Chapter 6

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Protozoa and Nematodes

Elaine R. Ingham

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Thus when a soil loses fertility we pour on fertilizer, or at best alter its tame flora and fauna, without considering the fact that its wild flora and fauna, which built the soil to begin with, may likewise be important to its maintenance.

Aldo Leopold

Two components of the wild fauna which Leopold may have never considered are major players in the important processes that build soil. They are soil protozoa and nematodes. These organisms are nonphotosynthetic, motile, eukaryotic predators of bacteria and fungi and each other. Protozoa are considered **microfauna** because they are usually < 200 μm long, while nematodes are usually considered to be members of the **mesofauna** (200 to 1,000 μm long). Other members of the soil mesofauna (e.g., mites, rotifers, springtails, and tardigrades) and **macrofauna** (> 1,000 μm long; e.g., vertebrates, earthworms, and large arthropods) also play critical roles in soil processes, particularly with regard to decomposition of organic matter, but are beyond the scope of this microbiology textbook. For more information on the mesofaunal and macrofaunal groups, see the *Soil Biology Guide* by Dindal (1990) and *Fundamentals of Soil Ecology* by Coleman and Crossley (1996).

Classification

Protozoa

Protozoa are unicellular, eukaryotic organisms which represent a group in which mitosis and meiosis became established. Certain members of the protozoa may have been hosts for photosynthetic prokaryotes. These internal "parasites" evolved into plastids, suggesting an extremely important evolutionary role for protozoa. In the past, scientists generally believed that soil protozoa evolved from aquatic species, but recent evidence shows some unique soil protozoa probably did not evolve from aquatic forms.

Box 6–1

Free-Living Soil Protozoa. Free-living soil protozoa fall into three categories:

- **Flagellates** (Phylum Sarcomastigophora, Subphylum Mastigophora) move by means of one to several flagella.
- **Amoebae** (Phylum Sarcomastigophora, Subphylum Sarcodina) move by protoplasmic extrusions called pseudopodia. Recently, the slime molds (Sporangia) have been classified with amoebae.
- **Ciliates** (Phylum Ciliophora) move by means of cilia on the surfaces of the cells.

Flagellates. These are the smallest members of the protozoa and are divided into two classes based on whether they contain chlorophyll (Phytomastigophorea) or not (Zoomastigophorea). Only nonchlorophyll-bearing flagellates occur in soil; the photosynthetic species are strictly aquatic. Soil flagellates, like *Oicomonas*, *Scytomonas* and *Peranema*, are morphologically similar to photosynthetic flagellates. Other flagellates are the nonchlorophyll-bearing Zoomastigophorea, such as *Bodo* (Fig. 6–1) or *Pleuromonas*. Flagellates sometimes display amoeboid movements, although they usually have at least one flagellum. Flagellates also have a resting stage, called a cyst, which enables them to survive stressful environmental conditions (Fig. 6–2).

Amoebae. These protozoa move by protoplasmic flow, either with extensions called pseudopodia or by whole-body flow. Within this group:

- Naked amoebae are differentiated by **pseudopodia,** or "false feet" (Fig. 6–3), which can be lobose (rounded), conical (thinner at the tip than the base), filiose (thin filamentous extensions), or reticular (netlike). The number and flow patterns of the pseudopodia also are important taxonomic characteristics for naked amoebae.
- Slime molds have an amoeboid stage that also forms a stage that resembles a slug. This "slug" then differentiates into a sessile stage with a fruiting body containing spores on a stalk.
- Testate amoebae live within a **test** (or shell) constructed of soil particles bound together by secretions. The size and shape of the test, the size and shape of scales and spines or horns on the surface of the test, and the placement and border pattern of the pseudostome or "false mouth" (the opening in the test that allows the amoeba to go in and out of its "mobile home") are important characteristics for identifying species.

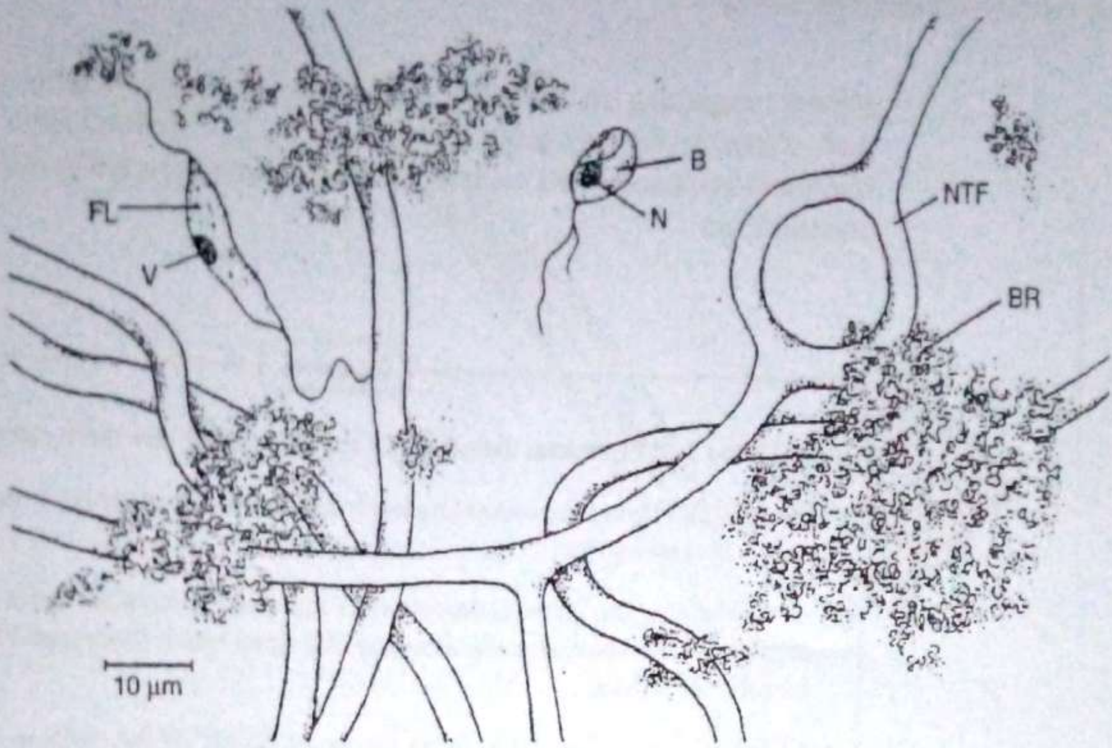

Figure 6–1 An example of two flagellates; *Bodo* sp. (B) with a flagellum emerging from anterior end, and a second flagellate (FL) with flagella arising at either end of the flagellate's body. Often the nucleus (N) or vesicles (V) can be seen within the cell. Compare the size of the two flagellates with the matrix of thousands of bacterial cells (BR) on which flagellates feed. A hypha of a large nematode-trapping fungus (NTF) is also visible. This fungus forms rings which produce compounds that attract nematodes. When a nematode contacts the inside of the ring, the ring constricts, holding the nematode so the fungus can grow into the nematode and consume it. *Drawing by Kim Luoma.*

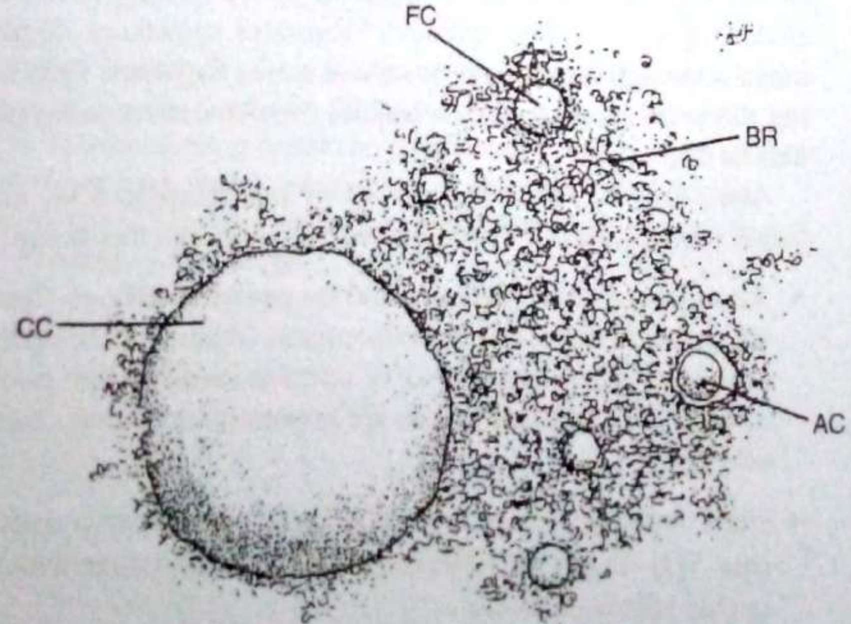

Figure 6–2 Flagellate cysts (FC) of the genus *Cercomonas*. Amoebal cysts (AC) have double-walls. Ciliate cysts (CC) are usually much larger. Cysts of all protozoa are usually spheroid, and often enmeshed in a matrix of bacterial cells and organic matter (BR). *Drawing by Kim Luoma.*

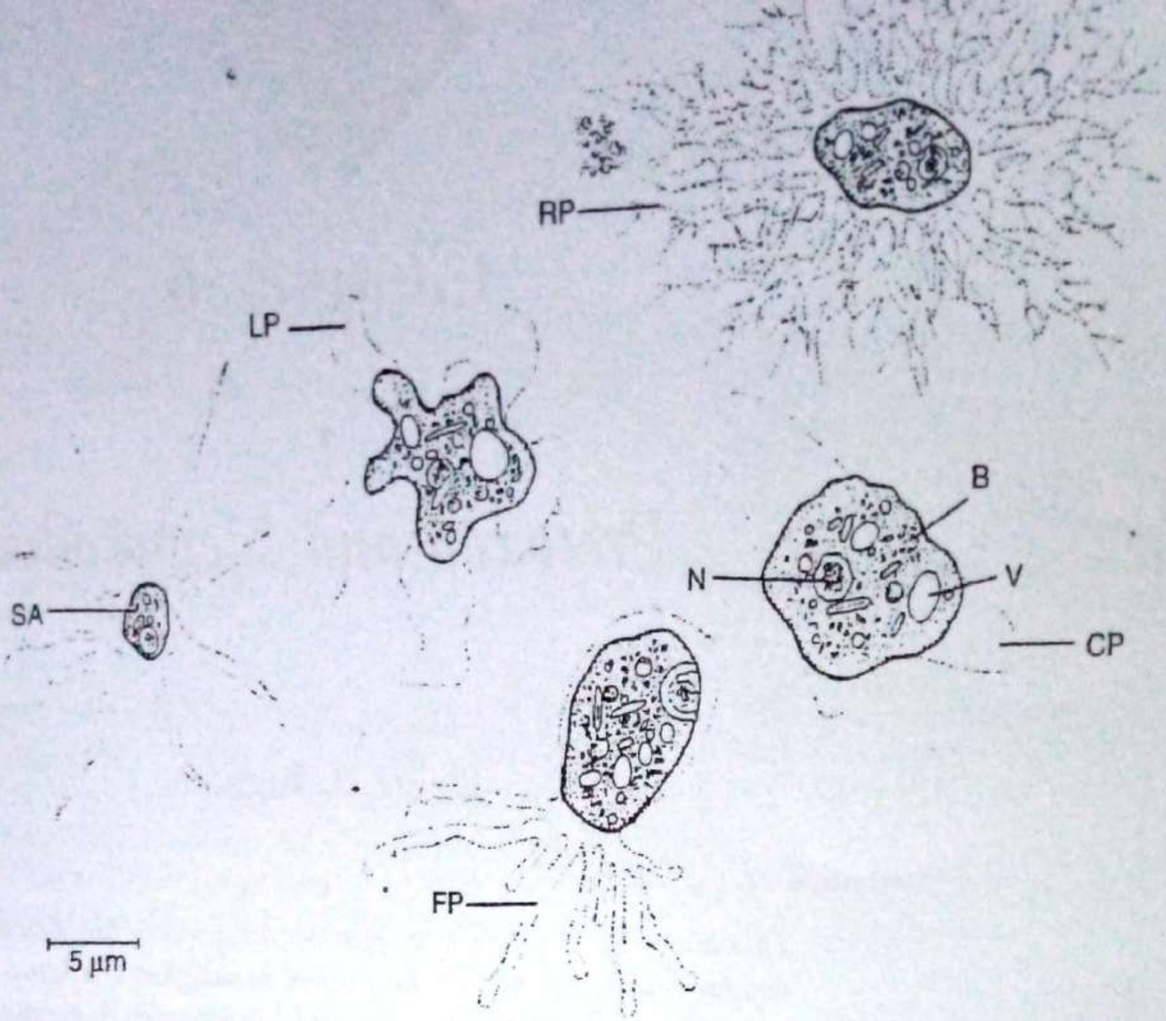

Figure 6–3 Soil amoebae form various types of pseudopods. The shape and the way the pseudopods are formed are important characters for identifying species. Lobose pseudopods (LP) are rounded and form by a smooth, streaming motion or by a more abrupt, "burping" motion. Conical pseudopods (CP) are pointed at the tip, and broader at the base. Filiose pseudopodia (FP) are slender, fingerlike, filamentous structures. Stellate amoebae (SA) form stiff, filamentous pseudopods and so look like floating stars. Reticular pseudopodia (RP) form weblike, fanciful structures. Pseudopods are used for locomotion, to pull or push the amoeba along the soil surface. Within the amoebal body, nuclei (N), vesicles (V), and ingested bacterial cells (B) can be seen. *Drawing by Kim Luoma.*

Ciliates. Ciliates move by beating short, numerous **cilia** (singular, **cilium**) on the surface of their bodies (Fig. 6–4). Distribution of the cilia, fusion of cilia into tufts called *cirri* or into membranelles (undulating membranes), and the placement of the cytostome (mouth) with associated membranelles are important features for species identification.

Sizes of protozoa. Flagellates, naked amoebae, and small testate amoebae normally are barely larger than their bacterial prey, while some forms are much larger (Table 6–1). Giant amoebae, which attack fungal hyphae, spores, or even sclerotia, are up to 1 mm in diameter. Ciliates are larger in size than flagellates, larger than most naked amoebae, and similar in size to large testate amoebae. The large forms are restricted to larger pore spaces and require thicker water films that occur at water potentials of 0 to −0.03 MPa to remain active, generally swimming free in the soil solution.

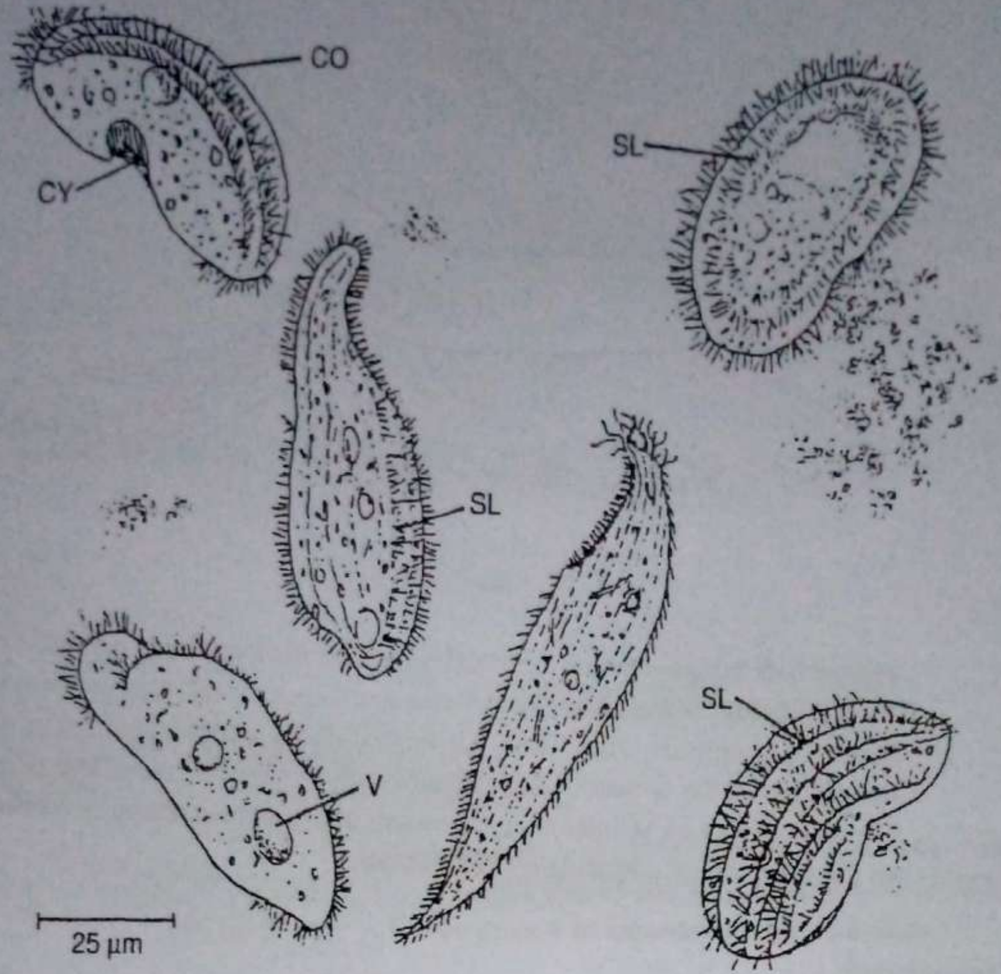

Figure 6–4 A group of soil ciliates. A typical genus is represented by *Colpoda* (CO), about 40 µm in length. The cytostoma (CY) or mouth cavity forms an indentation that makes the organism look like a kidney. Cilia around the cytostoma guide bacteria into the ciliate's mouth, but the major purpose for cilia is locomotion. One important characteristic for identification of ciliates is the pattern made by cilia attachment to the body surface, called "silver lines" (SL). Cilia can run the entire length of the body, part of the body, or occur in patches as shown in this drawing. Vesicles (V) for collecting waste products are also shown. *Drawing by Kim Luoma.*

Table 6–1 **Average length and volume of soil protozoa and nematodes as compared to bacteria.**

Group	Length (µm)	Volume (μm^3)	Shape
Bacteria	<1–5	2.5	Spherical to rod-shaped
Flagellates	2–50	50	Spherical, pear-shaped, banana-shaped
Amoebae			
Naked	2–600	400	Protoplasmic streaming, pseudopodia
Testate	45–200	1,000	Build oval tests or shells made of soil
Giant	6,000	4×10^9	Enormous naked amoebae
Ciliates	50–1,500	3,000	Oval, kidney-shaped, elongated and flattened
Nematodes	250–5,500	5,000	Long, slender, wormlike

Nematodes

Nematodes (Phylum Nematoda) are multicellular, eukaryotic, nonsegmented roundworms. They constitute as much as 90% of all multicellular animals in soil, and their numbers often exceed several million per m^2 of surface soil. Although the total biomass of nematodes may be less than that for other faunal groups (see Table 1–2), nematode metabolic activity is often higher.

Most nematode species are fusiform (tapered ends), while others are vermiform (rounded ends). Nematode digestive, nervous, excretory, and reproductive organs float in a fluid-filled cavity surrounded by a body wall. Nematodes lack circulatory, respiratory, and endocrine systems. The body wall consists of a cuticle, a hypodermis which produces the cuticle, and a layer of muscle. Placement of body openings, such as the stoma (mouth), anus, vulva, excretory openings, and sensory openings, and the structure of these openings are important in identification of nematode species. Also important are cuticular markings, such as striations, ridges, and apparent segmentation. In addition, the position of sensory papillae (small elevations), setae (hair-like projections), and spines help to identify species. The shape and placement of chemoreceptors (called *amphids*) are important in identification of certain species. Prolobae (ornate lip structures) occur in a number of bacterial-feeding species.

Nematodes are grouped into four or five trophic categories based on the nature of their diet, the structure of the stoma and esophagus, and their method of feeding.

- Bacterial-feeding nematodes do not have **stylets** (hard, spear-like structures in the mouth region used to puncture cell walls) but have a simple stoma in the form of a cylinder or cone, terminating in a valvelike apparatus which may bear minute teeth (Fig. 6–5).
- Fungal-feeding nematodes have slender stylets or small teeth used to puncture cell walls; they attack fungi instead of plant roots (Fig. 6–6). After puncturing the hyphal cell wall, the muscular esophagus provides the suction necessary to empty the cytoplasm from the fungus. The morphology of the esophagus in different species varies considerably, from a long, straight esophagus in some genera to those with large cuticularized pumping valves halfway between the mouth and the intestine.
- Root-feeding nematodes usually possess large stylets (Fig. 6–7) and are the most extensively studied group of soil nematodes because of their ability to cause plant disease and reduce crop yield.
- Predatory nematodes possess huge mouths (stoma) armed with powerful teeth (Fig. 6–8).
- Omnivores are sometimes considered as a fifth trophic category of soil nematodes because they may eat many types of food, including roots, fungi, bacteria, algae, and protozoa.

Feeding Behavior

Most soil protozoa are **phagotrophic,** engulfing bacteria, yeasts, and algae. Large protozoa, such as ciliates, may also engulf small protozoa. Some flagellates are