Limit of resolution of optical microscope

Optical Microscope uses system of lenses and visible light to sharply magnify small detailed samples which is projected directly to the eye.

In the 1870s, Ernst Abbe explained why the resolution of a microscope is limited. Since the microscope uses visible light and visible light has a set range of wavelengths. The microscope can't produce the image of an object that is smaller than the length of the light wave. Any object that's less than half the wavelength of the microscope's illumination source is not visible under that microscope. Light microscopes use visible light.

Limitations of Resolution

- The diffraction limits the resolution to approximately 0.2 µm.
- It is difficult to differentiate the four lines drawn within a 250 nm. Below this line lies the realm which is invisible to human naked eye: 200-250 nm approximately.
- The resolution of the light microscope cannot be small than the half of the wavelength of the visible light, which is 0.4-0.7 µm. When we can see green light (0.5 µm), the objects which are, at most, about 0.2 µm. Below this point, light microscope is not useful, as wavelength smaller than 400 nm is needed.

The waves that associate the electrons has smaller wavelength. Then we can use electrons using an electron microscope. Electron microscopes can be used to visualise viruses, molecules and even individual atoms.

Live cells generally lack sufficient contrast to be studied successfully, internal structures of the cell are colourless and transparent. The common way is to increase contrast by different structures with selective dyes, but it often involves killing and fixing the sample.

These limitations have overcome to some extent by specific microscopy techniques that can non-invasively increase the contrast of the image. In general, these techniques make use of differences in the refractive index of cell structures.

Links

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Bibliography

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