

# LAB 20: PARASITOLOGY

## A. Parasitic Protozoa

**Protozoans** are unicellular **eukaryotic** microorganisms belonging to the Kingdom Protista. They **reproduce asexually** by **fission** (one cell splits into two), **schizogony** (multiple fission; the nucleus divides many times and the nuclei are separated into daughter cells), or **budding** (pinching off of a bud from a parent cell). Some protozoans also **reproduce sexually** by fusion of haploid sex cells called gametes.

The vegetative form (motile, feeding, reproducing form) of a protozoan is called a **trophozoite**. Under certain conditions, some protozoans produce a protective form called a **cyst** that enables them to survive harsh environments. Cysts allow some pathogens to survive outside their host. Favorable conditions in a new host result in excystation that once again produces a trophozoite.

The parasitic protozoans can be divided into groups based primarily on their means of locomotion.

### 1. The Sarcomastigophora (Amoeboflagellates)

a. The **amoebas** (subphylum Sarcodina) move by extending lobelike projections of their cytoplasm called **pseudopodia**. Food is obtained by phagocytosis.

- [Photomicrograph of an amoeba.](#)

[YouTube movie of an amoeba feeding.](#)

[YouTube movie an amoeba moving by forming pseudopodia.](#)

An important pathogen in this group is ***Entamoeba histolytica***, the causative agent of **amoebic dysentery**. The organism is **transmitted** by the **fecal-oral route**. Cysts are excreted in the feces of an infected individual or carrier and ingested through fecally-contaminated food, water, objects, etc. After excystation, the trophozoites penetrate the walls of the large intestines causing ulceration and frequently causing the symptoms of dysentery. Involvement of the liver and other organs may occur if the protozoan invades the blood. The disease is **diagnosed by microscopically looking for cysts of *E. histolytica* in a fecal smear** ([see Fig. 1](#)).

*Acanthamoeba* can infect the **eye, blood, spinal cord, and brain** and is transmitted by waterborne cysts picked up while swimming in contaminated water, crossing the mucous membranes.

[YouTube movie of Acanthamoeba](#)

b. The **flagellates** (subphylum Mastigophora) move by means of **flagella**. Some also have an **undulating membrane**.

1. *T. gambiense* and *T. rhodesiense* cause the disease **African sleeping sickness** or **African trypanosomiasis**. They are **transmitted** to humans by the bite of an **infected tsetse fly** (a vector). The disease primarily involves the lymphatic and nervous systems of humans and is **diagnosed by microscopically looking for *Trypanosoma* in the blood** ([see Fig. 2](#)), **in aspirated fluid from lymph nodes, or in spinal fluid**.

[YouTube movie of Trypanosoma](#)

*T. cruzi* causes **South American sleeping sickness** or Chagas' disease and is **transmitted** by infected **Triatomid bugs (kissing bugs)**.

### 2. *Giardia lamblia*

*Giardia lamblia* (*G. intestinalis*) causes a gastroenteritis-type of disease called **giardiasis**. Giardiasis is the most common protozoan intestinal disease in the U.S. and is **transmitted** by the **fecal-oral route**. Cysts of the organism are ingested through fecally-contaminated food, water, etc. Giardiasis is **diagnosed by microscopically looking for cysts of *G. lamblia* in fecal smears** ([see Fig. 3](#)).

- Scanning electron micrograph of [Giardia](#); licensed for use, [ASM MicrobeLibrary](#).
- Scanning electron micrograph of [Giardia](#); courtesy of CDC.

[YouTube animation illustrating giardiasis](#)

### 3. *Trichomonas vaginalis*

This protozoan causes **genitourinary trichomoniasis**. There are an estimated 2.5 million cases per year in the U.S. In females, it usually appears as **vaginitis** with itching and a white discharge. In males it is often asymptomatic but may cause urethritis. It is **transmitted** mainly by **venereal contact** and is **diagnosed** by **microscopically looking for *T. vaginalis* trophozoites in vaginal discharge and urine** ([see Fig. 4](#)).

[YouTube movie \*Trichomonas vaginalis\*.](#)

[YouTube movie showing motility of \*Trichomonas vaginalis\*.](#)

## 2. The Ciliophora

This group of protozoans is characterized by a covering of **cilia** used for motility and direction of food particles into the mouth or cytosome. In the trophozoite, a large macronucleus, small micronucleus, cilia, and contractile vacuoles may be seen.

The only pathogen in this group is ***Balantidium coli***, which causes a diarrhea-type infection called **balantidiasis**. The protozoan is **transmitted** to humans by the **fecal-oral route** and invades the large intestines causing ulceration. It is **diagnosed by microscopically looking for *B. coli* in a fecal smear** ([Fig. 5](#)).

[YouTube movie showing motility of \*Balantidium coli\*.](#)

## 3. The Apicomplexans

The sporozoa are **not motile** in their mature forms, reproduce both asexually and sexually, and often have complex life cycles for transmission from host to host. They possess a complex of organelles at their apex (apical complexes) that contain enzymes used in penetrating host tissues.

### a. *Toxoplasma gondii*

This protozoan causes the disease **toxoplasmosis**. In adults, the disease is usually mild and resembles infectious mononucleosis. However, new-born infants who contracted toxoplasmosis *in utero* commonly have severe central nervous system damage. It also causes severe disease in immunosuppressed individuals such as people with AIDS. Domestic cats, that pick up the organism from eating infected rodents, may act as carriers of *T. gondii*, and their feces may contain oocysts of the protozoan. However, the organism may be found in practically every mammal. The disease is **transmitted** to humans by **ingesting raw meat of an infected mammal or by inhaling or ingesting cysts of *T. gondii* from cat feces**. Pregnant women should be especially careful to avoid raw meat and cat feces. The disease is **diagnosed** by **serologic testing and by growing the organism in cell culture**.

[YouTube video: \*Toxoplasma gondii\*](#)

### b. *Plasmodium*

Four *Plasmodium* species, *P. falciparum*, *P. malariae*, *P. ovale*, and *P. vivax* cause **malaria**. The vector involved in the **transmission** of the disease from human to human or from animal to human is an **infected female *Anopheles* mosquito**.

**Asexual reproduction** (or **schizogony**) of the *Plasmodium* occurs within liver cells and red blood cells of the **infected human**. With malaria caused by *P. vivax* and *P. ovale*, a dormant form or **hypnozoite** remains in the liver and may cause later relapses. The infected cells in which the organism is reproducing by schizogony are called **schizonts**. The **sexual cycle** (or **sporogony**) occurs in the **mosquito** (see [Fig. 6](#), the life cycle of *Plasmodium*). The typical **recurring malarial fever** is a result of the lysis of the infected red blood cells, causing release of **merozoites** and their metabolic by-products. Fever cycles of 24, 48, or 72 hours usually occur depending on the infecting species. Malaria is **diagnosed by microscopically looking for the parasite within infected red blood cells (schizonts)**. See [Fig. 7A](#) and [Fig 7B](#).

### c. *Cryptosporidium*

*Cryptosporidium* is an intracellular parasite that causes **diarrhea**, although in people who are immunosuppressed it can also cause respiratory and gallbladder infections. It is **transmitted by the fecal-oral route**.

[Medscape](#) articles on infections associated with organisms mentioned in this Lab Exercise. Registration to access this website is free.

- [Amebiasis](#)
- [Acanthamoeba](#)
- [African sleeping sickness](#)
- [giardiasis](#)
- [Trichomoniasis](#)
- [Balantidiasis](#)
- [Toxoplasmosis](#)
- [Malaria](#)

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## B. PARASITIC HELMINTHS

Helminthology is the study of worms, or **helminths**. Over one billion people worldwide are infected with intestinal helminths alone. Helminths are multicellular, often macroscopic worms having both rudimentary organs and organ systems. We will look at three groups of pathogenic helminths: **nematodes, cestodes, and trematodes**.

### 1. The Nematodes (Roundworms)

Nematodes are **elongated, unsegmented, cylindrical worms having separate sexes**. The various systems of the roundworms can be seen in [Fig. 8](#), and a generalized drawing of a nematode is shown in [Fig. 9](#). We will look at several pathogenic nematodes.

#### a. *Ascaris lumbricoides*

These worms range from 20-45 cm long and are 5 mm in diameter in the adult form, the female being larger than the male. The *Ascaris* life cycle is seen in [Fig. 10](#). The disease is called **ascariasis**. This is the most common helminth in humans worldwide. In the U.S., it infects over 4,000,000 people. Humans become **infected by ingesting water or food contaminated with feces that contains *Ascaris* ova or from fingers contaminated with polluted soil**. The disease is **diagnosed by microscopically looking for *Ascaris* ova in a fecal smear** ([see Fig. 11](#)). A similar roundworm, *Toxocara*, parasitizes dogs and cats. Visceral larva migrans is the migration of larvae of these worms in human tissues such as lung, liver, and brain, where they may cause tissue damage and allergic reactions.

[YouTube video: Colonoscopy showing \*Ascaris\*](#)

[YouTube video: Acute Intestinal Obstruction by Roundworms](#)

[YouTube video: Removing an Acute Intestinal Obstruction by Roundworms](#)

#### b. *Enterobius vermicularis* (pinworms)

*E. vermicularis* is a small worm, the female being 8-13 mm long and 0.3-0.5 mm wide; the male being 2-5 mm long and 0.1 mm wide. The life cycle of *Enterobius* is shown in [Fig. 12](#). Pinworms are the most common helminth infection in the U.S. with as many as 50,000,000 people infected. Humans, frequently children, become **infected by inhaling *E. vermicularis* ova or from transfer of ova to the mouth from fecally-contaminated fingers**. The female worm migrates to the perianal region of the infected individual, releasing masses of ova and causing an itching sensation. Each pregnant worm produces between 4600 and 16,000 eggs ([see Fig. 12A](#)). The disease is **diagnosed** by applying tape to the perianal region and **microscopically looking for pinworm ova that have stuck to the tape** ([see Fig. 13](#)).

[YouTube video: Pinworm](#)

[YouTube video: \*Enterobius vermicularis\*](#)

### c. *Trichinella spiralis*

*T. spiralis* causes a disease called **trichinosis**. Humans become **infected** mainly by **eating poorly cooked infected pork** containing the encysted larva (1-2 mm long). The larvae excyst and develop into adult worms in the intestines. After mating, the female releases larvae which enter the blood and are distributed throughout the body where they become encysted in muscle tissue. The life cycle of *Trichinella spiralis* is seen in [Fig. 14](#). The disease is **diagnosed by serological tests and microscopic examination of biopsy specimens** ([see Fig. 15](#)).

[YouTube video: \*Trichinella spiralis\*](#)

## 2. The Cestodes (Tapeworms)

Tapeworms are **flat segmented worms which are hermaphroditic** (contain both male and female sexual organs). Adult tapeworms have several distinct regions ([see Fig. 16](#)). The **scolex** ([see Fig. 17](#)) is a head-like structure with distinct suckers and possibly hooks used for attachment to the intestinal wall. Behind the scolex is a constricted neck region consisting of germinative tissue from which new segments, or **proglottids**, are formed. Finally, there is a long **strobila** or chain of proglottids of varying stages of maturity. Proglottids containing a uterus and thousands of ova are excreted in the feces ([see Fig. 18](#)). When ingested by intermediate hosts (such as cattle, pigs, and fish), the larva hatch from the ingested ova and migrate to muscle where they encyst as **cysticerci**. The life cycle of the pork tapeworm *Taenia solium* is shown in [Fig. 19](#).

Humans become **infected** with tapeworms by **eating poorly cooked infected beef, pork, or fish containing cysticerci**. *Taenia saginata*, the beef tapeworm often reaches 6 meters in length; *Taenia solium*, the pork tapeworm is normally 2-7 meters in length; and *Diphyllobothrium latum*, the fish tapeworm may reach 3-6 meters in length. These tapeworms are **diagnosed** by looking for **proglottids and ova in the feces**.

When humans ingest tapeworm eggs instead of cysts, embryos are released, penetrate the intestinal wall, and enter the blood. The embryos migrate to various tissues (frequently the brain) and develop into cysticerci. Humans also act as intermediate hosts for *Echinococcus granulosus* found in dogs and cats. Larva hatch from ingested ova and migrate to the liver and lungs and form hydatid cysts.

[YouTube video: Colonoscopy showing a Tapeworm](#)

[YouTube video: Colonoscopy showing \*Diphyllobothrium latum\* \(fish tapeworm\)](#)

[YouTube video: \*Taenia solium\* \(pork tapeworm\)](#)

[YouTube video: Trained tapeworm segment](#)

## 3. The Trematodes (Flukes)

Flukes are **unsegmented, flat, leaf-shaped worms** having a variety of organ systems ([see Fig. 20](#)). Most flukes are **hermaphroditic**. They attach to the host by means of an oral sucker and a ventral sucker. Flukes, as adults, may infect either the portal blood vessels, intestines, liver, or lungs of humans and are named according to the tissue they infect. Humans become **infected** with liver flukes, lung flukes, and intestinal flukes by **ingesting poorly cooked fish, crayfish, crabs, snails, or water vegetables infested with flukes**. Blood flukes **directly penetrate the skin**.

In the life cycle of flukes ([see Fig. 21](#)), ova leave the body of the infected human or animal by means of feces, urine, or sputum (depending on the type of fluke). The ova enter water and infect the first intermediate host, certain species of **water snails**. **A free swimming form of the fluke called the cercaria, then leaves the snail and infects second intermediate hosts (fish, crayfish, water vegetables, etc.) which are ingested by humans**. The cercariae of the **blood fluke *Schistosoma*** ([see Fig. 22](#)) **can directly penetrate the skin** of humans and cause **schistosomiasis**, a major problem in Africa, South America, and Asia.

[Medscape](#) articles on infections associated with organisms mentioned in this Lab Exercise. Registration to access this website is free.

- [Ascariasis](#)
- [Pinworms](#)
- [Trichinosis](#)
- [Tapeworms](#)
- [Flukes](#)

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## PROCEDURE AND RESULTS

1. Observe the prepared slides of the following **parasitic protozoans** and **compare them with the indicated figures in this lab exercise.**

### a. The Sarcodina

Fecal smear containing [cysts of \*Entamoeba histolytica\*](#) (the cause of amoebic dysentery). **Note** that it contains several nuclei.

### b. The Mastigophora

1. [Blood smear containing \*Trypanosoma gambiense\*](#) (the cause of African sleeping sickness).

**Note** the nucleus, the undulating membrane, and the red blood cells in the background.

2. [Fecal smear containing cysts and/or trophozoites of \*Giardia lamblia\*](#) (the cause of giardiasis).

**Note** the bilateral symmetry and macronuclei of the organism which look like "eyes."

3. [Vaginal discharge containing \*Trichomonas vaginalis\*](#) (the cause of genitourinary trichomoniasis).

**Note** the bundle of flagella, the undulating membrane, and the nucleus.

### c. The Ciliophora

[Fecal smear containing \*Balantidium coli\*](#) (the cause of balantidiasis). **Note** the large dumbbell-shaped macronucleus.

### d. The Sporozoa

1. [Sporozoites of \*Plasmodium\*](#) from the salivary glands of an infected mosquito.

2. [Blood smear containing red blood cells infected with merozoites of \*Plasmodium\*](#) (the cause of malaria).

2. Observe the prepared slides of the following **parasitic helminths** and **compare them with the indicated figures in this lab exercise.**

### a. The Nematodes (roundworms)

1. [Fecal smear containing ova of \*Ascaris lumbricoides\*](#) (the cause of ascariasis). **Note** the "bumpy" edge of the ova. [The life cycle is shown in Fig. 10.](#)

2. [Ascaris lumbricoides larva.](#) **Note** the [organ systems.](#)

3. [Fecal smear containing ova of \*Enterobius vermicularis\* \(pinworm\).](#) **Note** the "smooth" edge of the ova. [The life cycle is shown in Fig. 12.](#)

4. [Enterobius vermicularis larva.](#) **Note** the [organ systems.](#)

5. Muscle tissue containing [encysted larvae of \*Trichinella spiralis\*](#) (the cause of trichinosis). **Note** the spiral-shaped larva within the cyst. [The life cycle is shown in Fig. 14.](#)

### b. The Cestodes (tapeworms)

1. [Scolex of \*Taenia pisiformis\*](#) (dog tapeworm). **Note** hooks and suckers.

2. [Proglottids of the tapeworm \*Taenia pisiformis\*.](#)

3. [Gravid proglottid of \*Taenia pisiformis\*.](#) **Note** the uterus and ova. [Also see Fig. 19.](#)

### c. The Trematodes (flukes)

1. *Fasciola hepatica* (liver fluke). **Note** the organ systems. [See Fig. 20.](#)
2. *Schistosoma mansoni* (blood fluke). **Note** the oral and ventral suckers. [See Fig. 22.](#)

3. Observe the **preserved helminths**.

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## PERFORMANCE OBJECTIVES FOR LAB 20

After completing this lab, the student will be able to perform the following objectives:

### A. PARASITIC PROTOZOANS

#### DISCUSSION

1. Define the following: protozoan, trophozoite, cyst.
2. State how the following diseases may be transmitted to humans and briefly discuss how the diseases are diagnosed in the clinical laboratory:
  - a. amoebic dysentery
  - b. African sleeping sickness
  - c. giardiasis
  - d. genitourinary trichomoniasis
  - e. balantidiasis
  - f. malaria
  - g. toxoplasmosis
3. Describe the following in terms of the malarial life cycle: sporozoite, schizont, asexual cycle, sexual cycle.
4. State what causes the recurring fever of malaria.

#### RESULTS

1. Recognize the following organisms when seen through a microscope and state what disease they are associated with:
  - a. *Entamoeba histolytica* cysts in a fecal smear
  - b. *Trypanosoma gambiense* in a blood smear
  - c. *Giardia lamblia* cysts in a fecal smear
  - d. *Trichomonas vaginalis* in vaginal discharge
  - e. *Balantidium coli* in a fecal smear
  - f. *Plasmodium* merozoites in infected red blood cells

### B. PARASITIC HELMINTHS

#### DISCUSSION

1. Define the following: helminth, ova, hermaphroditic.
2. List the three classes of parasitic helminths and state the common name for each class.
3. State how the following diseases may be transmitted to humans and state how each disease may be diagnosed in the clinical laboratory:
  - a. ascariasis
  - b. pinworms
  - c. trichinosis

d. tapeworms

e. flukes

4. Compare protozoans and helminths in terms of their size and structural complexity.

## RESULTS

1. Recognize the following organisms or structures when seen through a microscope:

a. roundworms

b. *Ascaris* ova

c. pinworm ova

d. *Trichinella* in muscle tissue

e. tapeworms

f. scolex of a tapeworm

g. ova and uterus in a gravid proglottid of a tapeworm

h. flukes

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## SELF-QUIZ

[Self-quiz](#)

[Answers](#)

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