

Specimen preparation methods for Scanning Electron Microscopy

Sample preparation basics to high-quality imaging

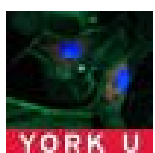
To find out more please navigate to <http://microscopy.info.yorku.ca>

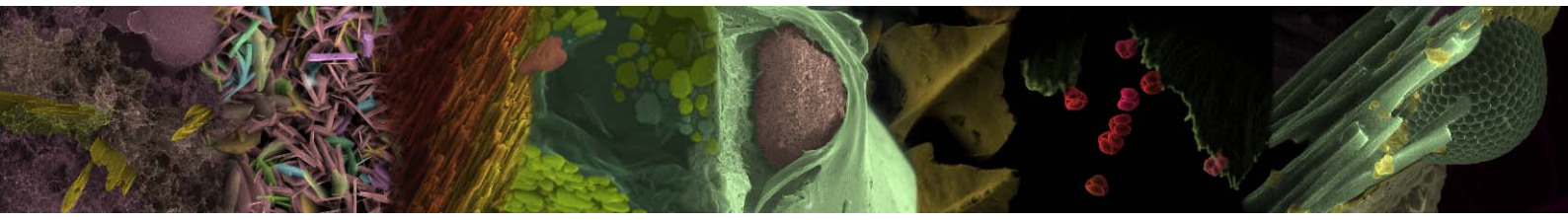
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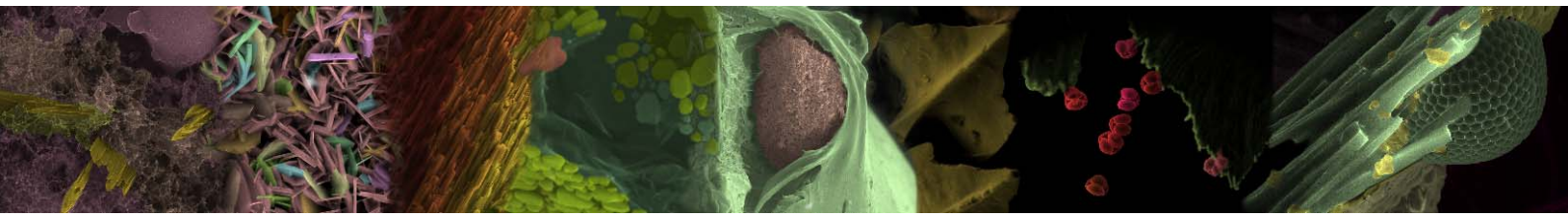
✉ microscopy@yorku.ca for any additional information





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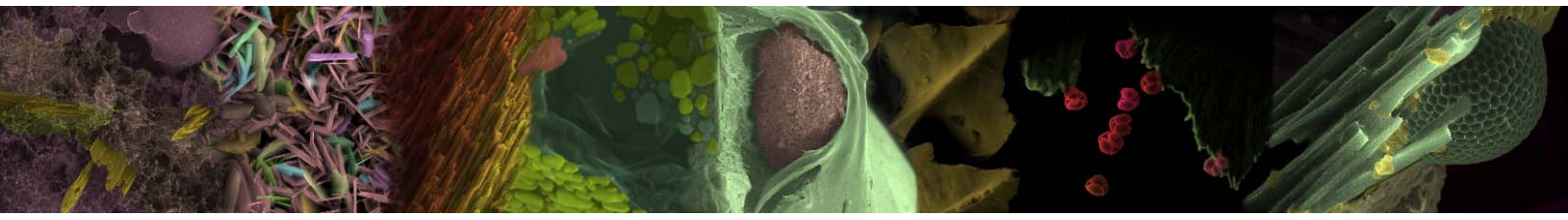
Overview

Quanta 3D is the most versatile electron microscope ever designed.

The Quanta can process a specimen in any form, however not all samples can be imaged without preparation.

The most common factors affecting imaging are: surface charging, degassing, mobility, beam sensitivity and magnetic properties.

Specimen harvesting, preservation and preparation are major factors impacting final SEM images.



Thermofisher Quanta 3D FIB FEG SEM with ESEM technology

Specifications: <http://microscopy.info.yorku.ca/quanta-3d>

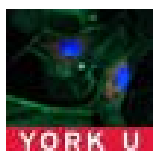
Detectors

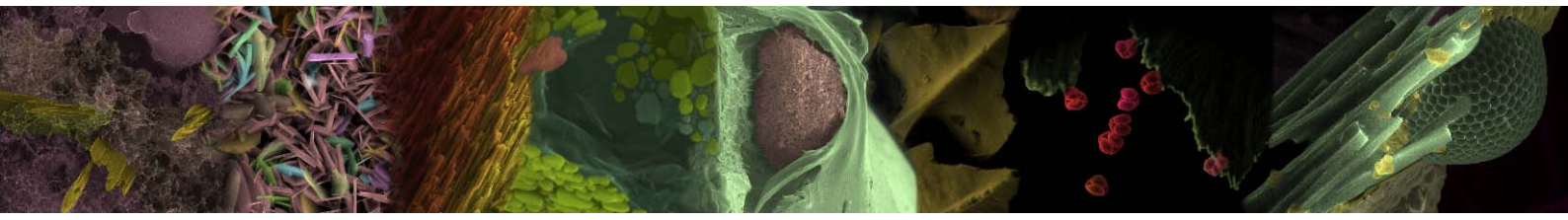
- Everhardt Thornley SED (secondary electron detector)
- Low vacuum SED
- Gaseous SED (GSED) (used in ESEM mode)
- Gaseous analytical BSED (GAD)
- Gaseous BSED
- Solid State backscatter detector BSED
- 14 segment STEM detector
- In-column detector (ICD) for BD mode
- EDS
- EBSD

System options

Beam deceleration

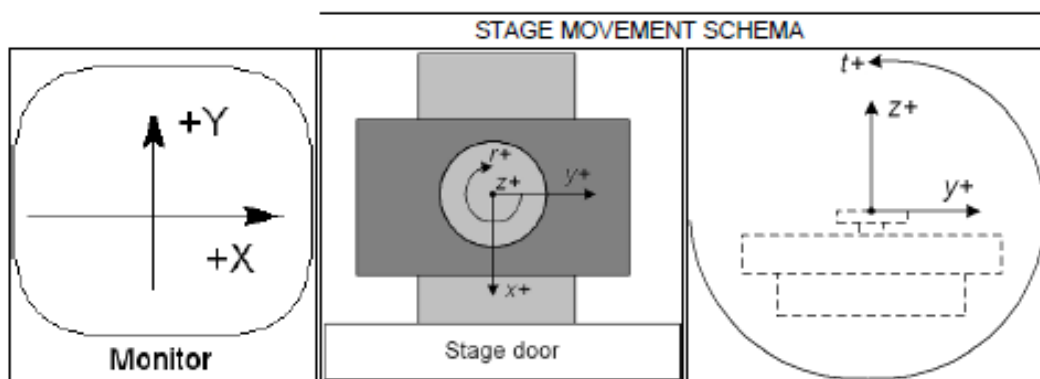
- SW controlled Peltier cooled specimen stage
- SW controlled 1400°C heating stage
- FEI Gas Injection System: Platinum





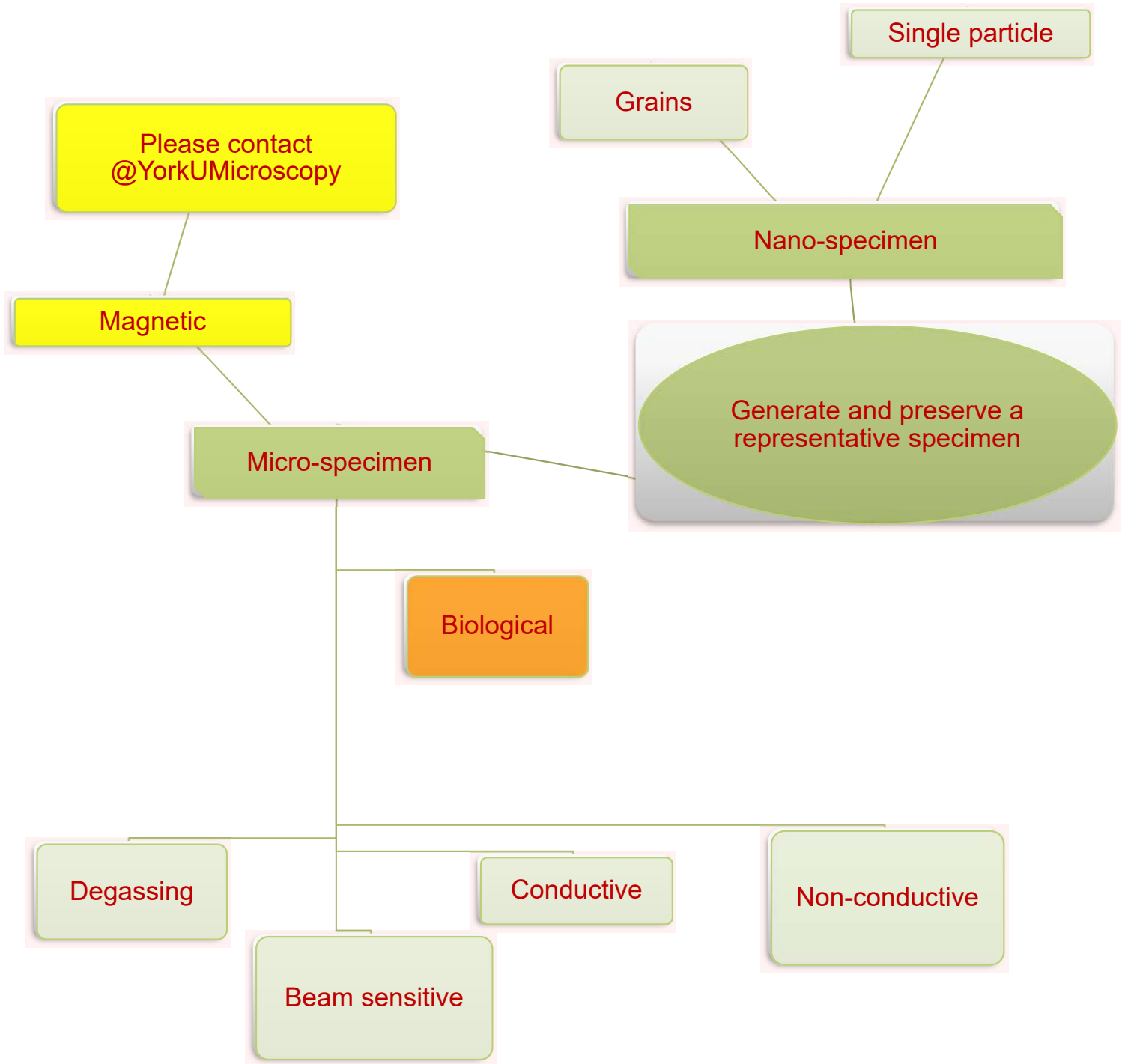
Standard specimen dimensions

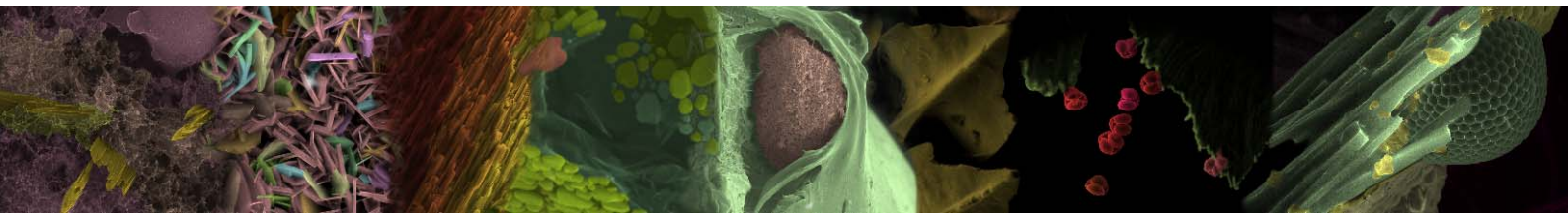
Please evaluate your specimen size and weight – your specimen must not be larger than the stage diameter and taller than the stage clearance (together with a holder).



Item	50 mm Stage
X	-25 to +25 mm
Y	-25 to +25 mm
Z	25 + 25 mm
R	360°
T	-15° to +75°
Eucentric Position	Electron column: WD = 10 mm Ion column: WD = 19 mm
Clamp	No
Maximum sample weight	250 g
	1 000 g

Specimens





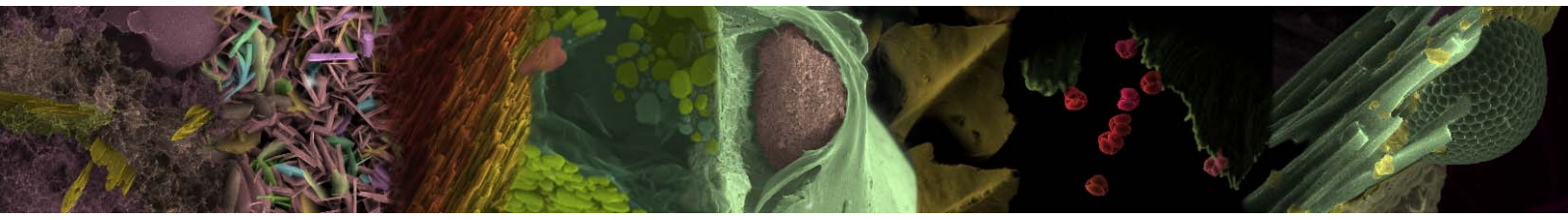
Basic sample preparation

1. Generate a representative specimen from the sample population
2. Prevent cross-contamination
3. Select a holder and a bonding medium
4. Quanta stage is equipped to house pin stubs

Note: Your specimen must not move in any direction once mounted. To execute you can use a conductive paint, a conductive tape or a special clamp.



Note: SEM Pin Stub Specimen Mounts and mounting media are readily available along with storage boxes

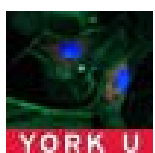


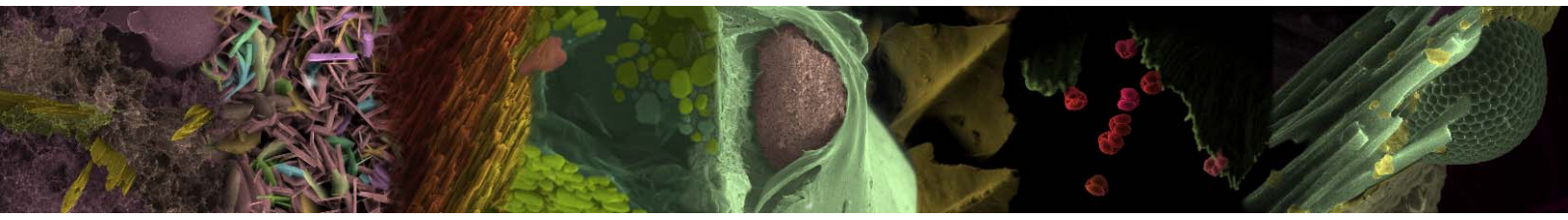
Specimen holders maintenance

Specimen holders are subject to contamination. They should be fully cleaned after every use.

Recommended cleaning procedures are given below. Infrequent/inadequate cleaning will result in low image quality and can potentially lead to the microscope chamber contamination.

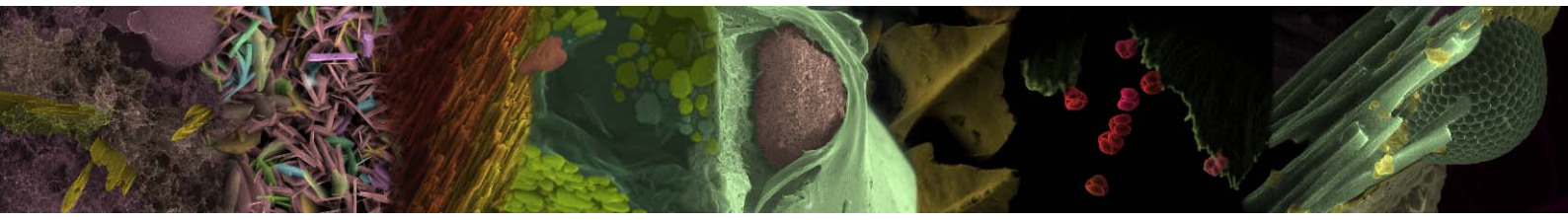
1. Extirpate your specimen and the bounding media from the holder
2. Rinse the holder in tap water with a mild abrasive domestic cleaner
3. Rinse in tap water
4. Clean in an ultrasonic cleaner for 5 minutes using distilled water.
5. Clean in an ultrasonic cleaner for 5 minutes using alcohol p/a or
6. Isopropanol (Do not place parts together in the beakers. Wash separately as damage can occur to the metal surfaces)
7. Rinse in alcohol p/a.
8. Blow dry the holder with a compressed air canister
9. Dry the holder under an infra-red lamp (15 min to 1 hr) at a temperature of between 80 °C and 100 °C





Conductive specimens

Mount the specimen exposing a selected surface for observation. Please note that rough, uneven surfaces may present a challenge in getting an evenly illuminated image.

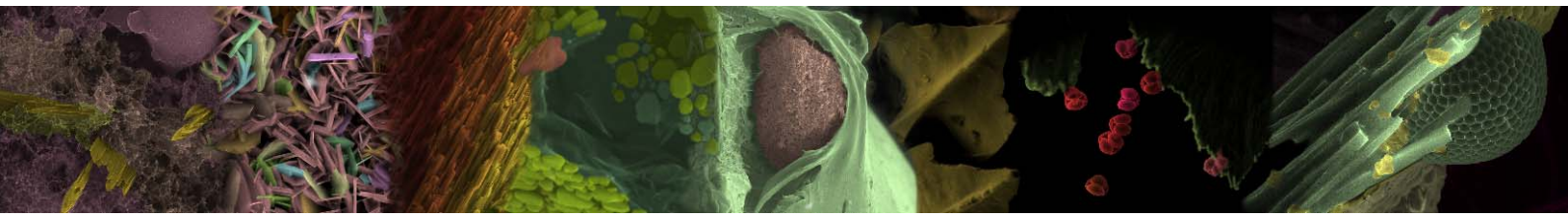


Non conductive specimens

A solid specimen can be mounted directly on a pin stub. An imaging surface should be minimized as possible by covering with a piece of conductive tape or a conductive paint.

A conductive bridge to the surface of a stub is needed to partially discharge the specimen.

Imaging in low vacuum/ESEM mode is recommended.



Non conductive specimens suitable for sputter coating

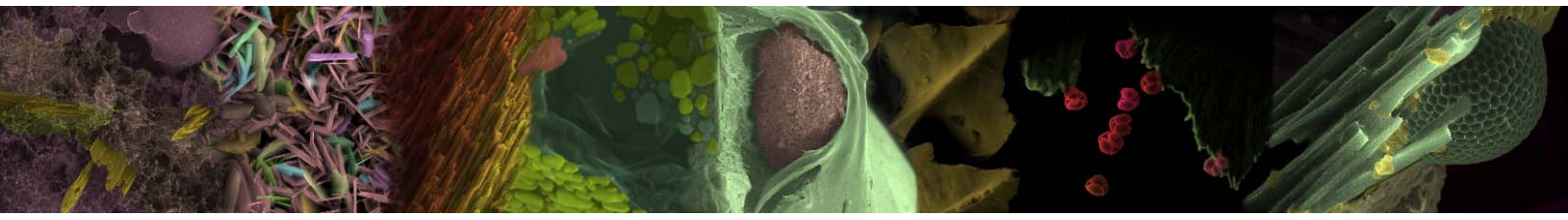
A solid, non-conductive specimen can be coated with a layer of a conductive material by means of a sputter coating.

Note: BS signal may be lost

The Facility houses a Denton V Sputter coater with a platinum target.

Specimens have to be mounted before coating.

If multiple specimens are mounted on the same stub – all specimens must be the same height.



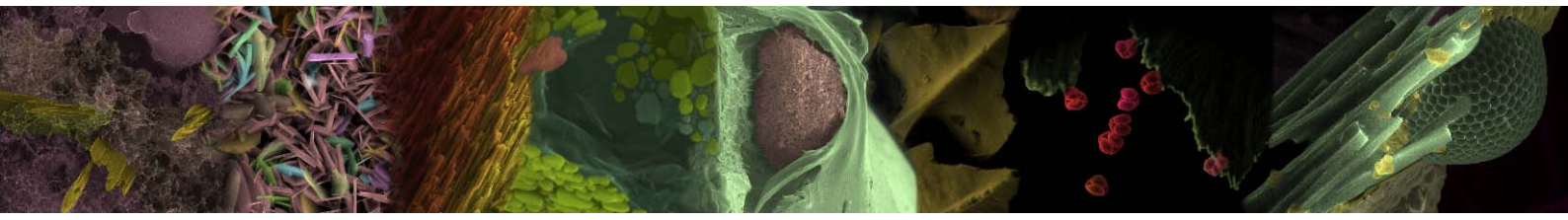
Beam sensitive samples

Example: thin foils, porous/fibrous polymer composites, gels

Please be advised that standard settings for high resolution work will result in degradation of the scanned area.

Recommended actions:

1. Sputter coating
2. Low voltage set-up and fast integration (the desired resolution may not be reached)
3. Low magnification approach is preferable
4. Imaging on a cooling stage (restriction in sample size will apply)

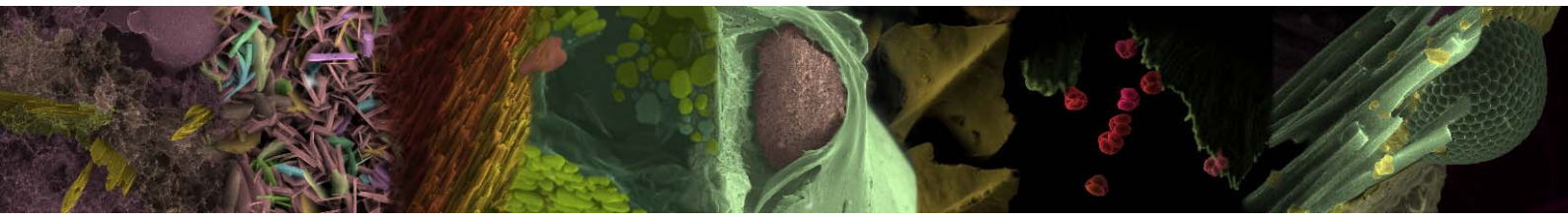


Powder samples

Only a dispersed, bounded monolayer will be accepted for the analysis.

Please assure that all particles are bounded to a substrate. The specimen will be sprayed with a compressed air to test the bound.

Please consider embedding and ultra-microtoming option.



Biological structures

Quanta 3D is fully equipped to handle a biological specimen without any special preparation.

Outgassing in wet specimen cannot be avoided - some microstructures provide acceptable results – in some cases additional sample preparation work is required.

Specimen should be sized up for a cooling stage holder (5mm by 5 mm) and cooled down to 5 C.

Imaging on a cooling stage should prevent the biological structure from collapsing.