### **KINETOPLASTA TRYPANOSOMES**



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#### **TRYPANOSOMES AND THEIR KINDS**

Infect humans to plants

3 families

**Bononidae** – coprozoic and free living **Crytobiidae** – parasites of fish and invertebrates **Trypanosomatidae** – human and veterinary pathogens

# **CELLULAR STRUCTURES**

- Mitochondria single, contains dark staining body – kinetoplast – contains the DNA, a network of linked circles
- **Microtubules** in cytoskeleton, beneath the plasma membrane
- Flagellar pocket paraxial rod along the axoneme, connecting it to microtubules of flagellum Undulating membrane – depends on the species Glycocalyx –present occasionally



### TRYPANOSOMATIDAE

1 nucleus and either elongated with 1 flagellum or rounded with non-protruding flagellum

#### Many are heteroxenous

- 1 stage in blood and fixed tissues and other stages in the intestine of blood sucking invertebrates (vectors)
   Hemoflagellates
  - requires blood for culture
  - Monoxenous
  - found only in a single arthropod host

### TRYPOMASTIGOTE FORM

- Characteristic of blood stream forms and metacyclic forms in tsetse fly vector
- Has undulating membrane
- Kinetoplast and kinetosome at the posterior end
- Mitochondria
  - Blood form may or may not have tubular cristae anterior to the kinetoplast
  - Insect form becomes larger and complex lamellar cristae
- Flagellar pocket
- Pellicular microtubules just beneath the plasma membrane
- RER and Golgi between nucleus and kinetosome
- See (fig)



#### Figure 5.3

# PROMASTIGOTE FORM 4---7

- Elongated body
- Flagellum as a functional organelle
- Kinetoplast and kinetosome at the anterior end, in front of nucleus
- Several species while in the insect host
  - *Leptomonas*See (fig)



#### Figure 5.3

# **EPIMASTIGOTE FORM**

- Characteristic of some life cycles
- Has short undulating membrane
- Kinetoplast and kinetosome between nucleus and anterior end
- Parasite of insects
  - Crithidia and Blastocrithidia

See (fig.e)



#### Figure 5.3

# PARA- AND OPISTHO-MASTIGOTE FORM

- Parasites mainly of flies
- No undulating membrane
- Flagellum runs the length of the body before exiting
- Paramastigote
  - Kinetoplast and kinetosome by the nucleus
- Opisthomastigote
  - Kinetoplast and kinetosome between nucleus and posterior end

See (fig.d)



#### Figure 5.3

# OTHER FORMS

- Amastigote see (a)
  - Spheroid
  - Very short flagellum
  - Leishmania
- Choanomastigote see (b)
  - Flagella moves out through a wide collar
  - Crithidia



#### Figure 5.3

# LIFE CYCLES

Trypanosomatid species are very diverse in their life cycles
 Leptomonas – insect is the sole host, multiply as
 promastigotes and spread by ingestion of an amastigote like cyst

*Leishmania* – multiply as promastigote in blood sucking insects but injected into vertebrate host when the insect feeds and it undergoes replication as amastigotes in a variety of tissues, usually in macrophages

*Trypanosoma* – all but T. equiperdum are heteroxenous or are transmitted by animal vectors

- Parasites of all classes of vertebrates live in the blood and tissue fluid
- T. cruzi occupies intracellular habitats
- Transmitted by blood feeding invertebrates (Insects)

### **TRYPANOSOMA SECTIONS**

#### 2 broad sections

- Salivaria develops in anterior portion or station of the digestive tract
- Stereocaria develops in posterior or station of the digestive tract



## SALIVARIA

#### • T. brucei has 3 subspecies

- Tb brucei infects introduced livestock not humans, bloodstream of native antelope and other ruminants
- Tb gambiense chronic form of African Sleeping Sickness; West and Central Africa
- Tb rhodesience acute form of African Sleeping Sickness; Central and East Central Africa
- Different pathological syndromes
- Widely distributed in tropical Africa in the fly belt
  - Glossina spp of tsetse fly

# HOST LIFE CYCLE

- Parasite morphology
  - Pleomorphic from long and slender to short and stumpy
  - Small kinetoplast very near the posterior end and undulating membrane is conspicuous(clear)
- Multiply as trypomastigotes in blood and lymph
  - Chronic disease may invade the CNS within the brain
- Live in blood, lymph nodes, spleen and CSF
  - Not in the cells but in the connective tissue spaces or the reticular spaces of spleen and lymph nodes
- Treatment with Suramin (anti- trypanosomal drug) will affect the respiratory system of the parasite

# **VECTOR LIFE CYCLE**

- Vector Glossina sp. (Vector Fly)
  - 90% of the flies are refractive to the parasite
  - Susceptible flies will take up the Tb where it will go to the midgut of the insect
  - Trypomastigote form for ~10 days, move into the foregut for 12 to 20 days, then migrate to esophagus, pharynx or hypopharynx then to the salivary glands, becomes epimastigote
    - Attach to cells or lie free in the lumen
  - Undergo asexual reproduction and then transform to metacyclic trypomastigotes short and stumpy
    - Infective to vertebrate host
    - Inject several thousand flagellates

In mammals



# **TRYPANOSOMA BRUCEI BRUCEI** (TBB)

- Infects horse, dog, mule, donkey and other ruminants
- Symptoms and signs
  - Anemia, edema, watery eyes and nose and fever
- Animals become emaciated(poor), uncoordinated and paralyzed followed by death
- Cattle can live for several months before death and swine will recover

# TRYPANOSOMA BRUCEI RHODESIENSE (TBR) AND TRYPANOSOMA BRUCEI GAMBIENSE (TBG)

- Human infection small sore or chancre forms at the site
- Invades nearly all organs of the body

TRYPANOSOMA BRUCEI RHODESIENSE (TBR)

- Tbr rarely in the nervous system
- Rapid weight loss
- Heart involvement
- Symptoms of African Sleeping Sickness are rarely seen as death happens rapidly before onset

# TRYPANOSOMA BRUCEI RHODESIENSE

- Trypomastigotes; peripheral blood smear.
- Note the undulating membrane (U),
- Anterior flagellum (F),
- and posterior location of the kinetoplast (K) relative to the nucleus (N).
- These would be indistinguishable from trypomastigotes of *T.b.gambiense*.
- The long slender form of trypomastigote is the dividing form (arrow),
- Whereas the short,
- Stumpy form (not shown)
- Is infective for the intermediate host.



# TBG

- Tbg enters the nervous system rapid course to death
  - LN (lymph node) swollen (Winter bottom's sign) and congested in neck, groin and legs
- Intermittent periods of fever, increase in pain and swelling LN, headache, weakness and cramps
- Invade CNS and the host is sluggish and apathetic
- Sleepiness increases until coma and death ensues



# **TRANSMISSION CONTROL**

- Influences on transmission
  - Reservoir hosts
  - Presence of vectors and environment for hosts to reproduce
- Controlling the vector will help to control the disease
  - Keep brush to a minimum
  - Remove the reservoirs
  - Avoid "fly-time"
  - Develop Trypano-tolerant cattle

## **TSETSE FLY**

- Glossina sp. (tsetse fly), the vector of African trypanosomiasis due to *Trypanosoma brucei brucei* in animals, and *T. b.* gambiense and *T. b.* rhodesiense in humans
  - Although most flies are not susceptible to infection,
    both males and females can serve as vectors



### **STEROCARARIA**

- Posterior portion of the gut of the vector
- Most prominent parasite Trypanosoma cruzi causative agent of Chagas disease – acute febrile disease of children
  - 12-19 million people in Central and South America
- Develops in the gut of the conenosed (blood sucking) bug or beetle as epimastigote type, passed in their feces
- Many types of wild and domesticated animals serve as reservoirs

# TRYPANOSOMA CRUZI



#### Trypomastigotes; blood smear

- Free flagellum, moderately long undulating membrane
- Posterior location of larger size of the kinetoplast (K)
  - Characteristic "C"-shape of several cells

Amastigotes develop in muscle and other tissues

 Now T cruzi is able to reproduce, loose flagella, undulating membrane and divide by binary fission, cell lyses and releases new parasites

Trypomastigote is non dividing, and serves instead to disseminate the infection to tissue cells and to the vector, reduviid bug

# LIFE CYCLE OF T.CRUZI

#### Life cycle:

intracellular amastigotes (dividing)  $\rightarrow$  burst cell  $\rightarrow$  trypomastigotes in blood (non-dividing)  $\rightarrow$  re-invade cell or ingested by kissing bug  $\rightarrow$ dividing epimastigotes in bug gut  $\rightarrow$  metacyclic trypomastigotes in reduviid bug feces  $\rightarrow$  rubbed into bite wound or eye  $\rightarrow$  intracellular amastigotes



# LIFE CYCLE IN THE VECTOR

- Triatomic bugs injest trypomastigotes that pass to posterior midgut – short epimastigote
- Replicate by longitudinal fission long and slender metacyclic trypomastigotes in their rectum





KISSING BUG, TRIATOMA GERSTAECKERI (FAMILY REDUVIIDAE, SUBFAMILY TRIATOMINAE)

- Potential vector of *Trypanosoma cruzi*
- Entire bug viewed from above
- Bug feeding on a mouse with an extended proboscis
  - cone-shaped head –
     "conenosed" bug

### EPIMASTIGOTES OF T. CRUZI



- Replicating stage found in the gut of the kissing bug
- Undulating membrane (U)
   and anterior location of the
   kinetoplast (K) relative to the
   position of the nucleus (N)
- These are indistinguishable from epimastigotes of other trypanosomes

# **CHAGAS DISEASE**

- Acute stage, disease in young children, sometimes fatal
  - Local inflammation at the bite site, causing a small red nodule Chagoma
  - Romana sign edema of the conjunctive because of the parasite entering through the eye
  - Anemia, loss of strength, nervous disorders, chill, muscle and bone pain and varying degrees of heart involvement
- **Chronic stage**, disease in the adult
  - CNS and PNS involvement
  - Gastrointestinal form megaesophagus and megacolon, due to destruction of autonomic ganglia

# CHAGAS DISEASE (CONT)

- Cardiac form cardiomegaly, ventricular aneurism (sac like widening), and arrhythmia( abnormal beating rate of heart), due to destruction of heart muscle and conducting cells
- Patients develop psuedocysts pockets of parasites in the muscle tissue
- **Diagnose** by trypanosomes in the blood, CSF, tissues or lymph
  - Xenodiagnosis allow triatominess to feed on patient and look 10-30 days later for intestinal flagellates
- Doesn't respond well to chemotherapy, kills only the extracellular forms

### Amastigotes of T cruzi



- Pseudocyst in a section of heart muscle
- Note necrosis in upper right corner.

### Amastigotes of T cruzi



- Spleen smear
  - Note the absence of an undulating membrane or emergent flagellum, the kinetoplast (K) is more darkly stained than the nucleus (N), and the parasite's cytoplasm is unstained
- Amastigotes of *T. cruzi* would be indistinguishable from those of L. donovoni