

Some advice for labs that are new to single particle cryo-EM structure determination on how to get started:

- 1) Learn about the theory and practical application of transmission electron microscopy, cryo-preservation and single particle reconstruction from: books, methods papers, online lectures (e.g. Grant Jensen's), online tutorials (e.g. for Relion by Sjors Scheres on YouTube), and by attending cryo-EM conferences (links to many of these resources are provided below).
- 2) If your student/postdoc doesn't have prior cryo-EM experience, it is helpful to send him/her to a hands-on workshop (not a "symposium" that only features talks). Some workshops are specific for only one component of the single particle workflow (e.g. image processing), others include all major components, such as hands-on training for sample preparation and low-dose imaging on the TEM. Usually each year, several workshops are being offered (e.g. see below).
- 3) Using UTSW "in-house" resources:
  - a) as soon as the single particle program of the Structural Biology Lab (SBL) is fully operational, labs are encouraged to utilize the expertise of the SBL. At a later time (starting 2018?) the SBL (with help from the CEMF and EM experts) will provide in-house courses/workshops related to cryo-EM.
  - b) attend the monthly Single Particle Discussion Group (aka "data clinic"): first Thursday of each month at 4-5pm in NB10.606;
- 4) Use "positive controls" to learn how "good data" look like, to put your procedures/skills to the test and troubleshoot possible problems:
  - a) for image processing: work through published data sets; many structures deposited on the EMDB database come with original micrographs – test if you can reconstruct the 3D structure with a similar resolution.
  - b) for the entire workflow: get a well-studied protein/complex (with published high resolution structures) as "standard" and process it from cryo-preparation to 3D reconstruction (e.g. GroEL can be ordered, the SBL might store some "standard proteins" in the future, or ask local labs specialized on protein like ribosomes).

### **General Online-Resources (no time-limit):**

- Relion 2.0 tutorial by the developer Sjors Scheres: 5 hour video on YouTube <https://www.youtube.com/watch?v=HLQTEYiIOaA> and written manual to read along: [ftp://ftp.mrc-lmb.cam.ac.uk/pub/scheres/relion20\\_tutorial.pdf](ftp://ftp.mrc-lmb.cam.ac.uk/pub/scheres/relion20_tutorial.pdf)
- Grant Jensen "Getting started in cryo-EM" <http://cryo-em-course.caltech.edu/>  
Plus in the fall a new set of practical videos that teach the workflow of single particle analysis from sample prep through microscope alignment and data collection. We will cover both the Krios and Arctica systems - more specifically, we are working on videos covering (i) introduction to the Krios and Arctica instruments and user interface, (ii) daily microscope check, (iii) plunge-freezing using a Vitrobot, (iv) sample insertion and screening, (v) choosing data collection parameters and (vi) actual data collection using EPU with both Falcon and K2 direct electron detectors. Finally we will also be recording a common troubleshooting module. Like my theory videos, these practical videos will assume that the viewers are completely new to cryo-EM. We are on course to start posting finished videos this fall, followed by formal training exercises and tests that students can do to develop and then demonstrate proficiency, step by step. This course will also form the basis for practical courses to come that will require completion of this online course as a prerequisite. In 2018 we plan to add similar instruction for tomography, plus a facility manager and "train-the-trainer" series that will cover full microscope alignment, performance checks, routine maintenance, advanced troubleshooting, phase plates and possibly even cryo-FIB. We hope cryo-EM students both new and old will find these videos helpful.

- Workshop on Computational Methods for CryoEM: <https://www.youtube.com/watch?v=XdKO1iaPP8o>
- Online talks: “Advanced Data Collection for High Resolution cryoEM: How to make the most of your National Facility Visit” [http://www.ccpem.ac.uk/training/biochemsoc\\_sep2016.php](http://www.ccpem.ac.uk/training/biochemsoc_sep2016.php)
- Classical book: Frank, Joachim (2006). Three-Dimensional Electron Microscopy of Macromolecular Assemblies. New York: Oxford University Press. ISBN 0-19-518218-9.
- LMB cryo-EM course: <ftp://ftp.mrc-lmb.cam.ac.uk/pub/scheres/EM-course>
- Test Datasets can be found in the EMDB database: <http://www.ebi.ac.uk/pdbe/emdb/empiar/> (one can download the dataset of a published structure and see if one can get a similar result as the one in the paper)
- YouTube videos about SerialEM: <https://www.youtube.com/playlist?list=PLGggUwWmzvs-DV4jCapS15XQ-hpAXtzMb>
- Comprehensive list of software tools: [https://en.wikibooks.org/wiki/Software\\_Tools\\_For\\_Molecular\\_Microscopy](https://en.wikibooks.org/wiki/Software_Tools_For_Molecular_Microscopy)
- 3DEM mailing list for workshop (and more) announcements: <http://3dem.ucsd.edu/maillinglist.shtm>
- Diamond Light Source is the UK’s national synchrotron science facility, events: <http://www.diamond.ac.uk/Home/Events>
- List of cryo-EM events: [http://www.emdatabank.org/3dem\\_events.html](http://www.emdatabank.org/3dem_events.html)

### **Workshops/Conferences (chronological):**

- Technical aspects in single particle analysis, one day symposium on **April 25th 2017**, Max Planck Institute for Biophysical Chemistry, Goettingen <http://www.mpibpc.mpg.de/events/9540/2169>
- NRAMM/SEMC Workshop on Addressing Challenges in CryoEM Grid Preparation, **April 26, 2017** – also life-stream and YouTube: [https://www.youtube.com/channel/UCgd1dW4HQ\\_yvpRjFc670htw](https://www.youtube.com/channel/UCgd1dW4HQ_yvpRjFc670htw)
- first SPHIRE workshop, to be held on 8-10 May, 2017 at the Max-Planck Institute of Molecular Physiology (Dortmund, Germany) <http://sphire.mpg.de> (deadline passed)
- The Swedish National Cryo-EM Facility Inaugural Symposium, Stockholm/Umea, **May 8-9th 2017** <https://www.scilifelab.se/events/cryo-em/>
- Symposium on Understanding Biology through Structure, **May 13-17, 2017**, Santa Fe [https://conferences.newmexicoconsortium.org/conferences/ubts\\_17](https://conferences.newmexicoconsortium.org/conferences/ubts_17)
- TEM Sussex Symposium invitation: From micromachines to cellular networks, Brighton UK, **15-16th May 2017**. <http://www.sussex.ac.uk/lifesci/internal/servicesandsupport/facilitiesandresources/electronmicroscopy/temsymposium>
- Annual Meeting of the American Crystallographic Association (ACA), to be held **May 26-30** in New Orleans, LA, <http://www.amerystalassn.org/2017-scientific-program> Special Session: Going Beyond PX with Cryo Electron Microscopy, Tomography, and Diffraction
- 3DEM Gordon Research Conference, Les Dablerets, Switzerland, **June 11-16 2017** <https://www.grc.org/programs.aspx?id=10866>

- Advanced Electron Microscopy for Cell Biology EMBL Heidelberg, Germany 13 - 23 Jun 2017 <https://www.embl.de/training/events/2017/EMM17-01/index.html> (registration closed)
- Cryo-EM symposium, Grenoble **6-7 July 2017** <http://www.esrf.eu/cryo-em.fr>
- M&M **August 6-10 2017** [http://www.microscopy.org/MandM/2017/program/short\\_courses.cfm](http://www.microscopy.org/MandM/2017/program/short_courses.cfm)
- 3rd International Workshop of Advanced Image Processing of Cryo-Electron Microscopy (IWAIP-CryoEM 2017) will be held in Beijing from **August 6-10, 2017**. <http://cbi.ibp.ac.cn/workshop/workshop2017/IWAIP2017/>
- AIC International School 2017, "Bridging the gap between cryo-EM and crystallography", Pavia, Italy, from **3-6 September 2017**. <http://www.cristallografia.org/aicschool2017/eng/detail.asp?idn=2949>
- Practical course: Image processing for cryo-electron microscopy , London **05–15 September 2017** <http://meetings.embo.org/event/17-cryo-em>
- Cryo-EM conference ‘Overcoming Barriers – Atomic-resolution and beyond: advances in molecular electron microscopy’, which will take place from **12-15th September 2017** at the Max-Planck affiliated research center caesar in Bonn (Germany). <https://www.caesar.de/en/events/2017/6th-international-caesar-conference-overcoming-barriers.html>
- Janelia Farm <https://conference.janelia.org>  
Challenges in Structural Biology: **15-18 October, 2017**
- NRAMM workshop on Advanced Topics in EM Structure Determination: Challenges and Opportunities, **29 Oct - 3 Nov 2017**, CUNY Advanced Science Research Center. <http://nramm.nysbc.org/4192-2/>
- <https://www.embl.de/training/events/2017/LEM17-01/index.html> CLEM