Electron microscope Diagnosis

• An electron microscope is a microscope that uses a beam of accelerated electrons as a source of illumination.

- Scanning electron microscopy (SEM),
- Transmission electron microscopy(TEM),
- Scanning transmission electron microscopy (STEM),
- Focus ion beam microscopy (FIB)

HOW A SCANNING ELECTRON MICROSCOPE WORKS

• The main SEM components include:

- Source of electrons
- □ Column down which electrons travel with electromagnetic lenses
- Electron detector
- Sample chamber
- Computer and display to view the images

• Electrons are produced at the top of the column, accelerated down and passed through a combination of lenses and apertures to produce a focused beam of electrons which hits the surface of the sample. The sample is mounted on a stage in the chamber area and, unless the microscope is designed to operate at low vacuums, both the column and the chamber are evacuated by a combination of pumps. The level of the vacuum will depend on the design of the microscope.

FUNCTIONS:

- Imaging (fluorescence, lattice-resolved and topography)
- Chemical analysis
- Structure determination
- Manipulation of atoms and molecules
- Nanolithography, e-beam lithography
- Spectroscopy: surface, electrical and optical properties

Transmission electron microscopy (TEM)

Sample preparation TEM

• For sample preparation three important steps are Processing, Embedding and Polymerization. Processing further includes fixing, rinsing, post fixation, dehydration and filtration.

Primary fixation of tissues

Double fixation is the most popular method of fixation, as it involves primary fixation in an aldehyde followed by secondary (post) fixation in osmium tetroxide.

Glutaraldehyde (dialdehyde) preserves ultrastructure well but penetrates slowly than the monoaldehyde, paraformaldehyde. Glutaraldehyde is used alone for small pieces of material, but a mixture of the two aldehydes may be used for perfusion fixation or fixation of larger items.