Address: 406035504, Password: The1ringismine!)
3. Login as Sauron (Password: The1ringismine!)
4. Open the 'Anaconda prompt'

5. Type ‘conda activate cs_tomo’
6. Type ‘jupyter lab’
7. In the jupyter lab file explorer, navigate to Documents/Tomography demo/ and open tomo_demo.ipynb

2.2 Install environment by file

From your own computer, create a conda environment using the attached cs_tomo.yml file.

1. Open the ‘Anaconda prompt’
2. Type ‘conda env create -f cs_tomo_env.yml’
3. Type ‘conda activate cs_tomo’
4. Type ‘jupyter lab’ and open the attached tomo_demo.ipynb notebook
5. (Note that in some cases, even with a preconfigured .yml file you may run into compatibility issues with numpy – if this is the case, run ‘pip uninstall numpy’ several times, and then run ‘pip install numpy’ and restart the notebook kernel to fix the issue)

2.3 Install environment from scratch

From your own computer, create a conda environment, choosing your own environments.

1. If you’re choosing this I guess you are already comfortable with python environments! You will need to make sure you install astra and hyperspy along with matplotlib, jupyter lab, numpy, scipy etc.
2. You will also need to clone the code from ToveyTomoTools and either add to your path or import it locally
3. Open the attached tomo_demo.ipynb notebook

3. Pre-process the data

Expected time: ~30 minutes

This is described further in the example notebook, but essentially involves:

- Correctly importing data
- Registering the pixel shift between images
- Calibrating the tilt axis shift and tilt of the series

4. Reconstruction

Expected time: ~1-15 hrs

This is described further in the example notebook, but essentially involves:

- Choosing reconstruction parameters
- Leaving algorithm to run to convergence

5. Extracting information

Expected time: ~10 minutes to 10 weeks…

This depends very much on the desired outcome, but typical options may involve material segmentation, surface thresholding, orthoslice visualisation, geometrical calculations etc.

Some useful programs for this include Avizo (commercial product), Dragonfly (commercial, but free for academic research), and Paraview (free and open-source)

References

- Seminal papers in tomography [1–4]
- HAADF-STEM principal [5]
- General overviews of tomography [6,7]
- Compressed sensing reconstructions [8,9]

[1] D.J. De Rosier, A. Klug, Reconstruction of Three Dimensional Structures from Electron Micrographs, Nature. 217 (1968).

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