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- Concept 6.1: To study cells, biologists use microscopes and the tools of biochemistry

Microscopy

- Scientists use microscopes to visualize cells too small to see with the naked eye

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- Light microscopes (LMs)
 - Pass visible light through a specimen
 - Magnify cellular structures with lenses

- Different types of microscopes

- Can be used to visualize different sized cellular structures

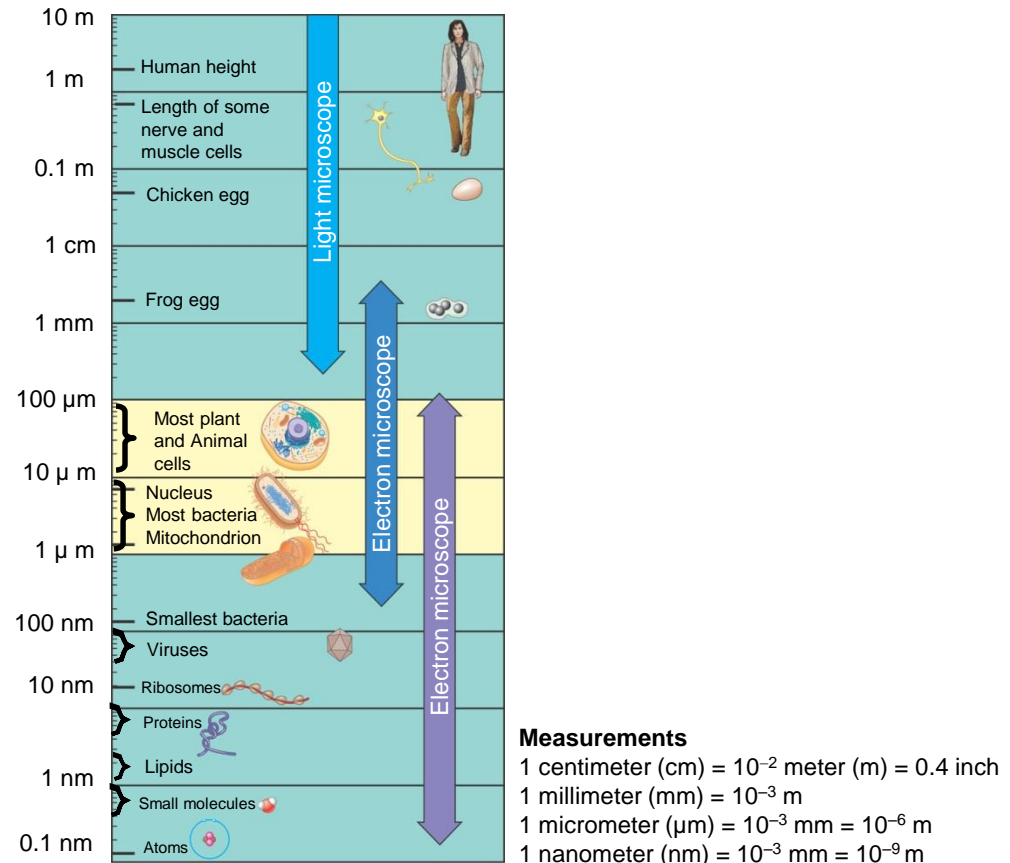


Figure 6.2

– Use different methods for enhancing visualization of cellular structures

TECHNIQUE

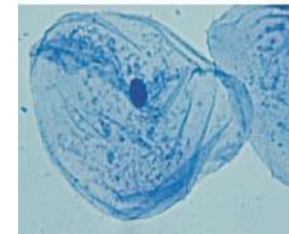
(a) Brightfield (unstained specimen).
Passes light directly through specimen. Unless cell is naturally pigmented or artificially stained, image has little contrast. [Parts (a)–(d) show a human cheek epithelial cell.]

RESULT



50 μm

(b) Brightfield (stained specimen).
Staining with various dyes enhances contrast, but most staining procedures require that cells be fixed (preserved).



(c) Phase-contrast. Enhances contrast in unstained cells by amplifying variations in density within specimen; especially useful for examining living, unpigmented cells.



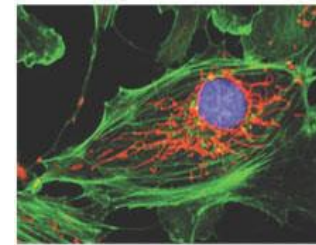
Figure 6.3

(d) Differential-interference-contrast (Nomarski).

Like phase-contrast microscopy, it uses optical modifications to exaggerate differences in density, making the image appear almost 3D.

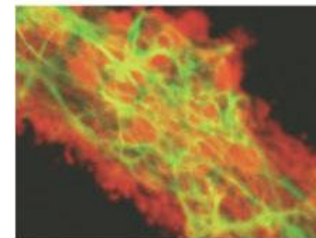
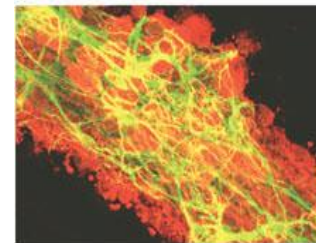


(e) Fluorescence. Shows the locations of specific molecules in the cell by tagging the molecules with fluorescent dyes or antibodies. These fluorescent substances absorb ultraviolet radiation and emit visible light, as shown here in a cell from an artery.



50 μ m

(f) Confocal. Uses lasers and special optics for “optical sectioning” of fluorescently-stained specimens. Only a single plane of focus is illuminated; out-of-focus fluorescence above and below the plane is subtracted by a computer. A sharp image results, as seen in stained nervous tissue (top), where nerve cells are green, support cells are red, and regions of overlap are yellow. A standard fluorescence micrograph (bottom) of this relatively thick tissue is blurry.



50 μ m

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- Electron microscopes (EMs)
 - Focus a beam of electrons through a specimen (TEM) or onto its surface (SEM)

- The scanning electron microscope (SEM)
 - Provides for detailed study of the surface of a specimen

TECHNIQUE

- (a) **Scanning electron microscopy (SEM).** Micrographs taken with a scanning electron microscope show a 3D image of the surface of a specimen. This SEM shows the surface of a cell from a rabbit trachea (windpipe) covered with motile organelles called cilia. Beating of the cilia helps move inhaled debris upward toward the throat.

RESULTS

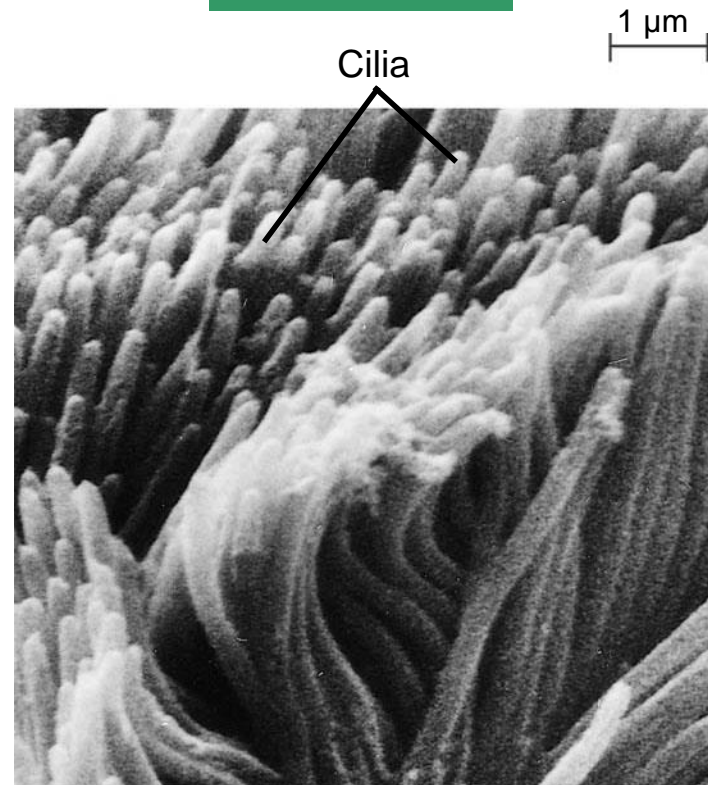


Figure 6.4 (a)

- The transmission electron microscope (TEM)
 - Provides for detailed study of the internal ultrastructure of cells

(b) Transmission electron microscopy (TEM). A transmission electron microscope profiles a thin section of a specimen. Here we see a section through a tracheal cell, revealing its ultrastructure. In preparing the TEM, some cilia were cut along their lengths, creating longitudinal sections, while other cilia were cut straight across, creating cross sections.

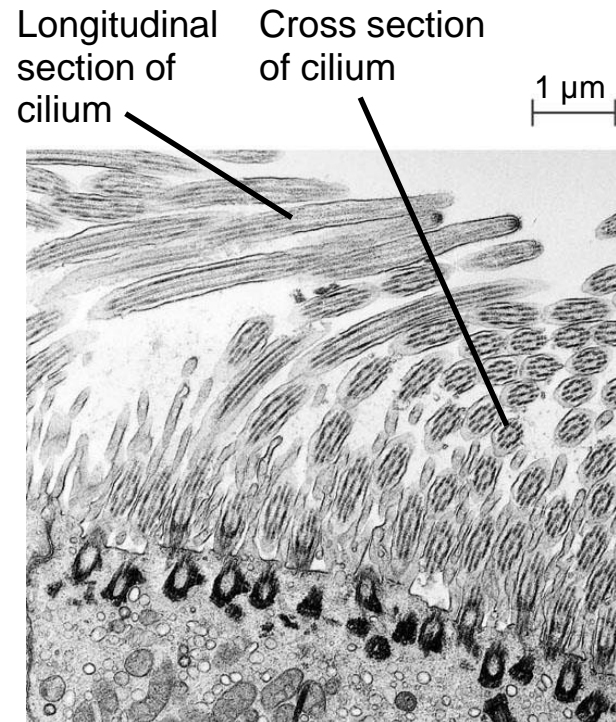


Figure 6.4 (b)