

# Basic Steps in single particle cryo-EM image processing and 3D reconstruction

## I. Pre-processing

- **image data acquisition (CCD/CMOS camera; digitization of micrographs / negatives)**
- **particle selection, « boxing »**
- **correction of the contrast transfer function**
- **band-pass filtering and normalisation of particle images**

## II. Structure determination

- **particle centering / alignments**
- **MSA (multivariate statistical analysis) + classification**
- **angle assignment**
  - **common lines / angular reconstitution**
  - **projection matching (correlation with reference); ML-based assignment**
- **3D reconstruction (first map)**
- **structure refinement**
- **resolution assessment**
- **map interpretation; fitting of known structures, atomic model building/refinement**