

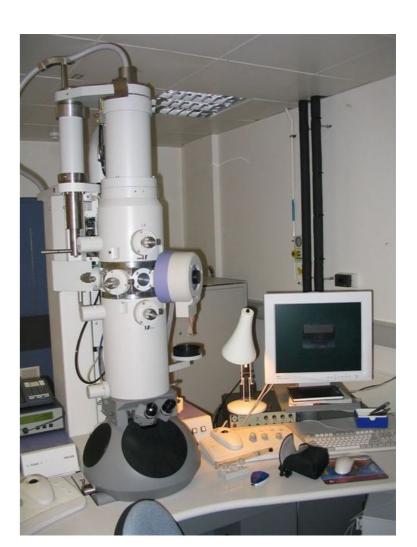


Electron microscopy

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Electron microscopy

Electron microscopy (EM) is a technique that uses a beam of electrons instead of visible light to observe the structure of small objects at very high magnifications. Electrons have much shorter wavelengths than light, allowing for the visualization of much smaller structures, including cellular components, viruses, and materials at the atomic level.

Transmission Electron Microscopy

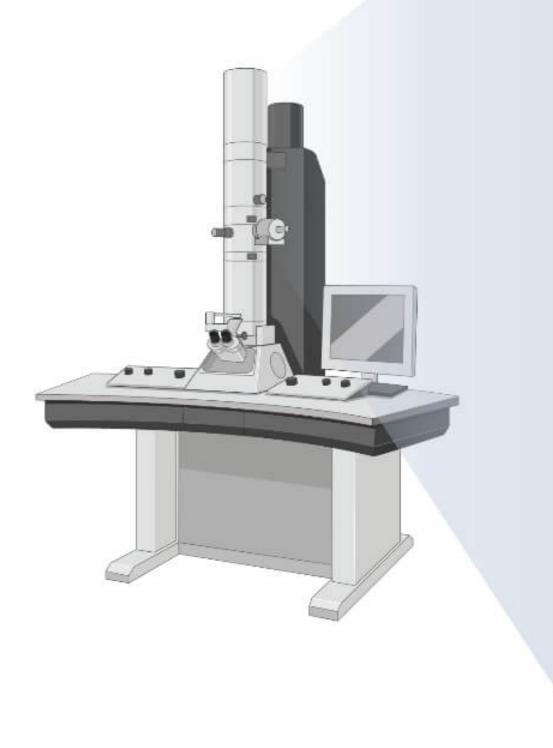
The transmission electron microscope (TEM) is an imaging system that permits

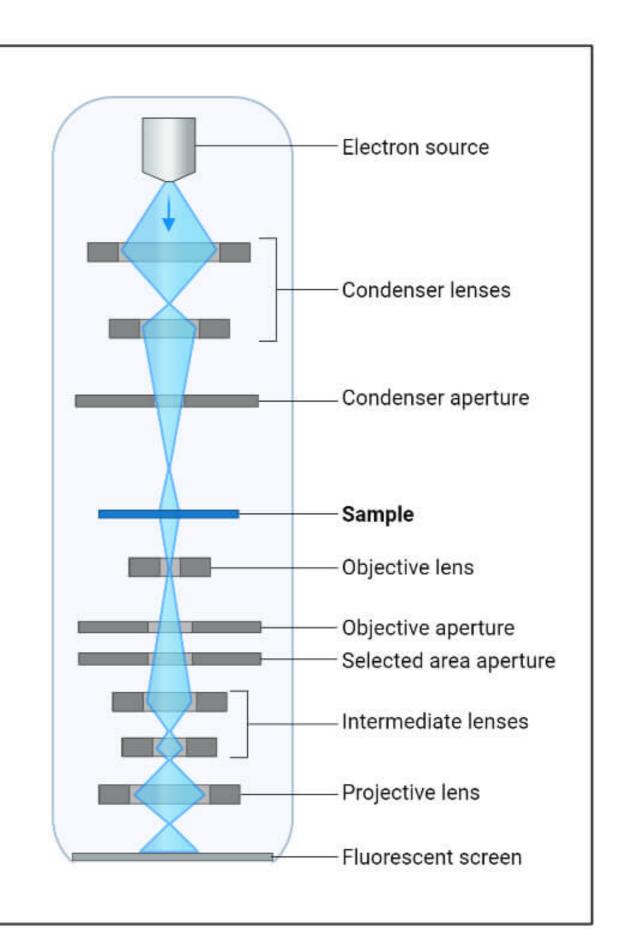
very high resolution (3 nm) This high resolution allows magnifications of up to 400,000 times to be viewed with details. Unfortunately, this level of magnification applies only to isolated molecules or particles. Very thin tissue

sections can be observed with details at magnifications of up to about 120,000

times.

Transmission Electron Microscopy (TEM)





deflected by electromagnetic fields in a manner similar to light deflection in glass

lenses. The beam is produced by a cathode at the top of the instrument and passes

down through the chamber in a vacuum. Because electrons change their path when

submitted to electromagnetic fields, the beam can be focused by passing through

electric coils which can be considered as electromagnetic lenses.

The first lens is a condenser focusing the beam of electrons on the specimen

section. Some electrons interact with atoms in the section and their course is

modified, while others simply cross the specimen without interacting.

Electrons

passing through the specimen reach the objective lens, which forms a focused,

magnified image that is then magnified further through other lenses and captured

on a viewing screen. The image of the specimen shows areas of white, black, and

shades of gray corresponding to areas through which electrons readily passed (appearing brighter or electron lucent) and areas where electrons were absorbed or

deflected (appearing darker or more electron dense). To improve contrast and resolution in TEM, compounds with beavy metal ions (like osmium tetroxide

resolution in TEM, compounds with heavy metal ions (like osmium tetroxide, lead

citrate) are often added to the fixative or dehydrating solutions used to prepare the

tissue. This will bind cellular macromolecules, increasing their electron density and visibility.

To provide a useful interaction between the specimen and the electrons,

TEM requires very thin sections (40–90 nm); therefore, embedding is

performed

with a hard epoxy and sectioning is done with a glass or diamond knife. The

extremely thin sections are collected on small metal grids and transferred to the

interior of the microscope to be analyzed.

Scanning Electron Microscopy

Scanning electron microscopy (SEM) permits pseudo-three-dimensional views of

the surfaces of cells, tissues, and organs. Like the TEM this microscope produces

and focuses a very narrow beam of electrons, but in this instrument the beam

does

not pass through the specimen . Instead the surface of the specimen is

first dried and coated with a very thin layer of metal atoms through which

electrons do not pass readily.

When the beam is scanned from point to point across the specimen it interacts with the metal atoms and produces reflected electrons or

secondary electrons emitted from the metal. These are captured by a detector and

the resulting signal is processed to produce a black-and-white image on a monitor.

SEM images are usually easy to interpret because they present a 3D view that

appears to be illuminated from above, in the same way that large objects are seen

with highlights and shadows caused by light from above.

