DATASHEET

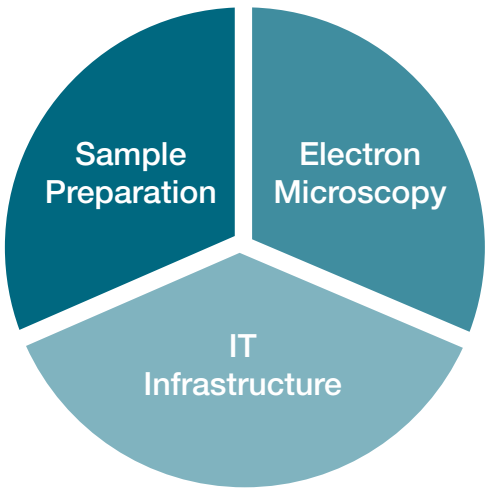
Guidance for planning a cryo-EM laboratory

Expert support and advice on laboratory organization

As there is no single rule on how a cryo-electron microscopy (cryo-EM) laboratory should be organized, Thermo Fisher Scientific provides support and advice to assist your planning in multiple ways:

- We provide specific guidance on site specifications for every microscope type with a dedicated Pre-Installation Manual (PIM).
- A site survey, conducted by one of our representatives, verifies that all site requirements are met.
- We provide extensive support for placing of Thermo Scientific™ products via different Site Preparation Service Portfolios. This service includes consultation for the building of new laboratories.
- A dedicated Thermo Fisher Scientific customer success manager is the single point of contact, responsible for coordinating the necessary resources.

In order to build a cryo-EM laboratory, three main infrastructure components should be planned, as illustrated below:



Thermo Scientific™ Vitrobot™ Mark IV System specimen preparation unit for cryo-EM

Glow discharge

Sample preparation

Prior to vitrification, the biochemical preparation of the sample can be done in any standard biochemical laboratory. Preparation of the sample for cryo-EM requires equipment for vitrification:

- Carbon evaporator to prepare the EM grid support
- Glow discharger to prepare the EM grid
- Vitrobot System to vitrify specimen
- LN_2 / liquid ethane (bottle) for freezing solutions, with a fume hood fitted with an O_2 alarm and special ethane valve

Minimal space is required to accommodate the required equipment (standard lab bench, see left photo above).

Thermo Scientific™ Arctic Express™ Transport Systems

Once the sample is frozen, it can be carried in LN_2, which means the sample preparation facility does not need to be located in close proximity to the electron microscope room. It is also possible to ship samples over longer distances in dry shippers.

Microscope facility

With respect to organizing the microscope facility, a regular existing laboratory can quite often be used with minimal modifications. A Cryo-EM lab can be part of BSL-1,2 facility. However, when planning a microscope facility, you do need to take into account the following essential requirements:

- Room dimensions
- Transportation route
- Environmental conditions

Room dimensions

Microscope facility room dimensions must comply with specifications to guarantee a trouble-free, safe installation and reliable operation.

The room space required for microscope installation is divided into two or three separate rooms:

- Microscope room
- Service room
- Operating room (recommended but not required)
- The Service room must be separated from the microscope room for both safety and comfort reasons. This also reduces building costs due to less stringent requirements than those for the microscope room.

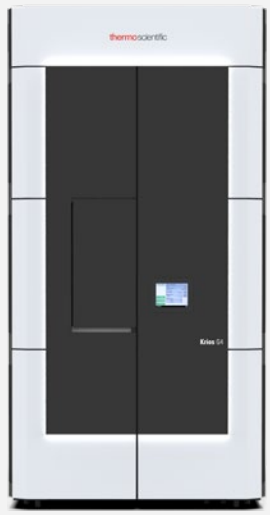
The room dimensions are microscope-dependent:

Dimensions	Tundra Cryo-TEM	Glacios Cryo-TEM	Krios G4 Cryo-TEM
Height	2.74 meters	2.8 meters	2.97 meters
Floor space	3.8 x 4.0 meters (with the cryo loading station in a neighbor room); 4.2 x 4.0 meters (with the cryo loading station in one room)	3.6 x 4.2 meters	6.7 x 5.1 meters

Tundra Cryo-TEM

Cryo loading station

Glacios Cryo-TEM



Krios G4 Cryo-TEM

Transportation route

Because some components being delivered are quite large, the full transportation route from the unloading area to the system space should comply with appropriate specifications. Such compliance will always be verified as part of the site survey.

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Environmental conditions

Generally, it is critical to make sure that the intended site for electron microscopy is not in the vicinity of:

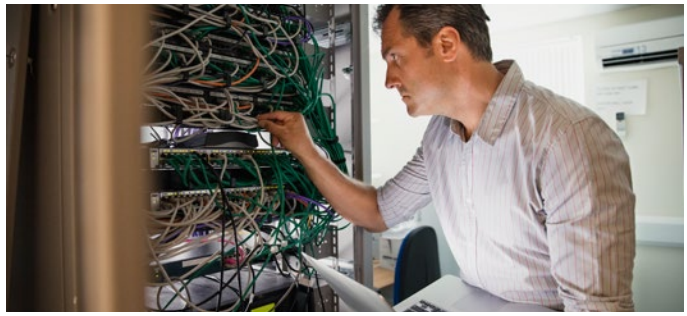
- Stray magnetic fields, e.g. locations near large motors and transformers, electric railways and tramways or any high-power or other EMI-radiating equipment
- Vibrations or high acoustic noise levels, e.g. locations near elevators, trains, shipping vehicles, busy roads, etc.

Additionally, each microscope room must comply with certain other requirements:

- Temperature and relative humidity
- Heat dissipation
- Ventilation, appropriate O_2 monitoring and room cleanliness
- Vibrations, acoustics and pressure waves
- Floor requirements

The precise specifications will depend on the microscope type and are provided with the PIM.

Most sites will meet the requirements for microscope installation, but if not, then technical solutions will be suggested to mitigate any issues. For example, if the proposed site does not comply with vibration specifications, an Integrated Vibration Isolation System (iVIS) could be offered; if electric and magnetic field requirements are not met, a third-party field-cancellation solution would be suggested.



IT infrastructure

Electron microscopy produces massive amounts of data within a very short amount of time (as much as 0.2-2 TB per day). As such, careful consideration needs to be taken when planning for IT Infrastructure:

- A 10G/Infiniband connection between all systems from which and to which data will be transferred.
- 100 TB of processing storage capacity is a good amount to begin the first year of instrument use.

3D reconstruction

With the introduction of GPU-accelerated tools, reconstruction packages have become significantly faster. It is possible to achieve atomic resolution reconstruction within days on a GPU workstation and within hours on a GPU cluster. A minimum of a four-GPU server or workstation is recommended.

Reconstruction packages can also run on a CPU cluster. A minimum of 64 GB of RAM is recommended and a cluster of at least 100-200 cores.

There are several commercial recommendations given on the www.SingleParticle.com site, where systems also come with pre-installed 3D reconstruction software. Another commercial option is to run 3D reconstruction on the Amazon EC2™ cloud.

Find out more at **thermofisher.com/EM-Sales**

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