Cell Motility

We often answer the question, "Is it alive?" by checking to see if it moves. All living cells exhibit movement of some type. Some cells move through their environment or move their environment past their "bodies"; all cells undergo intracellular movements of their cytoplasm and the organelles within it. This movement is based ultimately on cellular mechanisms that rely on one of two basic systems: actin (micro-)filaments or microtubules made of tubulin. Other proteins may be involved with actin and tubulin to produce or direct movement. We categorize the types of movement produced by these systems in one of three ways when it is utilized to move an organism or its extracellular environment: amoeboid movement, ciliary/flagellar movement, and muscular movement. Two of these are easily observable in protists; their characterization is the objective of this laboratory.

Amoeboid Movement is named for its observation in the unicellular protist - *Amoeba*. It is characterized by streaming of cytoplasm from the "body" of the organism into limb-like extensions known as pseudopods ("false feet") that can extend outward in any direction. The extended pseudopods can be anchored to the substratum and pull or push the cell through the environment. They can also be used to surround particles and engulf them in phagosomes. The later process of phagocytosis may be used in feeding by protists and in immune defenses by the leukocytes of vertebrates. Movement through the environment is easily observed in *Amoeba* although it is not limited to these unicellular organisms. Both leukocytes and embryonic cells move through the body of vertebrate animals as well.

The cytoplasm of *Amoeba* exists in two states. One is relatively solid and is known as the plasmagel; the other is very fluid and is called the plasmadol. The plasmagel exists in a thin peripheral layer of the cytoplasm (the ectoplasm) and there are very few organelles within it. The plasmadol phase is characteristic of the inner portion of the cell; it is called the endoplasm, has a granular appearance and contains the major organelles.

During amoeboid locomotion, there is a transformation between the gel and sol states of the cytoplasm at the anterior and posterior ends of the cell. (See fig. 16-31, p. 536 in text.) The advancing end of the organism is identifiable by a thick region of clear cytoplasm called the hyaline cap. Immediately posterior, the endoplasm streams laterally and posteriorly and changes from sol to gel. The cytoplasm at the trailing end of the organism, called the uroid, undergoes the opposite change, from gel to sol. Actin filaments have been demonstrated in cells that undergo amoeboid locomotion, and actin interacts in a way that is not yet clear with the actin-dependent motor protein, myosin, to produce movement. There is both a pushing force moving cytoplasm into the advancing pseudopod and a pulling force moving the "body" of the cell toward the anchored anterior extension. The same proteins, actin and myosin, occur in a very regular arrangement in skeletal muscle cells. There they produce movement by sliding along each other, shortening the cell and pulling on whatever structure attaches to the muscle tendon.

You will observe amoeboid locomotion in live *Amoeba* or *Pelomyxa*. These are rather large organisms and, in order to observe them without crushing them, you will need to make a
Vaseline chamber to elevate the cover slip. This also will inhibit drying of the preparation.

**Ciliary and Flagellar Movement** are based on a system of microtubules, composed of tubulin. The arrangement of these microtubules is identical in both organelles, which differ in their length, number, and pattern of movement. A cross-section of a flagellum from *Chlamydomonas* appears in the text in fig. 16-22, p. 529 along with a diagram identifying the components visible in the electron micrograph. The pattern seen is often referred to as "9 (x 2) + 2" and is characteristic of the portion of the cilium/flagellum that extends beyond the cell surface. At the base of each cilium/flagellum, within the cortex of the cell, there is a "basal body" in which the arrangement of microtubules differs. The basal body has the same structure as a centriole, with an array of "9 (x3) + 0." In addition to the tubulin, motile cilia and flagella have a second protein, dynein, attached as "arms" to the sides of the microtubules. Dynein is a "motor protein" that "walks" along the surface of a microtubule much as myosin does along actin filaments. Some invertebrates have non-motile cilia in photoreceptor cells; these non-motile cilia have the same arrangement of microtubules, but they have no dynein arms attached.

The movement patterns of cilia and flagella differ in that cilia demonstrate an oralike beat, with power and recovery strokes; flagella usually undulate with waves moving from the base to the tip, although other patterns may be visible.

You will observe ciliary locomotion and feeding in *Paramecia* and flagellar locomotion in *Chlamydomonas*. In another lab, you will observe and experiment with the process of flagellar assembly in the same organism, so note carefully what an "untouched" *Chlamydomonas* looks like. As you observe the paired flagella, look for evidence of variation in the pattern of movement. Because locomotion via cilia and flagella is often very rapid, it is sometimes difficult to make continued observations. The *Paramecia* can be slowed by addition of Congo red-stained yeast cells around which they will congregate in order to feed, allowing you to observe simultaneously the action of cilia during locomotion and feeding. Note that the cilia create a current of fluid and particles that is directed into the buccal cavity; this process is similar to the flow of mucus and trapped particles along respiratory surfaces in many organisms.

**Procedures:**

**Examination of Amoeba**

1. Use a dissecting microscope to locate individuals in the culture provided; with a dropper, gently transfer one or two to a clean slide. (Check with the microscope to make sure they’ve left the dropper.) Add a coverslip that has a ledge of Vaseline around the edges, deposited by scraping a small amount from the surface of your "greased" finger.

2. Under low power on a compound microscope, observe the movement of the organism(s) over a ten-minute period, making periodic drawings to show the different shapes and direction(s) of movement.

3. Observe cytoplasmic streaming within old and new pseudopods under low and high-dry magnification; how does the direction of intracellular movement differ? Does a change in direction of streaming occur first in the old or the new pseudopod? Where in the pseudopod is
movement first observed? Can you detect whether the movement is a push or a pull?

Examination of *Paramecium*

1. Place a drop of culture on a clean slide; add a small amount of Congo red-stained yeast suspension. The drop should be pink, not red. Add a coverslip and seal the edges with vaseline applied with a toothpick.
2. Before the paramecia begin feeding, note their normal swimming pattern. Identify the anterior and posterior ends and observe the rotation that occurs during swimming. Find a slow-moving organism and examine ciliary motion on various regions of the "body." Do all groups of cilia beat identically? If not, what are the differences?
3. Observe a feeding individual and note the collection of yeast cells in a food vacuole at the blind end of the cytopharynx. Do all yeast cells entering the buccal cavity get ingested? Follow the changes in a food vacuole over time. Does it move in the body? Does the color of the vacuole change? Congo red is a pH indicator: bright orange-red at/above pH 5, purplish between pH 3 and pH 5, and blue below pH 3.

Examination of *Chlamydomonas*

1. Place a drop of a *Chlamydomonas* culture on a clean slide; add a coverslip.
2. Observe the normal swimming motions under low power. You should be able to detect the movement of the paired flagella, even if you cannot see them clearly.
3. Make a fresh wet mount with a drop of Protosol added; observe this under low and high-dry objectives. You might have to close down the condenser iris in order to see the organelles. Note the action of the flagella in organisms that are moving forward (i.e., flagella leading.) Describe the motion of the flagella; does their coordinated movement resemble a swimming stroke used by humans?

In another laboratory, we will pH-shock some *Chlamydomonas*. This causes them to drop their flagella, which they will then re-assemble from tubulin monomers over time. We will track the re-assembly process under several different conditions. Observations will be made on organisms that have been killed and stained with Lugol’s iodine. Prepare one wet mount with a drop of Lugol’s iodine to familiarize yourself with the appearance of a flagellated organism after such a demise; locate several straightened flagella and measure them with an ocular micrometer.

**Points to Ponder**

1. Describe advantages and disadvantages of using the different types of locomotion you have observed today.

2. Both actin and tubulin are labile components of the cytoskeleton; of what significance is that
to the different types of locomotion?

3. What have you learned about feeding and digestion in the organisms used in this lab?