

C-CINA.org

How do you choose the optimal microscope/camera combinations?

What do you need? How do you validate your instrument performance? How many people does each microscope serve? How do you schedule time to optimize the instrument usage and performance?

> Henning Stahlberg Center for Cellular Imaging and NanoAnalytics (C-CINA) Biozentrum, University of Basel, Switzerland



Universität Basel The Center for Molecular Life Sciences



Negotiate your new position

- What type of instruments do you need?
 - What is your research focus?
 - Which instruments do you need on site for your research plans?
 - Who else will want to use your EM facility?
- Negotiate essentials with your host institution (startup package?):
 - Equipment (funds)
 - Is there a plan for covering the maintenance costs? How will you obtain new instruments in the future?
 - Is there a way to cover (semi-) permanent support persons?
 - Are there nice startup funds to cover the lab costs, until own funding is secured?
 - You will need the long-term commitment of your host institution, which should see your lab as a common institute effort.



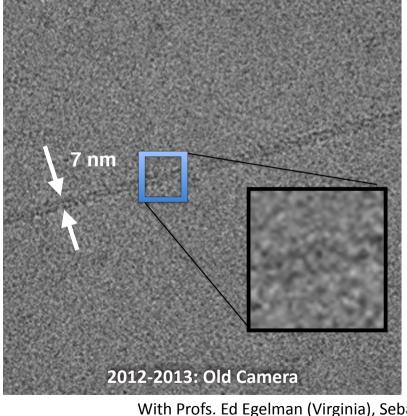
Choose your instruments:

- High-end flagship TEM -> High resolution requirements? Thin or thick samples? (e.g., 300kV FEG cryo-EM: Titan Krios (with GIF?) or JEM3200 (has energy filter))
- Preparatory TEM -> Grid optimization in cryo-EM, initial structural work (e.g., Talos or F20 or JEM2100, cryo sample holders (e.g. Gatan 626))
- Sample screening TEM -> Negative stain analysis of samples (e.g., T12 or JEM1400)
- Sample Prep -> automated grid plunger, HP-freezer, freeze substitution device, (cryo-) ultra microtomes, plasma cleaner, FIB-SEM, carbon evaporator, glow discharger.
- Tissue analysis -> 3View-SBF-SEM or FIB-SEM, CLEM setup?
- Choose your high-end cameras:
 - Gatan K2 Summit / FEI Falcon II or III / Direct Electron (DE-20, DE-64) / TVIPS CMOS (F416, F816)
 - What data collection time can you afford or do you want? (shorter=faster, longer=better coherence) How many dose-fractionation movie frames do you need? What software integration do you plan? What budget do you want to invest into the camera?



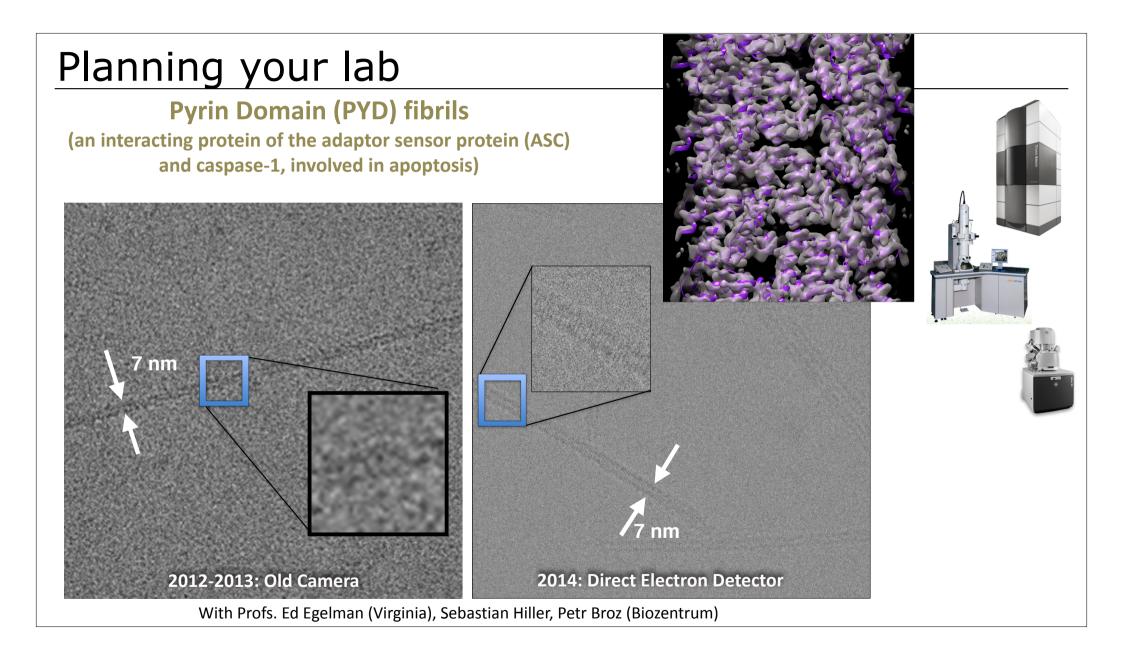
Pyrin Domain (PYD) fibrils

(an interacting protein of the adaptor sensor protein (ASC) and caspase-1, involved in apoptosis)





With Profs. Ed Egelman (Virginia), Sebastian Hiller, Petr Broz (Biozentrum)

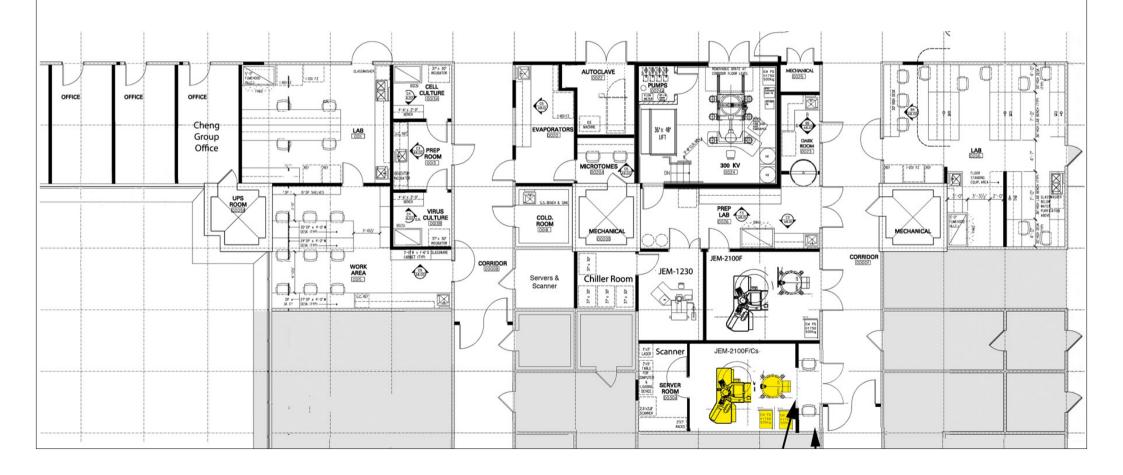


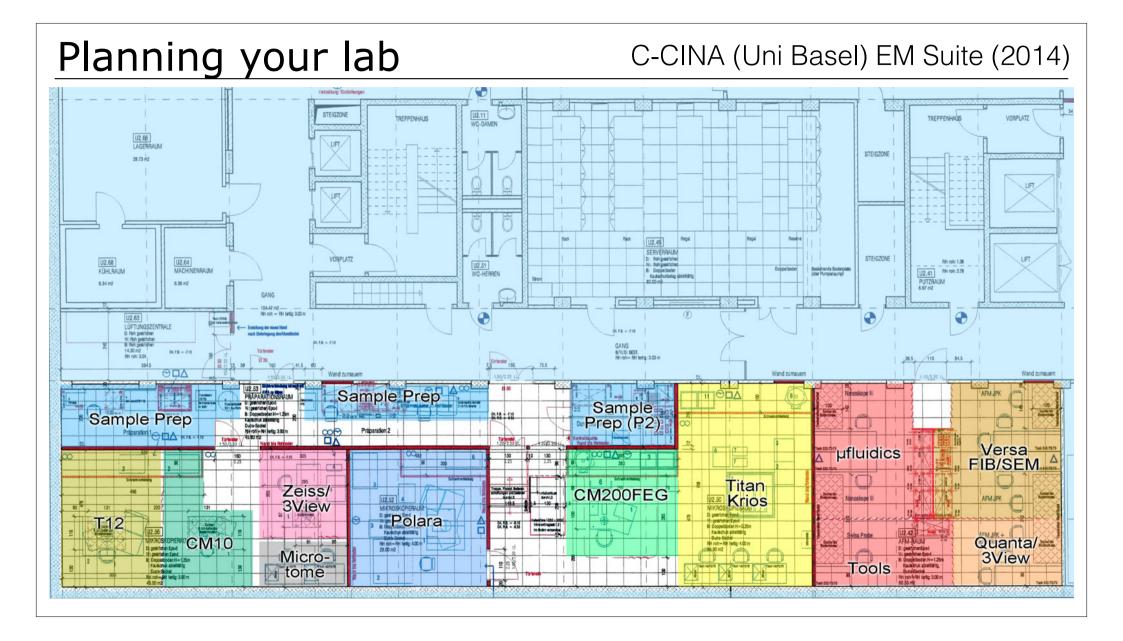
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- Visit other labs that have microscopes of interest.
- Speak with EM vendors about different options. Remember: They want you to be successful, they can help you. Work with them in order to commonly achieve your common goals.



UC Davis EM Suite (2008)

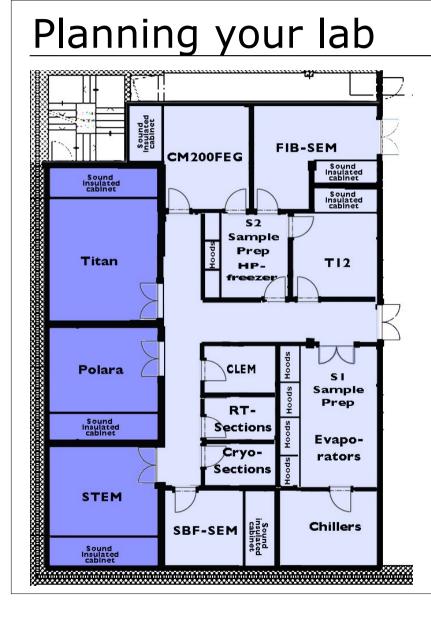
 Get drawings of other lab layouts, compare many different setups. Visit different labs





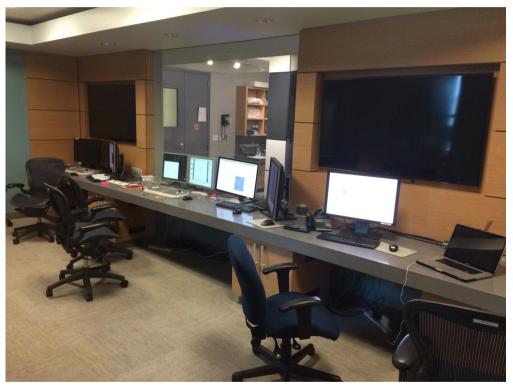
C-CINA (Uni Basel) EM Suite (2014)



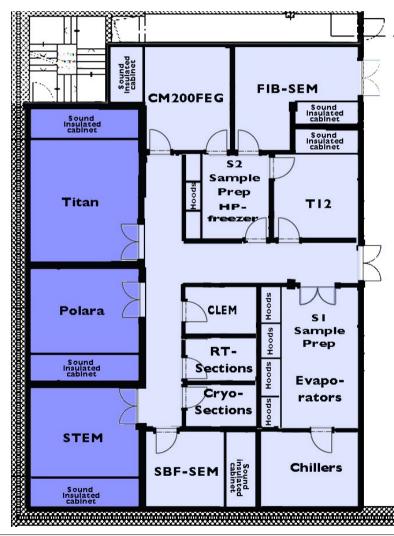


C-CINA (Uni Basel) EM Suite (2018)

Scripps EM facility control room



Future instruments will mostly operate automatically or with remote control.



C-CINA (Uni Basel) EM Suite (2018)

- Expected Problems:
 - Access for instruments ok? (Can doors, elevators, floors cope with the width, height and weight of the instruments?)
 - Ventilation often too loud.
 - Electromagnetic fields: Cancellation systems only work for one location in the room. µ-metal expensive. Best may be to look for another location for the suite.
 - Vibrations: Sometimes easiest to catch at the source.
 - House cooling water often not steady enough.
 - Will you need a UPS device?
 - Work with vendors when planning and preparing the site. Remember, they want to help you. (But don't sign waivers).

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Installation

- Define your specifications of the needed instrument (you)
- Negotiate with vendor about features, instrument details, warranty period, prices.
 Define acceptance test conditions. Speak with other instrument owners about possible weaknesses of the instruments, and put your conditions and specifications into ordering contract.
- Announce purchase intention for bidding at your home institution (several months)
- Order the instrument from vendor
- Vendor builds your instrument at vendor site (several months)
- Inspect and test the finished instrument at the vendor site
- Delivery of the instrument to your lab, installation (weeks to months)
- Acceptance test in your lab. When do you sign the acceptance protocol? Usually: Signing the acceptance protocol starts the clock for the warranty period, and releases the vendor from having to further improve the instrument performance. However, vendors also are interested in maintaining their good reputation, and usually have helped customers also after instrument acceptance. As long as you are not satisfied with the performance of the instrument, and don't sign the acceptance protocol, you may not be allowed to use the instrument for your research. Work with the vendors, in order to achieve the common goal: Satisfied and successful customers are their best advertisement.



Operation of your EM facility

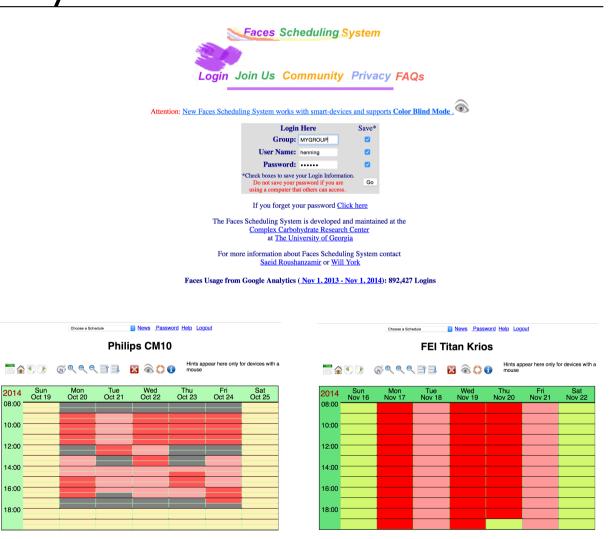
- For a large facility:
 - EM facility manager! Needs to have good social and instrument skills.
 - How do you cover instrument maintenance? How do you pay for it?
 - Wetlab manager (?)
 - Image processing manager ?
 - IT support is often provided by the host institutions.
 - How do you maintain long-term expertise in
 - Biochemistry, Sample preparation, EM handling, Image processing?

Running your EM facility

• A free online system is available at http://faces.ccrc.uga.edu

It features

- users assigned to user groups •
- instruments, instrument categories, and instrument times (day/night)
- accounting (if needed)



08:00

10:00

12:00

14:00

16:00

18:00