

Lesson 6:

Protozoa

Protozoa

Protozoans are found in all moist habitats within the United States, but we know little about their specific geographic distribution. They are unicellular organisms that range in size from a few microns to several hundred microns. Polluted waters often have a rich and characteristic protozoan fauna. The relative abundance and diversity of protozoa are used as indicators of organic and toxic pollution.

Although protozoa are frequently overlooked, they play an important role in many communities where they occupy a range of trophic levels. As predators upon unicellular or filamentous alge, bacteria, and microfungi, protozoa play a role both as herbivores and as consumers in the decomposer link of the food chain.

Both in organic pollution of the natural environment and in the biological processing of human and domestic animal sewage, the ceaseless activity of the protozoa, particularly the colonial ciliates, in the extraction and digestion of bacteria and other suspended particles is the main element of the natural process by which the water supply is rendered once again fit for consumption by humans and other creatures. Any change in our environment which threatens the life of a balanced community of protozoans threatens also the continuity of a clean water supply for humans. This is particularly relevant in the light of our current over-use of kitchen and lavatory disinfectants and their effect upon the ciliates at sewage processing plants and in the waterways beyond.

Protozoa are members of the Kingdom Protista. There are about 20,000 known species of protozoa that live in water and soil. Some feed on bacteria while others are parasites and feed off their hosts. Most protozoa are asexual and reproduce in one of three ways: fission, budding, and multiple fission. Some protists are sexual and exchange genetic material from one cell to another through conjugation, which is the physical contact between cells.



A protist can survive in an adverse environment by encapsulating itself with a protective coating called a **cyst**. The cyst defends the protist in extreme temperatures against toxic chemicals and even when there is a lack of oxygen, moisture, and food.

Factors Affecting Growth and Distribution

Most free-living protozoa reproduce by cell division (exchange of genetic material is a separate process and is not involved in reproduction in protozoa). The relative importance for population growth of biotic versus chemical-physical components of the environment is difficult to ascertain from the existing survey data. Protozoa are found living actively in nutrient-poor to organically rich waters and in fresh water varying between 0°C (32°F) and 50°C (122°F). Nonetheless, it appears that rates of population growth increase when food is not constrained and temperature is increased.

Nutrition

Protists receive nutrients by breaking down organic matter and can grow in both aerobic and anaerobic environments, such as protists that live in the intestine of animals. Some receive nutrients from organic matter and photosynthesis because they contain chlorophyll. These protists are considered both algae and protozoa. Protists obtain food in one of three ways: absorption, ingestion, and engulfing. Food is digested in the vacuole after the food enters the cell. The vacuole is a membrane-bound organelle. Waste products are excreted using a process called **exocytosis**.

Protozoa in the Wastewater Treatment Process

Most protozoa are free-living in soil and water and enter the activated sludge process through inflow and infiltration. The number of protozoa within the activated sludge process or mixed liquor varies greatly by process and operational conditions, especially hydraulic and organic loadings. The relative abundance of protozoa may be less than 100 per milliliter or greater than 100,000 per milliliter.

Protozoa, especially ciliated protozoa, perform several beneficial roles in wastewater treatment. These roles include cropping action, coating action, and recycling of mineral nutrients.

Bacteria are the primary food source for protozoa, and the consumption of suspended or dispersed bacteria by protozoa is referred to as "cropping" action. Cropping action removes many dispersed bacteria from the bulk solution.

Dispersed growth as well as colloids and particulate materials, collectively known as "fine" solids, also are removed from the bulk solution by the "coating" action of ciliated protozoa. This group releases sticky secretions that cover the surface of fine solids. Through coating action, the surface charge of fine solids is made compactible for adsorption to floc particles in the activated sludge process. The adsorption reduces the quantity of fine solids in the final effluent.

Protozoa also release excretions to the bulk solution. These excretions contain many mineral nutrients, including nitrogen and phosphorus, and help to recycle mineral nutrients in the activated sludge process. These nutrients then are available for bacterial activity in degrading wastes, the biochemical oxygen demand (BOD).

Protozoa in the activated sludge process commonly are placed in one of five groups: amoebae, flagellates, free-swimming ciliates, crawling ciliates and stalked ciliates. Amoebae and flagellates are considered "lower" life forms, while crawling ciliates and stalk ciliates are considered "higher" life forms. Free-swimming ciliates are considered "intermediate" life forms. Some treatment plant operators often perform routine microscopic examinations of the protozoa in the activated sludge to determine the health of the activated sludge process.

Typically, operators base the health of the process on the protozoan groups that are dominant as revealed by microscopic examination (Table below). For example, if lower life forms are dominant, the activated sludge is considered unhealthy and unacceptable. An unhealthy activated sludge produces an aeration tank effluent having a BOD greater than 30 mg/L. If the higher life forms are dominant, the activated sludge is considered healthy and acceptable. A healthy activated sludge produces an aeration tank effluent having a BOD less than 30 mg/L.

Protozoan Groups and Bioindicator Values	
Amoebae	Rarely predominant except for start-up conditions and conditions that mimic start-up such as over-wasting, recovery from toxicity, washout, and organic overloading
Flagellates,	Dominant under high organic loading, dispersion of floc particles, such

plant-like	as through chlorination, and start-up conditions or conditions that mimic start-up. Also may dominate in the presence of excess soluble phosphorus.
Flagellates, animal-like	Except for the presence of excess soluble phosphorus, these are dominant for operational conditions listed for plant-like flagellates and usually follow plant-like flagellates as the dominant group.
Free-swimming ciliates	Transition group that dominates between healthy and unhealthy conditions and proliferates when large numbers of free-swimming bacteria are present.
Crawling ciliates	Dominant in the presence of mature floc particles and low BOD in the bulk solution. Alternate with stalked ciliates as the dominant group.
Stalked ciliates	Dominant in the presence of mature floc particles and low BOD in the bulk solution. Alternate with crawling ciliates as the dominant group.

Operators should exercise caution when correlating final effluent quality with the health of activated sludge as indicated by the dominant protozoan groups. For example, loss of secondary solids from the clarifier due to a bulking condition would increase the BOD above that achieved in the aeration tank.

Diseases Caused by Protozoa

Several protozoa can cause waterborne diseases. The most common of these pathogenic protozoa are Cryptosporidium, Entamoeba, Giardia, and Toxoplasma. These species are all able to form cysts, or protective coatings, which allow them to survive outside a host for extended periods of time. Protozoan cysts also protect the organism from chlorine, so these species are not effectively controlled by chlorination. Instead, protozoa are usually removed from water by filtration.

Cryptosporidium

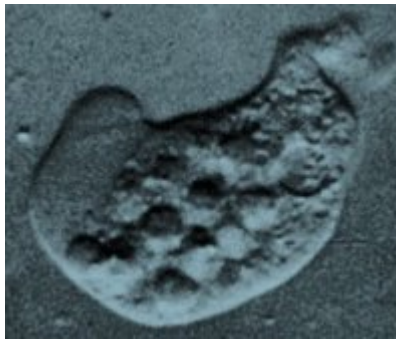


Cryptosporidium is a sporozoa which is one of the most common pathogens responsible for waterborne diseases in the United States. Scientists estimate that between 1% and

5% of Americans are infected with *Cryptosporidium* at any one time. *Cryptosporidiosis*, the disease caused by *Cryptosporidium* infection, results in diarrhea, abdominal cramps, and fever. Healthy adults usually do not require treatment for *cryptosporidiosis* since the body will heal itself naturally within a couple of weeks.

Cryptosporidium, like most other waterborne pathogens, is spread to new hosts through the ingestion of contaminated food and drinking water. In addition, several outbreaks have been found to result from fecal accidents in swimming pools. Water treatment plants find it difficult to prevent the spread of *Cryptosporidium* since the cysts are very resistant to chlorination and can sometimes pass through filters. Instead, the cysts can be killed by ozonation or by boiling the water for at least a minute.

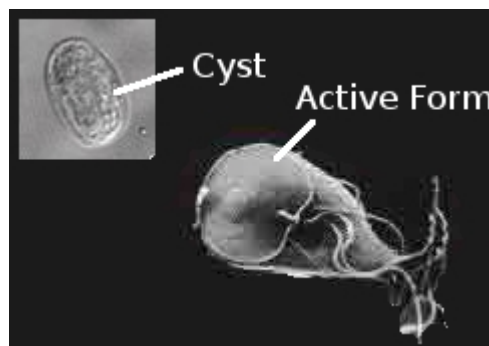
Entamoeba histolytica



Entamoeba histolytica is an amoeboid protozoan which lives in anaerobic environments. Like the other pathogenic protozoa, *Entamoeba* is capable of forming cysts which can remain dormant for extended periods of time in the water, in soil, or in food. These cysts spread to new hosts when we ingest contaminated food or water.

In humans, *Entamoeba* causes amoebic dysentery, which is usually treated with antibiotics. *Entamoeba* outbreaks are rare in the United States but are quite common in developing countries when raw sewage contaminates drinking water supplies or when the soil is fertilized with untreated wastes. In the water treatment plant, *Entamoeba* can be removed from water using a sand filter.

Giardia lamblia



Giardia lamblia is a flagellate protozoan which is capable of forming cysts. The species is the most important cause of waterborne disease outbreaks in the United States.

Infection with *Giardia lamblia* results in a disease called giardiasis, which is also known as traveler's diarrhea or Montezuma's Revenge. Symptoms of giardiasis include diarrhea, abdominal cramps, fatigue, and weight loss. Although most people who contract giardiasis heal naturally within a week or two, the illness sometimes lingers for up to a year, in which case antibiotics may be prescribed.

Giardia lamblia is spread when people ingest contaminated food or water. In the water treatment plant, chlorine is somewhat effective at inactivating the cysts at a dosage of 1.5 mg/L chlorine with a contact time of 10 minutes. Filtration and boiling are more effective at killing *Giardia*.

Toxoplasma gondii

Toxoplasma gondii is a sporozoa which causes a disease known as toxoplasmosis. In most cases, toxoplasmosis has mild flu-like symptoms which are often unnoticed or undiagnosed and do not require treatment. However, a pregnant woman may pass on the infection to her unborn child, in which case the child will have much more serious symptoms.

Toxoplasma gondii has a complex life cycle involving at least two different hosts. The protozoan reproduces sexually within its primary host, the cat, and releases cysts in the cat's feces. The cysts are then ingested by intermediate hosts, which include a large number of vertebrate species such as pigs, cows, and humans. In the intermediate host, the cysts develop into an active form of the protozoan which reproduces asexually. The offspring produced in the intermediate host then infect a cat, completing the cycle.

In most cases, humans contract toxoplasmosis by eating contaminated meat or by handling cats or changing cats' litter boxes. However, a recent outbreak of toxoplasmosis linked to drinking water in Canada has alerted us to the possibility of *Toxoplasma gondii* being spread in water.

Part 3: Amoebae