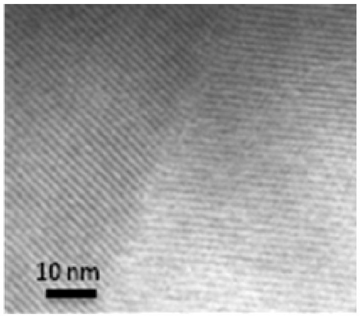
STEM-in-SEM – Analysis of Biological Specimens

SMART – POWERFUL – FLEXIBLE

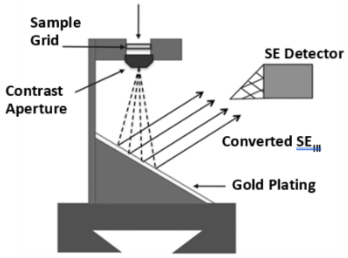
STEM-in-SEM (Scanning Transmission Electron Microscopy in an SEM) has become a popular technique for biologists, polymer scientists and materials scientists for its ease of use, cost effectiveness and high resolution. It is especially suited to investigating the internal structure of thin film (100-200nm) samples as well as size and shape of submicron to nanometer particles. With standard SEM imaging modes on bulk samples, there are limitations in the ultimate resolution that can be achieved due in part by the beam-sample interactions. With STEM-in-SEM, the sample is very thin and the interaction volume is small. Therefore, the resolution more closely approximates the diameter of the electron beam at the exit surface of the sample allowing for high resolution; using STEM with our state of the art FESEMs, sub-nanometer resolution is easily achieved.



STEM-in-SEM image of Faujasite zeolite with 7.4A lattice spacing.

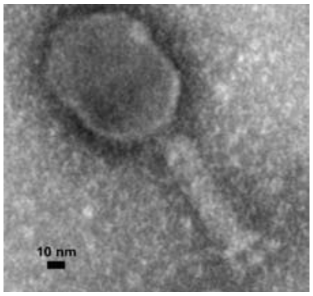
STEM-in-SEM is often referred to as a “low voltage STEM” technique. This is in reference to the 20-30kV typical operating energies used in the SEM versus the 100-300kV used with a traditional dedicated STEM or TEM. While the resolution of low voltage STEM cannot compete with that of dedicated STEM or TEM it does offer its advantages. Using accelerating voltages of 30kV or lower enhances image contrast, especially for low atomic number components or low density specimens. Enhanced image contrast at lower voltages make STEM-in-SEM an excellent method for the characterization of biological samples. Other advantages include reduced charging, less potential sample damage and reduced contamination.

There are two configurations traditionally used for STEM-in-SEM: 1) a STEM converter and 2) a dedicated STEM detector. The STEM converter offers a cost-effective and simple method for obtaining a bright field STEM image. It fits in an SEM like a standard sample holder and does not require an additional detector or hardware. A STEM converter holds a 3mm diameter sample (typical size for TEM/STEM holders) and has a hole through it. The beam scans on the sample and transmitted electrons strike a polished mirror surface below the sample. The transmitted electrons are converted to secondary electrons and these secondary electrons are detected by the in-chamber secondary electron detector.

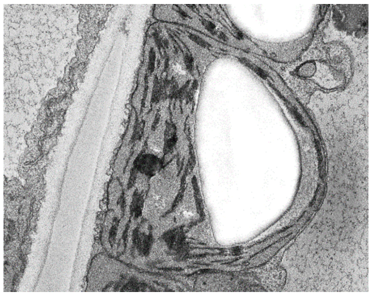


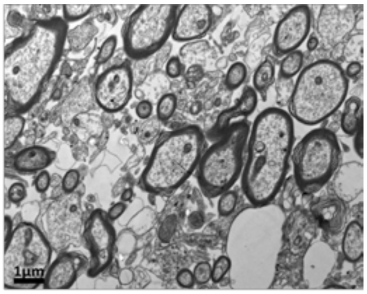
STEM Converter

STEM detectors can easily be interfaced with SEMs and ultrathin sections or nanometer range particles mounted on grids can be observed using the same optics as regular SEM operation. A dedicated STEM detector will allow for more flexibility such as both brightfield and darkfield STEM imaging or utilizing a multi- grid holder for higher throughput.



STEM-in-SEM 30kV image of negatively stained T4 bacteriophage.


STEM-in-SEM 30kV image of tobacco leaf cross section.



STEM-in-SEM 30kV image of a mouse brain- fixed and stained