Cryo-EM sample preparation and data collection

National course. The purpose of the course is to prepare and train our users in sample preparation methods, introduce users to the image data acquisition workflow, expand knowledge about cryo-EM methods among Swedish researchers and finally show that everyone can learn how to use cryo-EM.

The course is open for facility users or potential facility users, such as PhD students, postdocs, senior researchers and other employees within the life sciences at any Swedish university who are curious about structure biology and will profit from cryo-EM skills. To attend, the course participants must have experience in basic electron microscopy and be familiar with structure or cell biology.

This course is supported by SciLifeLab and run by the <u>SciLifeLab Cryo-EM facility</u>, both UmU and SU nodes together.

Important dates

Umeå, KBC : 2019-06-03 to 2019-06-05

Responsible teachers

Linda Sandblad and Marta Carroni

Teachers and course instructors

- Thomas Boesen
- Sonja Welsch
- Camilla Holmlund
- Jose Miguel de la Rosa Trevin
- Lars-Anders Carlson
- Linda Sandblad
- Marta Carroni
- Michael Hall

Course content

This course will cover practical aspects of cryo-EM sample preparation and data acquisition. Topics covered will include:

- Basic theory of vitreous water, how to handle a cryo sample and pitfalls
- Plunge freezing methods, tools and cryo-transfer
- Sample and instrument requirements for single particle methods and tomography
- Image data acquisition, concept of low dose and the cryo-specific workflow.
- Introduction to automatic data acquisition software, EPU, Tomography and SerialEM
- Introduction to cryo-EM image processing methods

The course consists of lectures, cryo lab work, demonstrations and practical exercises for all participants, handson experiences of microscopy operation and discussions.

Lecture schedule

Day 1: Monday 3th June, morning, 3 lectures (9:00-12:00 Stora Focussalen KBC KBF301)

- 9:00 Introducing the course, presentation of teachers and participants [all teachers]
- 9:30 Basic function of a TEM, and special requirements for cryo-EM [Camilla Holmlund]
- 10:30 Vitrification and how to work with cryo-EM samples [Linda Sandblad]
- 11:15 Increasing Automation and Throughput in cryoEM: news from Thermo Fisher Scientific [Sonja Welsch]
- 11:45 Brief introduction into each practical [Linda, Marta and Jose Miguel]

Day 2: Tuesday 4th June, morning 3 lectures (9:00-12:00 Stora Focussalen KBC KBF301)

- 9.00 Get ready for cryo-EM, biochemical methods and negative staining, and repetition for day 1 [Michael Hall]
- 9:45 Direct electron detectors, motion correction and CTF [Lars-Anders Carlson]
- 10:45 Practicalities of data acquisition, Single particle data collection [Marta Carroni]

Day 3: Wednesday 5th June, morning 3 lectures (9:00-12:00 Stora Focussalen KBC KBF301)

- 9:00 Starting single particle processing, pre-processing and introduction to 3D reconstruction methods, single particle methodology [Jose Miguel de la Rosa Trevin]
- 9:45 Tomography, sub-tomogram averaging and inspiration by cryo-EM projects [Marta Carroni]
- 10:45 >>> To be announced << [Thomas Boesen]