TEM

Transmission Electron Microscopy (TEM) is a microscopy technique in which a thin beam of rapidly moving electrons is transmitted through an ultra-thin specimen, interfering with the specimen placed in the tube and producing a black-and-white resulting image at very high resolution. The electron microscope consists of a cylindrical tube, which is devoid of air. The electrons are emitted by the cathode at the top of the tube and then accelerated by the anode. They then pass through a small aperture (metallic plate), which forms them into the beam in the vacuum inside the tube. The part of the microscope that generates the beam is sometimes called the electron gun. The beam is maintained along the tube by means of electromagnetic lenses. These are coils that surround the tube at given intervals. The electromagnetic field emitted by the coils focuses the beam at the center of the tube. Electromagnetic lenses are used to deflect the path of the electrons. Two condenser lenses control the "illumination" of the sample (how bright and how large the area illuminated) followed by the objective lens, which combines the scattered with unscattered electrons to give a contrasted image. After the objective lens follow a number of intermediate imaging lenses paired with apertures that are used to change the magnification from 50x to 400,000x. The image can be observed on a phosphorous screen or on a monitor.

In TEM, electrons have very high energies and are moving very fast, thus it cannot be used to observe "live" samples. Electrons encounter the specimen and are either absorbed (inelastically scattered, contributing to contrast), scattered (bounce off the nucleus and bent elastically, change direction but do not change speed or energy), or pass through it (no interaction with atom). Because different regions of the specimen are variously transparent to electrons, different amounts of electrons with changed energy pass through these regions. At the end of the tube the electrons are collected on fluorescent or photographic film or on screens that generate an image of the specimen. The beam that reaches the film consists of the different amounts of electrons that pass through particular regions of the specimen. This difference is responsible for the contrast in the film. The original image produced by the electron microscope is always black and white, and it is not possible to see it directly with the naked eye. Color may be artificially added later to the image for emphasis purposes.

Amplitude contrast is used to visualize sections of cells. TEM gives you a projection image at much higher resolution of ultra-thin (<100nm thick) samples that are introduced into the microscope, using elastically scattered electrons to generate contrast in the image. Elastically scattered electrons can be removed by means of an aperture. The aperture is placed after the objective lens and allows the unscattered electrons to pass through and blocks the elastically scattered ones, generating contrast in the image making regions where lots of scattering happen appear dark (electron dense), and regions where there was less scattering appear bright (electron lucent). Phase contrast is generated in images of crystalline structures such as protein and macromolecule complexes. The objective aperture of the microscope is utilized to make the elastically scattered electrons and unscattered electrons interfere, generating contrast. This is based on the principle that electrons come in waves and that when they pass through matter the scattered wavefront will suffer a phase shift, which will interfere with the unscattered wavefront. The observed image depends on several factors including amplitude of the electron beam and phase of the electrons.

Essential Components of TEM

- Electron gun
- Vacuum tube
- Sample stage
- Condensor lenses
- Imaging lenses with corresponding apertures
- Objective lens with corresponding aperture
- Display device

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