





### Protozoa

# **General Features**

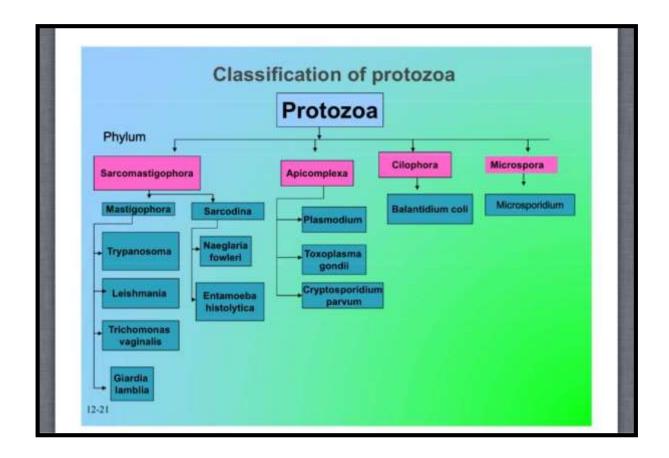
- Single-celled eukaryotic microorganisms belonging to kingdom protista are classified as Protozoa (Greek Protos: first; zoon: animal).
- The single protozoal cell performs all functions.
- Most of the protozoa are completely nonpathogenic but few may cause major diseases such as malaria, leishmaniasis, and sleeping sickness.
- Protozoa like *Cryptosporidium parvum* and *Toxoplasma gondii* are being recognized as opportunistic pathogens in patients affected with human immunodeciency virus (HIV) and in those undergoing immunosuppressive therapy.
- Protozoa exhibit wide range of size (1–150 μm), shape, and structure; yet all possess essential common features.
- Protozoa classes in to a-Amebas, b-Flagellates, c-Ciliates, d-Sporozoa
- Reproduction

a-Asexual Reproduction

- Binary fission: It is a method of asexual reproduction, by which a single parasite divides either longitudinally or transversally into two or more equal number of parasites. Mitotic division of nucleus is followed by division of the cytoplasm. In amoebae, division occurs along any plane, but in flagellates, division is along longitudinal axis and in ciliates, in the transverse plane.
- Multiple fission or schizogony: Plasmodium exhibits schizogony, in which nucleus undergoes several successive divisions within the schizont to produce large number of merozoites.
- Endodyogeny: Some protozoa like *Toxoplasma*, multiply by internal budding, resulting in the formation of two daughter cells.

**b-** Sexual Reproduction

- Conjugation: In ciliates, the sexual process is conjugation, in which two organisms join together and reciprocally exchange nuclear material (e.g. *Balantidium coli*).
- Gametogony or syngamy: In sporozoa, male and female gametocytes are produced, which after fertilization form the zygote, which gives rise to numerous sporozoites by sporogony (e.g. *Plasmodium*).

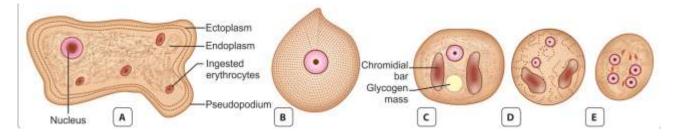


### Class: Amebas (Rizopoda)

### 1. Entamoeba histolytica (Intestinal Amoeba)

#### Morphology

*E. histolytica* occurs in 3 forms a. Trophozoite , b. Precyst , c. Cyst.



*Entamoeba histolytica*. A. Trophozoite; B. Precystic stage; C. Uninucleate cyst; D. Binucleate cyst; E. Mature quadrinucleate cyst

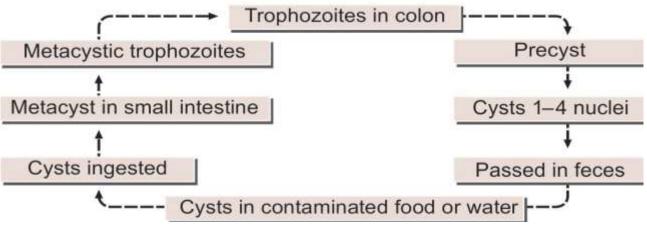
## Life Cycle

*E. histolytica* passes its life cycle only in 1 host-man

**Infective form:** Mature quadrinucleate cyst passed in feces of convalescents (نقاهة) and carriers. The cysts can remain viable under moist conditions for about 10 day.

Mode of transmission: Man acquires infection by swallowing food and water contaminated with cysts.

- As the cyst wall is resistant to action of gastric juice, the cysts pass through the stomach undamaged and enter the small intestine.
- **Excystation:** When the cyst reaches caecum or lower part of the ileum, due to the alkaline medium, the cyst wall is damaged by trypsin, leading to excystation.
- The cytoplasm gets detached from the cyst wall and amoeboid movements appear causing a tear in the cyst wall, through which **quadrinucleate amoeba** is liberated. This stage is called the **metacyst**
- **Metacystic trophozoites:** The nuclei in the metacyst immediately undergo division to form 8 nuclei, each of which gets surrounded by its own cytoplasm to become **8 small amoebulae or metacystic trophozoites**.
- If excystation takes place in the small intestine, the metacystic trophozoites do not colonize there, but are carried to the caecum.
- The optimal habitat for the metacystic trophozoite is the submucosal tissue of caecum and colon, where they lodge in the glandular crypts and grow by binary fission.
- Some develop into precystic forms and cysts, which are passed in feces to repeat the cycle.
- The entire life cycle is, thus completed in one host.
- In most of the cases, *E. histolytica* remains as a commensal in the large intestine without causing any ill effects. Such persons become carriers or asymptomatic cyst passers and are responsible for maintenance and spread of infection in the community. Sometimes, the infection may be activated and clinical disease ensues. Such latency and reactivation are the characteristics of amoebiasis.



Life cycle of *Entamoeba histolytica* (Schematic)

### Intestinal amoebiasis

Trophozoites can invade the colonic epithelium and produce ulcers and dysentery.

#### Extra - intestinal amoebiasis

Trophozoites migrate to other organs in the body such as liver, brain, pleura pericardium and other extra - intestinal sites.

#### **Pathogenesis and Clinical Features**

#### ✤ Intestinal amoebiasis

- Asymptomatic infection (cyst passers).
- Acute amoebic dysentery characterized by abdominal pain , tenderness , and tender hepatomegally.
- Non dysenteric colitis.
- Intestinal amoebiasis complicated by:- Toxic megacolon , (المنتشر) fulminant amoebic colitis , amoebic peritonitis ( التهاب "البيرتون" الصفاق) and perianal ulceration.

### 🛠 Extra intestinal amoebiasis

- Amoebic liver abscess .
- Pleura-pulmonary amoebiasis.
- Amoebic abscess of the brain .

### Complications and sequelae of intestinal amoebiasis

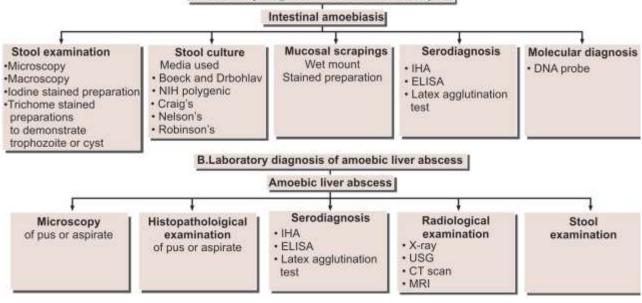
#### Fulminant amoebic colitis

- Toxic megacolon
- Perianal ulceration
- Amoeboma

#### **Extraintestinal amoebiasis**

- Amoebic hepatitis
- Amoebic liver abscess
- Amoebic appendicitis and peritonitis
- Pulmonary amoebiasis
- Cerebral amoebiasis
- Splenic abscess
- Cutaneous amoebiasis
- Genitourinary amoebiasis

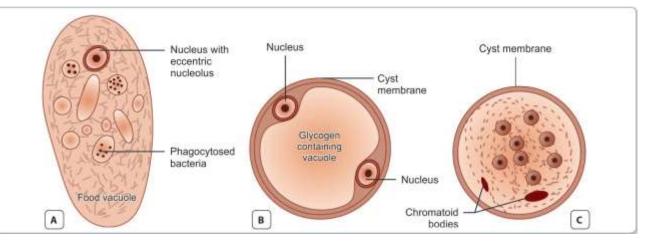
### Laboratory Diagnosis



A.Laboratory diagnosis of Entamoeba histolytica

#### Entamoeba coli

It is considered to be a **nonpathogenic** with worldwide distribution. That frequently exists as a **commensal parasite** in the human gastrointestinal tract. As with the other intestinal amoeba, *E.coli* is transmitted through the ingestion of the infected cyst through contamination food or water or drink. Its life cycle is similar to that of *E.histolytica* but **it does not have an invasive stage and does not ingest red blood cells** There is **no clinical symptoms** during and after infection by this parasite. This parasite have two stages:



Schematic diagram of the morphological forms of *Entamoeba coli* A. Vegetative form; B. Binucleate cyst; C. Eight-nucleate cyst

#### Trophozoite stage

It is move sluggishly by short pseudopodia, **non-progressive motility**. The single nucleus is consist of large irregular shape, eccentric karyosome and uneven

peripheral chromatin. The cytoplasm is coarse and contain food vacuoles filled with bacteria, yeast cell, but no RBCs.

### • Cyst stage

It is round to spherical in shape, surrounded by thick cell wall. The cytoplasm granular in appearance. Food vacuoles are not present. having (1-8) nuclei, There is large, irregular-shaped and eccentric position of the karyosome can be frequently distinguished even in unstained amoebae. uneven peripheral chromatin bars are thin with pointed end .

## The life cycle of *E.coli*

Human infected occurs through ingestion of food or water contaminated by cyst bearing faeces. Eight nucleated metacyst is excysted in intestine.

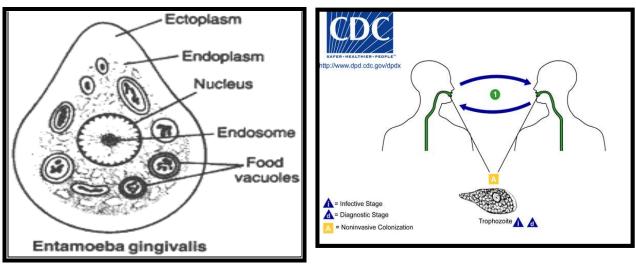
After a series of cytoplasmic divisions eight to fewer metacystic trophozoites are formed and develop into mature trophozoites in caecum. Trophs multiply by binary fission.

### Laboratory diagnosis

By examination of stool sample .There is non-pathogenic and no clinical manifestation –results in from into infection-to man.

### 3. Entamoeba gingivalis

It is first amoeba recovered from a human specimen, discovered in 1849. Infection of *E.gingivalis* occurring both in the mouth and in the genital tract. **Nonpathogenic**, there is **no clinical symptoms** during and after infection by this parasite. Trophozoites are frequently recovered in patients suffering from pyorrhea alveolaris. **Infection of** *E.gingivalis* **are 1.**contracted via mouth to mouth (kissing). **2.** droplet contamination . **3.** sex intercourse. **4.** by use of (IUDS) = intrauterine devices . which may be **5.** transmitted through contaminated drinking utensils. There is **no known cyst stage of** *E.gingivalis* **presence only trophozoite stage.** 



### Laboratory diagnosis:

Examination of **1.** mouth scraping, particularly from the gingival area. Also material from the **2.** tonsillar crypts **3.** pulmonary abscess as well as sputum may also be examined. In addition the examination of **4.** vaginal / cervical material may be performed to diagnose *E.gingivalis* present in the vaginal / cervical area.

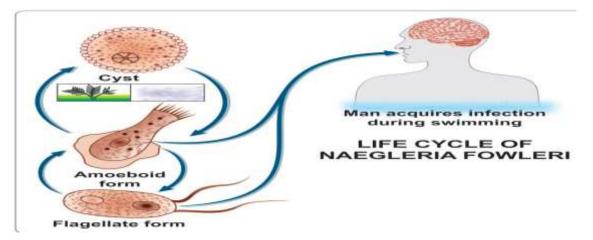
### 4. Naegleria fowleri

It is the only species of genus *Naegleria*, which infects man. *N. fowleri* causes the disease primary amoebic meningoencephalitis (PAM) التهاب الدماغ السحائي الاميبي, a brain infection that leads to destruction of brain tissue.

**Morphology** of *N*. *fowleri* occurs in 3 forms: 1. Cyst 2. Amoeboid trophozoite form 3. Flagellate trophozoite form.

The life cycle of *N. fowleri* is completed in the external environment.

- The amoeboid form of trophozoite is the feeding, growing, and replicating form of the parasite, seen on the surface of vegetation, طين mud, and water.
  It is the invasive stage of the parasite and the infective form of the parasite. multiplies by binary fission.
- Under unfavorable conditions, it forms a cyst and which undergoes excystation in favorable conditions.
- Flagellate form of trophozoite helps in the spread of *N. fowleri* to new water bodies. Since the amoeboid form is the invasive stage. The flagellate forms revert to amoeboid forms to become infective to man. hence *N. fowleri* is classified as amoeboflagellate.



**Laboratory Diagnosis**: The diagnosis of PAM is based on the finding of motile *Naegleria* trophozoites in wet mounts of freshly-obtained CSF. Cysts are not found in CSF or brain.

#### 5- *Acanthamoeba* Species

### **Morphology**

Acanthamoeba exists as active trophozoite form and a resistant cystic form.

## <u>Life Cycle</u>

\*Both trophozoites and cysts are infective.

\*Human beings acquire by inhalation of cyst or trophozoite , ingestion of cysts, or through traumatized ( المصابة ) skin or eyes.

\*After inhalation of aerosol (الرذاذ) or dust ( الغبار) containing trophozoites and cysts, the trophozoites reach the lungs and from there, they invade the central nervous system through the blood stream, producing **granulomatous amoebic encephalitis** (GAE).

**Granulomatous amoebic encephalitis (GAE):** It is a serious infection of the brain and spinal cord that typically occurs in persons with a compromised immune system. GAE is believed to follow inhalation of the dried cysts. The incubation period is long and the evolution of the illness is slow. Clinical picture is that of intracranial space-occupying lesions with seizures ( نوبات ) and mental deterioration ( تدهور عقلي ).

#### <u>Laboratory Diagnosis</u>

\***Diagnosis of amoebic keratitis** is made by demonstration of the cyst in corneal scrapings by wet mount, histology and culture. Growth can be obtained from corneal scrapings inoculated on nutrient agar, overlaid with live or dead *Escherichia coli* and incubated at 30°C.

\***Diagnosis of GAE** is made by demonstration of trophozoites and cysts in brain biopsy, culture, and immofluroscence microscopy using monoclonal antibodies. CSF shows lymphocytic pleocytosis, slightly elevated protein levels, and normal or slightly decreased glucose levels.

CT scan of brain provides inconclusive findings.

ad Trophozoite showing pinous acanthopodia

	Naegleria	Acanthamoeba
Disease	Primary amoebic meningoencephalitis (PAM)	Granulomatous amoebic encephalitis (GAE) and keratitis
Portal of entry	Nose	Upper Respiratory tract (?), comea
Clinical course	Acute	Subacute or chronic
Pathogenecity	Acute suppurative inflammation	Granulomatous inflammation
Morphological forms	3 stages: trophozoite, cyst and flagellate form	2 stages: trophozoite and cyst flagellate form absent
Trophozoite	10–20 µm, with a single pseudopodia	20-50 µm, with spine-like pseudopodia
Cyst	7–10 µm, round with smooth wall	15-25 µm, polygonal double-walled with wrinkled surface
Nuclear division	By promitosis, nucleolus divides, nuclear membrance persists	Nuclear membrance dissolves
WBC in CSF	Predominantly neutrophils	Predominantly lymphocytes