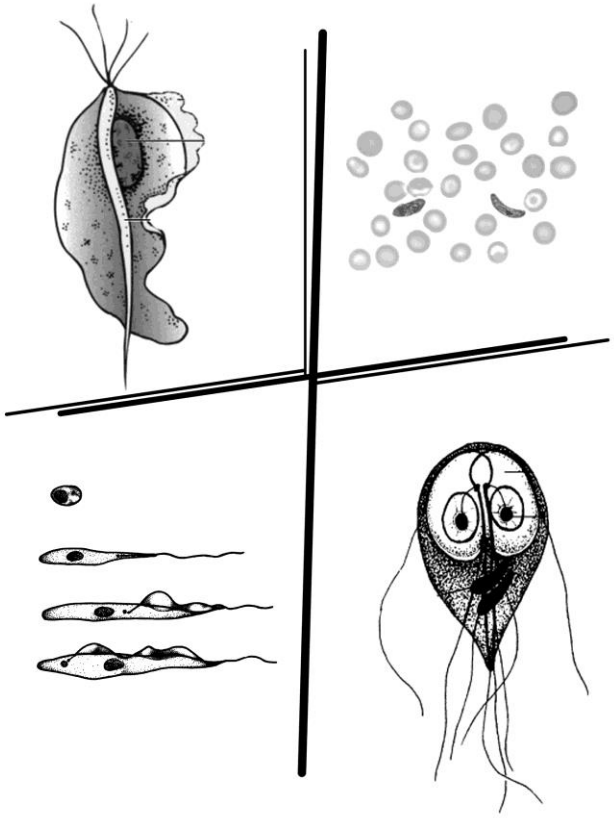
PROTOZOLOGY

Lecture Notes

University of Khartoum
Faculty of Medicine
Batch Qasawir 88
Academic Secretory



Dr. Ahmed Hassanein

INTRODUCTION

Parasitology:

is the study of parasites , their hosts & the relationship between them

- **Two main subjects:**
  1- protozoalogy: is the study of protozoa.
  2- helminthology : study of helminthes.

Ecology :

If 2 organism are living together :

- A pathogenic------→ pathogen is harmfull to the body (cause disease)
- A commensel---→ is harmless(one ben
- A symbiont ----→ both beneficial

- New definitions :
- Mutualism =symbiotic → both benefit
- neutralism = neither incur cost or benefit
- commensalism = one benefit but no cost on the other
- parasitism = one benefit & there is cost on the other
- external parasite (ectoparasite): live on skin or hair of the host
- internal parasite (endoparasite): line in body organs, tissues, cells & cavities .
- definitive host : is the host in which a parasite attains sexual maturity & harbours the sexual mature parasite .
- intermediate host: harbours the immature or sexual cycle
- reservoir: source of infection

- vector : the organism which carry the parasite to host (from reservoir to host)

GENERAL CONCEPT OF PROTOZOA

PROTOZOA :

- a taxonomic group of single celled organism with a nucleus (i.e eukaryotes) recently called acellular organism
- eukaryotic organism
- generally unicellular
- Major cause of human disease
- Usually in tropical or less developed country
- Are very dangerous to immunocompromised patients
- They rival anything in the animal kingdom for uniqueness & novelty manwell

Classification :

- Kingdom : animalia
- The phylum protozoa is classified into subdivisions according to the method of locomotion :
  1- The amoeba (sarcodina): move by mean of pseudopodia.
  2- The flagellates (mastigophora): move by long wipe like flagellae .
  3- The ciliates (ciliaphora): move by cilia.
  4- Sprozoa (sporozoans) :can't move also called apicomplexa (cuz it has apical structure that found on it)

LIFE CYCLE STAGES:

- Trophozoite : the stage of parasitic protozoa that actively feed, move & reproduce & it has flexible cell membrane & the it is the diagnostic stage .

- Cyst : stage with protective membrane or thickened wall & it is the infective stage . it has 2 types :
  - Reproductive : e.g amebae
  - Protective : e.g balantidium coli
- Encystation : transformation from trophozoite to cyst
- Excystation : rupture of the cyst & release of nuclei

Reproduction:

1- Asexually by :
   A- Binary fission (mitosis): the commonest
   B- Budding
   C- Schizogony : a multiple binary fission (nucleus undergo multiple division before the cell divide)
2- Sexually by :
   A- Conjugation
   B- Gamete formation (e.g malaria)

Transmission:

1- Person to person : Entamoeba gingivalis & trichomonas sp. .
2- Oro-fecal : E.histolytica .
3- Vector borne : malaria & leishminia

Diagnosis:

- Clinical & laboratory .
- Laboratory is used for detection & identification .
- Sample :

A- Blood :

- Can be
  - Drops of blood (finger stick)
  - Few mls (venipuncure)

- ❖ EDTA is used to anticoagulate the blood sample
- ❖ Parasite numbers decreased if processing is delayed (fter 4 hrs isn't useful)
- ❖ Methods of detection :
    - Direct microscopy : used in malaria & trypansomiasis
    - Stain with Giemsa :
      Thin film : for identification mainly, fixed & stained.
      Thick film : for detection mainly , stained without fixation .
- ❖ Serology :
    - Advantages : simple, quick, highly sensitive & specific.
    - Disadvantages : remains positive for long time
    - Useful in field & poor countries.
    - Usage :
      1-Difficult identification of the parasite (e.g differentiation between babesie & plasmodium).
      2-Low parasitemia .
      3-asymptomatics (e.g chagasic pateins)
    - Antigen detection :
      1- Dot ELISA
      2- Rapid diagnostic test (RDTs) based on immunochromatogrophic antigen detection .
      3- Complement fixation test (CF)

    - Antibody detection :
      1- enzyme linked immune sorbent assay (ELISA):
         A. falcon assay screening test ELISA (FAST-ELISA).
         B. Luciferase immune precipitation system (LIPS)
      2- Heamagglutination (HA) test .
      3- Indirect or direct immune fluorescent antibody (IFA or DFA) test .

4- Immunoblotting

❖ Molecular based methods :

- Nucleic acid based :
  - Greater sensitivity & specificity
  - Used in very low parasitized sample
  - Used in asymptomatics patients
  - Multiplexed PCR allows for the detection of several parasitic infections simultaneously .
- PCR & RT-PCR
- Loop mediated isothermal amplification
- LAMP & multiplex LAMP (mLAMP)

B- Stool :

- Wet film microscopy :
  - Quick & cheap
  - Not such useful for minute & smell number organism.
  - Used mainly for detection .
  - Best major criteria is the num of nuclei
  - Called wet cuz we add normal saline
  - Perminant stained stool used for identification
- Concentration methods :
  - Used to increase the sensitivity

1- Sedimentation method :

- Cyst are down
- We use Formal-ether & water to dissolve fats that found in stool

2-Floating method :

- Cyst are up
- By adding Zinc sulphate method

- Antigen detection :

- New widely used :
- DFA
- EIA
- Immunochromatogrophic test (rapid test)
    - PCR similar to that of the blood

❖ Diagnosis

- Tissue biopsies :
- Provide definitive evidence of invasion
- It is Invasive procedure not used as much except in leishmania
- Aspirate :
    - Useful for lymph

So mainly we use serology

AMEBIASIS

- 5 sp of Entamoeba.
- E.histolytica > most important = pathogenic.
- E.dispar, E.coli, E.hartmanni, E.gingivalis.

AMEBIASIS:

- Infection with E.histolytica
- 90 % asymptomatic cyst passers (carrier)

Morphology:

- Trophozoite
- Cyst (1,2,4 nuclei)

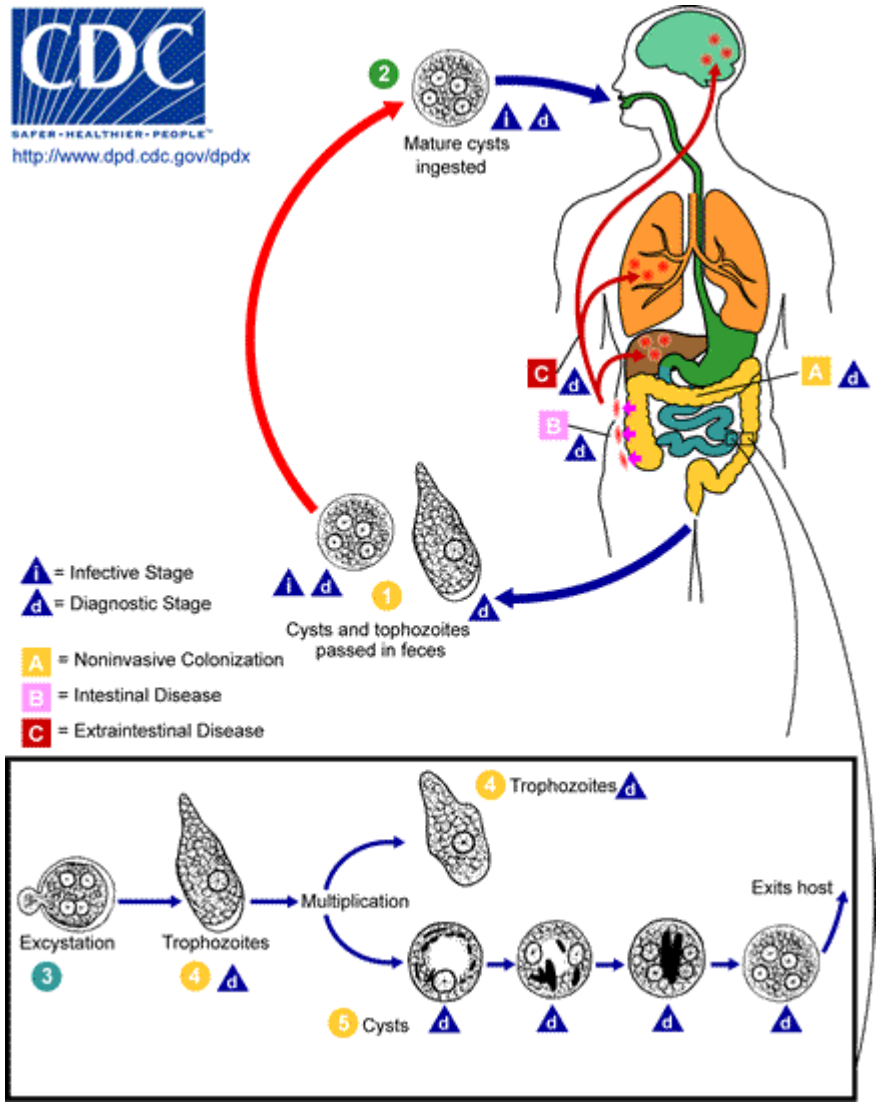
Trophozoite character:

- Contain ingested RBC.
- Large invasive form (magna)contain RBC.
- Noninvasive form small (minuta) no RBC.
- Different from cyst no flagella , no RBCs.

Transmission:

- Through contaminated food /drink with faces contain cyst of E.histolytica (fecal-oral).
- Sexually through anal route in homosexual.
- Release of Trophozoite inside small intestine , live in colon and multiple (carrier) , if immunity decreased it damage mucosa &cause (dysentery).

- It reach serosa (blood vessels)= hematogenous spread= extra-intestinal Amebiasis.



Pathogenesis:

- Lectin mediated adhesion
- Contact dependent cytotoxicity
- Children with mutated Lectin receptor more likely to get disease (increase affinity)
- Caspase 3 induce apoptosis and necrosis
- Cysteine protease for protein breakdown
- After adherence,Trophozoite> invade colon (flask shaped ulcers). Serosa and sub mucosa

- Spread of Amebiasis to right lobe of liver via portal blood
- Destruction of liver = necrosis (liver abscesses) with a cellular proteinaceous debris = anchory paste appearance.

Clinical presentation:

- Amebic colitis: amebic dysentery, acute.
- Gradual onset (unlike shigella sudden).
- Diarrhea watery without pus or blood or weight loss.
- Pt. is not toxin (unlike shigella).
- Fever, abdominal cramps and pain.

Shigella	**E.histolytica**
Sudden onset	Gradual onset
Pus	No pus
Toxic	Not toxic

- **Fulminant amebic colitis :**
  - Rare complication.
  - Rapid onset.
  - Sever fever.
  - Severe diarrhea.
  - Risk of perforation and peritonitis.
- **Toxic mega colon :**
  - With corticosteroid use (decrease immunity)
- **Chronic amebic colitis :**
  - Trophozoite remain as carrier because of high immunity
  - Recurrent episodes of bloody diarrhea (occasional diarrhea) and abdominal discomfort
- **Ameboma :**

- Formation of chronic granulation in response to the infecting amebae
- Right quadrant in abdomen
- Mistaken as cancer, lymphoma, TB
- Diagnosed by biopsy

– **Amebic liver abscess :**
  - Is the **most common form of extra intestinalAmebiasis.**
  - 5% of pt. with symptoms of dysentery.
  - More common in men.
  - Swinging fever (abscess) + pain in right hypochondrium.
  - Chest + shoulder pain (referred).
  - Associated with GI symptoms: nausea, vomiting, abdominalpain, diarrhea, weightloss, anemia (of chronic disease).

– **Pleuropulmonary Amebiasis:**

Contagious spread from liver abscess, pneumonia signs

– **Hepatooobronchial fistula:** anchory paste sputum

Trophozoite may be found in sputum sample

– Amebic peritonitis : from left lobe liver abscess
– Amebic pericarditis left lobe abscess rupture in pericardium, lethal.
– Cerebral Amebiasis :

Eating brain, rapid progression

Altered consciousness, focal neurologic signs

CT irregular lesion

Tissue biopsy >Trophozoite

- Skin Amebiasis

Differential diagnosis:??????????????????????

Laboratory diagnosis:

(E.dispar +E.histolytica)Trophozoite resembles each other, give treatment when there are symptoms

By immunological analysis

1. Detect E.histolytica antigen
2. Nucleic acid in PCR

If there is cyst it is just entamoeba

Immune and molecular diagnosis:

Antigen detection:

- Stool for colitis
- Blood for ALA

Diagnosis ALA:

- Symptoms and signs usually strongly positive serology sero converted during treatment
- Imaging
- Other tests (chest radiology, ultrasound, CT)

TREATMENT:

- Luminal agent: for none invasive
- **Paromomycin: 1st choice**
- Tissue amebicidal: netronidazole for invasive
- Then: luminal agent for prophylaxis
- Surgical intervention

ALA treatment:

Metronidazole or tinidazole + chloroquine phosphate or emetine followed by luminal drug.

Indication for drainage:

- Left side
- Ruptured
- Abscess don't response for treatment
- Diameter more than 10cm

PATHOGENIC FREE LIVING AMEBAE

Naegleria fowleri

- Causal agent of primary amebic meningeoencephalitis (PAM).
- Transmitted from contaminated water swimming.
- Life cycle has 3 stages : Trophozoite , temporary flagellar stage (ameboflagellate), cyst
- Diagnosis: by direct wet-mount, microscopy of CSF sample.
- Treatment: amphotericin B.

Acanthamoeba:

- Causes granuloma like TB.
- Keratitis ,blindness, lens contact.
- Disseminated granuloma.
- Granulation in brain.
- Treatment very difficult.

Intestinal parasites

Giardia intestinalis (giardia duodenalis, giardia lambia):
Most common protozoal intestinal parasite , in summer , in children (which become carrier)

Transmission:

- Waterborne (outbreaks)
- Homosexual anal-oral contact
- From person to person by food
- G.intestinalis is easily transmitted:
  1. Infective dose is very small >10 cyst

2. Resistant cyst (resistant to chlorine and other disinfectant)

Morphology:

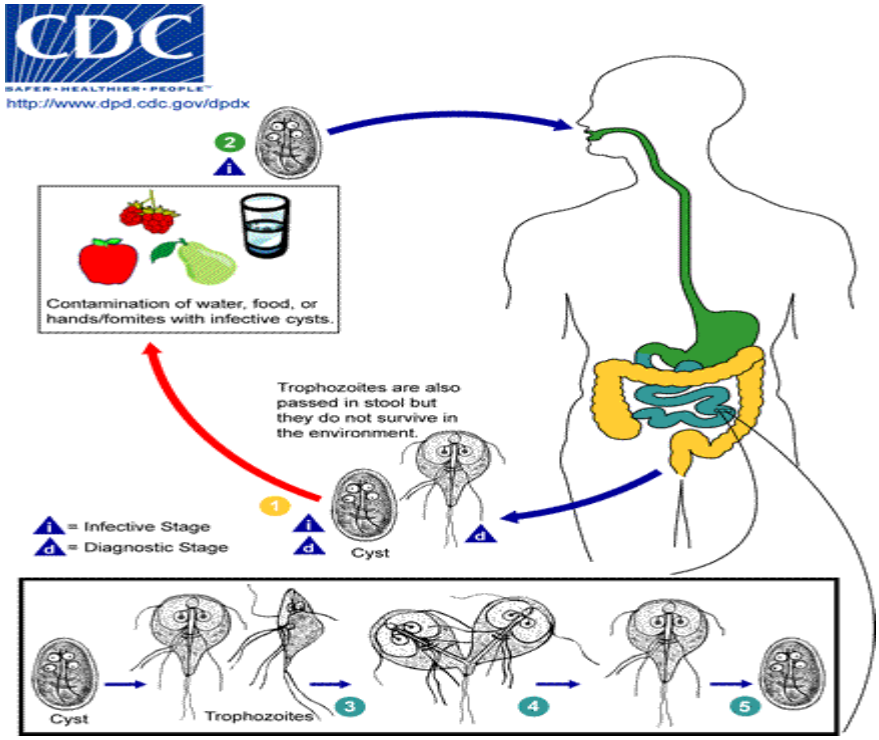
- Diagnosis very easy
- Trophozoite :
  - Are binucleated
  - Move by flagella
  - Median body (adhesion disk)(suction disk)
- Cyst
  - Rounded with 4 nuclei give 2 Trophozoite
  - There is axonemes (support) ,2 bars

Life cycle:

Excystation in duodenum (small intestine) and cause disease there

Pathogenic mechanism:

- Adhesive disk
- Cover all intestinal surface > barrier (easy eating)
- Interfere with enterocytes
- Loss of epithelial function
- Villus atrophy
- Destruct brush border enzymes (disaccharides)>sugar malabsorption (gluc)>osmotic diarrhea
- Also hypersecretion of Cl > watery diarrhea
- Interfere with bile salt metabolism
- Immune inhibition
- No blood



Symptoms:

- Incubation 6-15days
- 60-80% asymptomatic infections
- Diarrhea , foul smelling faeces
- Malaise
- Flatulence
- Anorexia, mild fever , weight loss

Extra-intestinal symptoms: as complication

- Disaccharidesdeficiency
- Malabsorption
- Growth retardation in children
- Weight loss

Diagnosis:

1. **Light microscopy: best method**
2. String (entero test) , aspiration or biopsy (villus atrophy) IgA deficiency& hypogammaglobulinemia with giardiasis > sever atrophy

3. Serology : sensitive > carrier's screening

- Direct fluorescence antibody :useful for screening
- Antigen detection (ELISA)
- PCR

Treatment:

- Metronidazole 1st choice
- (Tinidazole ,nitazoxanide) >teratogenic not used in pregnancy
- Paromomycin for pregnancy

Trichomonas

- Include a group of flagellated protozoa that infect human & man.
- 3 species of trichomonas found in human:- Pentatrichomonas hominis, T.tenax, T.vaganalis
- P.T.hominis, T.tenax not pathogenic & less important species.
- Are not distinguishable by wet film examination.
- T.tenax:- inhibit caries around teeth, not pathogenic.
- P.T.hominis :- inhibit large bowle, not pathogenic.

Trichomonas vagainalis

- Exclusively sexually transmitted (that's why no cyst form).
- Most important STD.
- In female organism inhabits in vagaina.
- In male found in urethra(prostate).

Morphology & life cycle:-

- Only trophozoite.

- Transmitted by direct contact during sexual intercourse.

Pathogenicity:- (has 4 flagella)

- Contact depends on cytotoxicity.
- Specterace (protenase) …… cell lysis(cytoskeleton)
- Form pore in epithelium.

Presentation:-

Sign & symptoms:-

- I.P 4_28 day

Female:-

- Vaginitis: with fowl smelling vaginitis , discharge is gray, but can be yellow or green & occasionally frothy or blood tinged + pruritis (white discharge in candidiasis).
- Vulvovaginitis
- Urethritis:

Painfull urination.

Male:-

- Is likely asymptomatic , include urethral discharge(range from scant to purulent) , dysurea , urethral pruritis & some experience burning immediately after coitus.

Complication:-

- Low birth wt & preterm babies , high risk of transmission of HIV.
- High risk of carcinoma of cervix.
- Very recently high risk of prostate cancer.

- Respiratory or genital infection on new born.
- Infertility.

There is co-infection with AIDS 3/1000.

Diagnosis:-

- Wet film examination.
- Culture(in pouch culture system)
- Whiff test (amine odor test): by adding several drops of 10% potassium hydroxide.
- Immunochromatographic test : rapid Ag detection kit for Ag (best method).
- PCR for researches.

Treatment:-

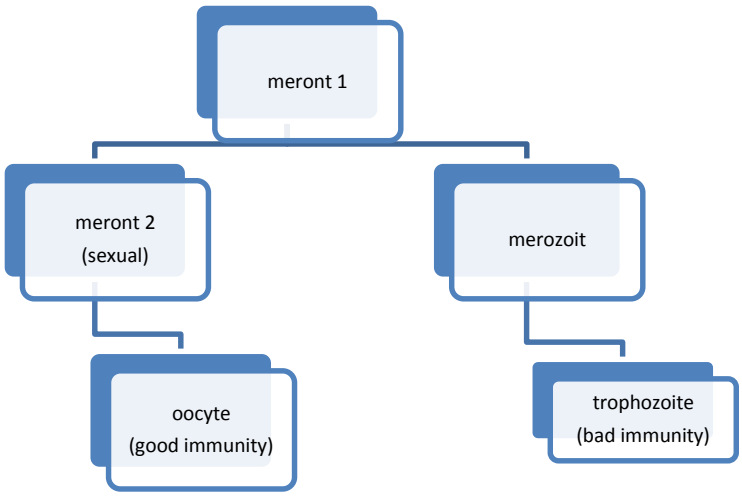
- Metronidazole for both partners.
- Other 5- nitrimidazole .

+ maintenance of low PH of vagina.

+ use of condom.

Cryptosporidium

- c.parvum causes disease
- waterborn illness with AIDS patient
- life cycle:-
- occyte infective & diagnostic stage
- macrogamete(female) unit with microgamete(male).....occyte....sporozoit..... trophozoit.....meront1



- Occyte : double walled:_
- 1) resistant to: chloride, drying, progressive freezing, salt& water.
- 2) only occyte in this stage can live invivo.
- Transmission:_
- Fecal_oral route , drinking water/ swimming pools
- Unpasterlize apple cider
- Animal contact

- Pathology:-
- Infective dose <10 organisms
- Attach to epithelial cell of GI
- Affect other tissue respiratory tract& conjunctiva.
- Cases:-
- Villous death/malabsorption
- Presentation:-
- Asymptomatic
- Watery diarrhea- fever- myalgia- fatigue
- In AIDS pts:-
- Infect lung……. Cough
- Infect gall bladder…….. cholecystitis
- Dehydration & weight loss
- Recovery:-
- Immunocompetent……. 1-2 wks
- Immunocompermise…….(month to year)
- Complication:-
- Pancriatitis
- Sclerosing cholangitis
- Calculous cholicystitis
- Pt with AIDS….. respiratory tract infection
- Diagnosis:-
- Microscopy with acid fast satined stool(occyte)
- Direct florescent antibodies
- Enzyme immunoassay for (screening)
- Treatment:-

- No effective therapy
- Nitazoxande for diarrhea
- Mainly Supportive with fluid & electrolyte
- If immunecompermise treat AIDS.

Di entameoba fragilis(flagellate without flagella)

- Amoeboid appearance but ultrastructural & molecular evidence show clearly that it is related to trichomonas.
- Doesnot form a resistant cyst
- Transmitted via entrobius eggs

• Transmition:-

- Via enterobius egg……. Case itching around anus in children.

• Symptoms:-

- Diarrhea
- Abdominal pain
- Anal pruritis
- Flatulence
- Urticaria
- Fatigue

_ Best method for diagnosis is microscopy

- Senidazole is best choice
- Metronidazole – less effective
- Tetracycline

• Diagnosis:-

- Trophozoite with egg

Malaria

- The most important tropical disease.

- **Species :**

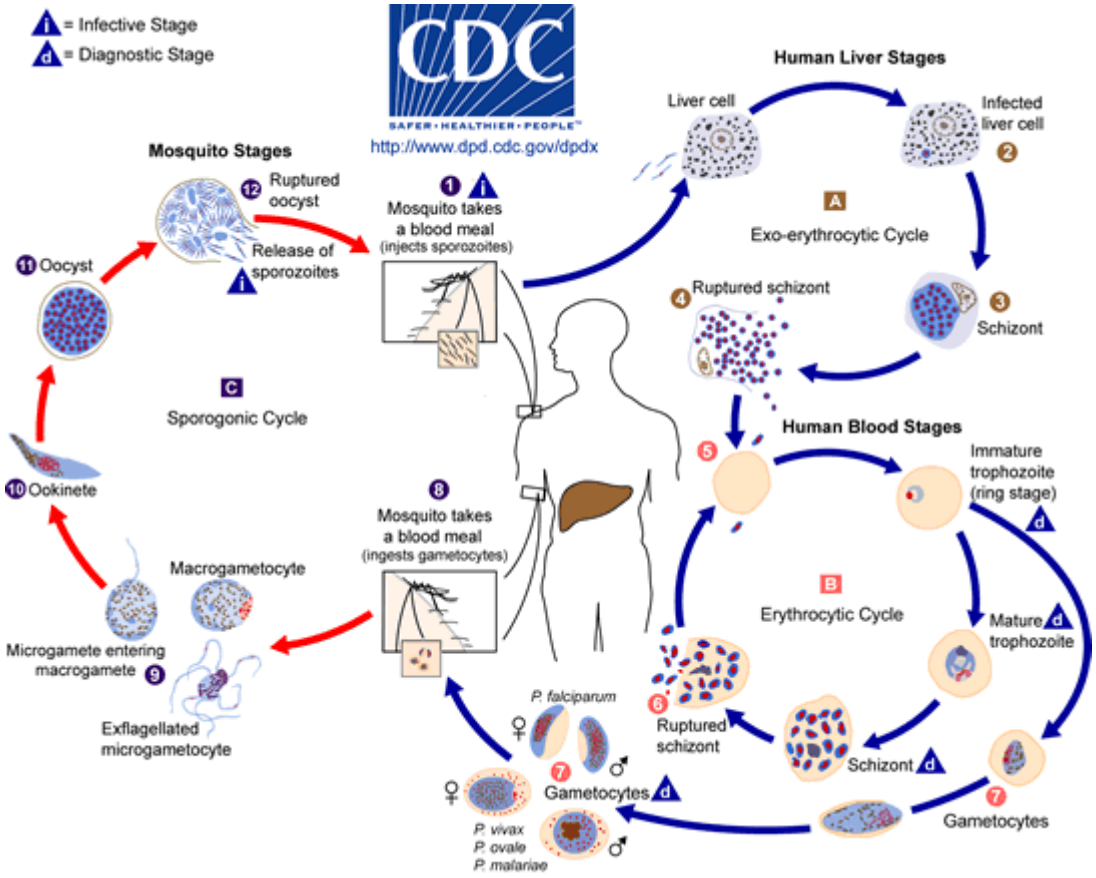
  - Plasmodium falciparum.
  - Plasmodium vivax “tertian malaria, i.e., fever every 3 days”.
  - Plasmodium ovale “tertian malaria”.
  - Plasmodium malariae “quartan malaria, i.e., fever every 4 days”.
  - P.Knowlsei : same as P.Malariae, new & not well described.

- **TRANSMISSION :**

  - Only Anopheles mosquitoes transmit human malaria.
  - Blood transfusion.
  - Transplacental (vertical / congenital).
  - All species have 3 modes of transmission.

- **Life cycle :**

  - In mosquitoes : sexual and sporogony.
  - Extraerythrocytic : schizogony in liver, asexual.
  - Erythrocytic : schizogony in blood, asexual.
  - Gamogony : sexual in blood and mosquito gut.



- **Extraerythrocytic "tissue" phase:**

  - Blood is infected with sporozoites about 30 minutes after mosquito's bite.
  - The sporozoite enters liver cells where they multiply forming pre-erythrocytic schizogony.
  - P.vivax & P.ovale sporozoites form parasites in the liver called hypnozoites.
  - P.falciparum & P.malariae develop directly into schizonts in the liver, takes 6-16 days, then schizonts rupture releasing merozoites & producing clinical symptoms "invade RBCs".

- **Relapsing malaria :**

- P.vivax & P.ovale hypnozoites remain dormant for months.
- They develop the schizonts rupture releasing merozoites & producing clinical symptoms.

- **Erythrocytic phase :**
  - Trophozoites early stages in the ring forms.
  - Trophozoites divide forming a schizont.
  - Segmentation of schizonts form merozoites.
  - Schizonts rupture, releasing merozoites into blood stream causing fever.
  - Merozoites invade other RBCs and schizogony is repeated.
  - Merozoites may develop to gametocytes, the sexual form of the parasite “when starting treatment or when patient close to die”.

- **Schizogenic periodicity and fever patterns :**
  - Schizogenic periodicity is the length of asexual erythrocytic phase.
  - 48 hours (in P.vivax & ovale) tertian malaria.
  - 72 hours (in P.malariae) quartan malaria.

- **Gametocytogenesis :**

- Sexual dimorphism : microgametocyte

  Macrogametocyte

  - No pathology.

- Infective stage for mosquito.

- **Sporogony :**
  - Occurs in mosquito intestine.
  - Fusion of micro and macrogametes.
  - Zygote ookinete (24 hours).
  - Ookinete transverses gut epithelium (trans-invasion).
  - Sporozoites released.
  - Sporozoites invade salivary glands.

- **Erythrocytic phase :**
  - Prepatent period : interval between date of infection and detection of parasites in peripheral blood.
  - Incubation period : interval between infection and first appearance of clinical symptoms, 8-9 days or more.

- **Pathogenesis of malaria :**
  - We will consider 2 : immune-toxin and sequestration\cytoadherence
  - TNFα : lead to the release of the mediators causing the symptoms of malaria.

- **Sequestration and Cytoadherence :**

  Rosetting : adhesion of the RBCs to other RBCs.

  Clumping : adhesion between infected cells.

  Are mediated by knobs.

  Knobs made up by PfEMP1.

- **Falciparum malignancy :**
  - Cytoadherence.
  - Each cycle produce 20 times more merozoites.
  - Multiple infestation of all RBCs ages.
  - Early hemolysis and endotoxin release.
  - Resistant to chloroquine.
- Note: P.vivax & P.ovale love new RBCs

  P.malariae love old RBCs

- **Types of infections :**
- **Relapse** : reactivation of hypnozoites form of a parasite in liver separate from previous infection with the same sp. (p.v , p.o).
- **Recrudescence :** exacerbation of persistent undetectable parasitemia, due to survival of erythrocytic form (p.f , p.m).
- **Recurrence or reinfection** : exoerythrocytic forms infect erythrocytes, separate from previous infection (all species) .

Clinical presentation :

1- Acute uncomplicated malaria.

2- Severe complicated malaria.

3- Congenital.

- **Acute malaria** : acute febrile illness may have periodic febrile paroxysms every 47-72 hours, with:

- Paroxysms that last 8-12 hours start between midnight and midday.
- Cold stage; rigors 1-2 hours.
- Hot stage; 39-41˚c , 2-6 hours.
- Sweating stage; 2-4 hours.
- **Early symptoms:**
- Headache
- Malaise
- Muscular pain
- Fatigue
- Nausea
- Vomiting
- **At night :**
- Mild diarrhea
- Mild jaundice
- Respiratory symptoms
- **Signs :**
- Pallor
- DIC (bleeding)
- Hepatosplenomegaly
- Jaundice
- Respiratory distress

- Mental status change

- **Severe malaria :**

  Life threatening condition of P.falciparum malaria.

- **Clinical :**

- Cerebral malaria
- Convulsions
- Dyspnea
- Bleeding disorders
- Jaundice
- Shock
- Hyperpyrexia
  - HYPERPARASITEMIA

- **Lab :**

- Hyperparasitemia
- Hypoglycemia
- Metabolic acidosis
- Heamoglobinuria
- Acute renal failure (insufficiency) , (ARF , AGN)
- Electrolyte acid/base imbalance

- **Cerebral malaria :**

- Common in children.
- Unarousable coma that's not attributed to any cause but the presence of P.falciparum.
- Pathology of cytoadherence.
- Pathognomic feature : excess of malaria pigments in brain (hemazoin).
    - Complications : blindness, deafness, hemiplegia, cerebellar ataxia.

- **ALGID malaria :**
    - Characterized by shock & GI symptoms (hypotension).
    - Presentation : pale, cold with shock and severe diarrhea.
    - Associated with Gram negative septicemia.
- **Black water fever :**
    - Syndrome of intravascular hemolysis lead to hemoglobinuria.
    - Presentation : fever, pallor, jaundice, dark urine, hepatosplenomegaly & acute renal failure.

- **Pulmonary edema :**
    - Microvascular obstruction , ↑ vascular permeability.
    - Presentation : dyspnea, cough, hypoxia.
- **Multi-organ failure :**

- Shock , DIC.
- **Common features of P.vivax infection :**
- Typical prodrome of acute symptoms.
- Milder that P.F (no severe malaria).
- Splenic rupture hypovolemic shock.
- Carries high mortality rate.
- **Common features of P.ovale :**
- Similar to P.vivax, but spontaneous recovery more common.
- Milder.
- Fewer relapses.

- **P.malariae :**
  - Similar to P.vivax, but prodrome may be severe and the rest of the disease is milder.
  - Can cause nephritic syndrome, prognosis is poor (proteinuria and loss of Abs leading to low immunity and increased susceptibility to bacterial infections).

- **chronic complications of malaria :**
  - tropical splenomegaly sundrome (in endemic area).
  - Burkitt's lymphoma.
  - Nephropathy (membranous type) by P.malariae.

- **Congenital malaria :**
  - Transplacental infection.

- All 4 species, most commonly in P.F in Sudan.
- 1st trimester abortion.
- 2nd trimester miscarriage.
- Presentation : fever, irritability, feeding problems, anemia, hepatosplenomegaly & jaundice.

- **Diagnosis :**

Clinical Diagnosis :

- Hyperendemic & holoendemic areas, fever or history of fever.

Specimen :

- Blood can be collected by finger pricks.
- If other test is being used, we can use vein puncture.
- Smear should be prepared and stained within an hour.

Microscopy :

- The gold standard.
- Thick film considered gold standard for detection.
- Thin film considered gold standard for identification.

Thick film	Thin film
• Large volume (10ml)	• Smaller
• Good screening test (detection)	• Good species identification
• Parasitic density	• Low density infection can be missed

- **Malaria parasite erythrocyte stages:**
  - Ringform : in RBCs.
  - Schizonts : in RBCs.
  - Trophozoites : in RBCs.
  - Gametocytes : inside & outside RBCs.
- **P.F :**
  - more than 1 ring stage inside the RBCs (more malignant).
  - Rarely merozoites are found in peripheral blood because of knobs protein.
  - Mature gametocyte looks like “banana”.
- **P.V :**
  - 1 or rarely 2 ring stages.
  - Deformed RBCs.
  - Gametocyte is round.
- **P.M :**
  - Trophozoite is typically band shaped.
  - 1 ring stage.
  - Gametocyte round with dark pigment.

- **Parasitemia & clinical correlates :**

%	Parasite/ml	Clinical correlates
2-5 %	100,000 – 250,000	Hyperparasitemia / severe malaria
10 %	500,000	Exchange transfusion

- **Fluorescent microscopy :**
  - Modification of light microscopy.
  - Fluorescent dyes detect RNA & DNA.
  - Expensive.
  - Malarial antigen detection :
  - Immunochromatographic or rapid diagnostic test "RDTs".
  - Used with blood.
- **Antigens :**
  - P.F histidine-rich protein 2 (pfHRP2)
  - Parasite LDH (pLDH)
- **Malarial serology :**
  - Antibody detection.
  - Abs may persist for months.
  - Positive test indicates past infection.
  - IgM specific immunoglobulin used in hyper malaria splenomegaly syndrome.
  - Immunoflourescence test (IFA)

- Radioimmuno assay (RIA)

- **Polymerase chain reaction (PCR) :**
  - Molecular technique to identify parasite's genetic material.
  - For screening blood donation.
  - Can differentiate between species.
  - Detect mutations (drug resistance).

- **Treatment :**
  - Uncomplicated malaria.
  - In 2004 the national drug policy for malaria was changed from mono-therapy to ACT (= artemisinin based combination therapy) , (tissue-cidal).
  - Artemether + lumefantrine (best).
  - Artesunate + fansidar (sulfadoxine – pyrimethamine) , used in Sudan (the worst!).
  - Others : artesunate + amodiquine

    artesunate +melfaquine

    artesunate + piperaquine

- **Failure :**
  - Treatment failure with 14 days of receiving drug ACT.
  - 2nd line drug :

- Artesinate + tetracycline or doxycycline or clindamycine , for 7 days.
- Quinine + tetracycline or doxycycline or clindamycine , for 7 days.

- **note** : quinine can cause severe hypoglycemia (given with dextrose) & nephrotoxicity.

- **Pregnant :**
  - Quinine + clindamycine (no artesunate) => 1st trimester.
  - ACT or artesunate + clindamycine or quinine + clindamycine => 2nd & 3rd trimesters => for 7 days.

- **Notes about treatment :**
  - ACT except as +sp (fansidar) is used for P.vivax, in combination with primaquine for radical cure.
  - Two antimalarial medicines have an effect specifically on gametocytes (gametocidal) :
    - Primaquine.
    - Artemisinin.

- **Radical cure :**

  When P.vivax & P.ovale malaria parasites are eliminated from the blood, the remaining hypnozoites must be removed from the liver by administration of a 14-day-course of primaquine.

- **Lactating women:**

ACT except primaquine and tetracycline.

- **Severe malaria :**
    - Intravenous artesunate :
- IV or IM can be used.
- Only in hospitals, not outpatient clinics.
    - 2nd line :
        - Quinine.
        - Artemether by IM route.

Given parentrally for 24 hours and completed by giving course of ACT.

- **Prophylaxis :**
- Against hypnozoites :
    - Primaquine.
    - Atovaguone with proguanil(malarone).
- Against RBCs stage :
    - Mefloquine (pregnant & children).
    - Doxycycline.

- **Vaccine :**
- No vaccine
- RTS or masquirix , and experimental vaccine for malaria(but not used).

- **People resistant to malaria :**

- People with abnormal hemoglobin “sickle-cell anemia”.
- G-6-P dehydrogenase deficiency.
- People who has not glycophrine A receptor which act as receptor for malaria parasite
- Duffy antigens.
- Changes in genes involved in MHC.

TOXOPLSMA GONDII

- T. Gondii was first discovered in 1908in desert rodent, the gondi.
- Most common chronic infection.
- Obligate intracellular.
- 3-4 % of aids patients develop cns toxoplasmosis.
- Congenital toxoplasmosis is a serious disease.

EPIDEMIOLOGY:

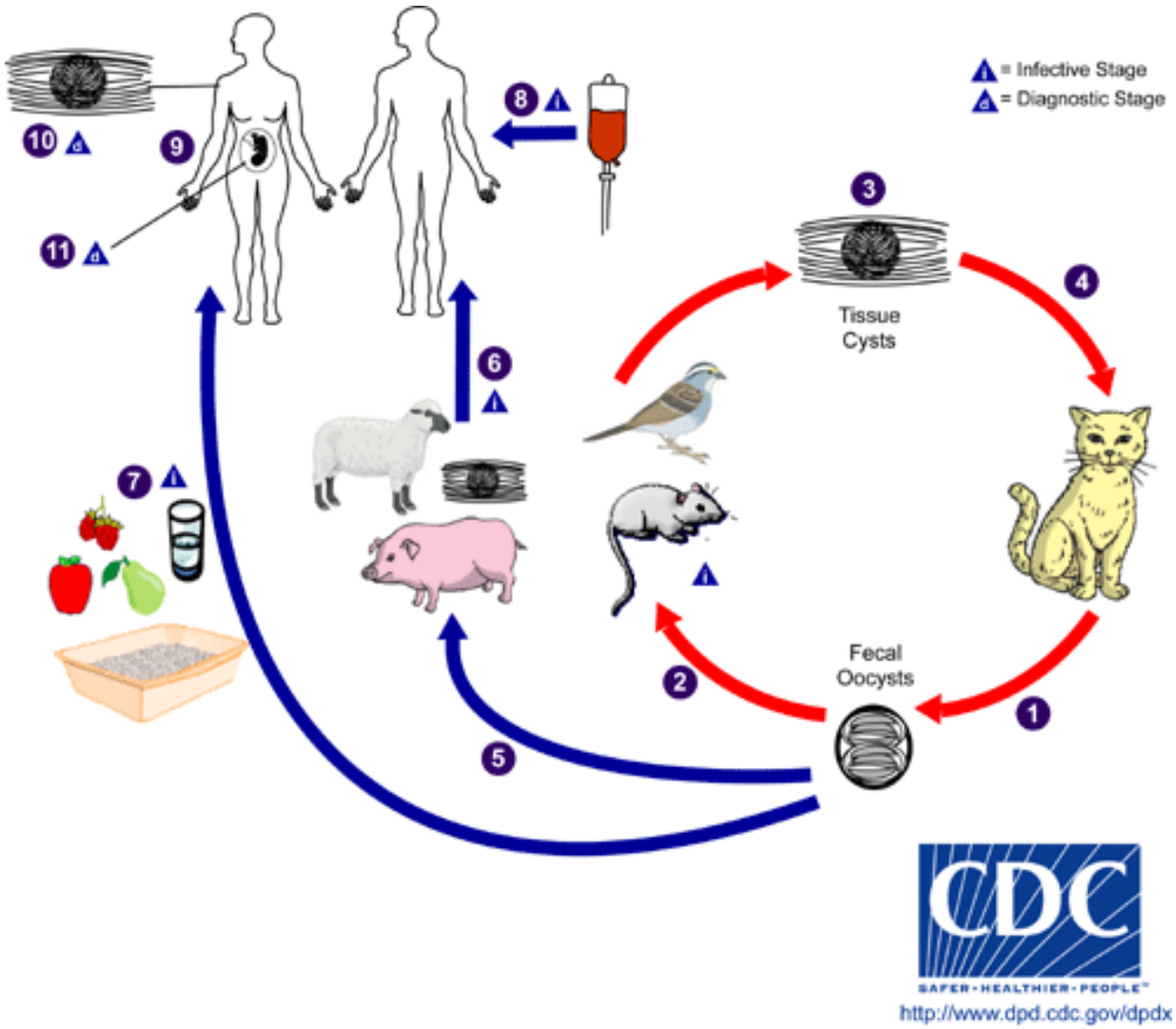
- Definite host is **cats**.
  sexual cycle → oocyst.
- Intermediate hosts are **humans**, muse, pigs and sheep.
  asexual cycle → tissue cyst.

TRANSMISSIN:

- **Oral :**
  1. Ingestion of undercooked pork or lamb meat (ingestion of tissue cyst)
  2. Exposure to oocyst :
     i. Ingestion of contaminated vegetables.
     ii. Direct contact with cat feces.

- Others :
  - Transplacental (the infective stage is tachyzoites)
  - Blood product transfusion.
  - Organ transplantation.

- The protozoa reproduce asexually in the intermediate host:
- Ingested oocyst goes to the digestive system.
- Engulfed by macrophages.
- Tachyzoites develop and travel to various parts of the body via blood steam (heart, spleen, liver, brain).
- Tachyzoites encyst to bradyziotes.
- All 3 stages are infective.



PATHOGENSIS :

Infective stage

↓

tachyzoites

↓

disseminate throughout the body

↓

Infection →any nucleated cell →multiplication →cell destruction →necrotic foci →inflammation

↓

tissue cyst → lifelong chronic infection

- Tachyzoites proliferate, producing necrotic foci surrounded by a cellular reaction (inflammation).
- In immuodeficient, the acute infection progresses, resulting in dissemination pneumonitis, myocarditis, and necrotizing encephalitis.
- Tissue cysts produce recrudescent disease in immunocompronised or congenital infected.

CLINICAL PRESINTATION OF AQUIRED INFECTIONS IN :

Immunocompetent :

- Asymptomatic.
- Mononucleosis like syndrome:
  1. Cervical lymphadenopathy.
  2. Sore throat.
  3. Fever.
  4. Malaise.
  5. Night sweating.
  6. Myalgias.
  7. Abdominal pain (mesenteric lymphadenopathy)
- Rare : pneumonia, myocarditis, polymyositis.

Immunocompromised (non AIDS patients):

- Encephalitis.
- Meningoecephalitis.
- Mass lesion.
- Myocarditis.
- Chororetinitis.
- Pneumonia.

In AIDS patients:

- **Cerebral :**
  - Manifested primarily as toxoplasmic encephalitis, include:

    Altered mental state, focal neurologic deficit, headache, fever, and seizures.

- **Extracerebral :**
  - Ocular:
    - Choreoretinitis ; confluent, thick, opaque.
    - Anterior uveitis.
  - Pulmonary:
    - Sepsis like syndrome.
  - Cardiac:
    - Cardiac tamponade.
    - Biventricular failure.

CONGENITAL TOXOPLASMOSIS :

Transmitted mainly with acute infection in 3rd trimester.

Outcome :

1. Abortion
2. still birth
3. congenital disease :

- Classical triad of **chorioretinitis**, **intracranial calcifications**, and **hydrocephalus**.
- Hepatosplenoegaly.
- Microcephaly.
- Mental retardation.
- Jaundice.
- Rash (macular or blueberry muffin lesion)
- Fever.

DIAGNOSIS :

1. **Isolation from :**

- blood.
- amniotic fluid (diagnosis of congenital infection).
- CSF.

Then inoculate in human cell lines or mouse.

2. **Serology** :

- IgM usually indicate acute infection (best for congenital infection).
- Rising IgG antibodies (best for adult infection).
  - Peaks within 2 weeks after infection.
  - Remain elevated for life.

    Sabin - Feldman dye test (gold standard)

    IFA

    ELISA
- IgG avidity test (best for pregnancy infection)
  - Low results indicate acute infection.
  - High results indicate past infection.
- PCR for detection of the organism in blood.

TREATMENT :

- First line: pyrimethamine **+** sulfodiazine **+** folic acid (the therapy cause folic acid deficiency).
- Second line: pyrimethamine **+** folic acid **+** clindamycin or clarithromycin.
- In pregnant patients:
  - Spiramycin (septrin **+** folic acid), **OR**
  - Pyrimethamine **+** sulfodiazine **+** folic acid

PROPHYLAXIS :

Indications :

- T. Gondii seropositive aids patients with cd4 t cell count of < 100 regardless of clinical status.
- T. Gondii seropositive aids patients with cd4 t cell count of < 200 if an opportunistic infection or malignancy develops.
- In these patients give trimethoprim /sulfamethazole [cotrimoxazole / septrin]

or dapsone **+** pyrimethamine **+** folic acid.

TRYPANOSOMA

HUMAN AFRICAN TRYPANOSOMIASIS (HAT) (AFRICAN SLEEPING SICKNESS)

EPIDEMILOGY:

- Fatal and neglected disease.
- 50,000 in Africa die because of this.
- 70 million are at risk.
- Human African Trypanosomiasis (HAT) affects 36 countries at sub-Saharan Africa.

CAUSATIVE AGENTS:

- Caused by the protozoan **Trypanosoma Brucei**.
- Has 3 subspecies:
  1. **Trypansoma brucei gambiense**.
  2. **Trypansoma brucei rhodesiense**.
  3. Trypansoma brucei brucei in animals only (not important clinically).
- extracellular parasite.
- Tsetse fly.

GEOGRAPHICAL DISTRIBUTION:

- Over 200 active foci of HAT between 5 degrees north and 15 degrees south ("tsetse belt").
- Trypansoma brucei gambiense is found in western and central Africa.
  - Over 95% of the cases of human infection in Sudan.
- Trypansoma brucei rhodesiense is found in eastern and southern Africa.
- Sudan is endemic of Trypanosoma.

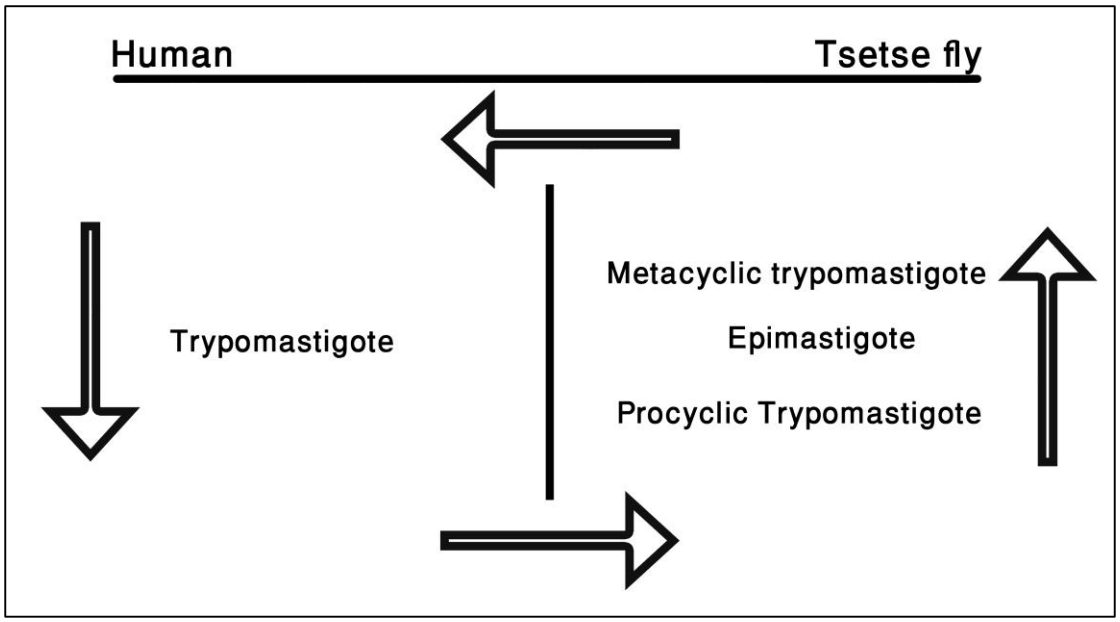
	T. b gambiense	**T. b rhodesiense**
Virulence	Less	More
Reservoir	Human	Animal
Zoonotic	Less	More
Vector	G. palpalis	G. mortisans
Distribution	Western Africa	Eastern Africa
Presentation	Chronic	Acute
Prevalence	90%	10%

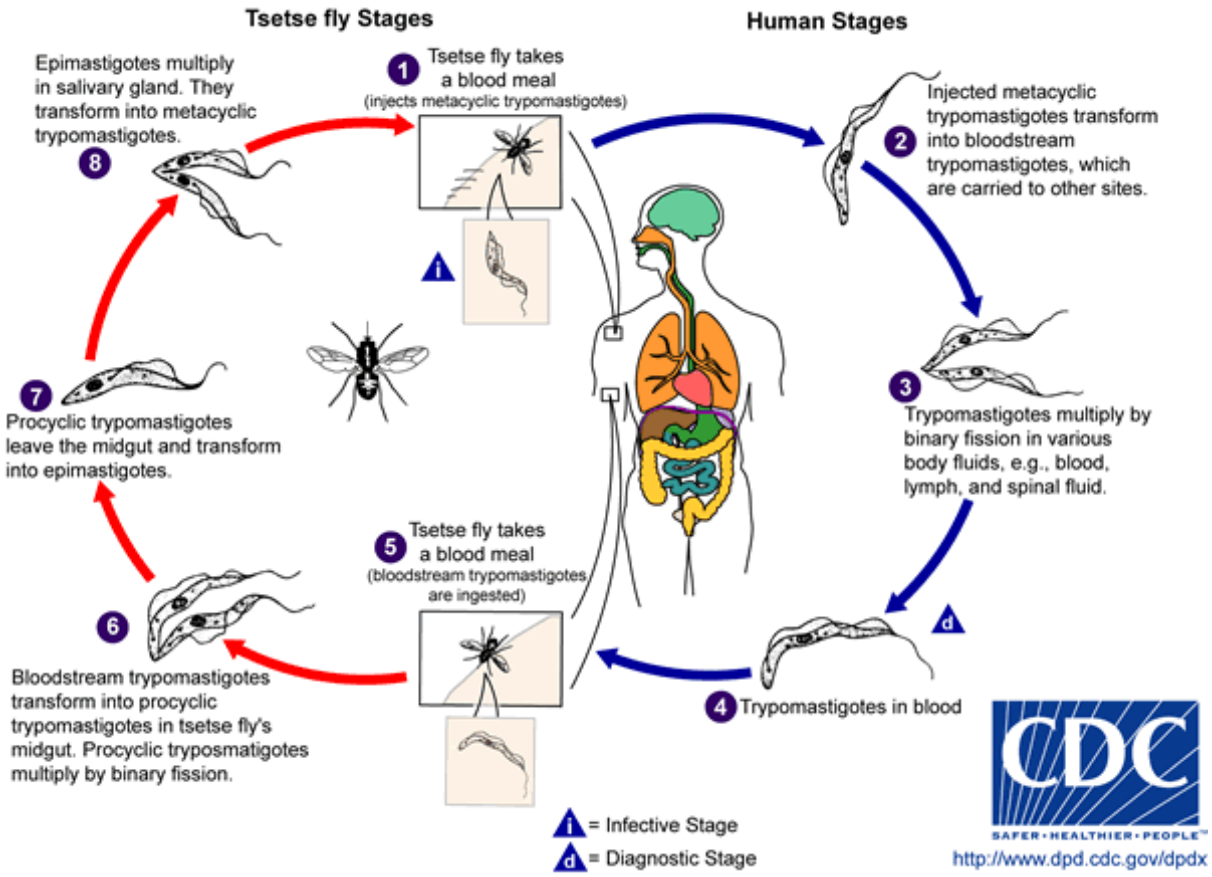
TRANSMISSION:

- Glossina palpiles (important in Sudan).
- Glossina marsitans.
- Bites during daytime.
- it both the male and female of the fly can transmit the disease.
- It can pierce the clothes.
- Vertical transmission (transplacental).

LIFE CYCLE:

The tsetse fly inject the infective stage which is the **metacyclic trypomastigote** ➔ converted to **mature trypomastigote** (extracellular) ➔ Tsetse fly take a meal of blood with the **trypomastigote** ➔ start reproduction and looks like a cycle, for that it's called **epimastigote** (is the stage of the trypanosoma inside the gut of the fly) ➔ travel to salivary gland and in the salivary gland is called **metacyclic trypomastigote**.





Mature trypomastigote → in human.

Metacyclic trypomastigote

Epimastigote

Procyclic trypomastigote

In Tsetse fly

PATHOPHYSIOLOGY:

- Affect: hematologic, lymphatic, Cardiac, GI, and central nervous systems.
- Hypersensitivity reaction type 1 & 2.
- Immune-invasion by antigenic variation of parasite surface glycoproteins: Variant Surface Glycoprotein (VSG).
- Gene rearrangement
- Antibodies because its extracellualr (Humoral response)
- This disease is never be self limited (you will never clear trypanosoma by immune response).

- VSG specific IR:
    - IgM response against VSG
    - IgM response often > IgG (Trypanosomisis is the only disease that causes IgM more than IgG).
    - Trypanosoma disappear within hours
    - VSG specific IgG appears not relavant (like brucella).
    - The level of the parasite will be fluctating, high parasaitemia, low parasitemia, high parasitemia... etc (like brucella)

CLINICAL PRESENTATION:

- Asymptomatic phase: incubation period at least 5 days.
- First stage (haemolymphatic phase)
- Second stage (neurological phase)
- **First stage:** (Heamolymphatic phase):
    - The parasite in the lymph and blood.
    - painless skin chancre (first presentation can be like this) that appears

    5-10 days after the bite, resolving spontaneously.
    - Intermittent fever
    - Malaise, myalgia, and headache.
    - Arthralgia.
    - **Winterbottum sign**:
        - Posterior cervical lymphodonepathy.
        - Characterisstic of Trypansoma brucei gambiense.
    - Urticarial, erythomatous or macular rash.

- **Second stage:**
    - Fatal

- Eventually, the disease will manage to cross the blood-brain barrier.
- persistent headache
- Daytime somnolence followed by nighttime insomnia (disturbance in sleep cycle).
- Behavioral changes, mood swings, and psychiatric symptoms. Aggressive, Depresion, and swinging in mood.
- Loss of appetite, wasting syndrome.
- Sleeping all time.
- Coma and death.

– **Signs:**
  - chancre (first sign).
  - Skin lesion. (which is called trypanids).
  - Lymphodonopathy: the classic Winterbottom sign is clearly visible.
  - Hepatosplenomegally
  - Kerandel sign (patient when sleeping will not respond to pain).
  - Fevers.

DIAGNOSIS:

– Depend on the stage of the disease:
– Direct microscopy: acute stage is diagnosed usually by direct microscopy:
  - Blood for first stage and for T b rhodiensae
  - Lymph node aspirate from winterbottum area for T b gambiense.
  - Lumbar puncture (double centrifugauation method) for the second stage.

Biopsy from ulcer if you are lucky
Intrathecal IgM is specific marker. Parasite
No serological screening test for T b rhodiensae.
The card agglutination test for trypanosomiasis/T b gambiense
CATT (like ICT in malaria).
ELISA: rarely used.
PCR: rarely used.

Diagnosis in other words:

- Acute stage is diagnosed by **direct microscopy** from the blood, lymph node or the aspirate of the chancre
- Acute stage can be detected by antigen detection in just T. gambiense and this by **Cord Agglutination Test for trypanosoma**.
- CNS stage (2nd stage) can be detected by **direct microscopy** by lumbar puncture (using double centerfiguation method).
- The other method for detection of CNS stage is the detection of **intrathecal IgM in the CSF**.
- Rarely ELISA and PCR can be used.

CHEMOTHERAPY:

- **Western African Trypanosomiasis:**
  - Stage 1
    - First line: pantamidine.
    - Second line: elforinthine or melarsoprol.
  - Stage 2:
    usually fatal.
    - First line: melaarspolol.
    - second line: elforinthine.
- **East African trypanosomiasis:**

- Stage 1:
  - First line: suramin .
  - Second line: melarsopol.
- Stage 2:
  - First: metarsoprol.
  - Second line: nifurtimox combined with melarsoprol.

PREVENTION:

- **No vaccine** against African Trypanosomisasis
- You have to fight the fly.

American trypanosomiasis
(Chagas Disease)

EPIDEMILOGY:

- 1960s
- 16-18 million people affected as of 2008.
- ~100 million at risk.
- Kills around 20,000 annually.
- Discoved by chagas and for that it's called Chagas disease.

The vector is the kissing bug which usually bites at night.

THE VECTOR:

- Blood sucking insect of the subfamily Triatominae:
  - Emerges and bites at night.
  - Bites, feeds, and then defecates.
  - Blood transfusion, Organ transplants, or breast milk.
  - 15% of intrauterine deaths are due to Chaga's disease.
- Congenital transmission:
  - 13% of stillborn deaths in parts of Brazil.
  - 15% of them die.
- Food contamination.

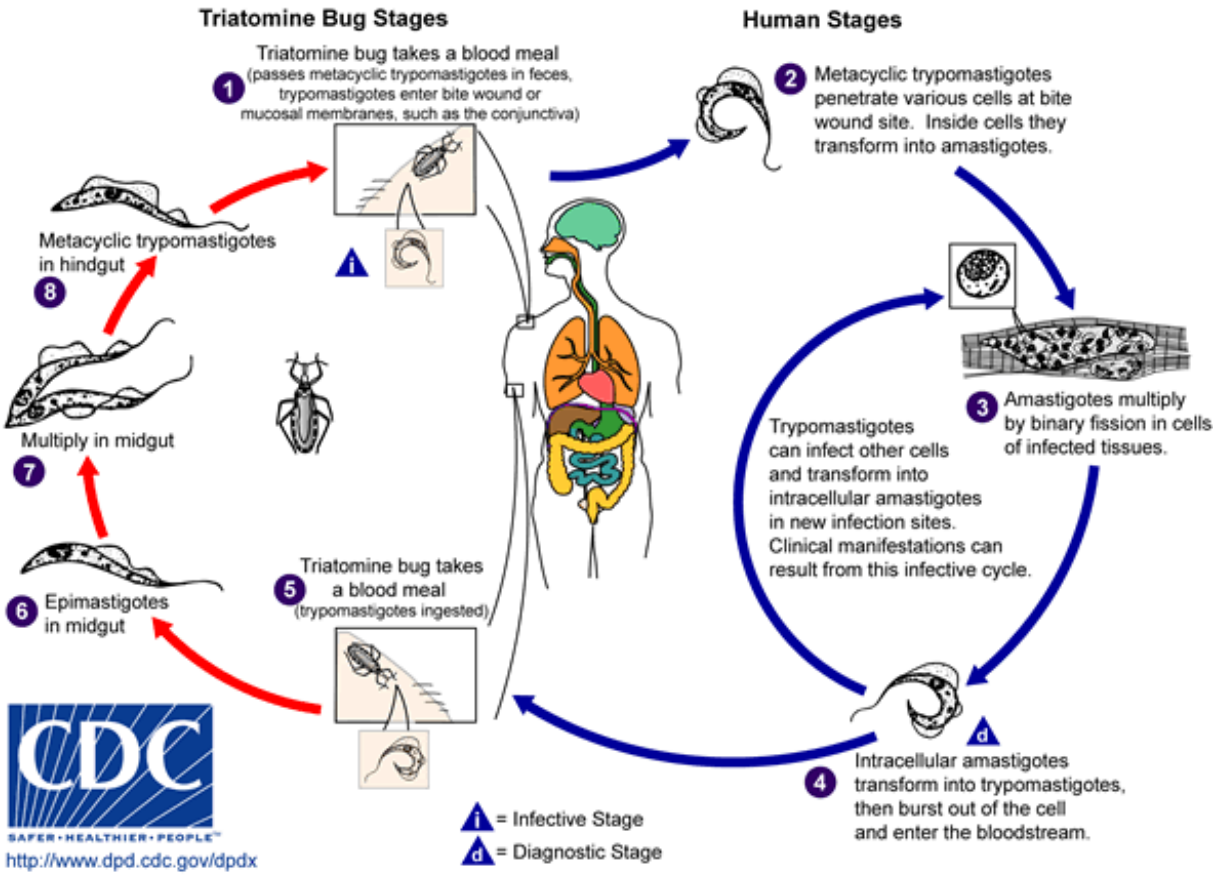
CAUSATIVE AGENT:

- Caused by **Trypanosoma Cruzi**:
  - Sylvatic cycles of transmission: raccoons and monkeys.
  - Domiciliary (dogs and cats).

LIFE CYCLE:

Trypomastigote enter your body through faeces of the buck, metacyclic trypomastigote will enter you, and stay both inside and outside the cell, inside the cells, trypomastigote will be converted onto amsastigote (looks like leishmania), when amastigote rupture will lead to trypomastigote, and continue the cycle.

The diffrence is that the trypomastigote will not enter the salivary gland, but its in the gut of the vector. that is mean the vector is not injecting.



Pathophysiology:

- Chronic inflammation.
- Intracellular parasitism of muscles.
- The GIT symptoms result from denervation of hollow viscera, reaches the mucosa as well as the muscles. (megacolon)
- Affect the heart #y chronic inflammation leading to myocarditis and dilated cardiomyopathy.

Signs and symptoms:

- Asymptomatic
- Incubation period: 7-14 days (1-2 weeks).
- **Symptoms of acute stage:**

- Non specific symptoms i.e. fever, diarrhea, vomiting.
- Swelling and inflammation.
- Chagoma (redness and inflammation at the bite).
- Roman's sign (unilateral painless palpebral edema). (unilateral periorbital edema).
- No chancre in Chagas disease.

Presentation:
Indetermine phase.
Does not cause any symptoms.(like syphilis).
In the this phase no treatment.

Symptoms of chronic stage:

- Fatal and irreversible.
- About 30% develop medical problems.
- Cardiac damage (dilated cardiomyopathy) causing atrial and ventricular rhythm disturbances, and congestive heart failure
- Mega syndrome.
- Digestive system damage
  - Megaoseophagus ➔ Dysphagia ➔ malnutrition ➔ weight loss ➔ fatal.
  - Megacolon (gastric diltation)➔ constipation and gas production.
- Megaureter ➔ obstructive uropathy ➔ Can't produce urine.
- Neurological disorders :
  - In late stage of the disease.
  - Because of infection of the parasite to the CNS as well as cardiac myopathy.

Signs of Acute stage:

- Roman's sign.
- Chagoma (erythema).
- Hepatomegally and **splenomegally**.
- Facial edema.
- Generalized **lymphodonopathy** (like visceral leishmaniasis)
- Schizotrypanides.

Signs of chronic stage:

- Cachexia.
- Hypertrophy of the salivary gland ➔ no eating.
- Pneumonia: because of megacolon, the food will enter into the larynx ➔ trachea ➔ Aspiration pneumonia.

Can lead to death because of:

1. Heart failure.
2. Aspiration pneumonia.
3. Shock.

DIAGNOSIS:

- **Acute Chagas disease:**
  - Direct microscopy of the blood (mainly).
  - PCR to ID strains (for conformation).
  - Xerodiagnosis nor Homoculture are not useful.

- **Chronic Chagas disease:**
  - Parasite inside the heart, esophagus, colon.
  - Biopsy is difficult.
  1. Serological diagnosis:
     - IIF.
     - ELISA.
  - Indirect haemoagglutination.

- The Chagas Radioimmune Precipiatation Assay (Chagas RIPA).

2. PCR.
3. Biopsy: if the patient died.
4. Others: can use imaging by CT and MRI for dilated esophagus as well as the colon and heart.

TREATMENT:

1. **Acute Chagas disease:**
   - First line: Benznidazole (for indetermine phase).
   - Second line: Nifurtimox.
2. **Indeterminate-phase Chagas disease:**
   - Children should receive either benznidazole or Nifurtimox (for adult not useful).
3. **Chronic symptomatic chagas disease:**
   - General medical treatment.
   - Surgery.
   - No cure.

   Colostomy, gastrotomy tube, cardiac transplantation, nephrotomy tube, anti peristaltic, Diuretics etc…

LEISHMANIASIS

- Parasitic disease, transmitted by SAND FLY (the vector).
- Found in 88 countries.
- 3 main forms of Leishmaniasis:
  1. Cutaneous (involving skin).
  2. Mucocutaneous (involving mucus membranes of mouth & nose).
  3. Visceral (liver, spleen, & bone marrow).

CUTANEOUS LESHMANIASIS

- About 1.5 million cases each year.
- It's distribution is divided into old world & new world (America).
- Spectrum of the disease:
    1- Localize L. (LCL).
    2- Diffused L. (DCL).
    3- Mucosal L. (destructive mucosal inflammation-mouth & nose).
- Different spp. of L. cause different types of disease.
- Found in exposed areas of the body (face, forearm, feet "rare").

TRANSMISSION:

- Old world: Phlebotomus sand fly (orientries الشرق الأوسط).
  Female hematophagus.
- New world: Lutzomyia sand fly.

CAUSATIVE AGENT:

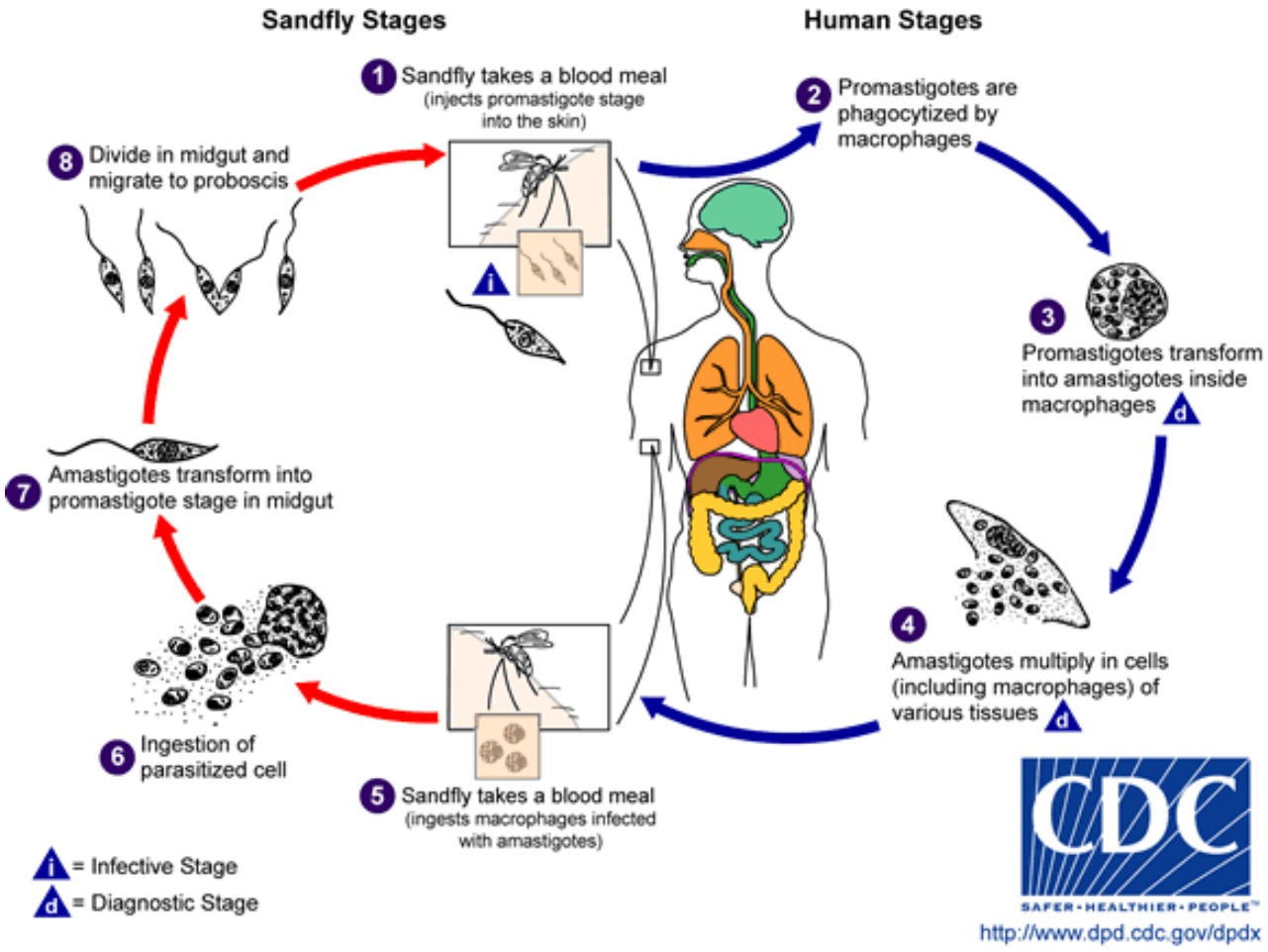
1- **L. aethiopica** (cause LCL & DCL, zoonophilic reservoir is animals, in Sudan: Camels & rodents).
2- **L. major** (major causative agent).
   Cause all 3 forms of the disease, most severe, & most virulent spp.
3- **L. tropica**:
   Cause localized disease, anthropophilic, benign, & no animal reservoir.

LIFE CYCLE:

1. **Promastigote**:
   - Insect stage.
   - Motile (flagella).
   - In the midgut of the insect.

2. **Amastigote**:
   - Mammalian stage.
   - None-motile.
   - Intracellular (inside macrophage) the parasite can`t live outside the cell.
   - The parasite cause infection, macrophages move toward the parasite (non-motile), & it cause rupture of macrophages.

   - S.F. -- enter the body --→ (promastigote) --- enter macrophage --→ (amastigote) --→ taken by S.F.



INITIAL INFECTION:

- Inoculation of promastigote.
- Receptor mediated phagocytosis into macrophages.
- Inflammation (ulceration)
- Pathogenesis appear to be related to T-cell cytotoxicity.

Forms of C.L.:

1- **LCL:** " one ulcer"

- Incubation period 2 weeks-month.
- Lesion starts as **erythema**, develops into a **papule** then a **nodule** that progressively ulcerates over a period of 2 weeks to 6 months, then heals by fibrosis causing **scar**.
- Skin sores (oriental/Bagdad – sores) of cutaneous L. can heal by their own (2-15 month).
- Sores can leave significant scars (healing by fibrosis), it can be disfiguring if they occur on the face.
- If the infection is by L. tropica it can spread to contiguous mucus membrane (Upper lip to nose).

Leishmaniasis Recidivans:

- Reactivation of dormant parasites or new inf. By a different spp.
- New ulcers & papules form over the edge of the old scar.
- If the immunity is low, parasites are reactivated in the normal skin around the old lesion.

2- DCL:

- Non-ulcerative nodules disseminated from the initial site of infection &cover thept.'s entire body.
- In immune-compromised & malnourished, DCL is difficult to be treated & the disease is not self-limited.
- Non-ulcerative. (As in M.Leprae).

- Differential Diagnosis of a face full of nodules:
  1. DCL.
  2. Leprosy.
  3. PKDL.

3- Mucocutaneous L.:

- Only in the new world.
- More severe than cutaneous L. & can cause death.
- Species:
  1. L. braziliensis (the most common).
  2. L. Mexicana.
     - ✓ The only spp. u should consider.
  3. L. venzwellensis.

    - All are zoonotic, first cause LCL & then it develops to MCL.
    - Affect cartilage, muscle, fibrous tissues (except bone & teeth).

- There is 2 types of MCL:

  1- Mild mucosal L.:
    - Benign with nasal infection & stuffiness.
    - Ulceration of the nasal mucosa & perforation of the septum.

  2- Severe MCL:
    - A hole in the soft palate (brain), pharynx (dysphagia), or larynx (pneumonia).
    - Never heal spontaneously.
    - Death from secondary bacterial infection.
    - Dysphagia → malnutrition.
    - Pneumonia & respiratory compromise.

IMMUNITY:

- Disease resolution is achieved by the cell mediated response.
- The primary activation of T-cell subsets is importantfor the development of:
- T-helper 1 ➔ LCL (مفيدة) activation of cell- mediated immunity -> killing the intracellular parasite -> more localized inf.
- T-helper 2 ➔ DCL (غير مفيدة) activation of A.B. production through B-cell -> more antibodies without killing the intracellular parasite.
- <u>Patients with any of the following findings should be referred:</u>
  - Big lesions (greater than 1 inch in size).
  - Many lesions (3 or more, DCL).
  - Sores in the face (MCL).
  - Sores in hands & feet (affect life style).
  - Sores over the joint (can cause stiffness& loss of function after healing by fibrosis).

DIAGNOSIS:

1- **Microscopic examination** (**gold standard-best method of <u>detection</u>**):
   - The sample is aspirate or biopsy.
   - By Gimsa stain: see amastigote inside macrophages.

2- Culture (for <u>identification</u>):
   - NNN (Nicolle-Novy-MacNeal) media.
   - See promastigote.

3- **PCR**: when? (indications)
   - With low parasite load (mucosal L.).
   - For therapy monitoring.

4- **Montenegro skin test** (Leishmanin test):

- Inject PPD (purified protein derivative of tuberculin) from the parasite → if there is swelling means the pt. has the disease in the past or now.
- Not effective!!! Because it can`t differentiate between acute & chronic.
- Like Tuberculin test (Mantoux test).

5- **Serological diagnosis** is rarely used in cutaneous L. diagnosis.

Treatment:

Cutaneous & Mucocutaneous L.:

- Cutaneous L. is treated to accelerate cure & to reduce scaring, otherwise it can heal spontaneously.
- **Antimony (Pentostam & Sodium stibogluconate) is the drug of choice**, given I.V. or Intra-lesion to act locally.
- Amphotericin B (antifungal).
- Paromomycin (anti parasitic).
- I.F.N ɣ (immune mediator to increase T-cell immunity).
- Fluconazole (antifungal).

Vaccines:

1- Leishmanisation:

- Currently not recommended by WHO.
- Inoculation of the virulent L. parasites.

2- Killed vaccine:

- Killed parasites.
- Had been developed in South & Central America, Sudan, & Iran.
- Not useful.

VISCERAL LEISHMANIASIS (KALAZAR)

CAUSATIVE AGENT:

- Leishmania Donovani complex
- Sub species:
  1. L.d. donovani (main).
  2. L.d. infantum (main).
  3. L.d. chagas.
  4. L.d. archibaldi.

EPIDEMIOLOGY:

- 500,000 new cases/ yr, >50,00 deaths
- Very common in Sudan
- Animal reservoir: Dogs, Jackels& foxes
- Transmission: by female phlebotomine sand flies.
- Digenic life cycle: promastigote in the sandflies &amastigotes in the human.
- Lifecycle similar to that of CL the difference in VL is the tissue tropism:
- L.d likes blood therefore expected to be found in the RES (liver, spleen, Bone marrow, lymphnodes) & the blood.

PATHOGENESIS:

- Inoculation of promastigotes into skin, internalized by dendritic cells & macrophages in the dermis & transform into amastigotes.
- Macrophages disseminate through the lymphatic & vascular system(dendritc cells stay in the skin) in:
  - Bone marrow → failure
  - Lymphnode → generalized lymphadenopathy
  - Liver → hepatomegaly

- Spleen→hypersplenomegaly

– Immunity is cell mediated (mainly Th-1 cells)

RISK FACTORS:

– Malnutrition.
– Immunosuppressive diseases (e.g. HIV, leukemia, immunosuppressive drugs).
– Young age.
– Decrease production of Interferon gamma (low immunity).
– Polymorphism (mutation in the gene that leads to more than one allele) in the TNF (low immunity).

SYMPTOMS:

– In Sudan >30% asymptomatic.
– Symptoms usually occur after months due to:
  1) division cycle of amastigote is slow (similar to M.TB).
  2) till it reaches the RES where proliferation occurs.

o Fever (any type but mainly low grade fever)
o Weight loss (due to high metabolic rate despite v.good appetite)
o Nausea, vomiting, low grade watery diarrhea
o Generalized lymphadenopathy
o Hepatosplenomegaly
o Hyper pigmentation (Black fever in India)
o Anemia: Chronic disease
  Bone marrow suppression (due to IL-1, IL-6, TNF)& infiltration (due to amastigotes proliferation)

Hyperspleenism (destruct RBCs, platelets)

o Leukopenia- complicated by 2nday bacterial infection.
o Thrombocytopenia: epistaxis & bleeding presentation.
o Elevated liver enzymes & presentation of Jaundice.
o Hypergammaglobulinemia (B-cell respond to different amastigotes in diffrenetmacrophages by proliferation&

polyclonal Abs production that are not useful bczleishmania is an intracellular organism that requires the CMI.

The importance of this is that the B-cells get exhausted & when the body is infected by an extracellular organism it can't defend against it by Ab production)

POST KALAZAR DERMAL LEISMANOID (PKDL):

Complication of Visceral Leishmaniasis.

Between 6 months-2 years after recovery.

Pathophysiology: Recrudescence (reactivation of un-detectable parasites in the dendritic cells that have not been destructed by the treatment- it works on macrophages of the RES more).

Macular, maculopapular& nodular skin lesions that start periorally& then extend to the whole body.

The main loci are in Sudan where PKDL follows healed in 50%.

The disease is a spectrum:

- Good immunity-low parasite number: Cut. L
- Low immunity- high parasite number: Visceral .L/ chronic cut.L (worst)
- better immunity than that of VL: PKDL
- V.good immunity- v.low parasite number: Asymptomatic

DIAGNOSIS:

- Specimen: aspirate & biopsy of LN/ BM (huge spleen/liver & low platelets causes severe bleeding that can lead to death so not taken from there.)
- Microscopy: visualization of amastigotes in macrophages (Leishman Donovan bodies) - "gold standard."
- Culture: on NNN media -promastigote
- Serology:

1. Ag detection:

- Latex agglutination test.

Two urinary Ags (CHO Ag).

The urine must bebolied to avoid false positive (to destroy the Abs)

Disadv: the pt. must be symptomatic.

Adv: easy, specific & sensitive.

2. AB detection:

- Anti – k39 by ICT (Immune-chromatographic test)

Takes 10 mins

Useful in the field.

- ELISA
- DAT (Direct Agglutination Test)
- IFAT (immune fluorescent Ab test)

Abs to Leishmania represent previous infection/ chronic disease.

Positive Ab test + clinical symptoms= disease.

Positive Ab test + no symptoms = prev. infection.

3. Others:

- DNA detection methods (PCR)
- CBC (Pancytopenia)
- Formal gel test: Polyclonal hypergammaglobulinemia

Disadv: non-specific (+ve also in Brucella)

TREATMENT:

- First line therapy:
  - Pentavalentantimonias (Pentostam)
  - Amphotericin B
  - Miltefosine: anticancer also
  - Paromomycin
  - Toll-like rec agonist (Imiquimod)-imrove T-cell immunity.

- Combination therapy:
  - AmBisome + Miltefosine
  - Ambisome + Paromomycin
  - Miltefosine + Paromomycin.

VACCINE:

Killed vaccine not recommended

Immunechemotherapeutic vaccines (drug + vaccine):

In Sudan, **BCG+ Pentostam**

Treatment of PKDL & for prevention of Visceral Leismaniasis.