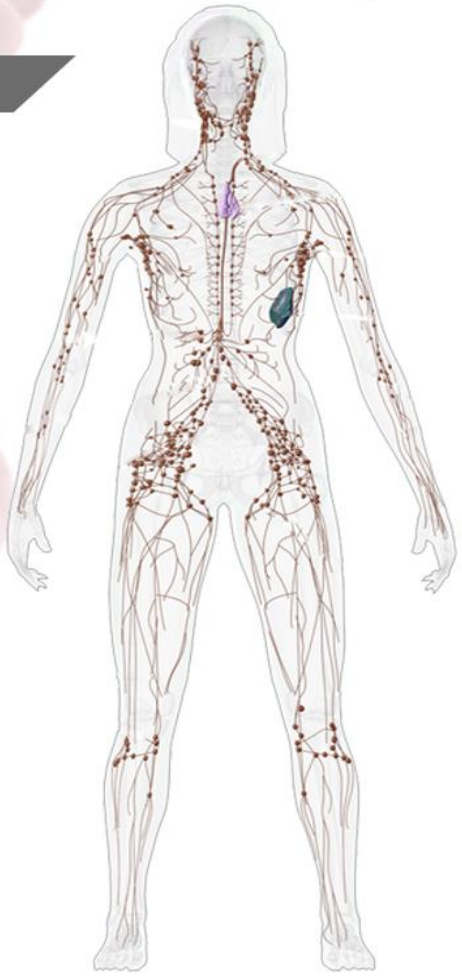
Sheet 2

Hematology and Lymphatic system



Subject | Microbiology

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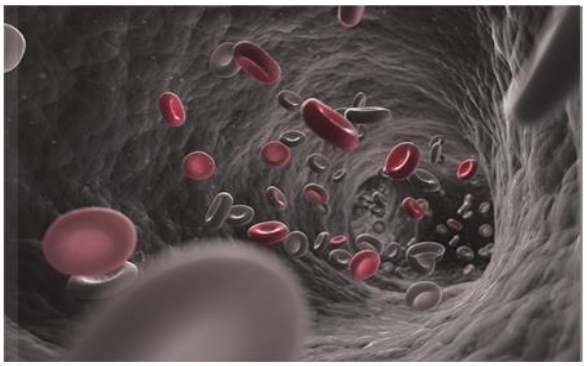


Haemflagellates

Trypanosoma & Leishmania

- ***Haemflagellate*** is subtype of ***protozoa***.
- They have a ***single flagellum*** which helps them moving.
- No sexual reproduction, ***binary fission*** instead.
- 2 examples of Haemflagellates:
  - ***Trypanosoma*** (the causative agent of ***trypanosomiasis***)
  - ***Leishmania*** (the causative agent of ***leishmaniasis***)

- Morphological, Trypanosoma and Leishmania fall under the order ***Kinetoplastida*** as they contain ***kinetoplasts*** in their developmental stages which flagella and undulating membrane arise from later on.

- Notice the developmental stages: Amastigote ➔ Promastigote ➔ Epimastigote ➔ Trypomastigote

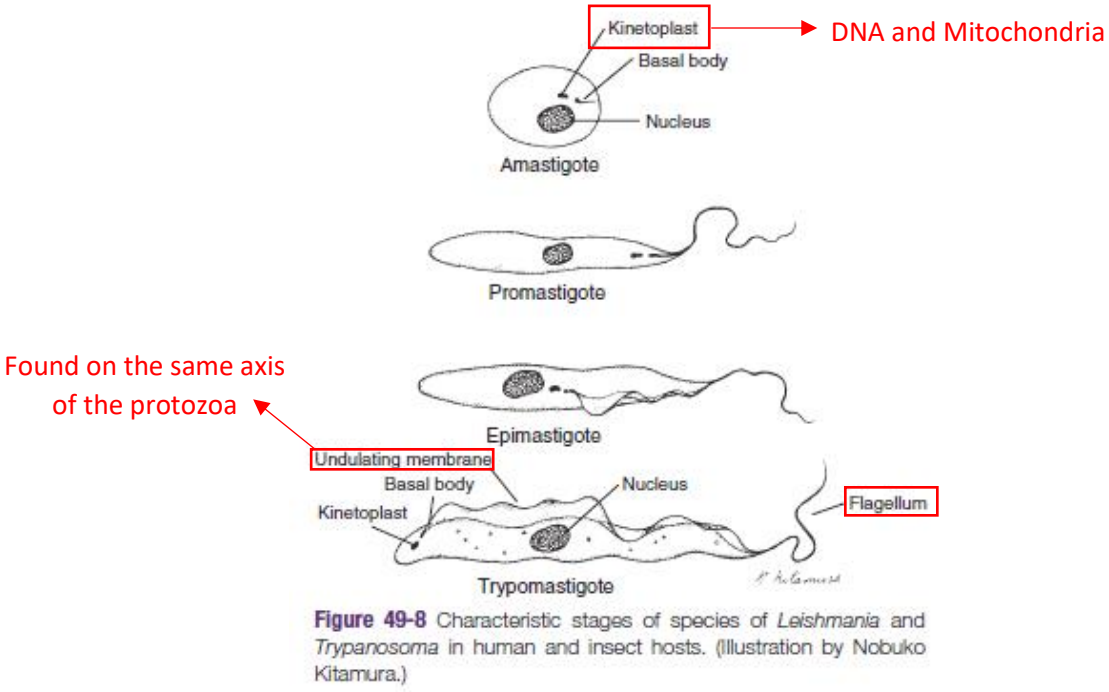


Figure 49-8 Characteristic stages of species of *Leishmania* and *Trypanosoma* in human and insect hosts. (Illustration by Nobuko Kitamura.)

Note: this is just an overlook there are a lot of differences between each organism or even the subtypes of the same organism.

Trypanosoma:

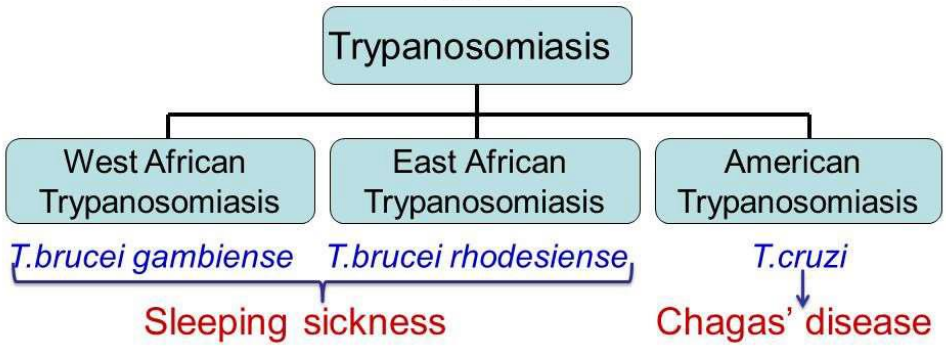
- ***Trypanosoma brucei complex (Gambiense and Rhodesiense)*** is the causative agent of African Trypanosomiasis (***African sleeping sickness***) in humans.
- ***Trypanosoma cruzi,*** the causative agent of American trypanosomiasis ***(Chagas disease)*** which occurs in humans and many vertebrate animals in Central and South America.

- Trypanosomiasis is a vector borne disease
  - African Trypanosomiasis is transmitted by the ***Glossina species "Tsetse fly"*** of both sexes.
  - American Trypanosomiasis is transmitted by the ***Triatominae, or "kissing bugs".***

Tsetse fly.

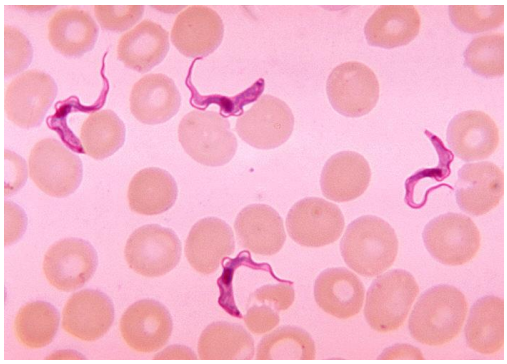
Triatominae.

- Trypanosomiasis classification according to geographical disease and vector distribution:



Morphology:

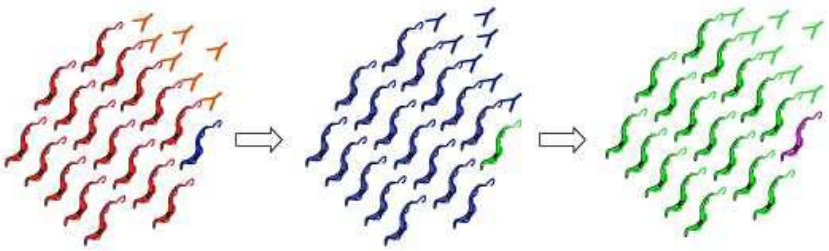
- The morphologically differentiated forms include spindly, uniflagellate stages (Trypomastigote, Epimastigote, Promastigote) and a rounded intracellular Amastigote form.
- In general, Trypomastigote and Amastigote forms can be found in humans while the Epimastigote and the Promastigote forms can be found in vectors.
- In African trypanosomiasis there is no intracellular form (Amastigote), it is only in the American Trypanosomiasis (preferably cardiac myocytes, that's why American Chagas patients suffer from cardiac problems).



African Trypanosomiasis (extracellular)

Antigenic variation of Trypanosoma brucei:

- A unique feature of African trypanosomes is their ability to change the antigenic surface coat of the outer membrane of the trypomastigote, helping to evade the host immune response.
- The trypomastigote surface is covered with a dense coat of ***variant surface glycoprotein (VSG),*** those VSGs are controlled by more than a hundred genes that forms a mosaic of them in which the antigens keep changing as soon as the immune system starts recognizes them.
- Each time the antigenic coat changes, the host does not recognize the organism and must mount a new immunologic response.

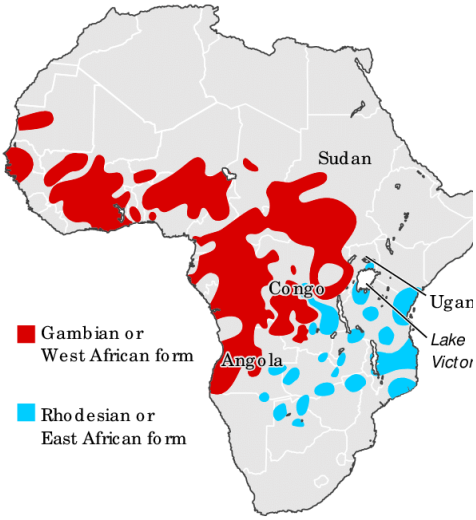


African Trypanosomiasis:

- T. brucei rhodesiense: East African trypanosomiasis, ***it is acute in nature, progresses faster, less frequent and animals are the reservoir.***
- T. brucei gambiense: West African trypanosomiasis, ***it chronic in nature, progresses slower, commoner and humans are the reservoir.***
- The vector Tsetse fly (Glossina species) is only found in rural Africa):
  - ***Glossina palpalis*** transmits T. b. gambiense.
  - ***Glossina morsitans*** transmits T. b. rhodesiense.

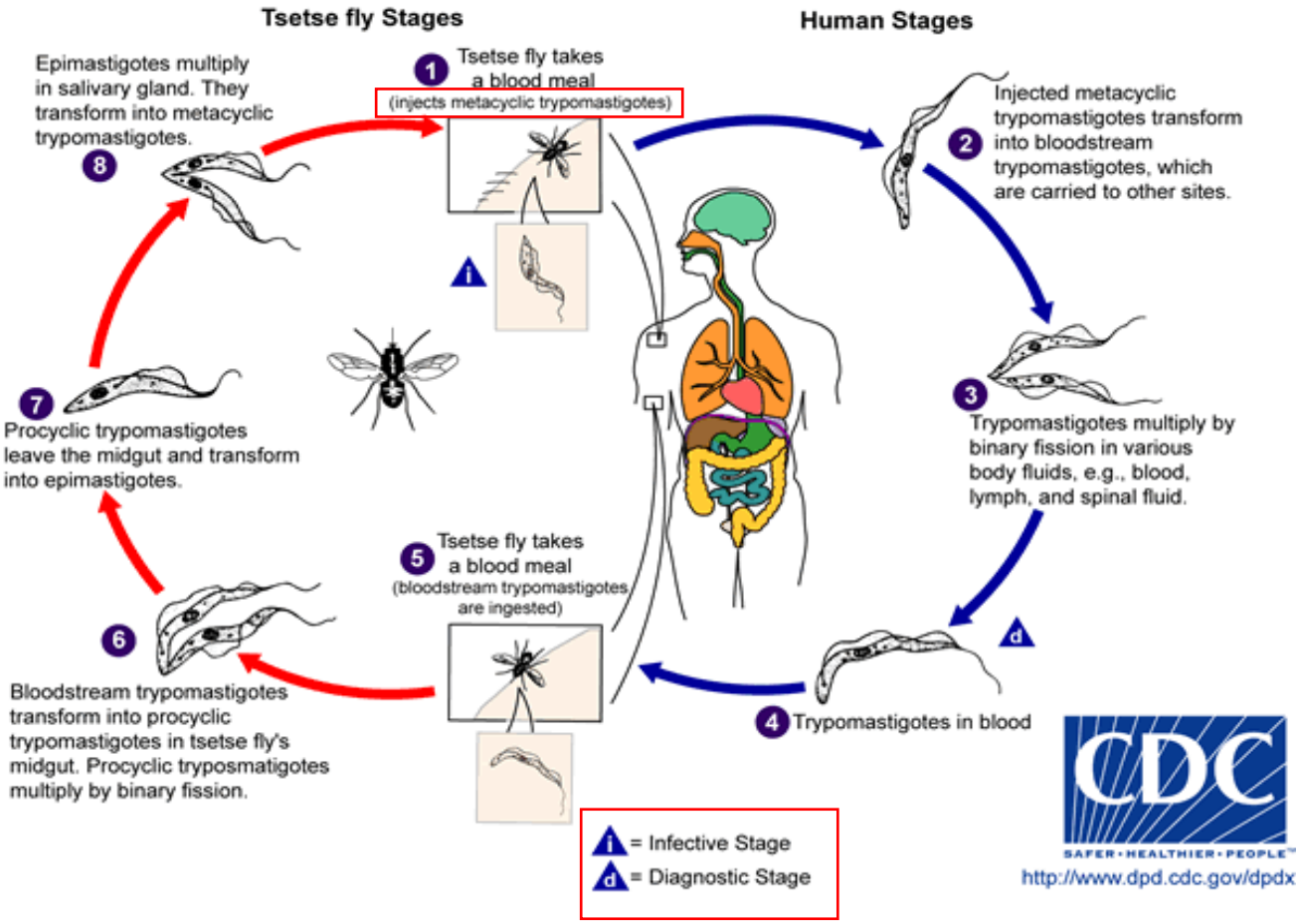
Epidemiology:

- There are epidemiological differences between T. gambiense and T. rhodesiense), the main one being that T. rhodesiense persists in a latent enzootic cycle in wild and domestic animals and is normally transmitted by Glossina from animal to animal, more rarely to humans.
- T. gambiense, on the other hand, is transmitted mainly from human to human by the tsetse flies, although various animal species have also been identified as reservoir hosts for T. gambiense strains.
- It is called African sleeping sickness as the patients start having CNS manifestations in the late stages of the diseases especially changes in character and personality.



Life cycle:

Note: it is very important to distinguish between the infective and the diagnostic stages.



- ***Metacyclic trypomastigotes*** which are transmitted by the vector's saliva is the infective stage, then they get transformed into ***trypomastigotes*** which are the diagnostic stage.
- Subsequent tsetse flies become infected by ingestion of blood that contains the trypomastigotes then they will be changed into promastigotes (procyclic trypomastigotes).

- Promastigotes then move into the saliva to be transformed again into metacyclic trypomastigotes.

Pathogenesis:

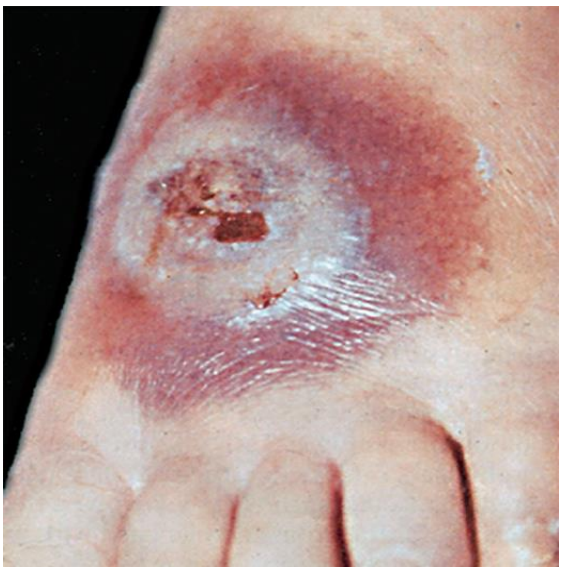
- When the patients get bitten by tsetse fly, they will first encounter a ***localized reaction*** to the metacyclic trypomastigotes. This reaction is called ***Trypanosomal chancre*** which is a painless nodule.
- When the parasite invades the blood or the lymphatics (parasitemia) symptoms starts to appear characterized by 2 stages:

First stage: the patient has systemic trypanosomiasis without CNS involvement.
The first symptoms appear and include: irregular fevers with night sweats, enlargement to liver and spleen, ***Winterbottom's sign. (only parasitemia, no CNS involvement)***

Second stage: organisms invade the CNS and reach the cerebrospinal fluid through the choroid plexus; the sleeping sickness stage of the infection is initiated characterized by change in character and personality.

At first, patients are agitated, have troubles with sleeping (restlessness), then they will have the over sleeping problems.

The patient becomes emaciated (because of encephalitis) and progresses to profound coma and death.



Trypanosomal chancre

Winterbottom's sign

(enlargement of the posterior cervical lymph nodes)

Laboratory diagnosis:

- Similar to American trypanosomiasis.

- **Specimen:** blood, serum, CSF, aspiration from lymph nodes
- **Routine Methods:** thick and thin blood films
- **Antigen Detection:** simple and rapid ***card indirect agglutination test***.
- **Antibody Detection**: Serologic by using ELISA Serum or CSF IgM concentrations
- **Molecular Diagnostics**: PCR-based methods to detect infections and differentiate species, but these methods are not routinely used
- **Definitive diagnosis:** requires seeing the parasite in the blood.

Treatment:

- If not treated, African sleeping sickness can be fatal, and the best result is when the disease is treated before stage 2 is reached (if CNS manifestations are reached, the prognosis will be very bad).
- No vaccines available.
- All drugs used in the therapy of African trypanosomiasis are toxic and require prolonged administration.
- Anti-parasitic drug selected depends on whether the CNS is infected or not:
  - ***Suramin or pentamidine isethionate*** can be used when the CNS is not infected
  - ***Melarsoprol***, a toxic trivalent arsenic derivative, is effective for both blood and CNS stages but is recommended for treatment of late-stage sleeping sickness.

- ***Eflornithine*** is a new drug that has been approved by the FDA to treat African trypanosomiasis but is still not fully recommended.

Prevention:

- Vector control by preventing flies from biting through the use of insecticide and bed nets will reduce the transmission of the parasite.
- Covering exposed skin areas.
- Screening of people at risk helps identify patients at an early stage.
- Treatment cases and should be monitored for 2 years after completion of therapy.

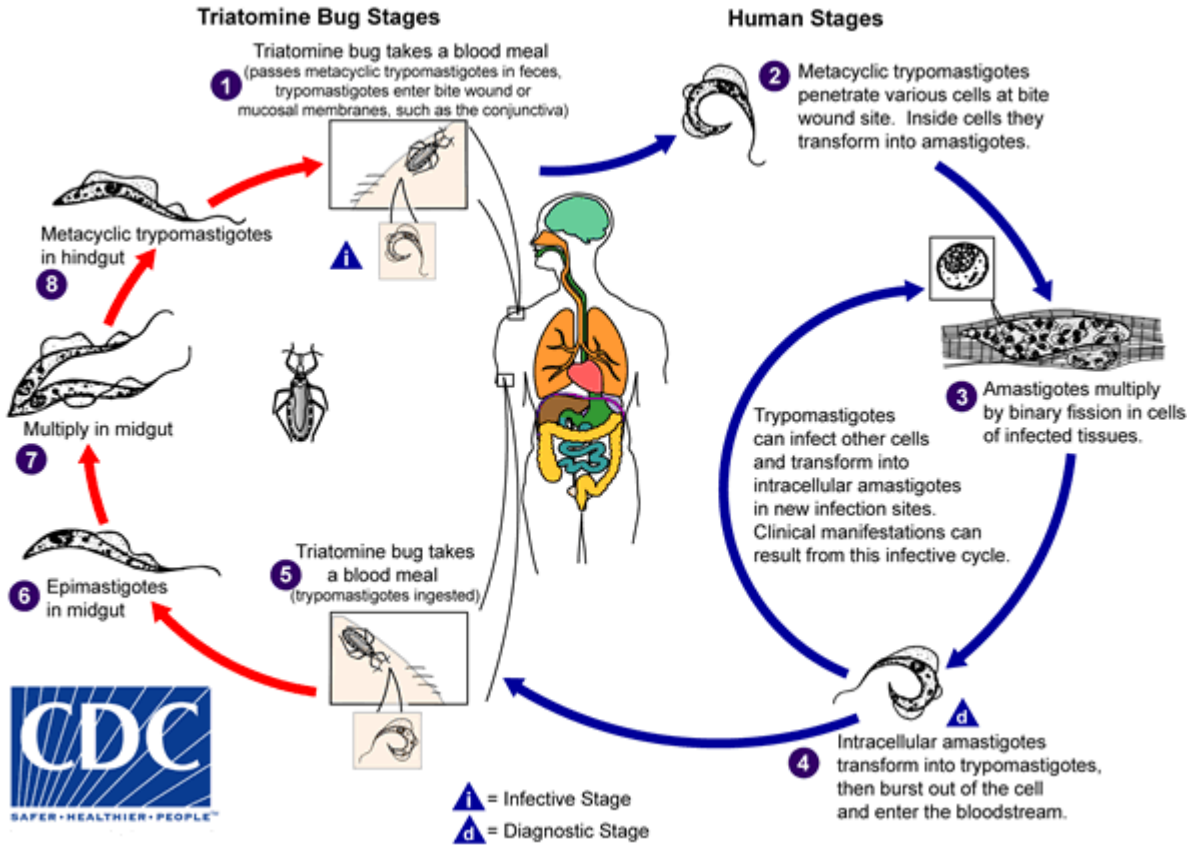
American Trypanosomiasis (Chagas disease):

- Caused by Trypanosoma cruzi.
- Zoonoses: transmitted by the vector Triatoma infestans (reduviid bugs) or “kissing bug “(named this way because they like to bite on the face). It is different from other bugs as it defecates after taking its blood meal and its feces contain the parasite. So, the life cycle may start if you have a skin abrasion in contact with the bug fecal material.
- Definitive host: humans, dogs, cats, rats, etc.
- In the definitive host it will take the trypomastigote form in the blood and the amastigote form in the tissues.

Epidemiology:

- Throughout central and south America.
- They have a shorter life expectancy as they suffer from sever heart issues because the amastigote intracellular form tends to select the cardiac muscles which causes cardiomyopathy, arrythmias and heart failure later on.

Life cycle:



- the infective stage is introduced by the feces usually in the face which will cause the patient to rub his eyes causing unilateral conjunctivitis (Romana's sign), but the parasite may be introduced by skin abrasion on other areas as well.
- Amastigotes need a heart biopsy to be seen.
- Although blood parasites need vectors in their life cycle but they also can be transmitted through blood transfusions or transplacental.

Pathogenesis:

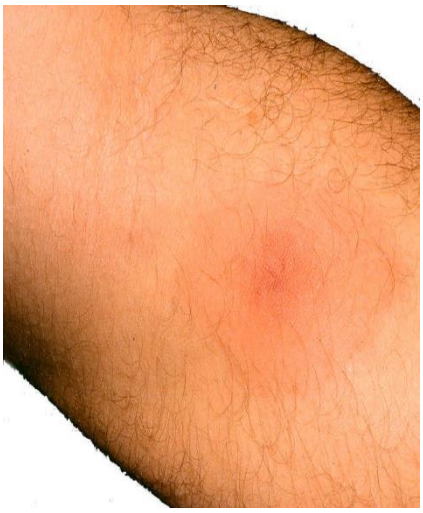
- Chagas' disease is categorized as acute, indeterminate or chronic.
- Localized reaction at the skin abrasion or the bite site is called ***nodule chagoma (red indurated swelling)***.
- The incubation period in humans is about 7-14 days.

Acute phase:

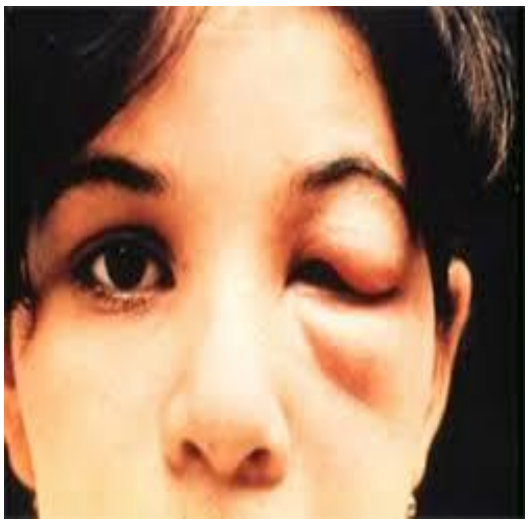
- 1 week after infection.
- Fever, lymph node enlargement, hepatosplenomegaly, unilateral swelling of the of eyelids because of conjunctivitis (Romana's sign) and acute myocarditis.

Chronic phase:

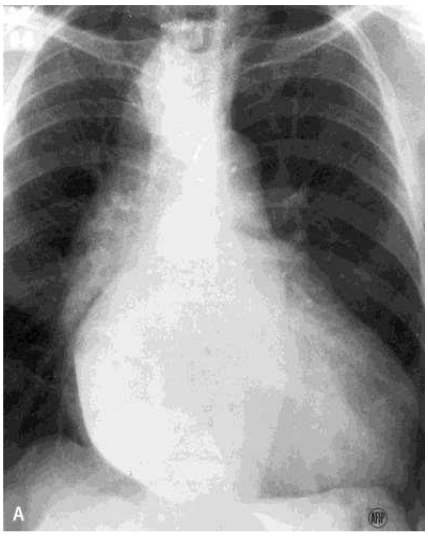
- Years after the diagnosis of acute disease.
- Mainly Involvement of the heart.
- Heart changes, heart enlargement, myopathies and heart failure.
- Colon enlargement (mega colon) or esophagus enlargement (mega esophagus)



Chagoma



Romana's sign



CXR showing cardiac enlargement in Chagas disease

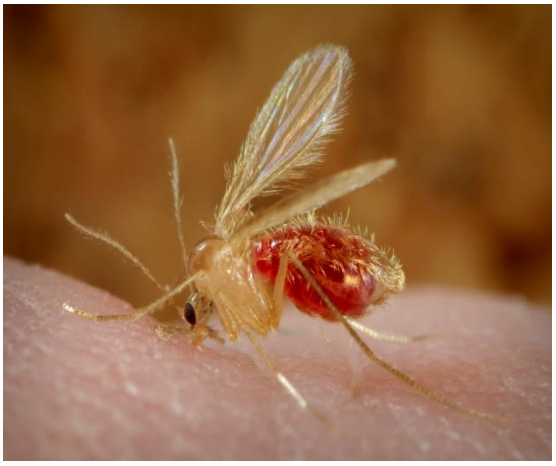
Treatment:

- Nifurtimox and benznidazole reduce the severity of acute Chagas disease.
- Both medicines are almost 100% effective in curing the disease if given soon after infection at the onset of the acute phase including the cases of congenital transmission.

Prevention:

- Vector control.
- Transfusion control and screening of blood donors.
- Testing of organ, tissue and cell donors and recipients.

Leishmania:



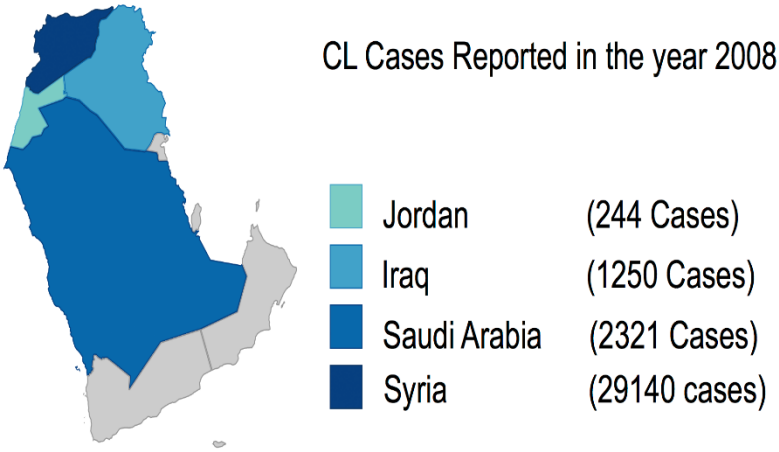
- Flagellated protozoan.
- Life cycle requires two hosts:
  - Vertebrate host: mammalian host.
  - Invertebrate vector: phelebotomus female sand fly.
- Obligate intracellular organism.
- Primary infects phagocytic cells and macrophages.
- The incubation period ranges from 10 days to 2 years.

Leishmania species:

Leishmaniasis is divided into clinical syndromes depending on the most affected part:

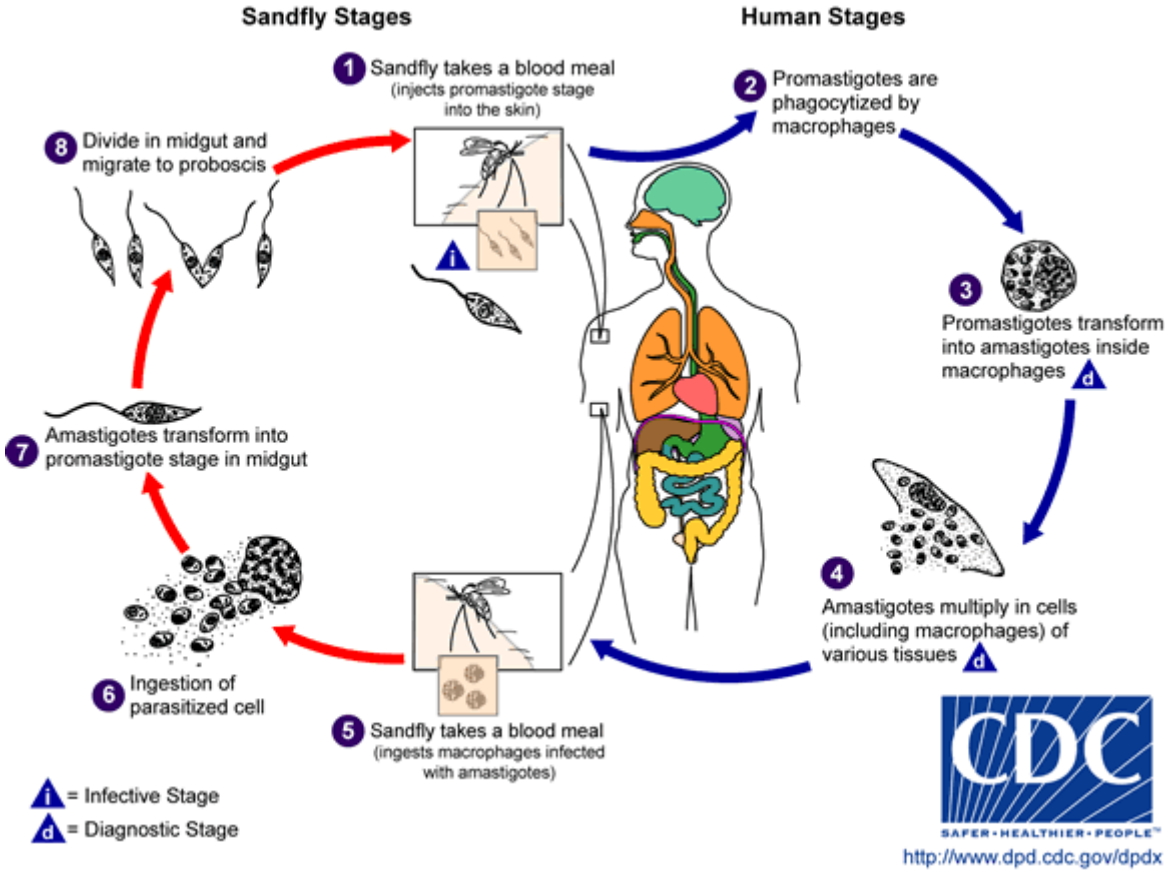
- Cutaneous Leishmaniasis (L. tropica, leishmania major) ➔ Skin and dermis.
- Mucocutaneous Leishmaniasis (L. braziliensis) → nasopharyngeal leishmaniasis
- Visceral Leishmaniasis (L. donovani) ➔ kala-azar leishmaniasis (black fever (hyperpigmentation)).

Leishmania in Jordan:



- Middle east countries and especially Iraq were very endemic for leishmaniasis but the incidence has dropped with modern medicine.
- In Jordan there are several species of Leishmania; Leishmania infantum, Leishmania tropica, and Leishmania major.
- Leishmania tropica (cutaneous) is the major type of Leishmaniasis in Jordan followed by the mucocutaneous form. ***(the doctor said major is the most common while slides say it is tropica)***
- In Jordan, it is found in al-aqaba (Al-qweera) region.

Life cycle:



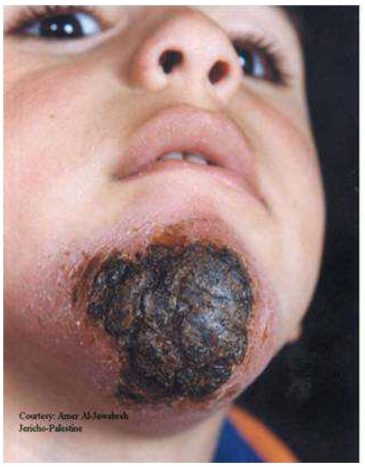
- Infective stage is the ***promastigotes***, while ***amastigotes*** is the diagnostic stage in the macrophages.

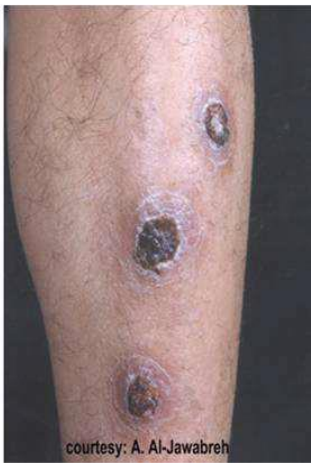
Routes of transmission:

- Bite of sand fly. -***The commonest***-
- Blood transfusion and organ transplantation.
- From mother to baby.
- Direct contact, from man to man through nasal secretions.

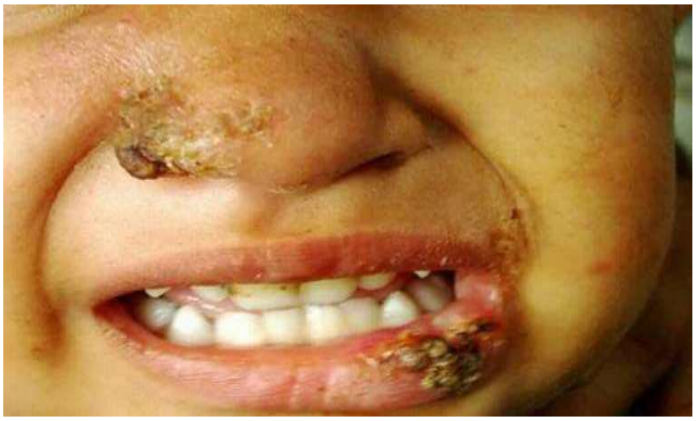
Cutaneous leishmaniasis:

- Caused by L. major, L. infantum and L. tropica (can also cause mucocutaneous form).
- first sign is a lesion (The lesions begin as reddish, soft and itchy papular then gradually enlarge, raise and firm with serous discharge at the bite site.
- Found in Middle east and south America.
- It causes painless ulcers and they may heal spontaneously but it may take months or it may leave a permanent scar (depending of the immune status). usually patients seek medical care for cosmetic purposes.





Mucocutaneous leishmaniasis:



- Caused by L. braziliensis and L. Mexicana (in central and south America).
- tends to affect the nasopharynx mucosa and may destruct the nasal septum if not treated.
- The primary lesions are similar to those found in cutaneous leishmaniasis.
- Dissemination to the nasal or oral mucosa may occur from the active primary lesion or may occur years later after the original lesion has healed.
- Samples are taken from the erosions for diagnosis

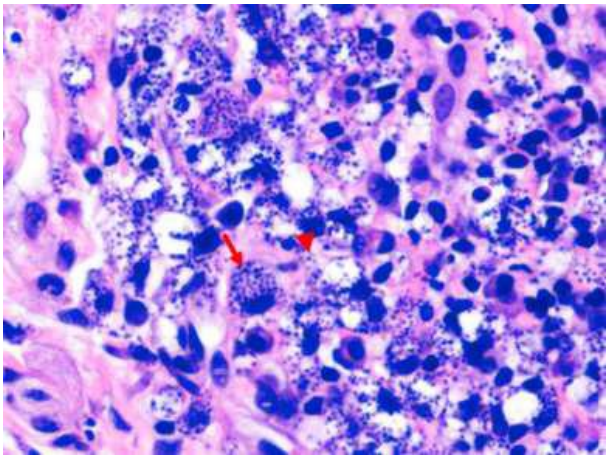
• These mucosal lesions do not heal spontaneously, and secondary bacterial infections are common and may be fatal.

Visceral leishmaniasis

- Its target is the macrophages and other APCs which they use to reach the reticuloendothelial system and lymph nodes.
- Causes hyperpigmentation of the skin.
- Is the most severe form of leishmaniasis.
- The parasite migrates to the internal organs such as the liver, spleen (hence "visceral"), and bone marrow.
- The incubation period: 10 days to 2 years.
- Symptoms: fever, anorexia, malaise, weight loss, and, frequently, diarrhea.
- Clinical signs: enlarged liver and spleen, swollen lymph nodes, occasional acute abdominal pain and abdominal distention.
- If left untreated, will almost always result in the death of the hos.t
- Epidemiology: Bangladesh, Brazil, Ethiopia, India, South Sudan and Sudan.
- Bone marrow sample by aspiration (from the sternum) must be taken for diagnosis.

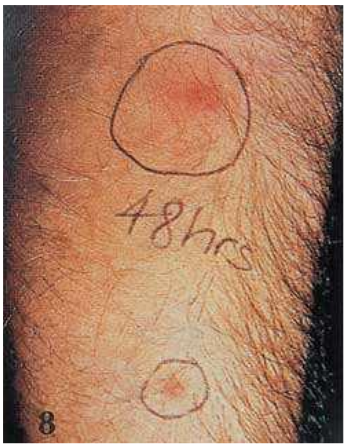
Laboratory diagnosis:

You look for amastigotes (for definitive diagnosis)



- Stained blood smear: aspiration, scraping or splenic puncture for visceral.
- Culture using special techniques.
- ElISA, IFA or direct agglutination give useful indication of active or recent kala-azar.
- PCR methods have excellent sensitivity and specificity for direct detection.

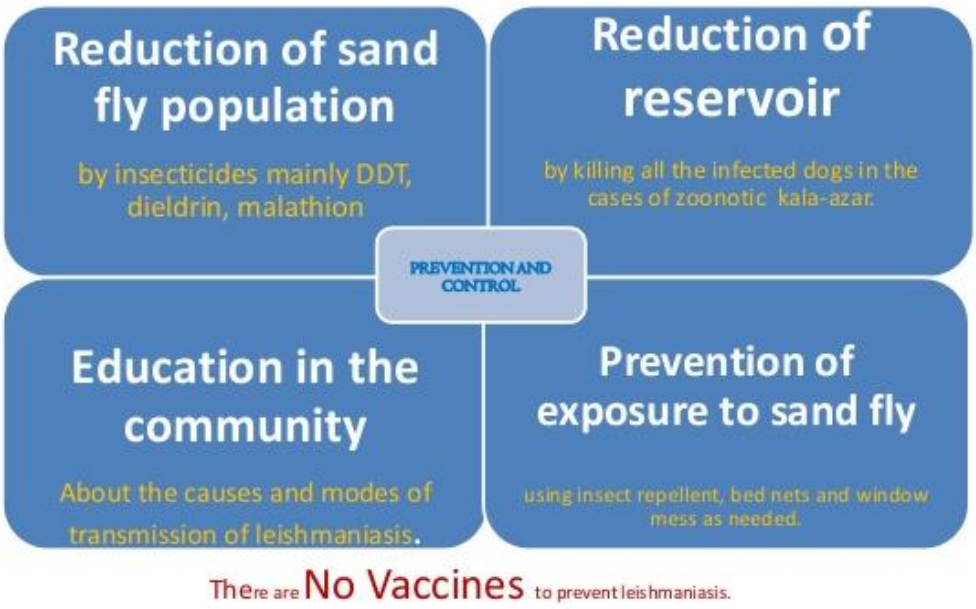
- ***Intradermal Montenegro test***: Injection of intradermal antigen which is prepared from cultured promastigotes of Leishmanian species. This produces a typical cell-mediated response (delayed type 4 hypersensitivity reaction), you examine the induration diameter just like TB PPD test.
- Histologic examination by biopsy from tissue to demonstrate the presence of organism in the tissue.



Treatment:

- The patient response varies depending on the Leishmania species and type of disease.
- In simple cutaneous leishmaniasis, lesions usually heal spontaneously, drugs are prescribed only for cosmetic purposes.
- ***Antimony, sodium stibogluc***onate drugs of choice for the treatment of visceral and mucocutaneous leishmaniasis.
- ***Pentamidine*** can be used for serious visceral leishmaniasis.

Prevention:



The End