Intestinal and urogenital Protozoa

Lecture 2
Medical Parasitology Course (MLAB 362)

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- 65,000 species
- Heterotrophic
- Eukaryotic
- Most are unicellular, colonies are rare
- Most have locomotive structures
  - flagella, cilia, or pseudopods
- Vary in shape
- Typically inhabit water or soil
Protozoa

- **Trophozoite**
  - Motile feeding stage
- **Cyst**
  - A dormant resistant stage
- **Asexual and sexual reproduction**
  - Most propagate by simple asexual cell division of the trophozoite
  - Many undergo formation of a cyst
  - Others have a complex life cycle that includes asexual & sexual phases
- **Majority are NOT pathogens**
- **Some are animal parasites & can be spread by insect vectors**

Zoonosis

- An infectious disease in animals that can be transmitted to people.
- The natural reservoir for the infectious agent is an animal.
Reproduction in protozoa

Asexual reproduction:
- Replication of chromosomes and the splitting of the parent into two or more parts by:
  - Binary fission or Multiple fission or Budding
  - Protozoans are problematic in their associations as colonial forms
  - The commonest form of reproduction is binary fission in which two essentially identical individuals result.
  - In some ciliates budding occurs in which a smaller progeny cell is budded off which later grows to adult size.

Sexual reproduction:
- All protozoa reproduce asexually, but sex is widespread in the protozoa too.
- In ciliates such as Paramecium a type of sexual reproduction called conjugation takes place in which two Paramecia join together and exchange genetic material

Movement in Protozoa

- Protozoa move mainly using cilia or flagella and by using pseudopodia
- Cilia also used for feeding in many small metazoans.

- No real morphological distinction between the two structures although cilia are usually shorter and more abundant and flagella fewer and longer.
- Each flagellum or cilium is composed of 9 pairs of longitudinal microtubules arranged in a circle around a central pair.
- The collection of tubules is referred to as the axoneme and it is covered with a membrane continuous with the rest of the organism’s cell membrane.
- Axoneme anchors where it inserts into the main body of the cell with a basal body.
### Taxonomic classification of protozoa

<table>
<thead>
<tr>
<th>Sub kingdom</th>
<th>Phylum</th>
<th>Sub-phylum</th>
<th>Genus-examples</th>
<th>Species-examples</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protozoa</td>
<td>Sarcomastig-ophora</td>
<td>further divided into Mastigophora move by flagella</td>
<td>Entamoeba</td>
<td>E. histolytica</td>
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<tr>
<td></td>
<td>Sarcodina—move by pseudopodia</td>
<td></td>
<td>Giardia</td>
<td>G. lamblia</td>
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<tr>
<td></td>
<td>Apicomplexa—no organelle of locomotion</td>
<td></td>
<td>Plasmodium</td>
<td>P. falciparum, P. vivax, P. malariae, P. ovale</td>
</tr>
<tr>
<td></td>
<td>Ciliophora—move by cilia</td>
<td></td>
<td>Balantidium</td>
<td>B. coli</td>
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<td></td>
<td>Microspora—Spore-forming</td>
<td></td>
<td>Enterocyto-zaa</td>
<td>E. bienus</td>
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</tbody>
</table>

### Transmission of Protozoa

- In intestinal and luminal protozoa, usually infective stages pass directly from host to host through food and water.
- In blood and tissue parasites, they pass either directly from host to another or indirectly via an arthropod host (alternation of generation).
Classification of Protozoan according to site of infection

- **Intestinal**
  - amebiasis  
    - *Entamoeba histolytica*
  - giardiasis  
    - *Giardia lamblia*
  - balantidiasis  
    - *Balantidium coli*
  - cryptosporidiosis  
    - *Cryptosporidium parvum*
- **Urogenital**  
  - trichomoniasis  
    - *Trichomonas vaginalis*
- **Blood and Tissue**  
  - malaria  
    - *Plasmodium spp*
  - meningoencephalitis  
    - *Naegleria fowleri*
  - toxoplasmosis  
    - *Toxoplasma gondii*
  - trypanosomiasis  
    - African Sleeping Sickness  
      - *Trypanosoma brucei*
    - Chagas Disease  
      - *Trypanosoma cruzi*
    - leishmaniasis  
      - visceral leishmaniasis( Kala-azar)  
        - *Leishmania donovanii*
      - cutaneous leishmaniasis  
        - *Leishmania topica/brazilensis*

### INTESTINAL PROTOZOA

**Flagellates:**
- *Giardia lamblia*
- *Dientamoeba fragilis*
- *Chilomastix mesnili*
- *Trichomonas hominis*
- *Enteromonas hominis*
- *Retortamonas intestinalis*

**Ciliated:**
- *Balantidium coli*

**Ameba:**
- *Entamoeba histolytica*
- *Entamoeba dispar*
- *Entamoeba coli*
- *Entamoeba hartmanni*
- *Endolimax nana*
- *Iodamoeba bütschlii*

**Apicomplexa:**
- *Cryptosporidium hominis*
- *Cryptosporidium parvum*
- *Cyclospora cayetanensis*
- *Isospora belli*
Amoeboid Protozoa

- Amoeba
  - Pseudopods

- *Entamoeba histolytica*
  - Incites dysentery, abdominal pain, fever, diarrhea & weight loss
  - Carried by 10% of world population
  - Asymptomatic in 90% of patients

**Entamoeba histolytica**

(a) Trophozoite

(b) Mature Cyst

(c) Excystment
Entamoeba histolytica

- Alternates between a large trophozoite, motile by means of pseudopods & a smaller nonmotile cyst
- Humans are the primary hosts
- Cysts are swallowed & enter the small intestine; alkaline pH & digestive juices stimulate cyst to release 4 trophozoites
- Trophozoites attach, multiply, actively move & feed
- Ameba may secrete enzymes that dissolve tissues & penetrate deeper layers of the mucosa
**Entamoeba histolytica**

- cosmopolitan distribution
- typical fecal-oral life cycle
- inhabits large intestine
- facultative virulent pathogen
- Cyst of *E. h.* survival at 2-5 weeks at room temperature.
- Metacystic amoeba in small intestine but Precystic amoeba in large intestine.

- *E. dispar* ~10-fold > *E. histolytica*
- *Entamoeba dispar* morphologically identical and non-pathogenic
Life cycle of *Entamoeba histolytica* and the clinical manifestations of infection in humans.

Life Cycle of *Entamoeba histolytica*

**Quadrinucleated cysts**
(Infected stages)

**Contamination of food & H₂O**

**Excystation in Small / large Intestine**
(alkaline pH & digestive juices stimulate cyst to release)

4 Metacyclic Forms (Amoebules)

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**Amoebules in Intestinal lumen**

**Binary Fission** ➔ 8 Amoebae ➔ Invade intestinal tissues (Trophozoites)

Asexual reproduction by binary fission
(Colonies of Amoebae: Increase in Population)

Precystic stage
Trophozoites Round up, Expel with food particles

Encystment & Cysts in lumen

Cysts in faeces ➔ Environmental contamination
(Soil/Ground)
Epidemiology of Amoebiosis

- **Ubiquitous**: Common in tropical & subtropical countries
- **Longevity of Cysts**
- **Tetranucleated cysts**: Infective stages
- Survive for two weeks in wet surrounding or Cyst of E. h. survival at 2-5 weeks at room temperature.
- No effect of water chlorination - viable for several weeks
- Very susceptible to desiccation
- Cysts are susceptible to heat (Thermal death point 40-55°C), freezing (below −5°C.), and drying
- Infection occurs when infective cysts are ingested in food or water that has been contaminated with human feces.
- Thus, this parasite is transmitted from human to human via **fecal contamination**

Epidemiology of Amoebiosis

- **Other Vital Factors**:
  - Flies
  - Vegetables
  - Food Handlers
  - **Faulty Plumbing** (Metropolitan Cities)

- Human Beings: Travelers disease
- Complete elimination is difficult & relapses are common
Prevention and Control of Amoebiosis

- Good Sanitation (sewage system).
- Avoidance of contamination of food & H₂O.
- Personal Hygiene (Hand washing after defaecation and before eating).
- Preventing faecal contamination of the environment by using latrines and protecting water supplies from faecal contamination.
- Covering food and water to prevent contamination from flies which can act as cyst carriers.
- Not eating green salads or other uncooked foods which may contain cysts, usually as a result of fertilization with untreated human faeces.
- Boiling drinking water (E. histolytica cysts are killed at 55 °C).
- Health education, particularly of food handlers, and also in schools and community health centres.

Pathogenesis of Amebiasis

- **NON-INVASIVE**
  - ameba colony on intestinal mucosa
  - asymptomatic cyst passer
  - non-dysenteric diarrhea, abdominal cramps, other GI symptoms

- **INVASIVE**
  - Invasion of tissues :
  - Secretion of proteolytic enzymes Phosphohydrolase
    - necrosis of mucosa → ulcers, dysentery
    - ulcer enlargement → dysentery, peritonitis
    - metastasis → extraintestinal amebiasis
  - Incites dysentery, abdominal pain, fever, diarrhea & weight loss to infected patient
Intestinal Pathogenesis of *E. histolytica*

- Invasion of Intestinal mucosa
  - Multiply asexually: Binary fission
  - Invasion in deeper layers (Submucosa)
    - Spread laterally
  - Flask shaped ulcers: Caecum & Ascending Colon

Exteraintestinal Pathogenesis of *E. histolytica*

- Invasion in intestinal mucosa/submucosa
  - Spread to other organs via circulation
  - Right lobe of liver via portal circulation
    - Trapped in interlobular venules
    - Lytic necrosis *Hepatic Amoebic Abscess*
    - Other organs: Lungs & Brain
Flask Shaped Ulcers in Colon

- flanked-shaped ulcer in large intestine (Colon)
- trophozoites at boundary of necrotic and healthy tissue
- trophozoites ingesting host cells
- dysentery (blood and mucus in feces)

Entamoeba histolytica pathology

I- INTESTINAL AMEBIASIS
(Amoebic dysentery)

II- EXTRA-INTESTINAL LESIONS occur in 3 ECTOPIC SITES
A. HEPATIC AMEBIASIS
B. PULMONARY AMEBIASIS
C. CEREBRAL AMEBIASIS (Amoebic brain abscess)
D. Cutaneous amoebiasis

Perianal ulcers with irregular borders and a necrotic yellowish dirty base with purulent exudates.
Clinical Signs of Amoebic Dysentery

Extra intestinal Phase

• **Liver**: Amoebic liver abscess, Pain on right side of abdomen, Enlarged tender liver Fever, Vomiting, Anorexia.

• **Lungs**: Chest Pain & Respiratory Distress.

• **CNS**: Epilepsy & Nervous signs.

Clinical Signs of Amoebic Dysentery

• **Chronic Amoebiosis** :
  • Recurrent GI symptoms.
  ➢ Diarrhea or Dysentery
  ➢ Abdominal Pain (Abdominal discomfort)
  ➢ Nausea
  ➢ Flatulence
  ➢ Anorexia
Amoebic dysentery

- Amoebic dysentery occurs when *E. histolytica* trophozoites invade the wall of the large intestine and multiply in the submucosa, forming large flask-shaped ulcers.
- The amoebae ingest red cells from damaged capillaries. Compared with bacillary dysentery, the onset of amoebic dysentery is less acute, lasts longer, and there is usually no significant fever.
- Without treatment dysenteric attacks may recur for several years.
- ameboma (=amebic granuloma)

- ameboma = inflammatory thickening of intestinal wall around the abscess (can be confused with tumor)

Diagnosis of Amoebiosis

- Intestinal Disease
- Stool Sample Examination
- Demonstration of cysts (tetranucleated).
- Differential diagnosis (1% Lugols Iodine)
- Trophozoites in loose stools.
- Immunodiagnosis: Invasive disease
Diagnosis of Amoebiosis

- Extraintestinal Disease:
  - Immunodiagnosis
  - Radiology
  - MRI
  - CT scan

Diagnosis of Extraintestinal Disease

- symptoms associated with specific organ
- history of dysentery
- hepatic
  - right upper quadrant pain
  - enlarged liver
- serology
- imaging (CT, MRI, ultrasound)
- abscess aspiration
  - only select cases
  - reddish brown liquid
  - trophozoites at abscess wall
**Entamoeba histolytica cysts**

- Cyst nuclei possess even peripheral chromatin and a central endosome (endosome position may be off-center in some stained specimens)

![Diagram of cysts](image)

- molecular probes used to survey for E. dispar and E. histolytica

**Cysts of E. histolytica/E. dispar**
- Round, measuring 10–15µm.
- Contain, 1, 2 or 4 nuclei with a central karyosome (special staining techniques are required to show details of nuclear structure).
- Chromatoid bodies (aggregations of ribosomes) can be seen particularly in immature cysts. They do not stain with iodine but can be stained with Burrow’s stain.
Entamoeba histolytica trophozoites
**Entamoeba histolytica cysts**

- Uninucleate cyst
- Binucleate cyst
- Quadrinucleate or mature cysts – diagnostic in feces

**Non-Pathogenic Amoeba**

Several species of protozoans may be mistaken for *E. histolytica*. Care must be taken to correctly identify the infection so that the correct treatment can be administered or other infectious agents sought after.
Commensal amoebae
(non pathogenic)

Entamoeba coli, E. hartmani, E. dispar, Endolimax nana, Iodamoeba butchlii, E. gingivalis.

They inhabit the large intestine and are transmitted by their cysts except E. gingivalis which inhabits the buccal cavity and has a direct method of transmission since it lacks the cystic stage.

Entamoeba dispar: it differ from E. histolytica in being non-invasive, living in the lumen of large intestine only. It must be separated by isoenzymatic, immunologic or molecular analyses.

Non-pathogenic Amoebae

Entamoeba coli life cycle stages

1. TROPHOZOITE - 20 to 30 μm in diameter
   - granular endoplasm is coarser than E. histolytica

   - lives in large intestine and feeds on bacteria and any other cells available to it; does not invade tissue
**Entamoeba coli** life cycle stages

2. CYST - encystment is similar to that of *E. histolytica*
   - immature cysts are rare in fecal smears
   - mature cyst is large, 10 to 33 μm, has 8 nuclei
   - chromatoidal bodies, if present, have splinter-like ends (disappear in most cysts)
   - cyst is released in the feces into the external environment
   - importance of human infection?

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**Entamoeba dispar**

- *E. dispar* was proposed as the name for non-pathogenic *E. histolytica*.
- Substantial biochemical and molecular data has been accumulated showing that the non-pathogenic isolates of *E. histolytica* are genetically distinct from the pathogenic isolates
- Identical to *E. histolytica* but does not invade tissue so is non-pathogenic.
<table>
<thead>
<tr>
<th><strong>Entamoeba dispar</strong></th>
<th>morphologically identical to <em>E. histolytica</em>. It must be separated by isoenzymatic, immunologic or molecular analyses.</th>
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<tbody>
<tr>
<td><strong>Entamoeba hartmanni</strong></td>
<td>some consider this a separate species. It differs from <em>E. histolytica</em> by being smaller in size.</td>
</tr>
<tr>
<td><strong>Entamoeba coli</strong></td>
<td>distinguished from <em>E. histolytica</em> by having an eccentric endosome, and mature cysts with 8 nuclei. If chromatoidal bodies are present, they have splintered ends, rather than rounded as in <em>E. histolytica</em>.</td>
</tr>
<tr>
<td><strong>Endolimax nana</strong></td>
<td>this is a very small amoeba (6-15um) with a large, eccentric endosome and thin nuclear envelope. Mature cysts contain 4 nuclei.</td>
</tr>
<tr>
<td><strong>Iodamoeba butschlii</strong></td>
<td>both the trophozoite and cyst have one nucleus with a large endosome. The cyst contains a large glycogen vacuole that stains darkly with iodine.</td>
</tr>
</tbody>
</table>

*E. coli* trophozoite: Note the eccentric endosome in the nucleus (centric in *E. histolytica*).

*E. coli* cyst: Note the number of nuclei - 5 clearly visible in this plane compared to only 4 in *E. histolytica*.

*Endolimax nana* trophozoite: Note the large endosome and thin layer of pigment around the nucleus.

*Iodamoeba butschlii* cyst: Note the large endosome in the nucleus and the large glycogen vacuole.
Flagellated Protozoa

- Motility by flagella alone or by both flagella & amoeboid motion
- Several parasites
  - *Giardia* (intestinal)
  - *Trichomonas*
  - *Trypanosoma*
  - *Leishmania*
Giardia lamblia

Trophozoite

- Small pear-shaped flagellate Measures 12–15 x 5–9μm
- Has a large concave sucking disc on the ventral surface

Cyst

- Small and oval measuring 8–12 x 6 μm.
- Internal structures include four nuclei grouped at one end (sometimes difficult to see)
**Giardia lamblia**  
*G. duodenalis, G. intestinalis*

- Unique symmetrical heart shaped cells
- Cysts are small, compact, and multinucleate
- Cysts can survive for 2 months in environment
- Cysts enter duodenum, geminate, & travel to jejunum to feed & multiply
- Spread through contaminated water & food (Fecal oral contamination)

**Adhesive Disk and Attachment**
In the tropics and subtropics, in areas where water supplies and the environment become faecally contaminated. In endemic areas.

- young children are more frequently infected than adults,
- *G. lamblia* has a direct life cycle
- Infection with *G. lamblia* is called *giardiasis*

**Giardiasis**

- Although many infections, particularly in adults are without symptoms, *G. lamblia can cause abdominal pain, severe diarrhoea*, flatulence, vomiting, weight loss, malabsorption with **lactose intolerance**.
- in children, impairment of growth, Symptoms can be severe particularly in children under 3 years of age, and in the undernourished. Those with reduced immune responses, gastrointestinal disorders or intestinal bacterial infections, tend to be more susceptible to *Giardia infection*.

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**Giardia lamblia**

- **worldwide distribution**
  - higher prevalence in developing countries (20%)
  - 1-6% in temperate countries

- **most common protozoa found in stools**
  - ~200 million clinical cases/year
Giardia and life cycle

- Cysts enter duodenum, geminate, & travel to jejunum to feed & multiply
- The trophozoites live on the small intestine mucosa (less frequently on the gallbladder mucosa as well).
- Reproduction is by means of longitudinal binary fission of the trophozoites.
- Trophozoites are excreted in stool but the cyst rarely excreted in the stool.
Clinical Features and Symptoms

**Acute Symptoms**

- 1-2 week incubation
- sudden explosive, *watery diarrhea*
  - Steatorrhea (*fatty diarrhea*), bulky, frothy, greasy, *foul-smelling stools*
  - no blood or mucus
- upper gastro-intestinal uneasiness, bloating, flatulence, belching, cramps, nausea, vomiting, anorexia
- usually clears spontaneously (undiagnosed), but can persist or become chronic

Giardiasis is different from amebiasis and bacterial dysentery

- No granulocytosis and no fever
- No mucous, blood

**Possible Mechanisms**

- mechanical irritation
- obstruction of absorption
LABORATORY DIAGNOSIS

- Sreatorrhea (fatty diarrhea). Faecal specimens containing *G. lamblia* may have an offensive odour and are pale colored, fatty and float in water.
- examination is described under Direct examination of faecal specimens
- *G. lamblia trophozoites in fresh diarrhoeic specimens particularly in mucus. They are often difficult to detect because they attach themselves to the wall of the intestine.
- *Finding G. lamblia cysts in more formed specimens. The cysts are excreted irregularly.*

**Note:** In giardiasis, the number of trophozoites or cysts present in faecal specimens cannot be taken as an indication of the severity of infection.

- ELISA kits are now also available to detect Giardia-specific structural and soluble antigens in stool samples.
- Duodenal aspirates: Occasionally giardiasis can be diagnosed by detecting *G. lamblia trophozoites in duodenal contents.*

**Prevention and control of *G. lamblia infection***

- Giardiasis may be reduced by improving environmental sanitation
- and personal hygiene to prevent food, water, and hands becoming contaminated with faeces containing cysts.
- The cysts are not killed in food or water stored at 4–6 °C.
- cysts of *E. histolytica, Giardia are resistant* to the concentrations of chlorine normally used for the treatment of domestic water supplies.
The Non-Pathogenic Intestinal Flagellates

- All inhabit large intestine. All are harmless commensals.

1- *Embadomonas (Retortamonas) intestinalis*

**Trophozoite**: elongate, pyriform, 6×4μ in size.
- Anterior cleft-like cytostome.
- One vesicular nucleus.
- Two blepharoplasts give rise to 2 flagellae (one directed anteriorly & the other pass through cytostome)

**Cyst**: uninucleated, pyriform, 5×4μ in size.

2- *Chilomastix mesnili*

**Trophozoite**: asymmetrical pear shaped, about 15×8μ in size.
- Anterior cleft-like cytostome.
- One vesicular nucleus.
- Six blepharoplasts (near nucleus) give rise to 3 free flagellae anteriorly, 2 surround the margins of the cytostome & the last pass through it.

**Cyst**: uninucleated, lemon-shaped, 6×8μ in size.
**Chilomastix mesnili**

*Trophozoite:*

*3- Enteromonas hominis*

*Trophozoite: oval in shaped,*

About 6×4μ in size.
- *No cytostome.*
- One vesicular nucleus.
- Four blepharoplasts (near nucleus)
  give rise to 3 free flagellae anteriorly,
  & the fourth pass posteriorly.

*Cyst: ovoidal in shaped,*

5×4μ in size.
Cysts can have one or two polar nuclei.
Mature cyst is quadrinucleated.
**Apicomplexan Protozoa**

- Non-motile in mature stage
  - Male gametes are motile
- Alternate between sexual & asexual phases & between different animal hosts
- All members are parasitic
- Most form specialized infective bodies that are transmitted by arthropod vectors, food, water, or other means
  - *Plasmodium*
  - *Toxoplasma*
  - *Cryptosporidium* (intestinal)
Human intestinal coccidia:

- *Isospora belli, C. parvum and C. cayetanensis* are thought to have a worldwide distribution.
- *Cryptosporidium parvum* is particularly prevalent in tropical countries.
- *I. belli, Cryptosporium, and Cyclospora cayetanensis* are transmitted by the faecal-oral route with infective oocysts being ingested.
The oocysts of *I. bellii* and *C. cayetanensis* are not infective when passed in faeces. They require several days in the environment in which to mature.

But the oocysts of *Cryptosporidium* are infective when excreted in the faeces.

Water-borne outbreaks of *C. parvum* and *C. cayetanensis* have been reported from tropical countries and elsewhere.

*Cryptosporidium* oocysts are particularly resistant to disinfectants, including chlorine at concentrations normally used to treat drinking water supplies.

*Cryptosporidium* infection rates are higher in the warm wet season, and lower in the drier cool months.

**life cycle of intestinal coccidia**

The life cycle of *I. bellii*, *Cryptosporidium* and *C. cayetanensis* is typical of sporozoa,

i.e. there is an asexual reproductive stage (*schizogony*) and a sexual development cycle (*sporogony*),

They complete their life cycle in a single host.

*Cryptosporidium parvum* infects a wide range of domestic animals and wildlife.

*I. bellii*, *C. parvum* (*C. hominis* and other species) and *C. cayetanensis* are being increasingly reported as causes of enteritis and as opportunistic pathogens in immunocompromised persons.
Intestinal coccidiosis

- *I. belli* infection is rarely serious and can be asymptomatic but severe diarrhoeal disease can occur in AIDS patients.

- *Cryptosporidium parvum* is widely spread in the environment. It has been reported as an important cause of diarrhoeal disease in young children and toddlers in developing countries.

- In persons with depressed immune responses, particularly those with AIDS, infection with *Cryptosporidium* can cause acute and often fatal diarrhoeal disease and also respiratory disease.

- Autoinfection can occur with infective oocysts sporulating in the intestine.

- Disseminated disease may occur. Person to person transmission is common and animal hosts are also important sources of human infections.

- *C. caytanensis* has been described as a cause of prolonged diarrhoea in tropical countries and elsewhere. It can cause severe prolonged diarrhoea in those infected with HIV.

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**TRANSMISSION**

1. Infective oocysts ingested.

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**ENVIRONMENT**

7. Faeces containing oocysts contaminate water supplies, food, etc.

*Note: C. parvum* oocysts are infective when passed in faeces. *C. caytanensis* and *I. belli* oocysts become infective in the environment after 3–5 days.

**HUMAN HOST**

2. Oocysts excyst. Sporozoites infect intestinal cells.


5. Sporozoites produced in oocysts by sporogony.

6. Oocysts passed in faeces.

Transmission and life cycle of intestinal coccidia
Cryptosporidium

- An intestinal pathogen
- Infects a variety of animals
- Exists in tissue & oocyst phases
- Causes enteric symptoms
- AIDS patients may suffer chronic persistent diarrhea

Cryptosporidium

- Infectious form = oocyst
- Sporozoites ‘invade’ intestinal epithelial cells
- fecal-oral transmission
- two species infecting humans
  - C. parvum: cattle and other mammals
  - C. hominis: only humans
  - now known to be common human pathogen
- self-limiting diarrhea in immunocompetent persons
- profuse, watery diarrhea associated with AIDS (life threatening)
**Pathogenesis**

**DIARRHEA**
- enterocyte malfunction (osmotic)
  - impaired absorption
  - enhanced secretion
- inflammatory diarrhea
  - mucosal invasion
  - leukocytes in stools
- secretory diarrhea
  - toxin associated
  - watery

**Laboratory diagnosis of Coccidia**
- Oocysts of *Cryptosporidium and C. cayetanensis* can be detected in wet preparations but they are more easily identified in smears stained by the modified Ziehl-Neelsen (Zn) method following concentration by the formol ether oocyst concentration technique
- *I. belli* oocysts can be easily identified in an unstained wet preparation

Cryptosporidium oocysts in Ziehl-Neelsen stained faecal smear.

Cyclospora oocysts in unstained preparation.

Cyclospora oocysts in Ziehl-Neelsen stained faecal smear.

Ciliated Protozoa

Trophozoites have cilia

Majority are nonpathogens except Balantidium coli
Balantidium coli

- intestinal symptoms
- Healthy humans are resistant
- Rarely penetrates intestine or enters blood
- An occupant of the intestines of domestic animals such as pigs & cattle
- Acquired by ingesting cyst-containing food or water

Trophozoite of B. coli

- Large, easily seen oval shaped ciliate with a rapid revolving motility, measuring 50–200 x 40-70 µm.

Cyst of B. coli

- Large, round, thick walled, measuring 50–60 µm in diameter.

- It is the only ciliate that can parasitize humans.
- causes intestinal necrosis and inflammation with ulceration.
- The disease is transmitted through spherical cysts (50–60 µm) from host to host on the fecal-oral route.
Life Cycle:

- Cysts are the parasite stage responsible for transmission of *balantidiasis*.
- The host most often acquires the cyst through ingestion of contaminated food or water (*fecal-oral route*).
- Following ingestion, *excystation* occurs in the *small intestine*, and the trophozoites *colonize the large intestine*.
- The trophozoites reside in the lumen of the large intestine of humans and animals, where they replicate by *binary fission*, during which conjugation may occur.
- Trophozoites undergo *encystation* to produce infective cysts. Some trophozoites invade the *wall of the colon* and multiply.
- Some return to lumen and disintegrate.
- Mature cysts are passed with feces.
**Balantidium coli**

Causative agent of balantidiosis

- *Balantidium coli* is a **worldwide distributed** ciliate of highly **variable size** (30–150 µm long).
- It is frequently found as an inhabitant of the **large intestine** of monkeys, rats, and in particular **pigs**. It is also found, more rarely, in humans.
- Human infections have been reported mainly from Central and South America, the Philippines and other tropical areas among those who **keep pigs and use pig faeces as fertilizer**.
- *B. coli* is **transmitted by the ingestion of infective cysts** in food or water or from **hands contaminated with pig faeces**.
- *B. coli* has a **direct life cycle similar to that of E. histolytica and G. lamblia** except a pig, not a human is the natural host. *B. coli* causes balantidial dysentery.

**Balantidal dysentery**

- Infection with *B. coli* can be **asymptomatic**.
- **Balantidal dysentery** occurs when the ciliates invade the wall of the large intestine, causing **inflammation and ulceration with blood and mucus** being passed in the faeces. Intestinal perforation is a serious complication of balantidiasis.

**LABORATORY DIAGNOSIS**

- Diagnosis involves detection of **cysts or vegetative forms** in fecal samples.
- Balantidal dysentery is diagnosed by finding the motile trophozoites of *B. coli in a fresh dysenteric* faecal specimen,
- examined in the same way as described for amoebic dysentery.
- In chronic infections, **B. coli cysts can be found in formed or semi-formed faeces**.
- Diagnosis is based on detection of **trophozoites in stool specimens** or in **tissue collected during endoscopy**. Cysts are less frequently encountered.
- **Balantidium coli is passed intermittently and once outside the colon is rapidly destroyed.** Thus stool specimens should be collected repeatedly, and immediately examined.
Prevention and control of balantidiasis

- not eating food which is likely to be contaminated with pig faeces,
- protecting water supplies from faecal contamination,
- improving personal hygiene.
- *B. coli* cysts are infective as soon as they are excreted in the faeces. They are rapidly killed by drying but in moist conditions they can remain infective for several weeks.
Human Trichomonads

There are 3 species:
1- *Trichomonas vaginalis*.
1- *Trichomonas hominis (intestinalis).*
2- *Trichomonas tenax (buccalis).*

- They have 4 anterior flagellae, an undulating membrane, an axostyle and a cytostome.
- There is no cyst formation but exist only in the trophozoite stage (The infective stage).

*Trichomonas hominis*  
*(T. intestinalis)*

It is non pathogenic & inhabit the large intestine.
- Small, 12x8μ, pyriform.
- Has small antero-lateral cytostome (mouth), one nucleus, four free flagella anteriorly and one marginal forming undulating membrane along 2/3 length of the parasite with free end.
- Thick axostyle (flagellum)
  - Transmission is direct, through contaminated food and drink.
**Trichomonas tenax**

*(T. buccalis)*

- It is non pathogenic parasite that inhabits the *mouth* especially in carious teeth & gums.
- It measures 10x6μ, pyriform in shape.
- Has cytostome, one nucleus, 4 free flagella and one marginal with undulating membrane 1/2 the lateral margin of the parasite with no free end.
- Slender axostyle and faint parabasal body.
- Transmission is by the use of contaminated utensils with the trophozoite or direct by kissing.

**Dientamoeba fragilis**

It was considered to be an amoeba, but its flagellate nature was discovered by electron microscope.

It is now classified among the trichomonads.

**Geographical distribution:** worldwide.

**Morphology:**

- It has no cyst stage.
- *Trophozoite*: 7 × 12 μ, characterized by the presence of mainly two nuclei (1-3 nuclei).
- *(The rosette shaped nuclei containing granules at times may resemble that of E. nana).*
- Cytoplasm contains food vacuoles (contain bacteria).
- Pseudopodia are leaf-like → active in fresh stool.
Morphology of *Dientamoeba fragilis* from a stool sample. Trophozoites exhibit an ameba-like morphology and are often bi-nucleated.

*Dientamoeba fragilis* Life Cycle

1. Trophozoites in feces
2. Transmission via fecal-oral route
3. It has been postulated that transmission occurs via helminth eggs, such as *Ascaris* and *Enterobius.*
4. Trophozoites ingested

= Infective Stage
= Diagnostic Stage

No cyst stage has been identified.

Binary fission
Trophozoites in lumen of colon
**Habitat:** mucosal crypts of large intestine of man.

(It may ingest RBCs but never invades the mucosa).

**Infective stage:** trophozoite.

**Mode of transmission:**
Transmission might occur via eggs of *Entrobius vermicularis* or *Ascaris* (trophozoite inside the egg) as oral transmission failed.

**Clinical picture:** mucus diarrhea, abdominal pain, flatulence and weakness.

**Diagnosis:** careful examination of fresh saline smear of stool. Staining with iron haematoxylin is preferable.

**Treatment:**
- Idioquinol
- Tetracycline
- Paromomycin
**Trichomonas**

- is a pear shaped protozoon about 10–20 µm long and 2–14 µm wide.
- Five flagella emerge from a basal body at the anterior pole, four freely extend forwards and one extends backwards, forming the outer edge of the undulating membrane.
- Exists only in *trophozoite* form
- The commonest species infect humans
  - *T. vaginalis*
**Trichomonas vaginalis**

- In average populations of developed countries, infection rates are about 5–20% in women and usually below 5% in men.
- Trichomonads are anaerobic protozoa which are specialized organelles producing H2 as a metabolite.
- *T. vaginalis* colonizes the mucosa of the urogenital tract and reproduces by longitudinal binary fission.
- Trichomonads do not encyst, although rounded, nonmotile forms are observed which are degenerated stages.
- It causes vaginitis in women and urethritis in men.
- Causes an Sexually Transmitted Disease (STD) called trichomoniasis.
- Reservoir is human urogenital tract
- 20-50% of infected are asymptomatic
- Strict parasite, cannot survive long outside of host

**Female symptoms:**

- vaginitis can develop after an incubation period of 2 - 24 days.
- The infection results in production of a purulent, thin yellowish discharge in which trichomonads, pus cells, bacteria and *candida albicanis* are found.
- Foul-smelling, green-to-yellow discharge
- Growth of organism is favored by high pH more than 5.9 (normal in vagina 3.5-4.5)
- May make intercourse painful.

**Male symptoms:**

- Infections in men are for the most part asymptomatic (50–90%).
- Urethritis, thin, milky discharge, occasionally prostate infection and seminal vesicles as well.
- Urinary frequency & pain
- but only rarely infect the urinary bladder.
• **Life cycle:**
  - trophozoite (infective stage) occur in the urinogential system, the stage multiplied by **longitudinal binary fission**.
  - trophozoite are present in the vagina, prostatic secretions and urine (diagnostic stage).
  - Infection by sexual contact

• **Diagnosis:**
  - *fresh specimen of vaginal or urethral secretion* is mixed with *physiological saline* solution and examined under a microscope for trichomonads.
  - Normally identified from *vagina or urethral swab*.
  - May found in *urine tests*.
  - May make *intercourse painful*.

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**Control and prevention**

• Treat infected *woman, her sexual partner*.

• Use *condom during future sexual intercourse till full recovery*.

• **Diagnosis**
  • *'wet mount' of vaginal secretion, urine sediment or prostatic massage ; characteristic fast (darting) movement.*
  • *In stained preparation (Geimsa stain)*
  • *Culture in special media*
  • *Serological test.*
<table>
<thead>
<tr>
<th>Protozoa found in human stool specimens</th>
<th>Flagellates</th>
<th>Ciliate</th>
<th>Coccidia</th>
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<tbody>
<tr>
<td><em>Trichomonas hominis</em></td>
<td><em>Chlamastix meytii</em></td>
<td><em>Giardia lambia</em></td>
<td><em>Balantidium coli</em></td>
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