Sample preparation for Scanning electron microscopy (SEM)

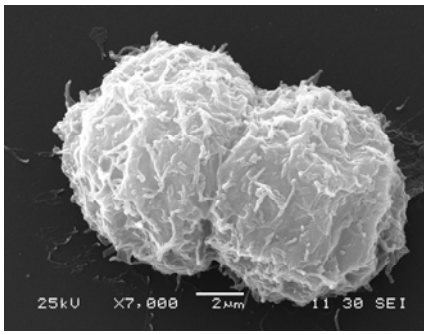
SEM is primarily useful for giving a three-dimensional image of the surface of the specimen and is for viewing large objects.

As with TEM, specimens are imaged with a beam of electrons, but instead of the electrons being transmitted through the specimen, the beam is "scanned" across, creating an image of the surface of the sample, with exceptional depth of field. This image is achieved via the detection of "secondary" electrons that are released from the specimen as a result of it being scanned by very high energy "primary" electrons (ie. those emitted from the electron "gun" in the SEM). As most biological specimens are made up of non-dense material the amount of secondary electrons produced is too low to be of much use in creating an image and therefore they are usually coated with a very fine layer of a metal which readily produces secondary electrons. The large depth of field achievable can produce an image of great visual depth with a three-dimensional appearance.

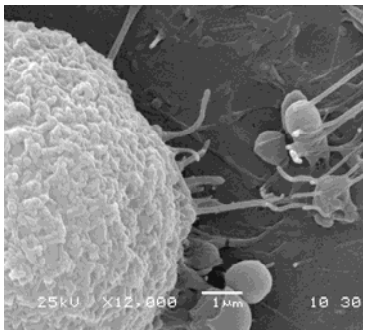
The operating environment of a standard scanning electron microscope dictates that specialist preparation techniques are used. Typically, a biological specimen is chemically fixed, dehydrated through an acetone or ethanol series and then dried at the critical point - a method used to minimize specimen distortion due to drying tensions. For dry samples, this process is not necessary. SEM can also be used to investigate smooth surfaces of industrial samples.

The samples are mounted on a stub of metal with adhesive, coated with 40 - 60 nm of metal such as Gold/Palladium and then observed in the microscope.

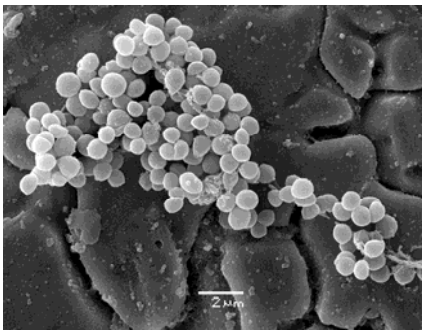
Every sample is different. Please consult with the EM Staff before starting a project.



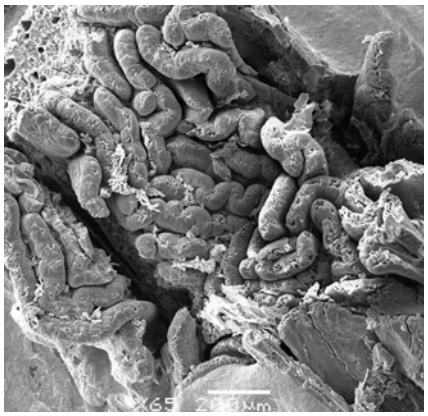
Epidermoid carcinoma cells. Bar: 2 µm



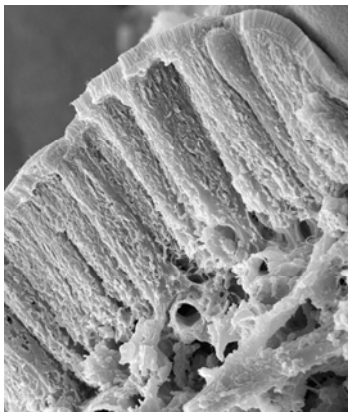
Epidermoid carcinoma cells. Bar: 2 µm



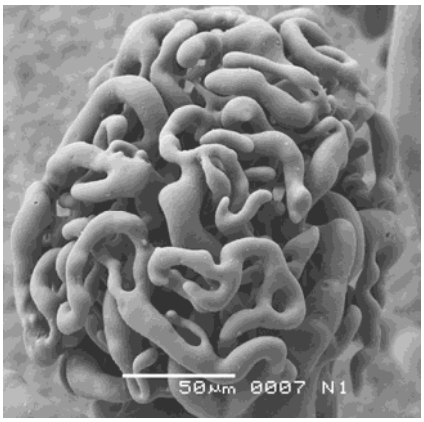
Staphylococcus Aureus. Bar: 2 µm



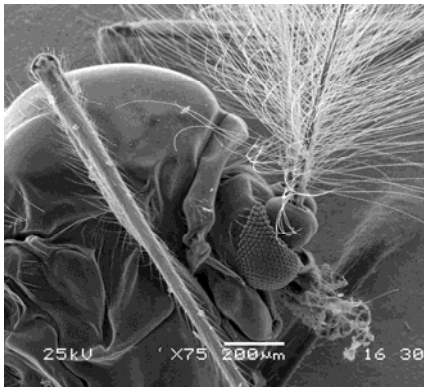
Small intestine, villi.
Bar: 200 µm.



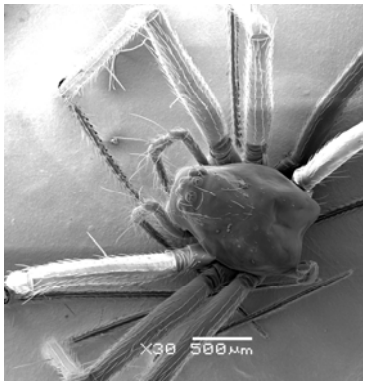
Small intestine, microvilli on the surface of a villus.



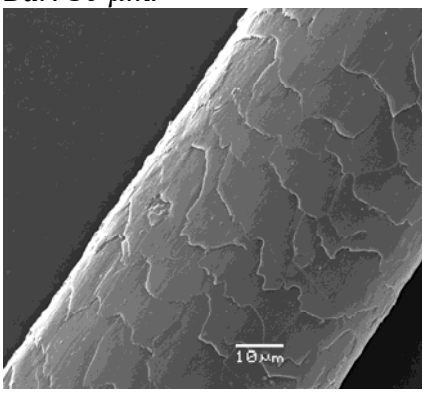
Plastinated glomerulus from rat. Tissue is removed.
Bar: 50 µm.



Fly
Bar: 200µm.



Spider
Bar: 300µm.



Hair – on the surface.
Bar: 10µm.

SEM services include:

- Fixation and dehydration
- Critical point drying
- Drying with hexamethyldisilazane (HMDS) and t-Butanol
- Coating with Gold/Palladium using Sputter coater
- Image processing (software Scandium)