

What Life Scientists Should Know About Molecular Imaging

Optical Imaging, Ultrasound, Photoacoustics

Advanced Microscopy Technologies

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Learning Objectives:

- Derive what limits the resolution in microscopy
- List several high- and super-resolution techniques
- Discuss pros and cons of advanced fluorescence microscopy techniques

More than 100 years ago, Ernst Abbe has theoretically shown that diffraction of the detected light in any optical microscope fundamental limits the resolution to about half the wavelength used for imaging. Using visible light (with a wavelength between 400 and 700nm), fine object structures below 200nm could not be resolved in these instruments. In recent years however, various high- and super-resolution microscopy devices have been developed in order to measure and visualize structural details of fluorescent objects with unprecedented quality down to the nanometer range. Starting with considerations of light as a wave and of its propagation, we will take a close look at some of the optical elements that make up a microscope in order to get an idea of the image formation process.

We will then point out which parts are limiting the resolution of the final images, and what physical effects can be used to overcome this limitation. Several examples of high- and super-resolution microscopes will be given with an explanation as to how each of these particular microscopic techniques circumvents the classical resolution limit. Example applications of these microscopes to biological specimens will be shown, illustrating the enormous progress that far field optical microscopy has recently made.