

3D-IMAGING OF BIOLOGICAL SPECIMENS BY ELECTRON MICROSCOPY

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New techniques are becoming available to study the complex structures of Life Science specimens. 3D-imaging using confocal microscopy in the late 80ties and early 90ties caused really a breakthrough in understanding the cell functions. The next logical step will be to resolve the ultra-structural changes down to the nanometer level in relation to functionality of a cell or cell organelles.

Software intelligence to facilitate the use of different microscope techniques is the key to results in modern microscopy. Imaging is no longer a 2D-presentation of the 3D-structure of the specimen. Software enables the user to acquire a whole series of images resulting in a 3D-reconstruction of the specimen. The 3D-visualization makes it a lot easier to understand the complex ultra-structure of cells or cell-organelles.

The results obtained at high resolution contain the necessary information to understand the processes and related structural changes during the many biological processes within a cell. The ultimate goal is specimen protection to achieve ultra-structural details down to Angstrom level. Many features are designed and re-designed to maintain specimen integrity during preparation, transfer and observation. New techniques are constantly developed.

Once in the microscope, the clean vacuum assures a contamination free observation for long working sessions. At the low temperature, the microscope can be used close to liquid nitrogen-temperature. In combination with the low dose software this gives the best protection for beam sensitive specimens. Important other improvements made are the optimization of the low magnification high-resolution performance.

Finally, in recent years a significant change took place in the recording of the images. Highly sensitive CCD cameras are used to record the digital images; a darkroom is no longer needed. Automation offers the possibility to obtain results without the involvement of the user and to improve the statistics of results by taking more images during the time that the microscope is not heavily used. The system's reliability assures researchers a high throughput of results. The specially designed low dose software and the automated 3D-imaging software make a microscope session relatively short. The recording of a data set is automated and results can be recorded without the user being present.

In this contribution, the entire route –including description of critical steps and factors- for obtaining 3D-images at high resolution will be exemplified by showing data of biological specimens.