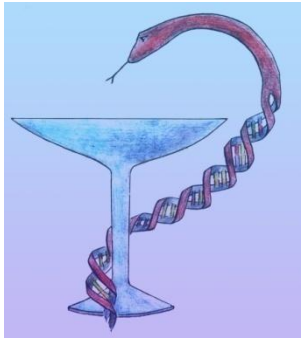
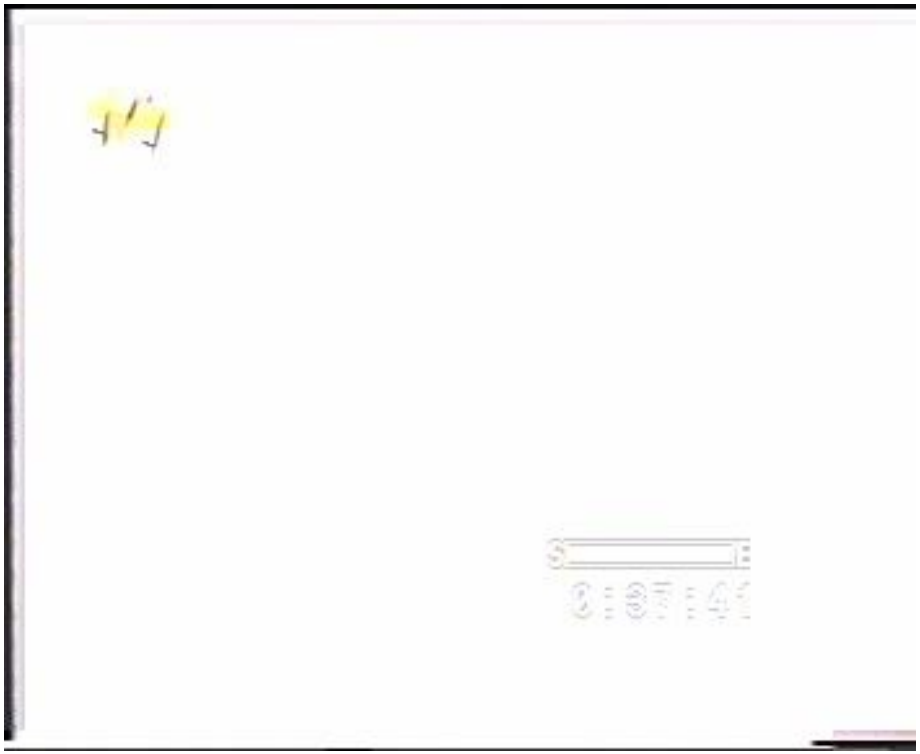


MEDICAL PROTOZOOLOGY

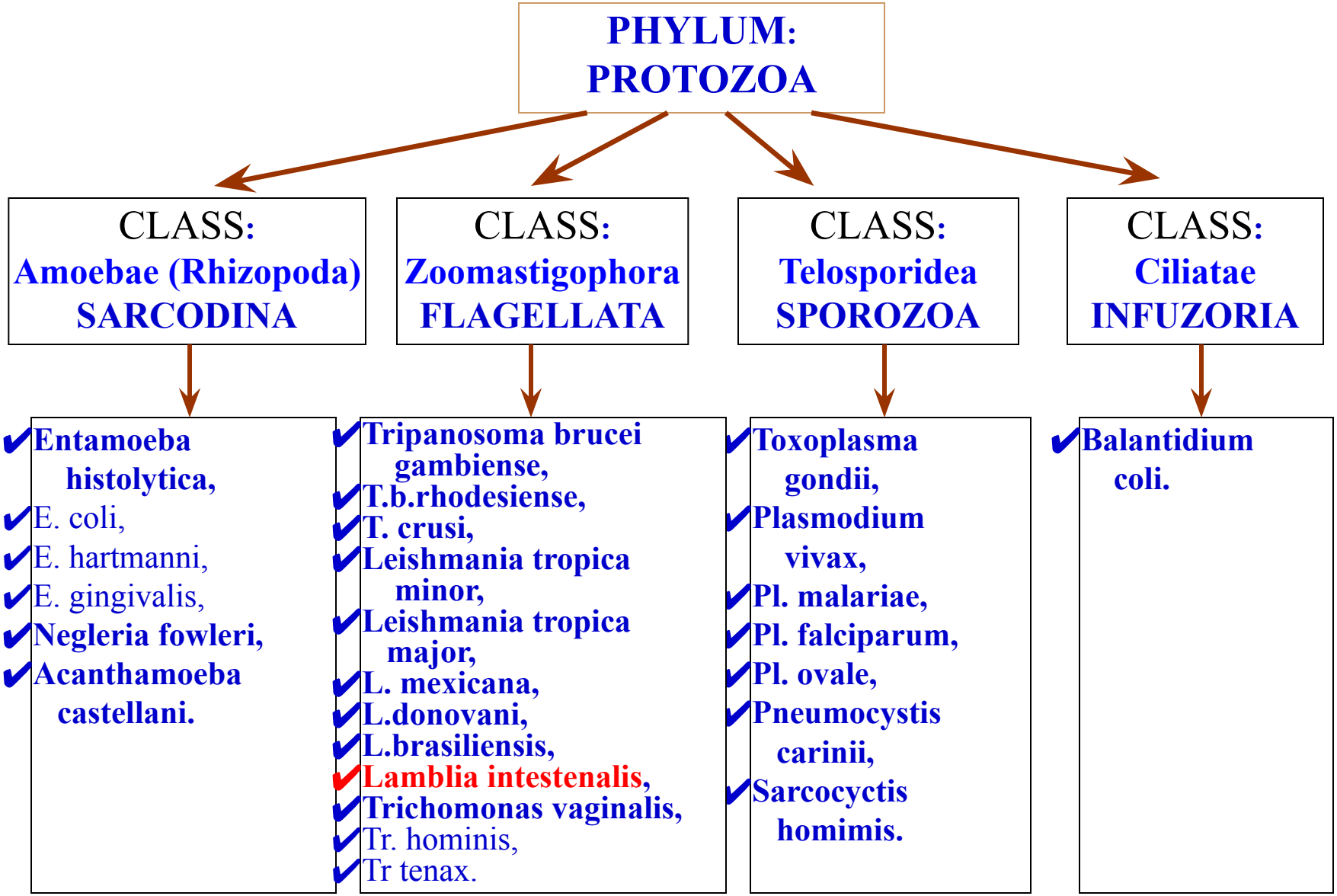
SARCODINA AND FLAGELLATA



General definitions

- Protozoa consist of a vast set of single-cell microorganisms that belong to protozoa phylum.
- Their morphology consists of cytoplasm and nucleoplasm. The cytoplasm consists of ectoplasm and endoplasm. The ectoplasm function consists in the protection, locomotion, and digestion of food, excretion and respiration. The endoplasm takes part in metabolism. It contains the nucleus and many organelles. The reproduction and maintenance of life are performed by the nucleus.
- The protozoa that have medical significance to humans include: Amoebas, Flagellata, Ciliata and Sporozoa.
- Many protozoan species are not pathogenic. However, they may be difficult to be differentiated from pathogenic species. For this reason, a laboratory person must be familiar with characteristics of pathogenic as well as non-pathogenic species.

BIOLOGICAL CLASSIFICATION



MEDICAL-PRACTICAL CLASSIFICATION

PHYLUM: PROTOZOA

protozoa inhabiting the gastrointestinal tract:

- In oral cavity
- In the small intestine
- In the large intestine

protozoa inhabiting the tissues:

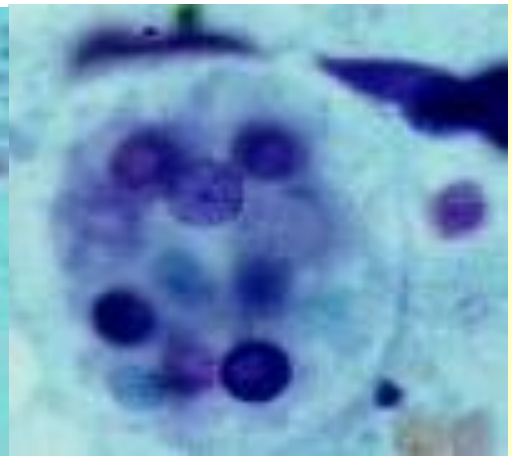
- Are transmitted by carriers
- Are not transmitted by carriers

CLASS
Amoebae (Rhizopoda)
SARCODINA

ENTAMOEBA HISTOLYTICA

Kingdom	Animalia
Phylum	Protozoa
Class	Rhizopoda
Genus	Entamoeba
Species	E. histolytica

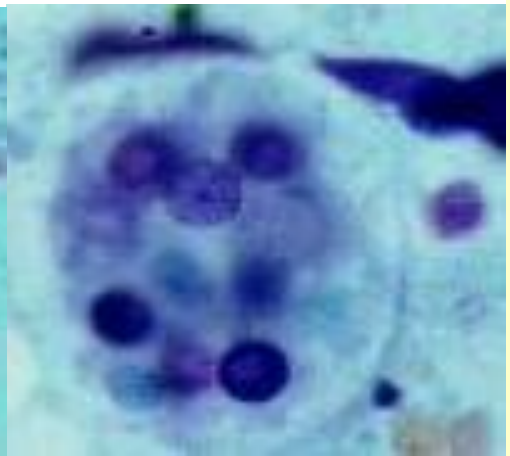
ENTAMOEBA HISTOLYTICA is the causative agent of the anatropous disease *amoebiasis* (amoebic dysentery). Amoebiasis is characterized by frequent watery stools mixed with blood and mucus, abdominal pain, fever, and dehydration of the body.

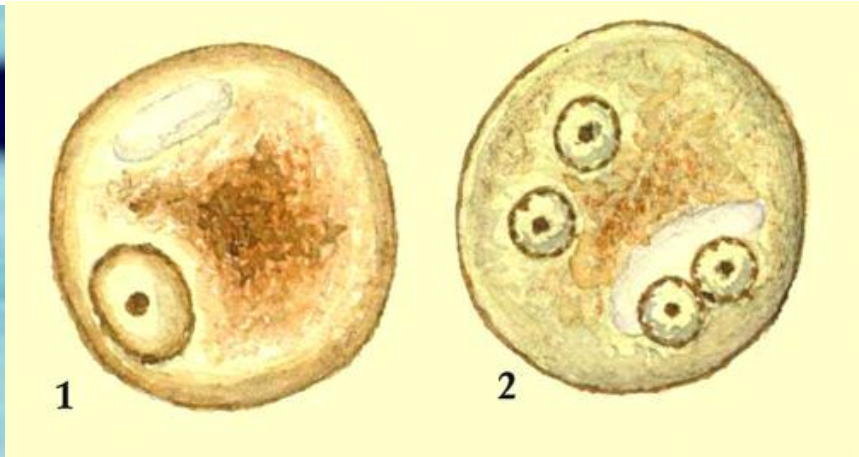


ENTAMOEBA HISTOLYTICA

Geographical Distribution: Cosmopolitan distribution, mainly in the tropics and subtropics. It is mainly related to an inadequate personal hygienic environmental sanitation, lack of safe water supply, and poor socioeconomic situation.

Habitat: Large intestine, liver abscesses and other extra-intestinal organs





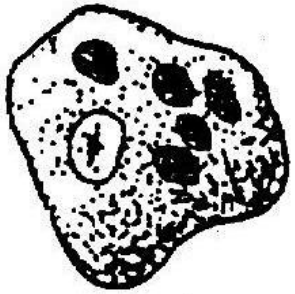
LIFE FORMS OF ENTAMOEBA HISTOLYTICA

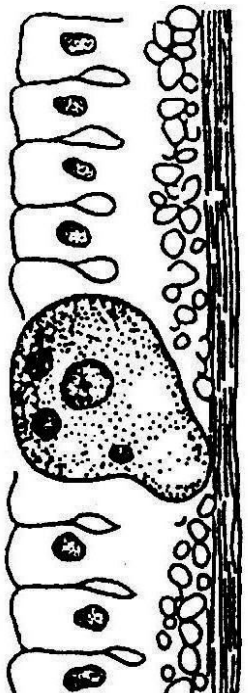
In the life cycle of this parasite, there are the following life forms: cyst and several vegetative forms that differ from one another by morphological features and pathogenic properties.

CYST

F. MINUTA

F. MAGNA

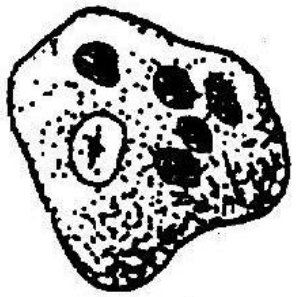




TISSUE FORM

A cyst is a round non-movable formation which has 8-15 microns in the diameter. The cyst has 4 nuclei. It is a diagnostic sign of Entamoeba histolytica. Cysts are found in the feces of patients. Daily, up to 8 million cysts are released from the human body. Cysts survive in water up to 2 months, and remain viable after the exposure to disinfectants, but they are killed by boiling.

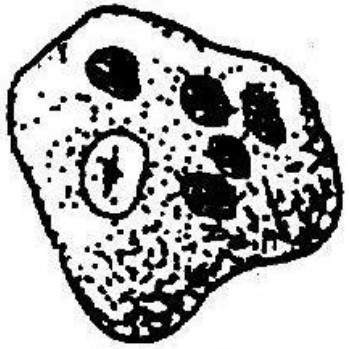
A small vegetative form (luminal form, forma minuta) is the main form of the existence of E. histolytica. It inhabits the lumen of the colon. This form of dysenterial amoeba is not pathogenic. This parasite does not cause any clinical signs of disease. In the lower part of the colon, the forma minuta is capable of forming cysts. Small vegetative forms are detected in the feces of cyst-carriers or in patients with chronic amebiasis. A luminal form has the dimensions from 7 to 25 microns. The movement of amoebae is slow. Their pseudopodia are short and in the digestive vacuoles phagocytized bacteria are found.



A large vegetative form (forma magna) is a large cell which has the size from 20 to 60 mkm. An ectoplasm and an endoplasm are clearly distinguishable in the cytoplasm of this parasite. This is a moveable form. It forms a "finger-like" pseudopodia. This is a pathogenic form. It feeds on erythrocytes, so this stage is called erythrophage. The large vegetative form is derived from the luminal form of a dysenteric amoeba, and this process may contribute to violation of the diet, vitamin deficiency, changes in the intestinal microflora, and decreased immunity. The large vegetative forms are found in freshly isolated liquid feces of patients with acute amebiasis.

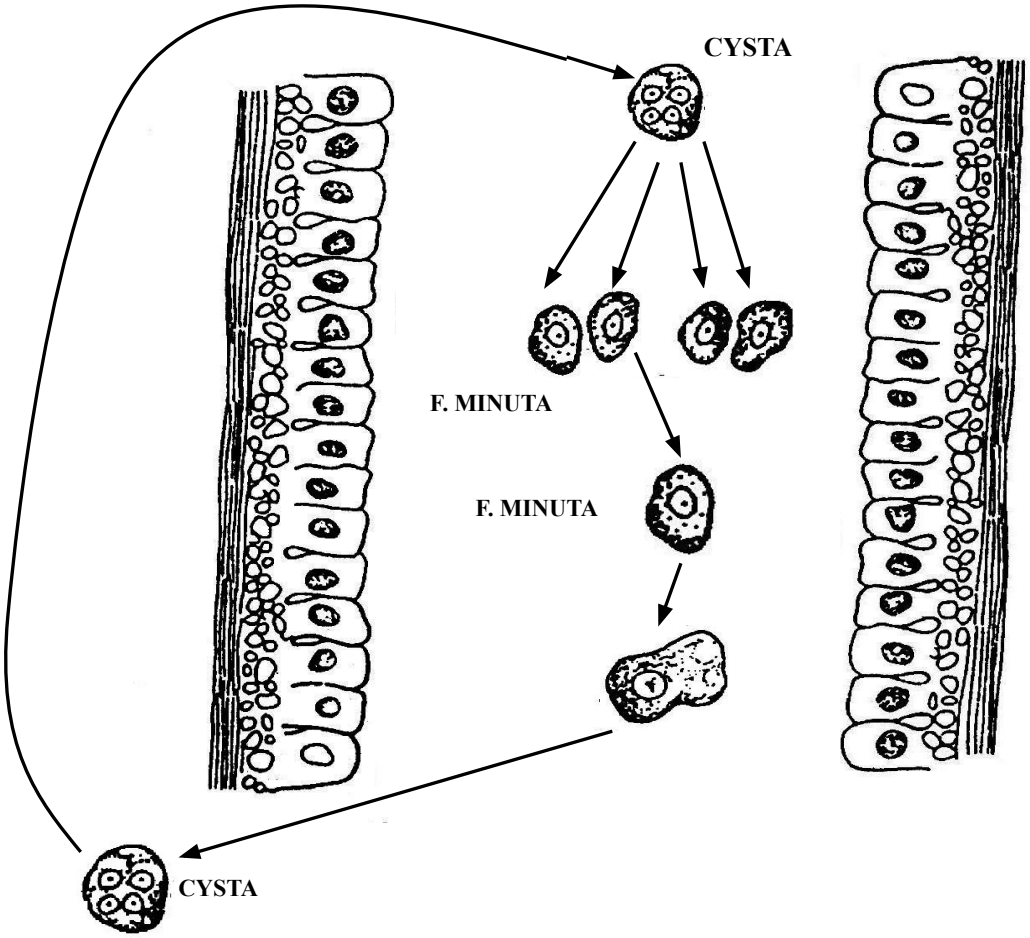
The tissue form is also a pathogenic stage of dysenteric amoeba. Its size is 20-25 microns. The localization of a parasite is the tissue of the mucous membrane of the colon. The tissue form causes specific damage, that is the formation of ulcers. Often, the large vegetative form and the tissue form are combined in the common name (tissue form or large vegetative form), but it is not quite correct.

The penetration of the large vegetative form from the intestinal lumen into the tissue is facilitated by the inflammation of the intestinal mucosa, by damage of the intestinal epithelium, hypothermia, hyperthermia, and vitamin deficiency, etc. The tissue form secretes the proteolytic enzymes that destroy the cell- to cell contacts of the mucosal epithelium, as well as necrotoxins causing the tissue necrosis of the intestine. The tissue form can penetrate into lymphatic or blood vessels and can be transported to other organs, most commonly into the liver, lungs, spleen.



LIFE CYCLE OF ENTAMOEBA HISTOLYTICA

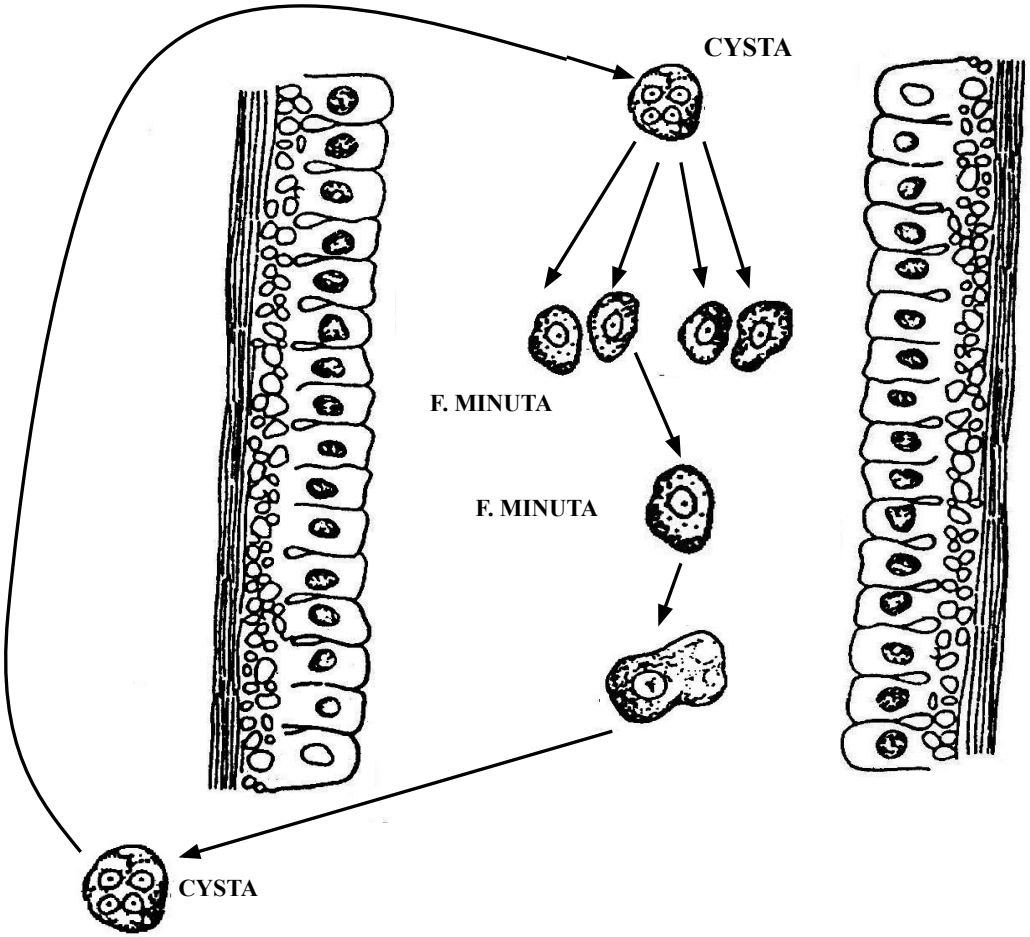
in asymptomatic carriers



The way for invasion is fecal-oral. The source of invasion is cysts that come into the body through the mouth. The mechanical carriers (flies and cockroaches) are involved in the spread of the cysts. In the small intestine, the cyst envelope undergoes the dissolution and 4 small vegetative forms are developing.

LIFE CYCLE OF ENTAMOEBA HISTOLYTICA

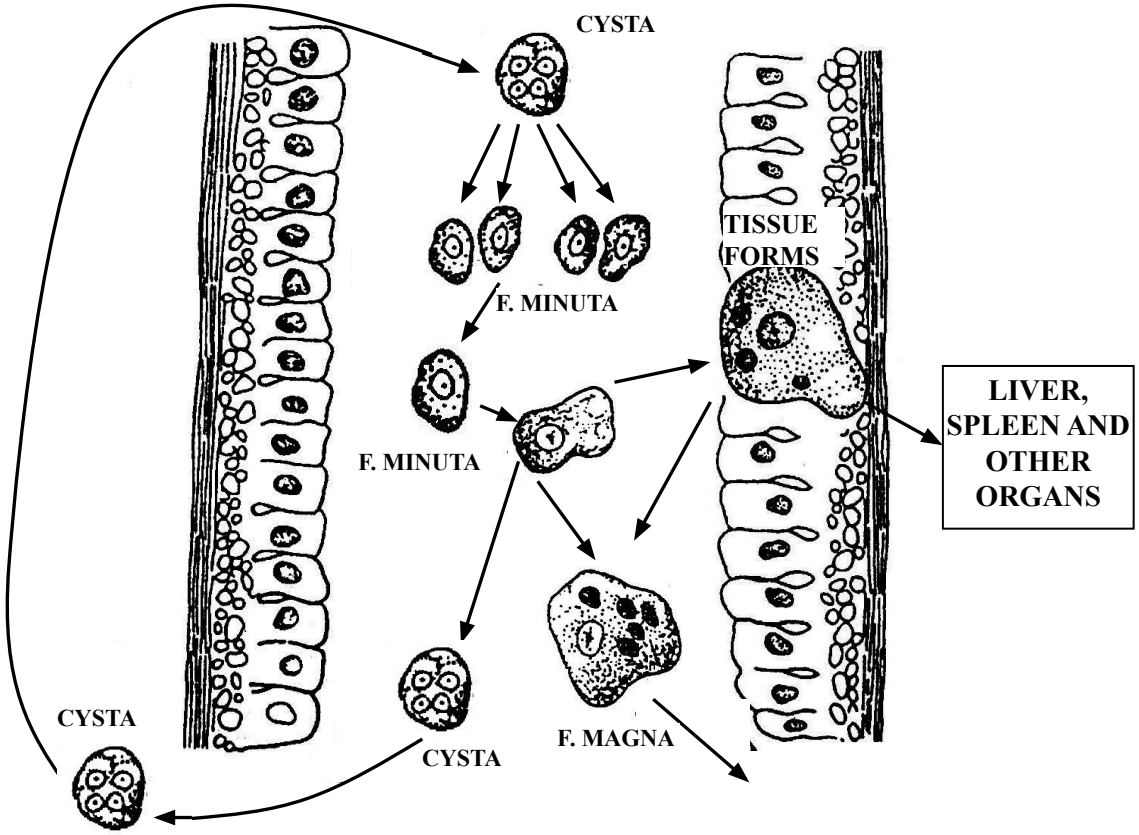
in asymptomatic carriers



These small vegetative forms feed on bacteria and cellular debris. They passively move with the intestinal contents. In the distal intestine, the small vegetative forms pass into the stage of cysts. Further, the cysts come out with feces. At this variant of the amoeba life cycle, humans do not get sick and they are cyst-carriers.

LIFE CYCLE OF ENTAMOEBA HISTOLYTICA

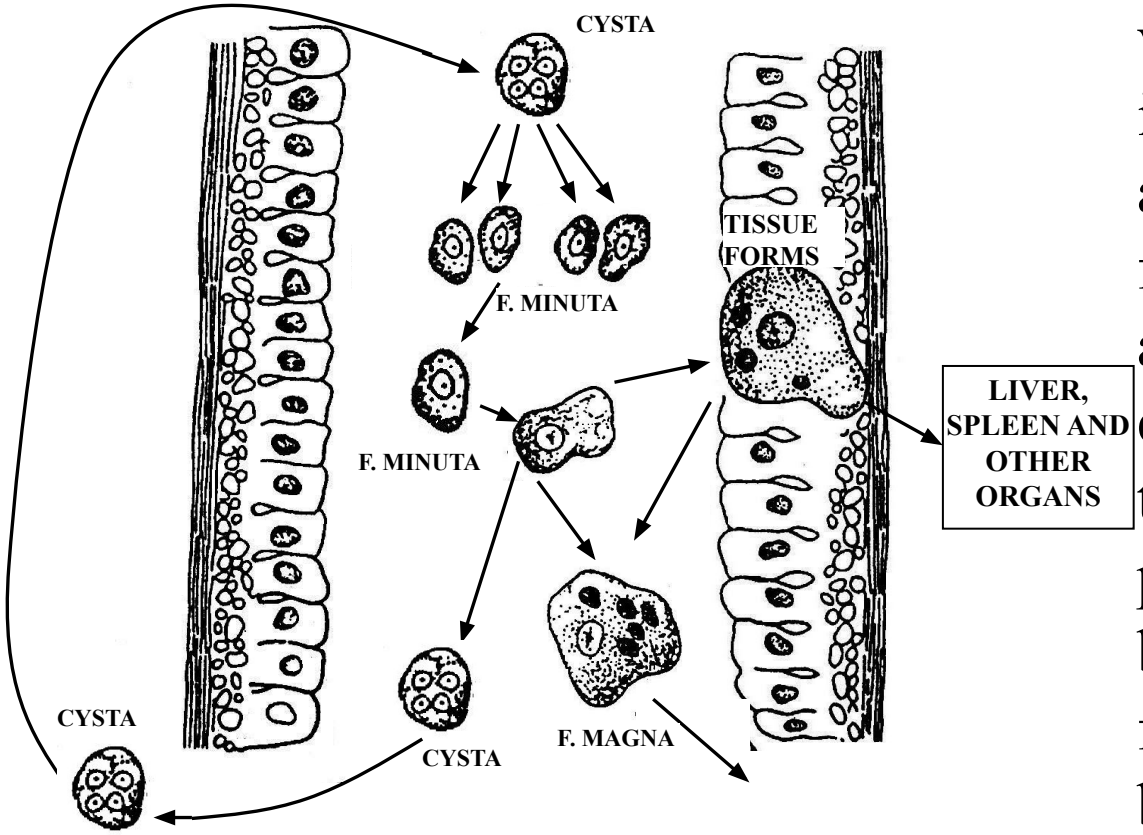
IN PATIENTS WITH AMEBIASIS



In a number of people with poor health, the small vegetative form develops into a large vegetative form, which destroys the intestinal wall, forms ulcers and causes bleeding. Such patients have a severe abdominal pain and frequent diarrhea mixed with blood (up to 15 times or more per day).

LIFE CYCLE OF ENTAMOEBA HISTOLYTICA

IN PATIENTS WITH AMEBIASIS



A certain part of the large vegetative forms penetrates into the intestinal mucosa and is converted into tissue forms. The tissue forms are also pathogenic. They enhance the destruction of the colon wall and can penetrate into the lumen of blood vessels. The tissue forms together with the bloodstream can penetrate into the liver, spleen and other organs and can cause their damage.

- Patients with amoebic dysentery must be hospitalized. In the absence of proper treatment, such patients have a variety of complications of the disease: anemia, dehydration, disturbance of electrolytic composition of the blood etc. These abnormalities may cause death. Spontaneous recovery rarely occurs.

Diagnostics of amebiasis

- During the acute form of the disease many forma magna with ingested erythrocytes are found in patient's feces.
- In the chronic form of the disease many cysts and a little of forma magna are found in patient's feces.
- During cysts-carriage many cysts are found in patient's feces.

PREVENTION

of amebiasis can be personal and public. The personal prevention is the activities that each patient should carry out himself.

RECOMMENDATIONS FOR PERSONAL PREVENTION:

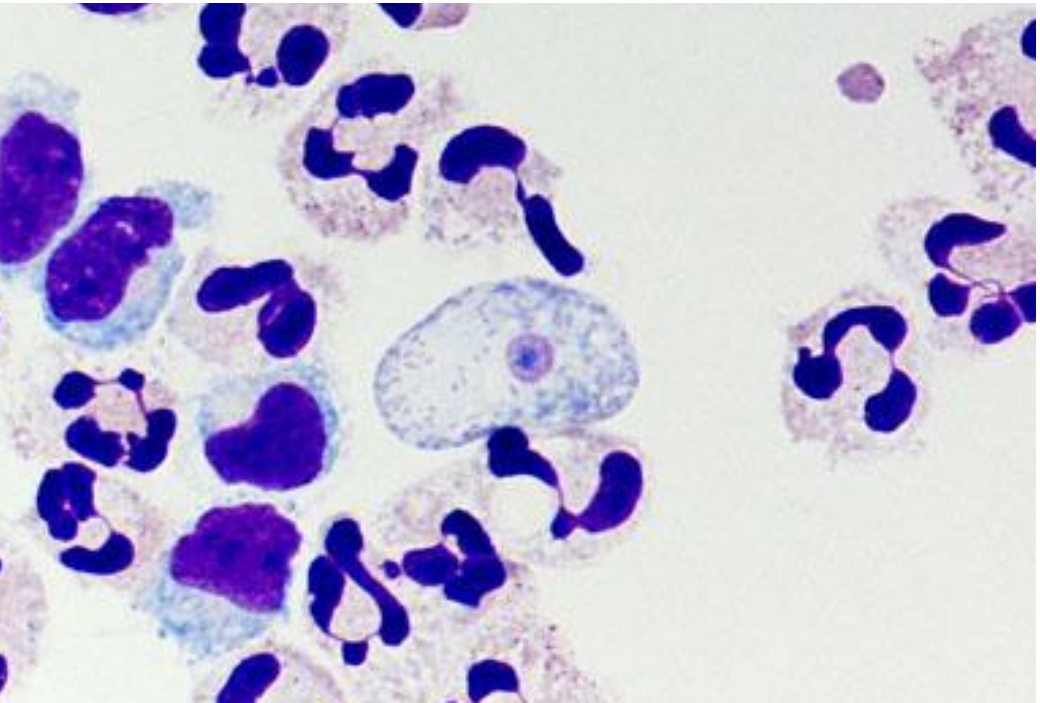
- use boiled water,
- wash hands before eating and after using the toilet,
- scald fruits and vegetables,
- protect products from flies and cockroaches.

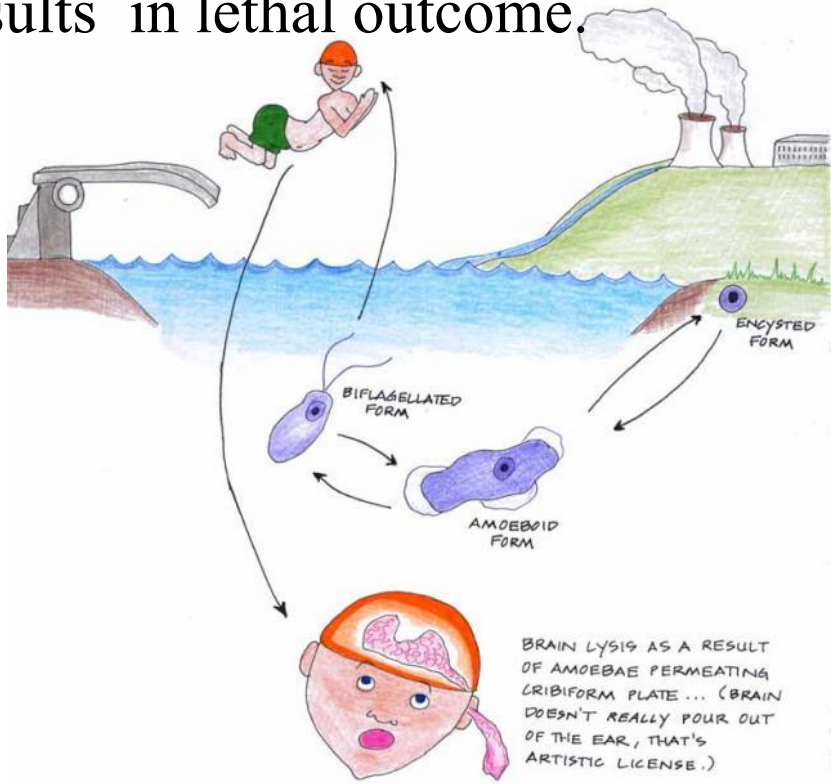
The public prevention is the measures which are carried out by a sanitary doctor.

- RECOMMENDATIONS FOR PUBLIC PREVENTION:
- closing of access to local water-sources,
- import of fresh water,
- identification and treatment of patients and humans, that are carriers of cysts,
- disinfection of water closets,
- sanitary-educational work in the community.

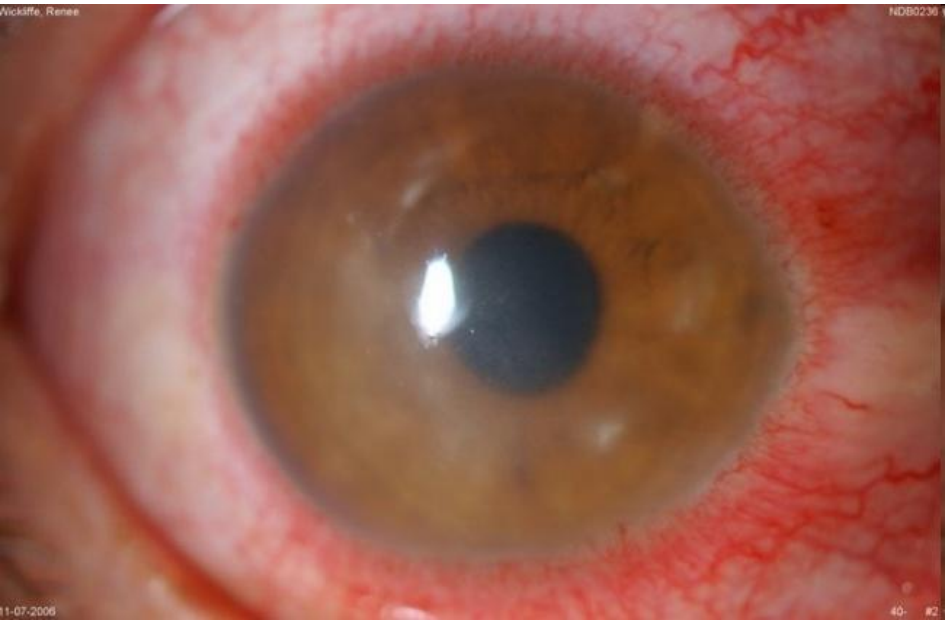
In phylum protozoa there are facultative parasites. These are Negleria fowleri and Acantamoeba castellani.

When a human being contacts with water (pond, lake, pool) and with earth, parasites penetrate into the blood and, further, into the cerebrospinal fluid. The parasites cause severe meningoencephalitis which usually results in lethal outcome.





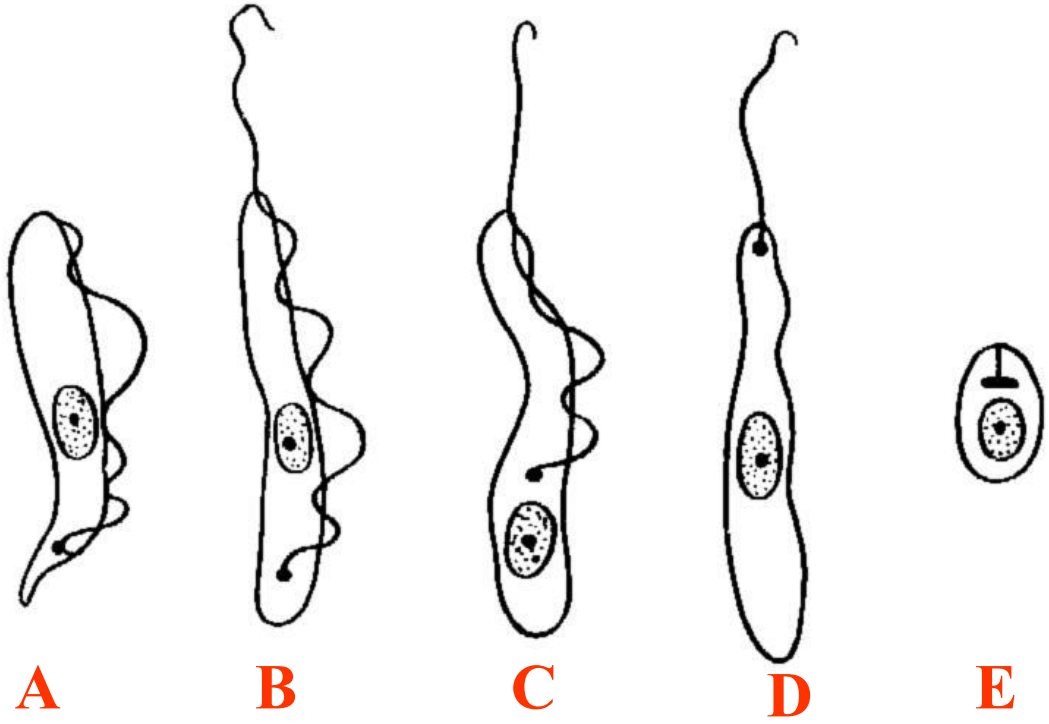
Амёбный менингоэнцефалит (*Acanthamoeba keratitis*) - острый гнойный менингоэнцефалит, проявляющийся поражение глазного яблока



CLASS
ZOOMASTIGOPHORA
(FLAGELLATA)

- All members of the flagellata class can be divided into two groups: parasites which have one flagellum and parasites which have many flagella.
- The parasites which have only one flagellum is called also the oro-intestinal and urogenital flagellata.
- The parasites which have many flagella are also called hemo-somatic flagellata.
- Our acquaintance with the parasites of class flagellata we will begin with oro-intestinal and urogenital flagellata.

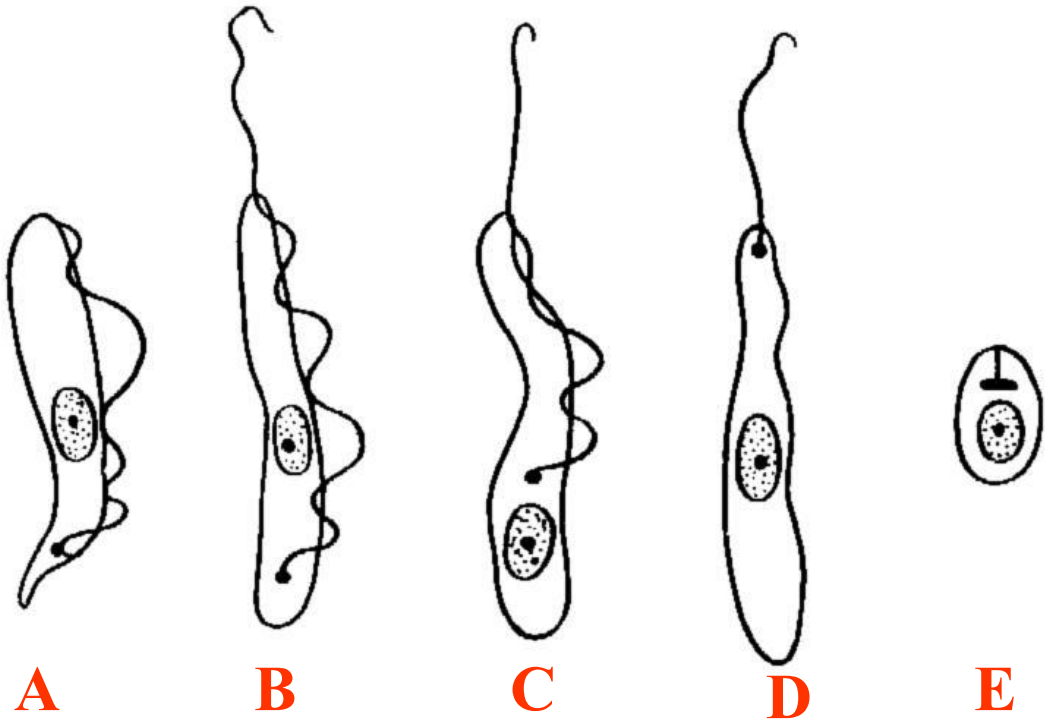
Flagellata with one flagellum have different life forms.



A- Metacyclic form; B- Trypomastigote (Trypanosomal) form; C- Epimastigote (Crithidial) form; D- Promastigote (Leptomonad) form; E- Amastigote (leishmania) form.

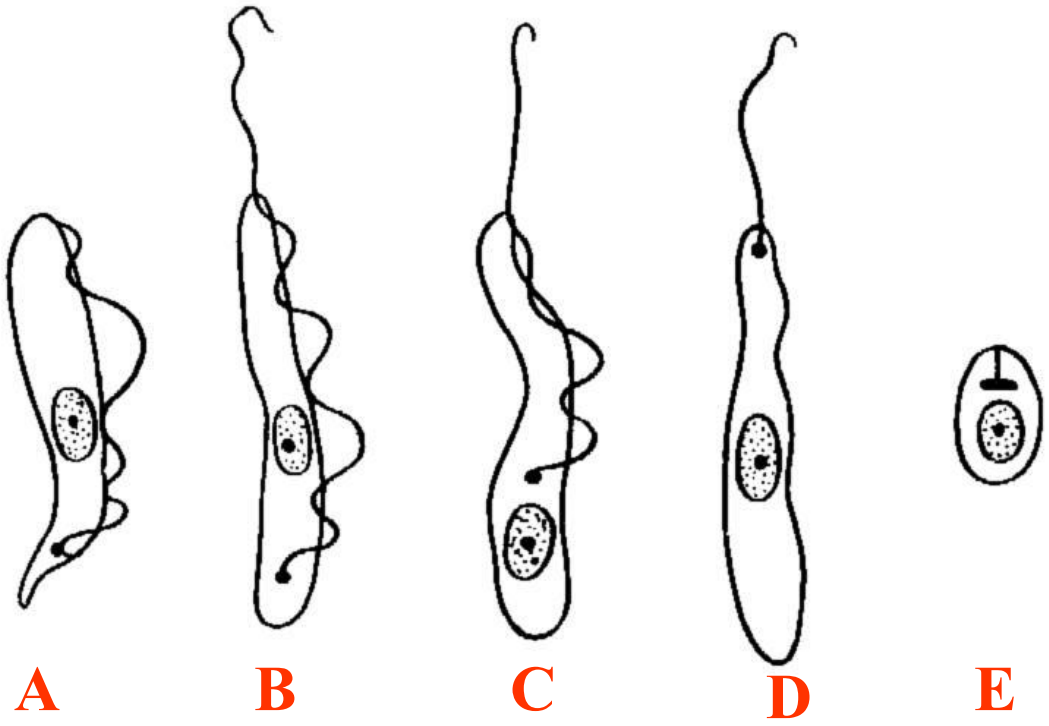
The different life forms of flagellates differ from one another by a cell shape, the presence or absence of an undulating membrane and a flagellum, as well as a kinetoplast localization (basal body).

E - Amastigote (Leishmania) form



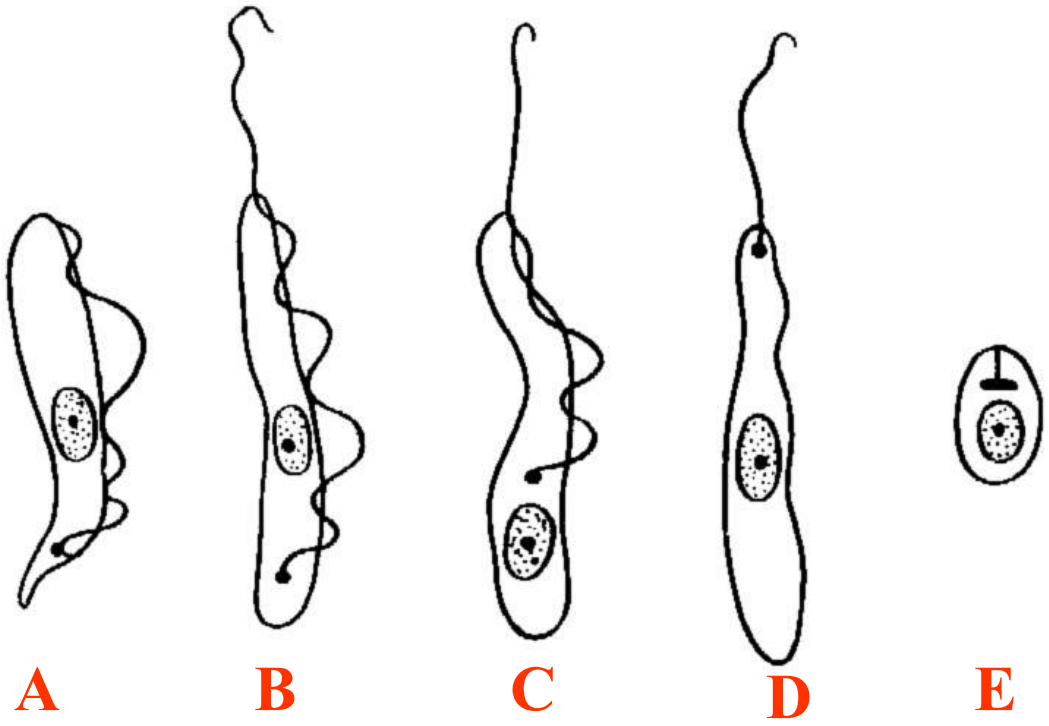
The amastigote form is an intracellular spherical form. It has no flagellum and has no undulating membrane. The amastigote form is the **intracellular form of all *leishmania* species and *Trypanosome cruzi*.**

D - Promastigote (Leptomonad) form



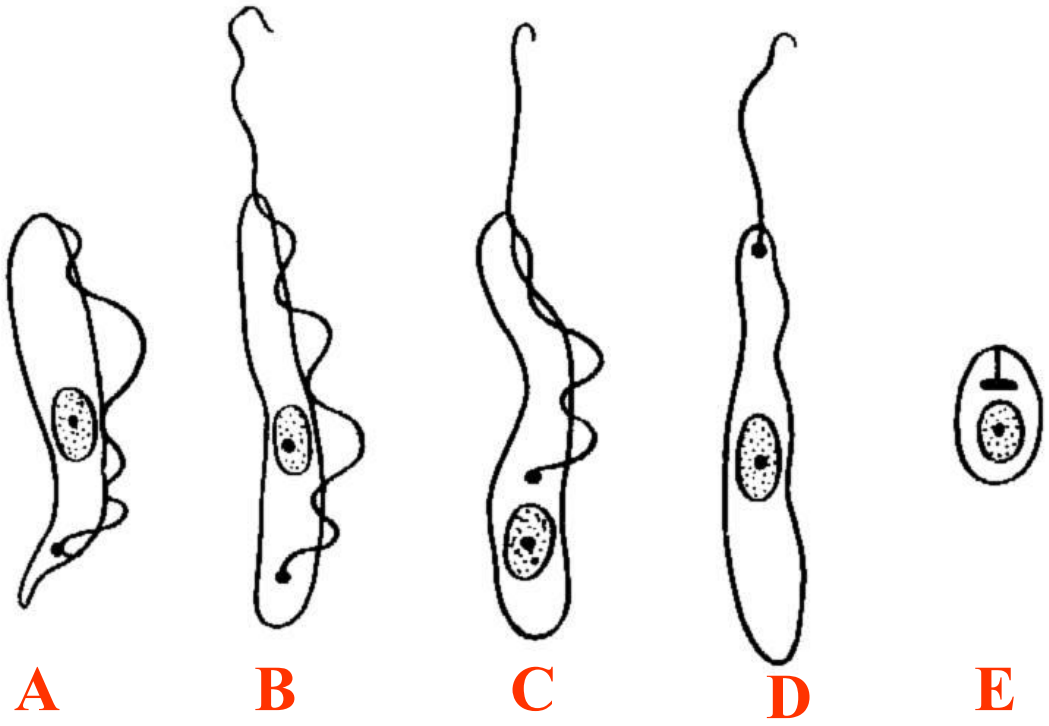
The promastigote form has an elongated body and a free flagellum without undulating membrane. The kinetoplast is in the anterior part. This form is found in the invertebrate host, and in the culture medium (of all L*eishmania* species) and in humans as a transitional form for *Tryponosoma cruzi*.

C- Epimastigote (crithidial) form



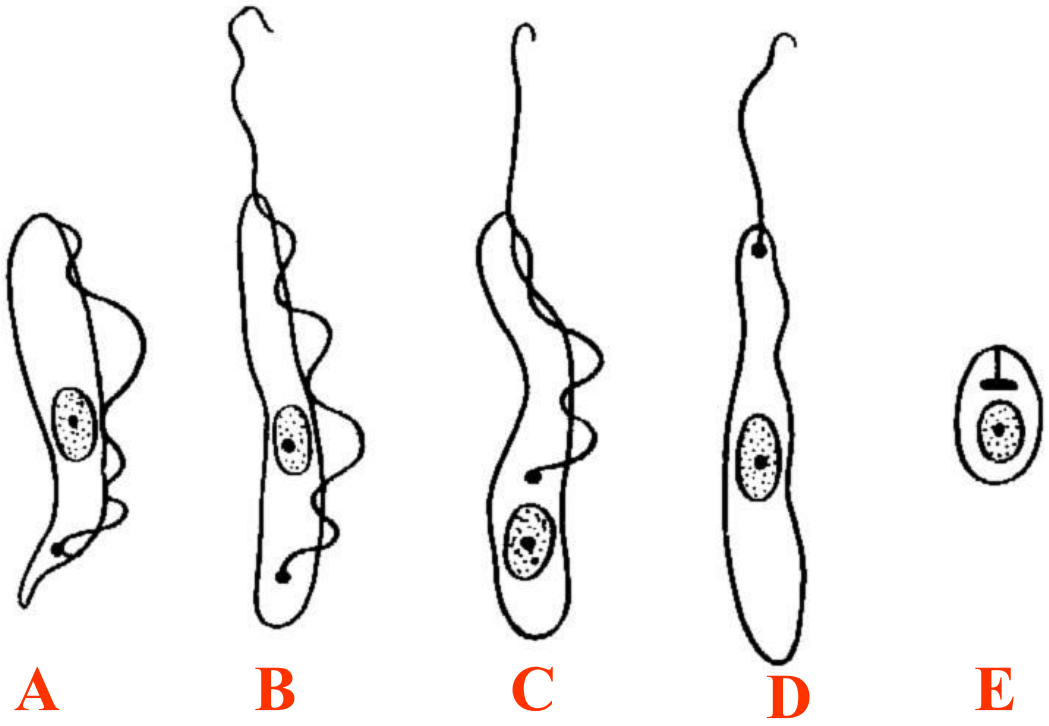
The epimastigote form has an elongated body, single free flagellum and a short undulating membrane. The kinetoplast is anterior to the nucleus. The epimastigote forms are found in the invertebrate host and in the culture medum (of Trypanosome species).

B- Trypomastigote (Trypanosomal) form



The trypomastigote form has an elongated body, single free flagellum, and a long undulating membrane. The kinetoplast is located behind the nucleus at the posterior end of the body of the parasite. This form is found in the peripheral blood of vertebrates and is a diagnostic stage of Trypanosome species.

A- Metacyclic form



The metacyclic form is morphologically similar to trypomastigote stage but it has no free flagellum. It is the final developmental stage in the guts of insect carriers and is an invasive stage for the transmission from an insect to humans.

The causative agents of leishmaniasis

Causative agents of leishmaniasis are members of the genus Leihmania species.

All leishmaniasis can be divided into three groups:

Cutaneous leishmaniasis

Mucocutaneous leishmaniasis

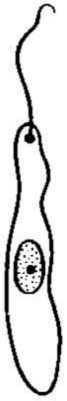
Visceral leishmaniasis

Different types of leishmaniasis are caused by different Leishmania.

L. tropica major
L. tropica minor
L. mexicana
L. brasiliensis

L. brasiliensis brasiliensis

L. donovani
L. infantum

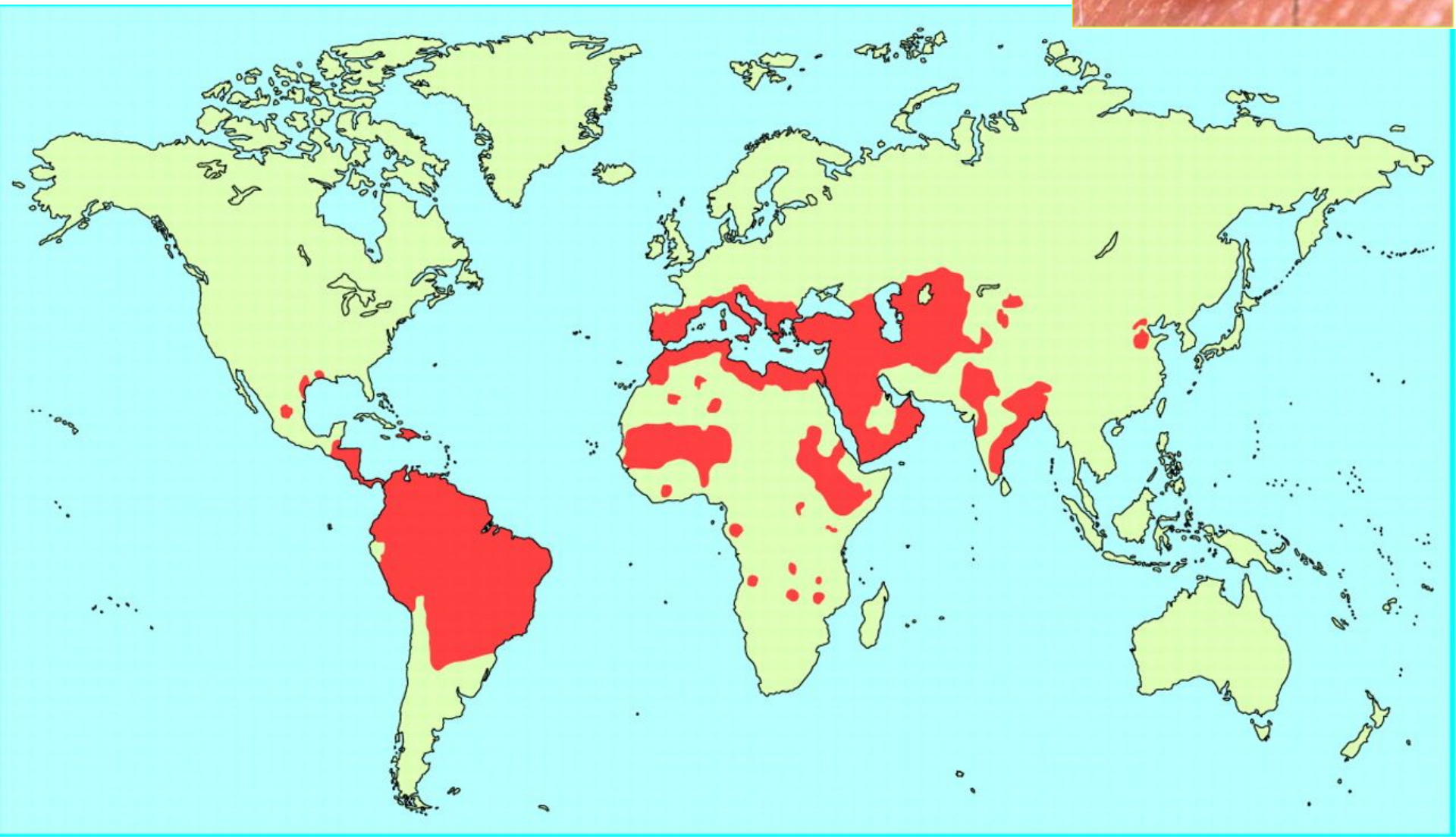


All Leishmania have only

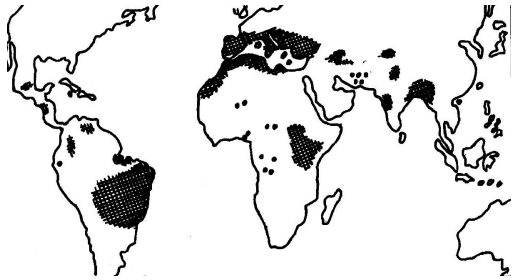
Promastigote and **Amastigote** 

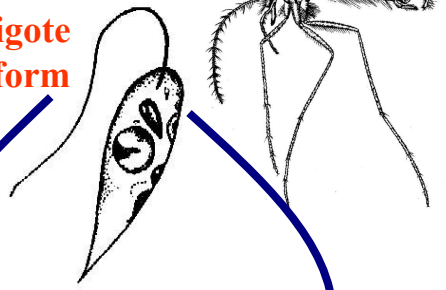
life forms

The main foci of leishmaniasis.



The life cycle of the causative agent of cutaneous leishmaniasis on the example of L. tropica minor





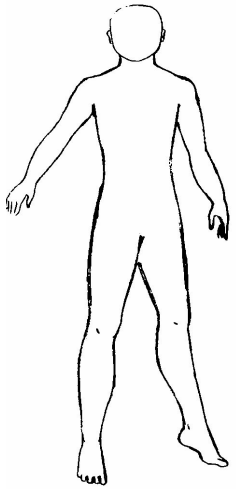
genus: female sandfly Flebothomus

Promastigote form

specific vector (carrier)

Intermediate and reservoir hosts

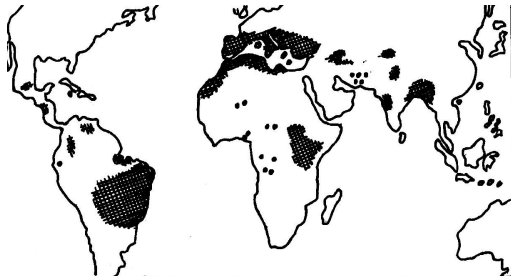
The parasite carriers are female sandy flies. Promastigote forms are located in the guts of sandy flies. The carrier inoculates promastigotes into the cutaneous tissue of the intermediate and reservoir host while sucking blood.

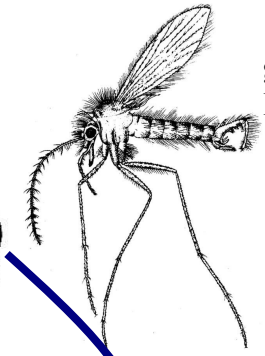


Amastigote form

The intermediate host of **L. tropica minor** is a human being. The reservoir hosts are various rodents such as mice, hamsters, gophers, gerbils, rats. Amastigote forms of the parasite are localized in the endothelial cells of the dermis and hypodermis of mammals.

The life cycle of the causative agent of cutaneous leishmaniasis by the example of L. tropica minor





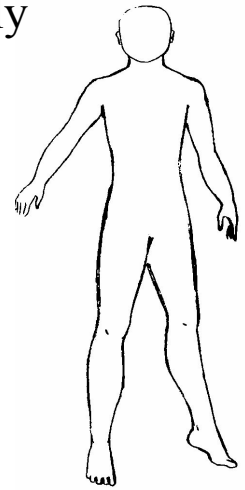
genus: female sandfly Flebothomus

Promastigote form

specific vector (carrier)

Intermediate and reservoir hosts

The amastigotes multiply and are ingested by a female sandfly carrier when it sucks blood.



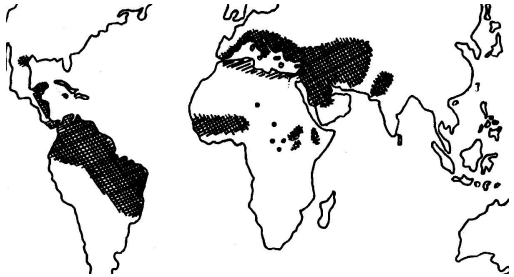
Amastigote form

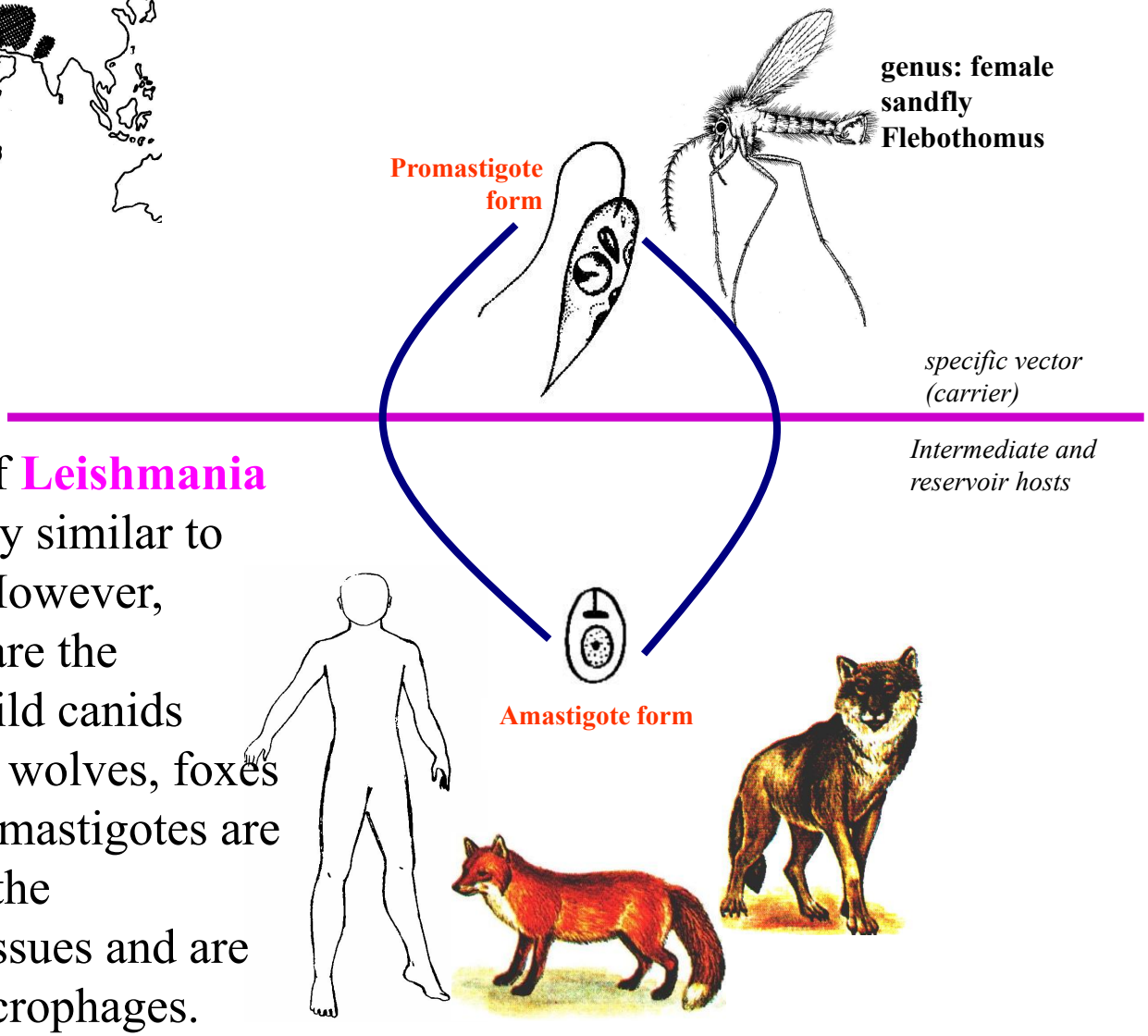
The amastigotes become promastigote (flagellated) in the midgut of the sandfly. The promastigotes multiply and fill the gut of the insect carriers. If the sandfly bites a human, promastigote forms penetrate into the skin of a human being.

At the place of a bite, there develops dry painless ulcer, 25-70 mm in diameter, usually self-healing after 1-2 years, often leaving a disfiguring scar. The infection usually spontaneously heals and forms long-lasting immunity to reinvasion.

THE CUTANEOUS LEISHMANIASIS

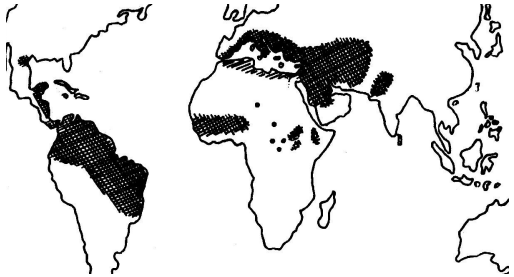
The life cycle of the causative agent of visceral leishmaniasis on the example of L. donovani

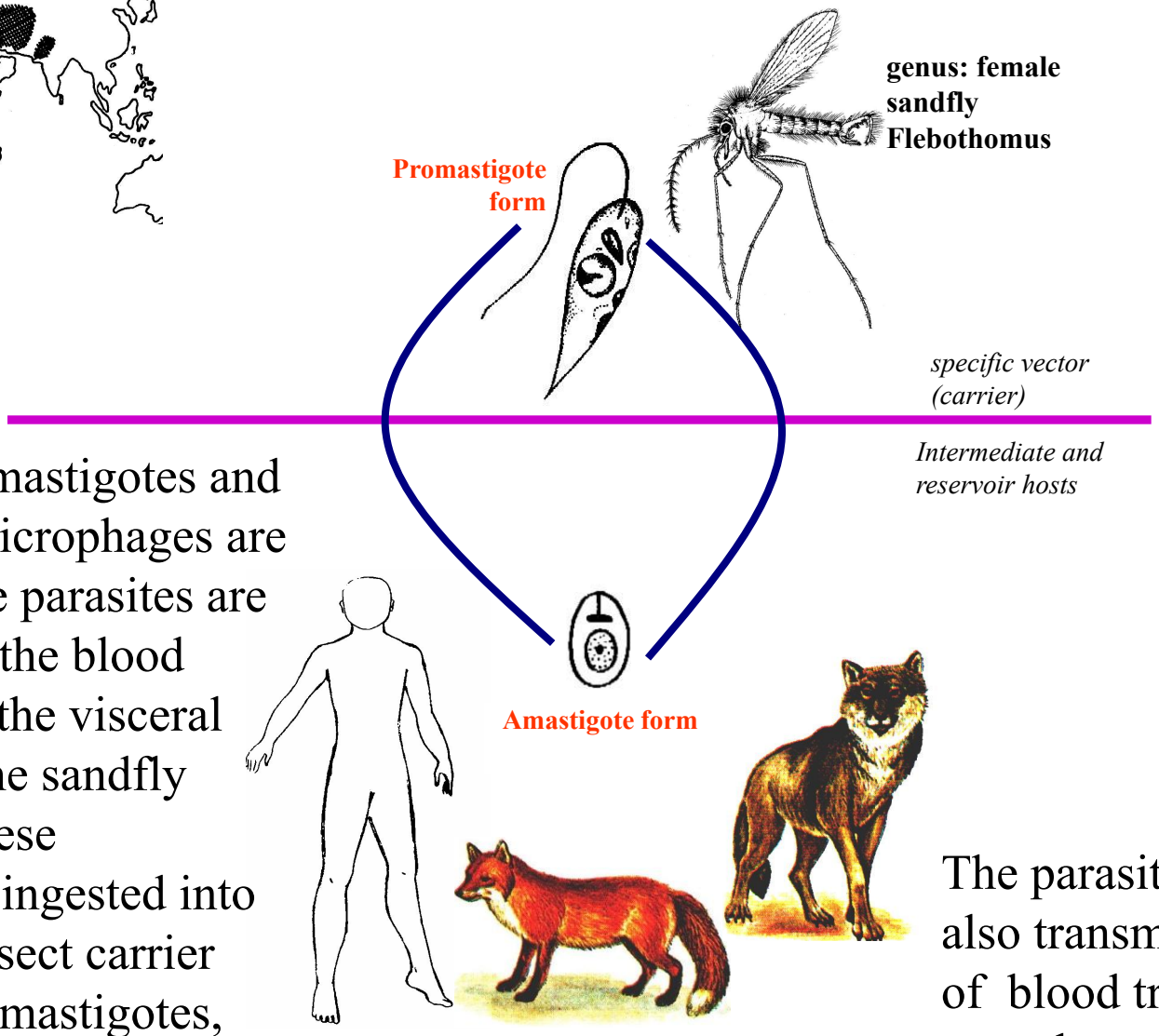




The life cycle of **Leishmania donovani** is very similar to the L. tropica. However, reservoir hosts are the domestic and wild canids (canines): dogs, wolves, foxes and jackals. Promastigotes are inoculated into the subcutaneous tissues and are captured by macrophages.

The life cycle of the causative agent of visceral leishmaniasis on the example of L. donovani





They become amastigotes and multiply. The microphages are invaded and the parasites are carried through the blood circulation into the visceral organs. When the sandfly sucks blood, these amastigotes are ingested into the gut of the insect carrier and become promastigotes, and then they multiply.

The parasites can be also transmitted by way of blood transfusion, sexual contact.

Leishmania braziliensis braziliensis

- Geographical Distribution: Tropical forests of South America and Central America.
- Reservoir hosts are rodents and some domestic animals.
- Habitat: Amastigote: in the reticulo-endothelial cells of muco-cutaneous tissues of the nose, mouth, lips, larynx. Promastigote: in the gut of Lutzomyia sandflies
- Life cycle: Lutzomyia sandflies are the main carriers, and man acquires infection from an enzootic area.
- Pathology: Mucocutaneous leishmaniasis (espundia). Chronic ulceration of the mucus membrane of the mouth nose, throat, etc. with the destruction of bones and cartilages.

Laboratory Diagnosis of Leishmania species:

Cutaneous and mucocutaneous leishmaniasis:

- **1. Amastigotes are revealed in stained smears taken from ulcers**
- **2. Promastigotes are revealed in the culture medium.**
- **3. Immunologic tests are used for this purpose.**

Visceral leishmaniasis:

- **1. Amastigotes are revealed in the bioptates of the spleen, bone marrow, the enlarged lymph nodes, and in the peripheral blood monocytes.**
- **2. Promastigotes are revealed in the culture medium**
- **3. Immunologic tests are used for this purpose. .**

PREVENTION

- RECOMMENDATIONS FOR PUBLIC PREVENTION :
- Treatment of infected individuals,
- Destruction of specific carriers,
- Destruction of reservoir hosts,
- Health education in the community
- RECOMMENDATIONS FOR PERSONAL PREVENTION :
- Avoiding endemic areas,
- Avoiding insect bites.

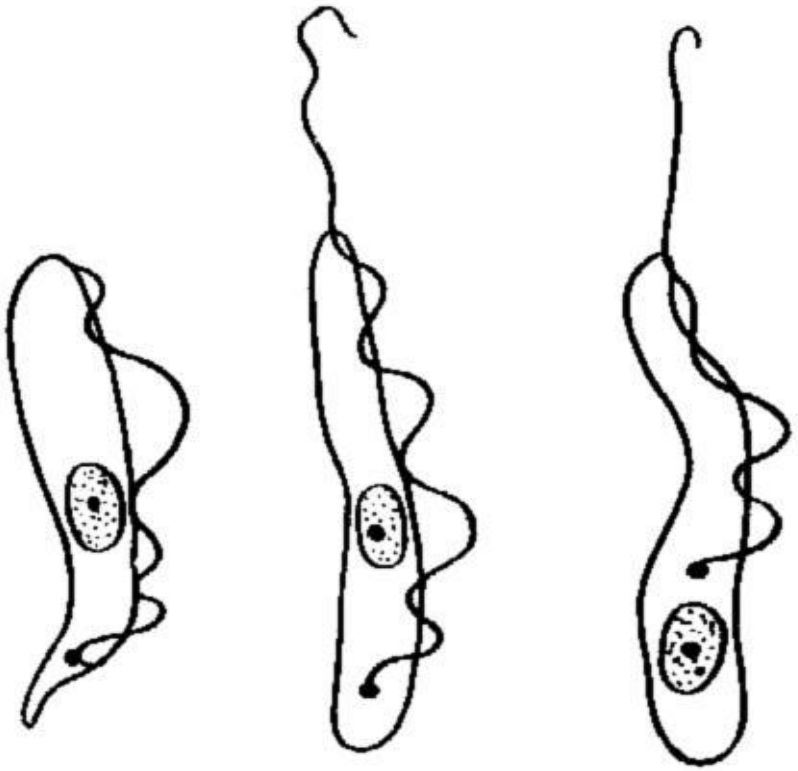
The causative agents of African sleeping sickness (African trypanosomiasis)

Causative agents of African sleeping sickness are members of the species Tripanosoma brucei.

There are two subspecies of Tripanosoma brucei, which are pathogenic for humans: ***Tripanosoma brucei gambiense*** and ***Tripanosoma brucei rhodesiense.***

Tr. b. gambiense – is the cause of the West African variant (chronic trypanosomiasis or Gambian version)
Tr. b. rhodesiense - is the cause of the East African variant (acute trypanosomiasis, Rhodesian version)

The causative agents of African sleeping sickness have three forms of life:

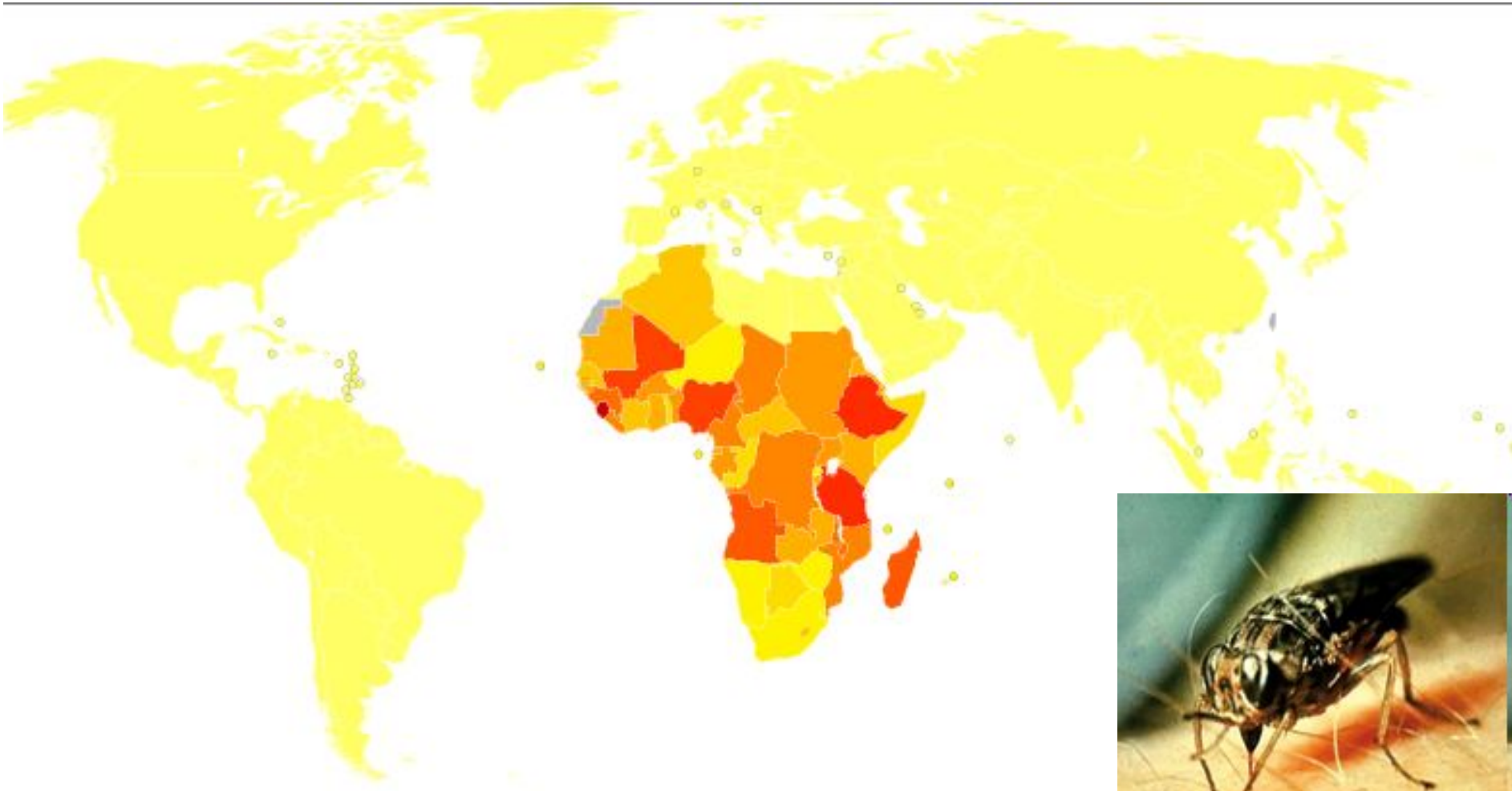


Metacyclic form

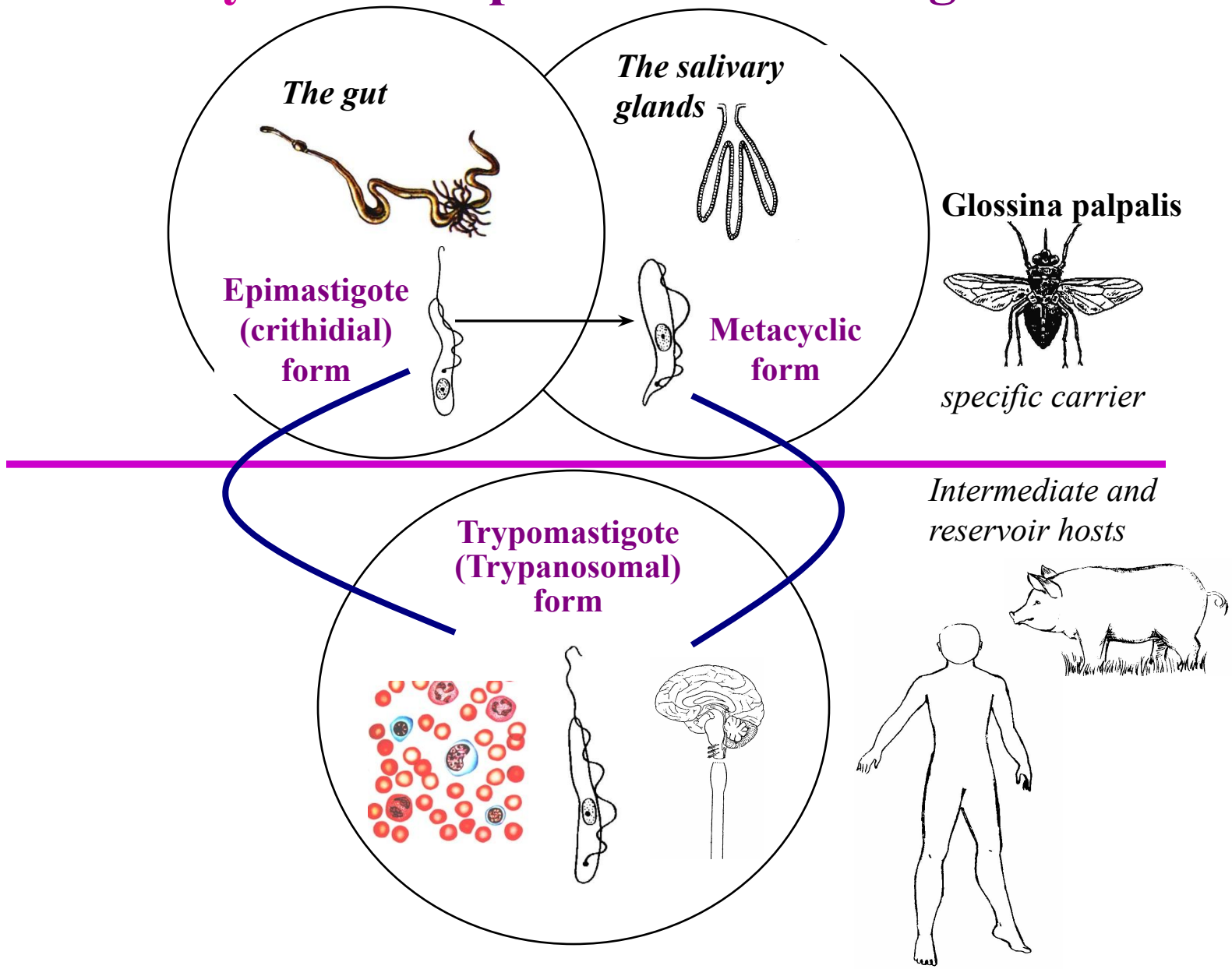
Trypomastigote (Trypanosomalform)

Epimastigote (crithidial) form

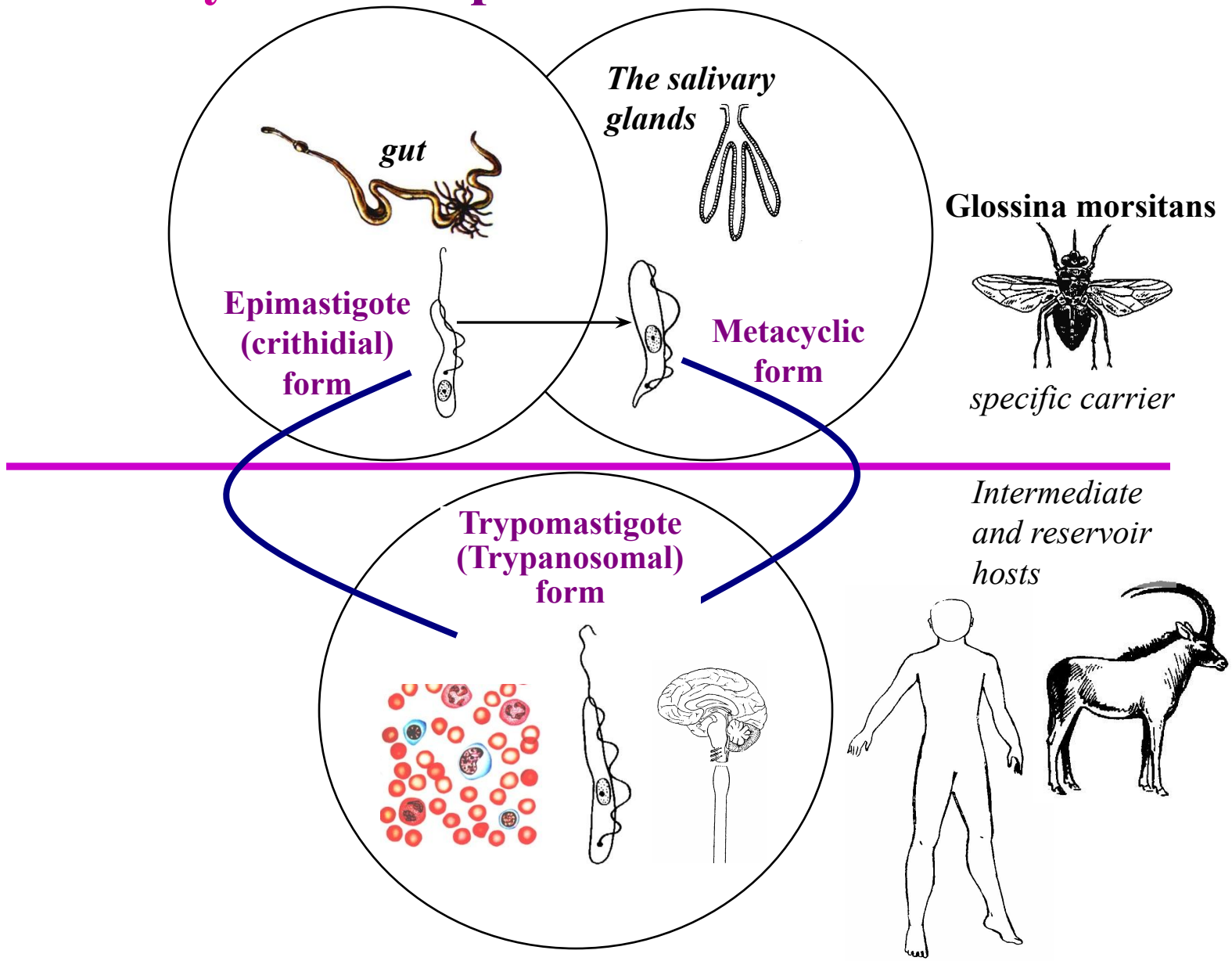
The African sleeping sickness is a natural-focal disease. It is typical of the East, West, and Central Africa, extending from Senegal across to Sudan and down to Angola.



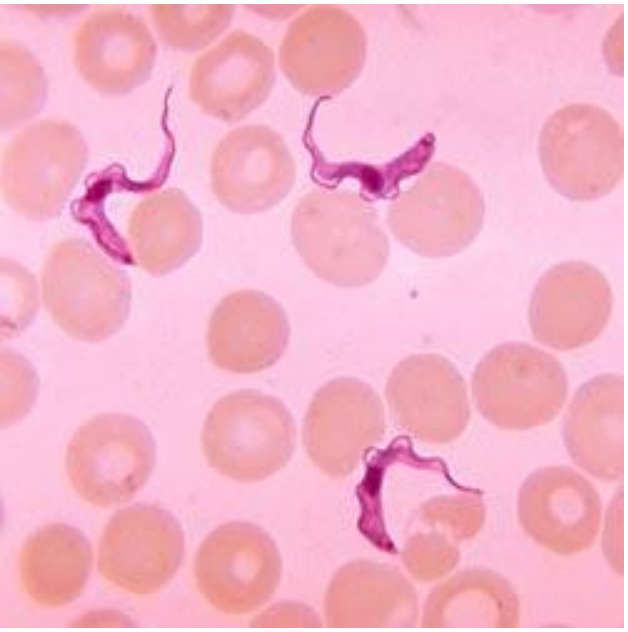
The life cycle of Tripanosoma brucei gambiense



The life cycle of Tripanosoma brucei rhodesiense



Patients with African sleeping sickness

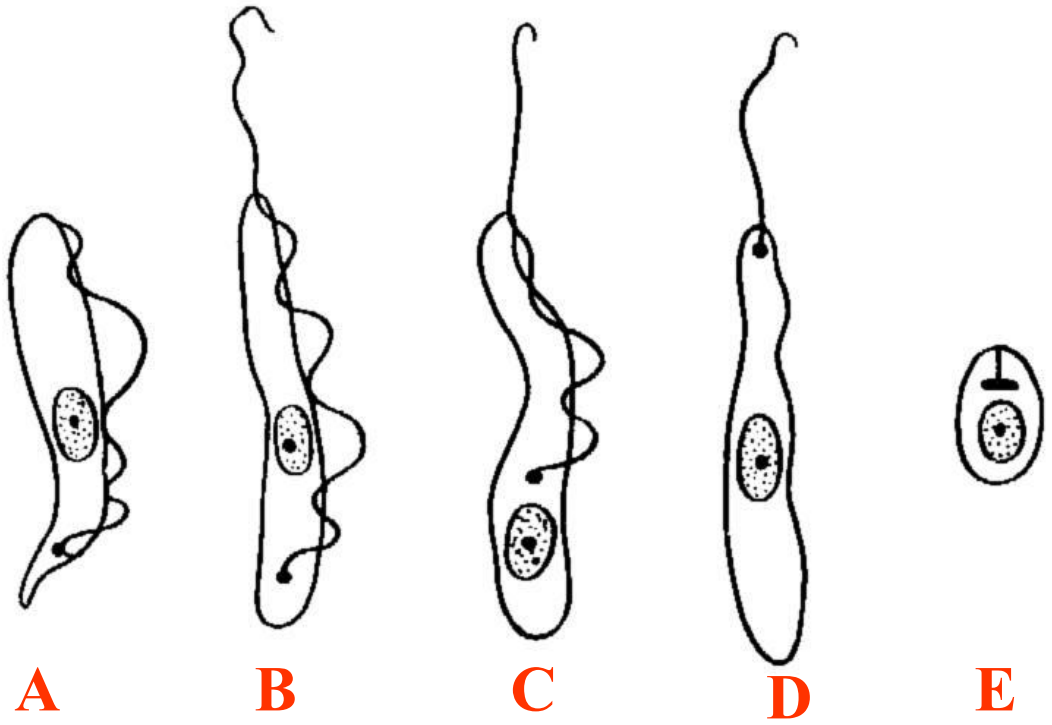


They are not physically active. They stay for a long time in a typical pose, and are sleepy.

African sleeping sickness

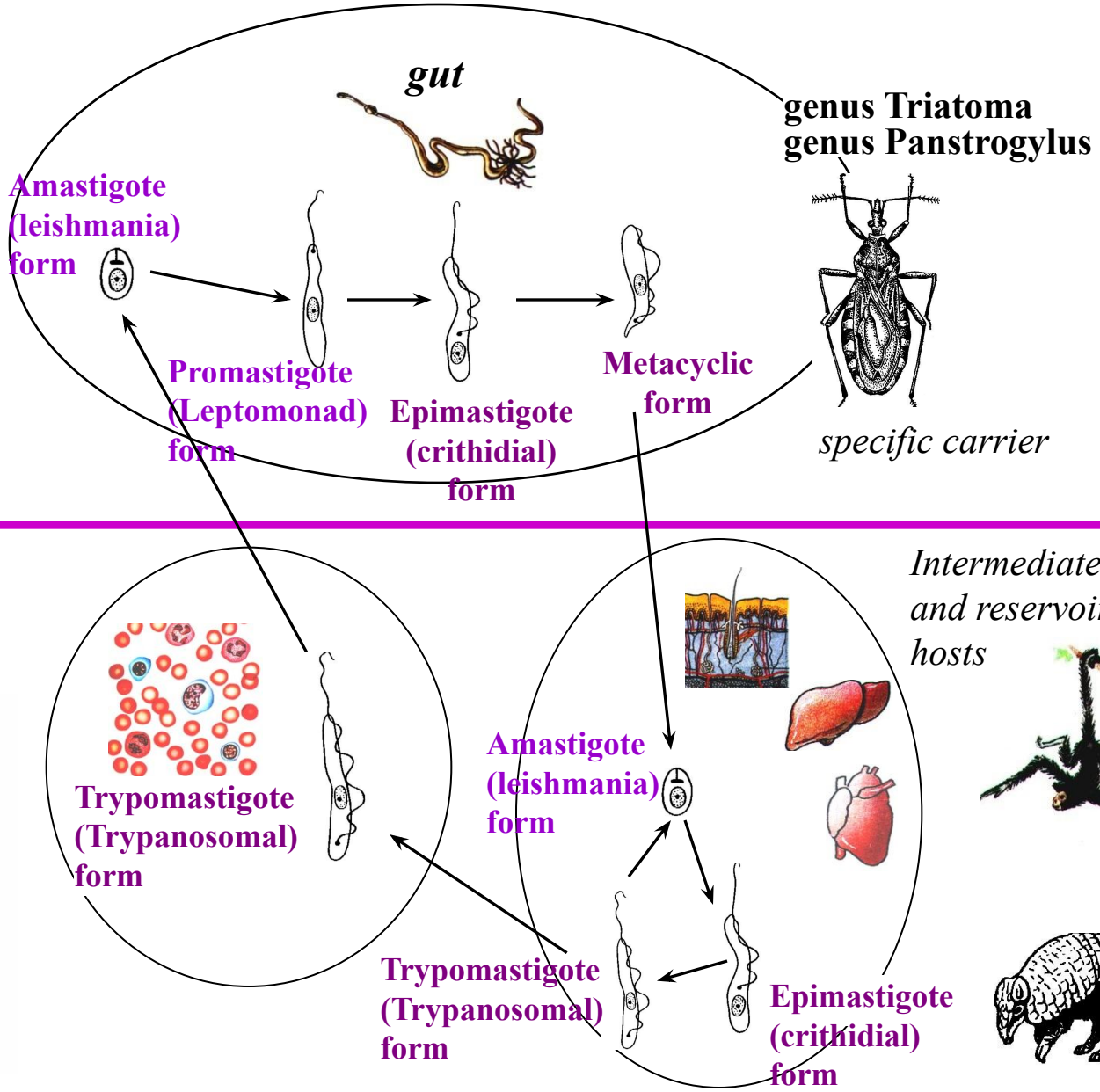
THE CAUSATIVE AGENT OF LATIN-AMERICAN TRYPANOSOMIASIS (CHAGOS` DISEASE) IS **TRIPANOSOMA CRUSI**

IT HAS FIVE LIFE FORMS



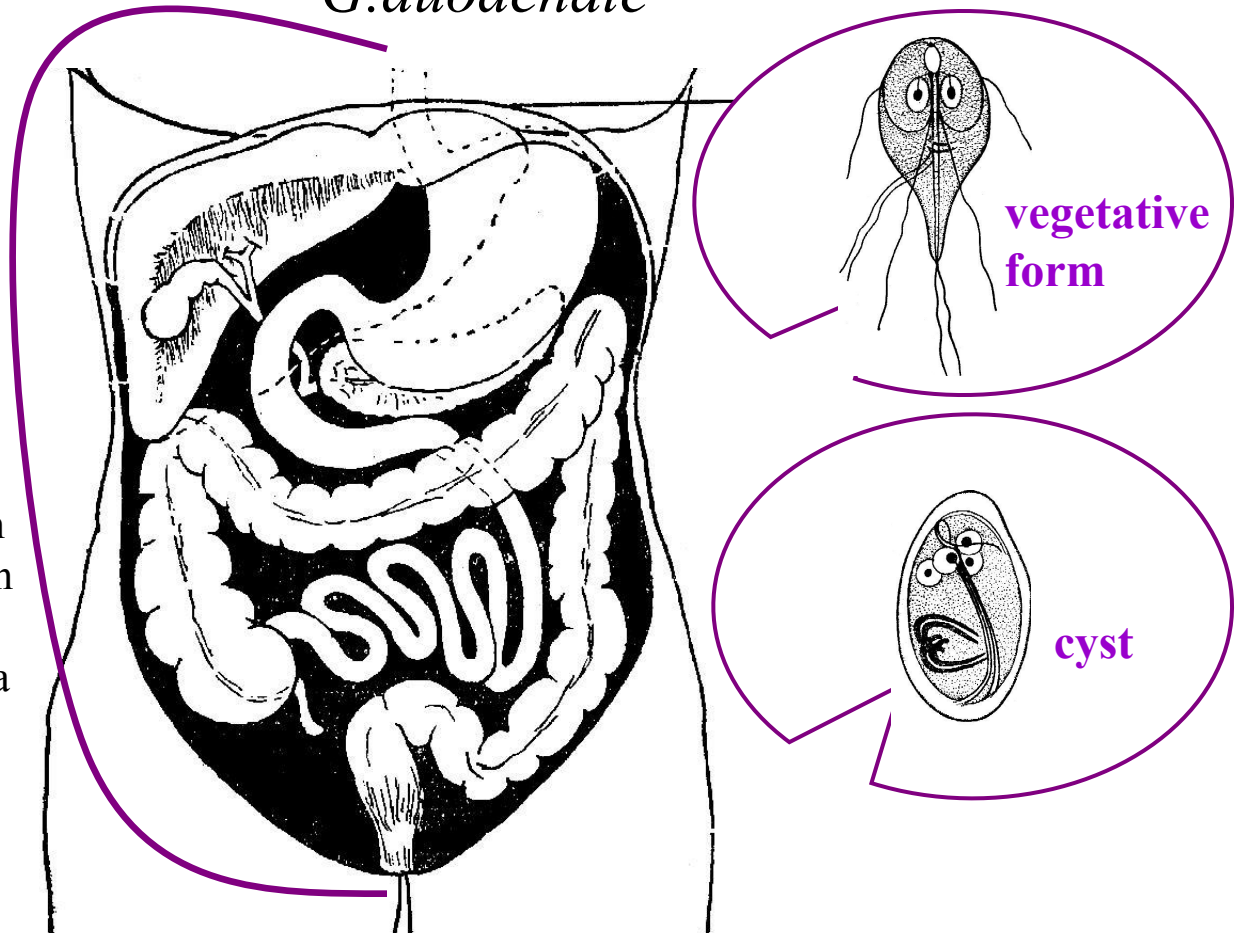
A- Metacyclic form; B- Trypomastigote (Trypanosomal) form; C- Epimastigote (crithidial) form; D- Promastigote (Leptomonad) form; E- Amastigote (leishmania) form.

The life cycle of Tripanosoma crusi



Lamblia intestenalis

This species is also called *Giardia intestinalis* or *G.duodenale*

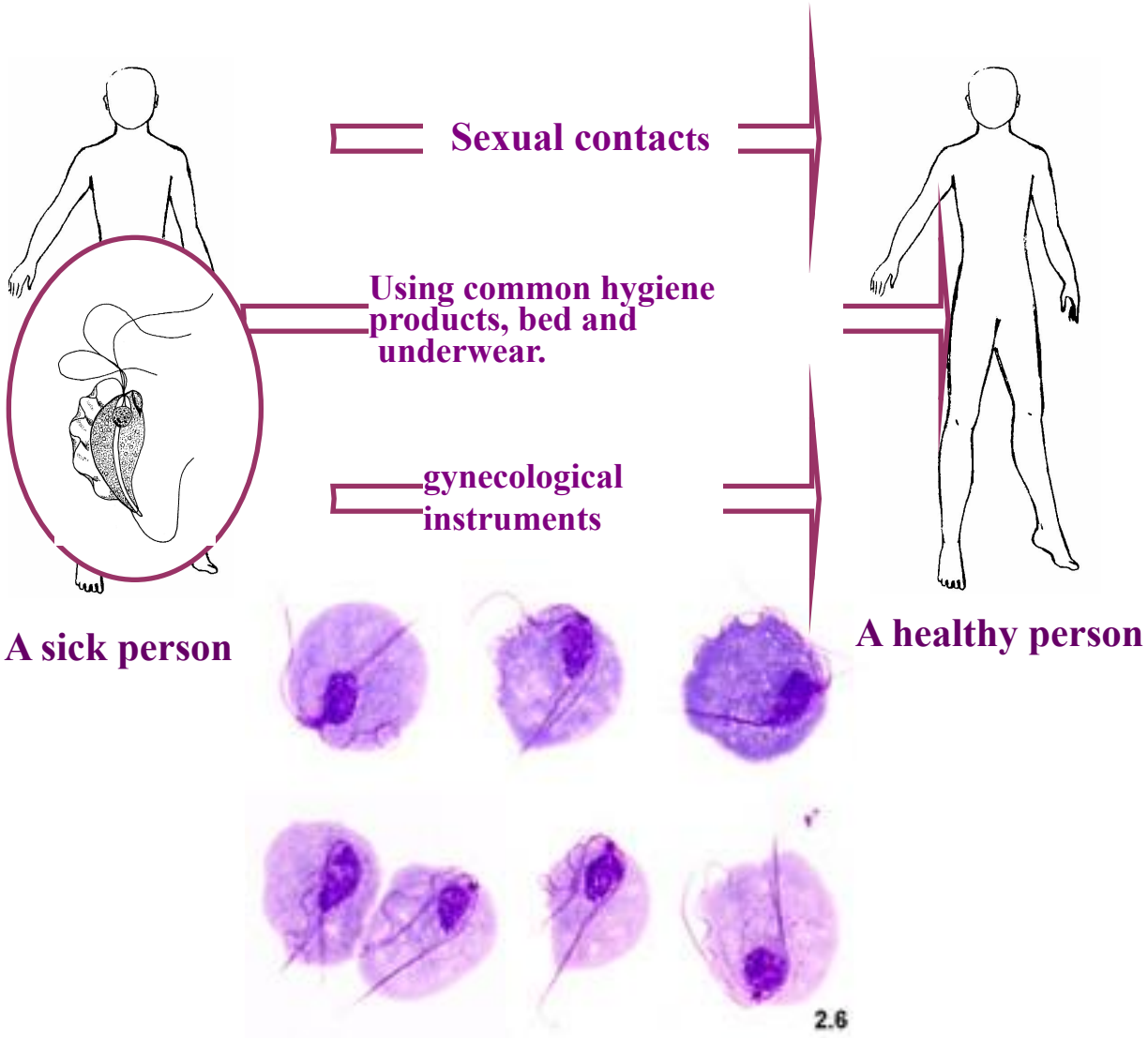


Geographical Distribution: The cosmopolitan distribution in a warm climate is more prevalent in children than in adults. This unicellular organism from the class of flagellata is most commonly diagnosed in the human intestinal tract. Its high prevalence occurs in young, malnourished children in large families, orphan asylums, and elementary schools.

Habitat: Upper parts of the small intestine, mainly in the duodenum and jejunum.

Manifestations of lambliosis

Trichomonas vaginalis



TRICHOMONAS HOMINIS