

**University of Virginia
Environmental Health & Safety (EHS)
Radiation Safety Office**

Radiation Safety Requirements for Use of Electron Microscopes

▶ Source of Radiation from an Electron Microscope

X-rays are produced in the electron microscope whenever the primary electron beam or back scattered electrons strike metal parts with sufficient energy to excite continuous and/or characteristic X-radiation. In terms of X-ray hazards, two aspects are important: the composition of the parts which are struck and their efficiency as X-ray sources and the effectiveness/integrity of the shielding provided by the metal casing of the microscope around these.

The higher the voltage and atomic number of the “parts”, the greater the efficiency of X-ray production.

The degree of X-ray “leakage” also depends on the shielding provided by the metal casing. A poorly designed microscope may have weak points where X-rays can escape, for example, between the gasket-sealed junction of two sections of the column.

▶ What are the radiation safety concerns?

The radiation safety concerns are related to the **electrons that are backscattered** from the sample, as well as **X-rays produced** in the process. Most modern electron microscopes are extremely well shielded and do not produce exposure rates greater than background. However, electron microscopes are radiation-generating devices and should be at least inventoried. It is also important that the integrity of the shielding is maintained, that all existing interlocks are functioning, and that workers are aware of radiation safety considerations.

▶ Rules for Safe Use

1.0 Applicability

The rules in this section apply to the following miscellaneous X-ray producing equipment: electron microscopes, electron microphones, luminoscopes, and cold-cathode gas discharge tubes. These requirements are in conformity with the Radiation Protections Regulations included in the Virginia Administrative Code (VAC). These regulations can be found on our webpage at: [12VAC5-481](#).

2.0 Registration

There are no requirements to register your electron microscope with the State of Virginia, however, maintains an inventory of X-ray producing equipment at UVA. **Please notify Radiation Safety if you install, move or dispose of an electron microscope.**

3.0 Posting

No area posting is required for electron microscopes.

4.0 Warnings and Labels

A label bearing the statement: "CAUTION: THIS EQUIPMENT PRODUCES RADIATION WHEN ENERGIZED - TO BE OPERATED BY QUALIFIED PERSONNEL ONLY" should be posted on the electron microscope.

5.0 Training

Individuals who wish to operate electron microscopes are not required to complete documented radiation safety training. However, all individuals should review this document and receive hands-on instruction and training (e.g. working with experienced users, reading the manufacturer's operation manual) before independently using the equipment.

6.0 Operating Procedures

Operating procedures shall be written and available to all users.

Each machine should be key controlled when not in use. Interlocks, if present, must remain operational unless approved by the RSO.

7.0 Radiation Limits

Radiation emitted from electron microscopes shall not exceed a dose equivalent rate (averaged over 10 square centimeters) of **0.25 mrem** (at **5 cm** from any accessible surface of the equipment).

8.0 Personnel Monitoring

Personnel monitoring is not required for users of electron microscopes.

9.0 Notifications & Emergency Procedures

The Radiation Safety Office must be notified at 2-4919 if any modifications are made to the interlocks or any other safety devices or in the event of an emergency involving the equipment. The microscope user should also keep a copy of operating and emergency procedures at the operator panel.

10.0 Inspections & Recordkeeping

Safety evaluations (including radiation surveys) should be performed initially when equipment is installed and after equipment has been moved.

The Radiation Safety Office will keep inventory and survey information on file. The electron microscope user should keep a logbook of any maintenance done on the equipment.

▶ Background - Types of Electron Microscopes

TEM (Transmission Electron Microscope)

A beam of [electrons](#) is transmitted through an ultra-thin specimen, interacting with the specimen as it passes through it. The electron beam is accelerated by an [anode](#) typically at +100keV (40 to 400 keV) with respect to the cathode, focused by [electrostatic](#) and [electromagnetic](#) lenses, and transmitted through a specimen that is in part transparent to electrons and in part [scatters](#) them out of the beam. An image is formed from the electrons transmitted through the specimen, magnified and [focused](#) by an objective lens and appears on an imaging screen, a fluorescent screen in most TEMs, plus a monitor, or on a layer of photographic film, or to be detected by a sensor such as a [CCD camera](#).

The TEM is used heavily in both material science/metallurgy and the biological sciences. In both cases, the specimens must be very thin and able to withstand the high vacuum present inside the instrument.

For biological specimens, the maximum specimen thickness is roughly 1 micrometer. To withstand the instrument vacuum, biological specimens are typically held at liquid nitrogen temperatures after embedding in vitreous ice, or fixated using a [negative staining](#) material such as [uranyl acetate](#) or by plastic embedding. In material science/metallurgy, the specimens tend to be naturally resistant to vacuum, but must be prepared as a thin foil, or etched so some portion of the specimen is thin enough for the beam to penetrate. Preparation techniques to obtain an electron transparent region include [ion beam](#) milling and wedge polishing.

SEM (Scanning Electron Microscope)

Type of [electron microscope](#) that images the sample surface by scanning it with a high-energy beam of electrons in a [raster scan](#) pattern. The electron beam, which typically has an [energy](#) ranging from a few hundred [eV](#) to 40 keV, is focused by one or two condenser lenses to a spot about 0.4 nm to 5 nm in diameter. Unlike the TEM, where electrons of the high voltage beam form the image of the specimen, the SEM produces images by detecting low energy secondary electrons which are emitted from the surface the specimen due to excitation by the primary electron beam. The electrons interact with the atoms that make up the sample producing signals that contain information about the sample's surface [topography](#), composition and other properties such as [electrical conductivity](#). The types of signals produced by an SEM include [secondary electrons](#), back scattered electrons (BSE), [characteristic x-rays](#), light ([cathodoluminescence](#)), specimen current and transmitted electrons. These types of signal all require specialized detectors for their detection that are not usually all present on a single machine.

The SEM micrographs have a very large [depth of field](#) yielding a characteristic three-dimensional appearance useful for understanding the surface structure of a sample. Because the SEM uses electromagnets rather than lenses, the researcher has much more control in the degree of magnification.

For conventional imaging in the SEM, specimens must be [electrically conductive](#), at least at the surface, and [electrically grounded](#) to prevent the accumulation of [electrostatic charge](#) at the surface. Metal objects require little special preparation for SEM except for cleaning and mounting on a specimen stub. Nonconductive specimens tend to charge when scanned by the electron beam, and especially in secondary electron imaging mode, this causes scanning faults and other image artifacts. They are therefore usually coated with an ultrathin coating of electrically-conducting material, commonly gold, deposited on the sample either by low vacuum [sputter coating](#) or by high vacuum evaporation. Conductive materials in current use for specimen coating include [gold](#), gold/[palladium](#) alloy, [platinum](#), [osmium](#),^[5] [iridium](#), [tungsten](#), [chromium](#) and [graphite](#). Coating prevents the accumulation of [static electric charge](#) on the specimen during electron irradiation.

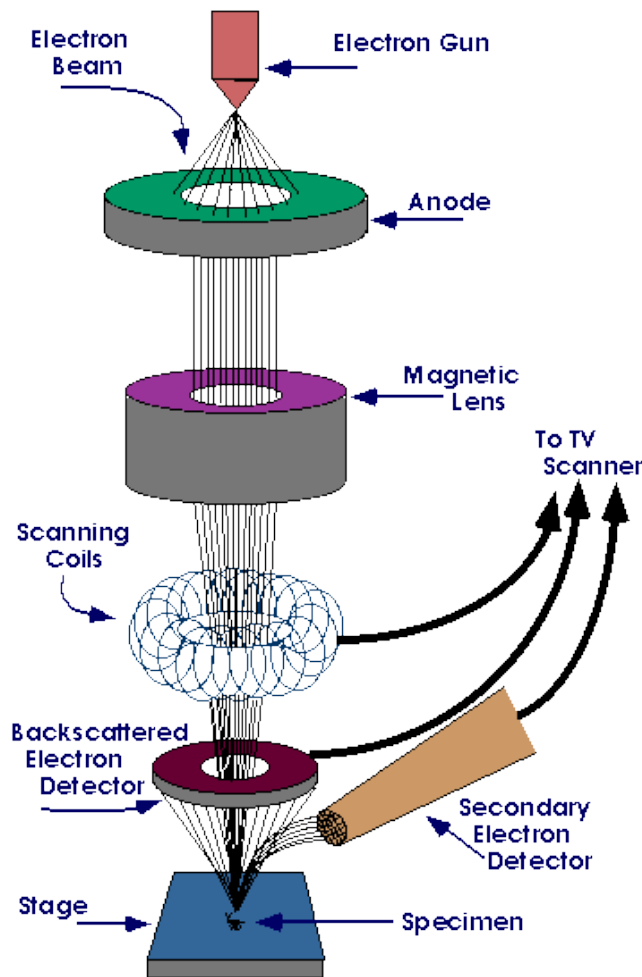
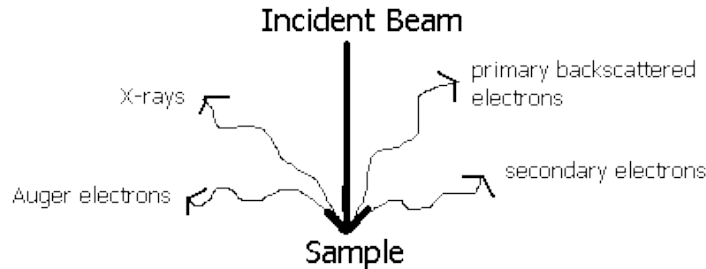


Diagram courtesy of Iowa State University SEM Homepage

A beam of electrons is produced at the top of the microscope by an electron gun. The electron beam follows a vertical path through the microscope, which is held within a vacuum. The beam travels through electromagnetic fields and lenses, which focus the



beam down toward the sample. Once the beam hits the sample, electrons and X-rays are ejected from the sample. Detectors collect these X-rays, backscattered electrons, and secondary electrons and convert them into a signal that is sent to a screen similar to a television screen. This produces the final image.

REM (Reflection Electron Microscope)

In the **Reflection Electron Microscope (REM)** as in the TEM, an electron beam is incident on a surface, but instead of using the transmission (TEM) or secondary electrons (SEM), the reflected beam of [elastically scattered electrons](#) is detected. This technique is typically coupled with [Reflection High Energy Electron Diffraction](#) and *Reflection high-energy loss spectrum (RHELS)*. Another variation is Spin-Polarized Low-Energy Electron Microscopy (SPLEEM), which is used for looking at the microstructure of [magnetic domain](#)

STEM (Scanning Transmission Electron Microscope)

A **scanning transmission electron microscope (STEM)** is a type of [transmission electron microscope](#). With it, the [electrons](#) pass through the specimen, but, as in [scanning electron microscopy](#), the electron optics focus the beam into a narrow spot which is scanned over the sample in a raster. The STEM rasters a focused incident probe across a specimen that has been thinned to facilitate detection of electrons scattered **through** the specimen. The motivation for STEM imaging of biological samples is particularly to make use of dark-field microscopy, where the STEM is more efficient than a conventional TEM, allowing high contrast imaging of biological samples without requiring staining.