

# BIO 475 - Parasitology

## Spring 2009

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<http://www4.nau.edu/isopod>

### Lecture 5

## Discovery of the Disease

In 1924 the Kala-Azar Commission noted that the distribution of a sandfly (*Phlebotomus argentipes*) in India closely overlapped the distribution of the disease.

In 1939 Smith, Haldar and Ahmed discovered that if sandflies after taking a bloodmeal were fed on raisins instead of being given additional blood meals, the flagellates grew so numerous that they blocked the pharynx as happens with plague bacilli in fleas.

These workers then subjected hamsters to the bite of blocked sandflies and each became infected.

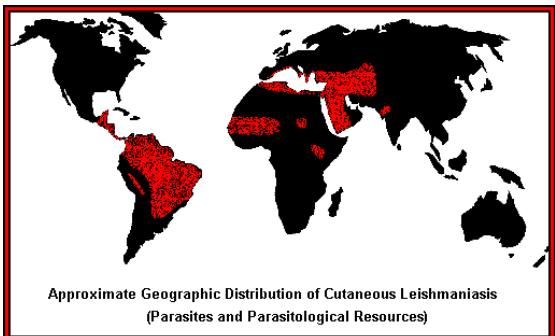


## Discovery of the Disease



Human transmission of leishmaniasis was demonstrated in 1941 when Adler and Ber successfully infected volunteers with *L. major* by the bite of *P. papatasii*.

In 1942 Swaminath, Shortt and Andersen allowed 6 human volunteers to be bitten by infected *P. argentipes* and all developed kala-azar.



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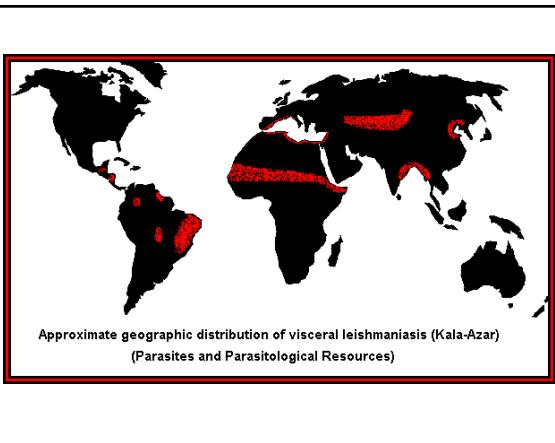
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## Leishmania-like Parasites

1. Life cycle is most similar to ancestral trypanosomes.
  1. *Leptomonas*
    - a. monoxenous, replication of promastigotes in gut,
    - b. formation of amastigote like cysts that are evacuated in feces
    - c. other insects ingest the cysts.

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## Leishmania Vectors

- a. Heteroxenous, replication of promastigotes in insect gut.
- 1. Usually in blood sucking insects like sand flies.
- 2. most common genus is *Phlebotomus* (Old World)
  - a. also *Lutzomyia* (New World).

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*Phlebotomous*

## Sandflies

*Lutzomyia*



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## Life cycle of Leishmania

- A. Injection of *promastigotes* into vertebrate host.
- 1. These forms are phagocytised by macrophages.
  - a. Within *parasitophorous* vesicles, become amastigotes
  - b. Like viruses, take over cellular/organelle machinery.

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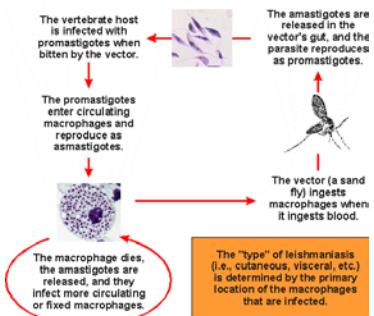
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### THE LIFE CYCLE OF *LEISHMANIA* spp. (VARIOUS FORMS OF LEISHMANIASIS)



## Pathology of *Leishmania*

1. Varies depending on species
  - a. May be *cutaneous* - forming lesion on skin.
  - b. May be *mucocutaneous* - lesion on mucous membranes with subsequent tissue erosion.
  - c. can be *visceral* - erosion of viscera, usually fatal.

## Species of *Leishmania*

1. Many in lizards, but also in mammals
2. Some mammals (dogs, gerbils) serve as *reservoir hosts*.
  - a. Can carry the infection and maintain it, permitting later infection of humans.

## *Leishmanias Infecting Humans*

1. *L. donovani* - visceral leishmaniasis
  - a. also known as "kala azar" and dum-dum fever.
  - b. Named for co-discoverers (Leishman-Donovan) 1900-03
  - c. Diagnosis of "L-D bodies" is after them.

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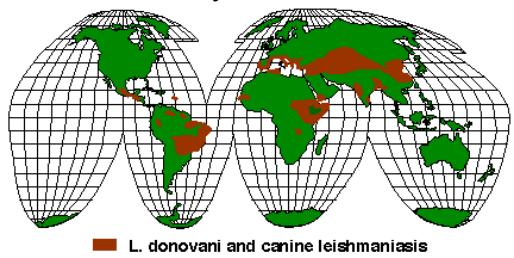
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## *Leishmania donovani*

1. Mainly restricted to Old World.
2. Appears to have been transmitted to New World by slave trade?



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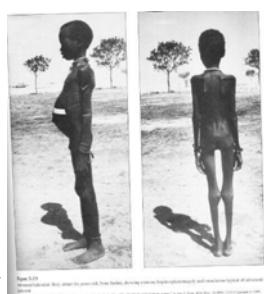
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## *L. donovani* Pathology

1. Slow onset of fever, later involvement of viscera,
2. Later abdominal edema, splenic enlargement.
  - a. Often fatal, but also can spontaneously clear.



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## However,

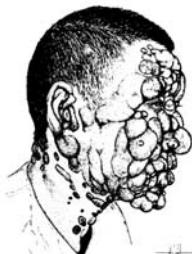


FIG. 37. A severe lympho-proliferative case of kala-azar infection caused by Leishmania donovani. (Source from a photograph. Courtesy of Dr. Robert Kania.)

1. Cleared cases can lead to Post-kala-azar dermatoid.
  - a. Horrible disfiguring
  - b. Can be cured with drugs.

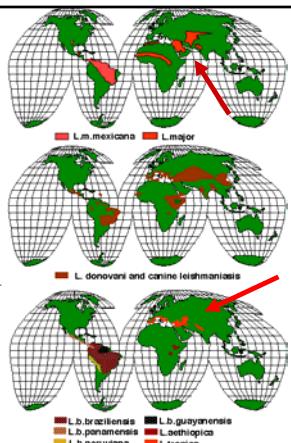
## Leishmanias Infecting Humans

1. *L. tropica*, *L. major* - oriental sore - (Old World cutaneous).
  - a. Widely distributed, mostly in Middle East
  - b. Lesions on exposed areas - subject of sand fly bites.

## *L. tropica*, *L. major*

Old World cutaneous

- a. Widely distributed, mostly in Middle East
- b. Lesions on exposed areas - subject of sand fly bites.



### CUTANEOUS LEISHMANIASIS:

#### Old World Cutaneous Leishmaniasis (CL)

##### *L. major*

causes a moist, cutaneous, ulcerlike lesion at the site of the bite; it starts as a papule that runs an acute course (1-3 weeks) with early ulceration and a surrounding zone of inflammation, that usually heals in two months to a year leaving a depressed unpigmented scar, and lasting immunity.



FIGURE 45-1 Clinical cases on face and forehead. (Courtesy of Ash and Saha, *Pathology of Tropical Diseases*, Armed Forces Institute of Pathology, #82.717.)

It is transmitted by *Phlebotomus spp.* from gerbil, dogs or rodents to human or human-to-human. It is generally found in sparsely populated rural areas.

### Leishmanias Infecting Humans

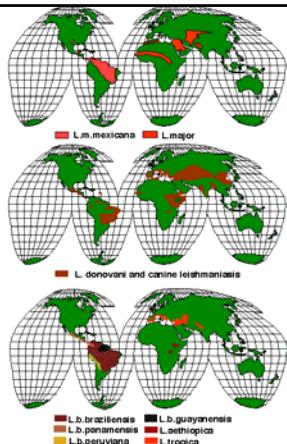
1. Also, flies hang around lesions
2. transfer may also happen by Muscoid flies (ick!)
3. However, may not be true since few amastigotes are found here.
4. Often heals with lifetime immunity
5. Some cultures innoculate their kids.

### Leishmanias Infecting Humans

2. *L. braziliensis* (New World cutaneous).
  - a. Mainly in the new world
  2. In woodland areas
- b. Also vectored by sand flies (*Lutzomyia*)

## *L. braziliensis*

- c. Lesion usually heals up with no problem.
- d. But, occasionally metastasizes if it involves mucous membranes.
- 1. can erode face, resp. structures
- 2. condition is known as "uta" or "espundia."



Mucocutaneous leishmaniasis, Peru 1983

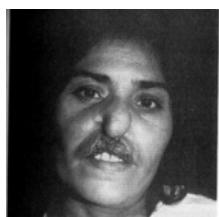


Figure 4-13. Mucocutaneous leishmaniasis in a Honduran female, probably due to *L. b. braziliensis* or *L. b. panamensis*.

## *Leishmania mexicana*

- 1. Mainly in central Mexico, Caribbean.
- a. Responsible for a condition called "chiclero" after harvesters of gum tree, *chiclé* (Chicklets)



Fig 13.5 "Chiclero ulcer" due to *L. (L.) mexicana*. Almost total destruction of the the external ear in a chicleo from Belize with an infection of many years duration. (From Lainson and Silveira-Orive 1983.)

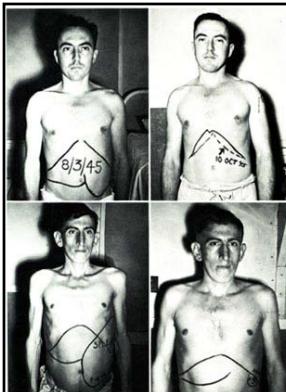
## Leishmania Medications

1. Usually with *antimonials* (mercury containing compounds).
  - a. some plants are useful too (dogbanes, gentians).



## Leishmania Medications

- Kala azar is treated today essentially as it was in 1940. The major drug is Sodium **stibogluconate** or **Pentastam**, a derivative of antimony, which was developed in 1930! Severe reactions including death occur in 10% of those treated. It is very expensive, and the recommended one month treatment costs around \$150. Drug resistance has also developed. Up to 70% of infected patients in India have infections that are resistant to this drug.
- **Amphotericin** is used with or after an antimony compound for visceral leishmaniasis unresponsive to the antimonial alone.
- **Pentamidine isotionate** has been used in antimony-resistant visceral leishmaniasis, but although the initial response is often good, the relapse rate is high and it is associated with serious side-effects.
- Recently a new drug was developed, **miltefosine**. This is a membrane signaling pathway inhibitor. This can be taken orally and is very effective against visceral leishmaniasis. In clinical trials has a 95% cure rate!
- **Pentamidine** is also used for New World cutaneous leishmaniasis, but it usually heals spontaneously.
- **There is no treatment for muco-cutaneous leishmaniasis.**



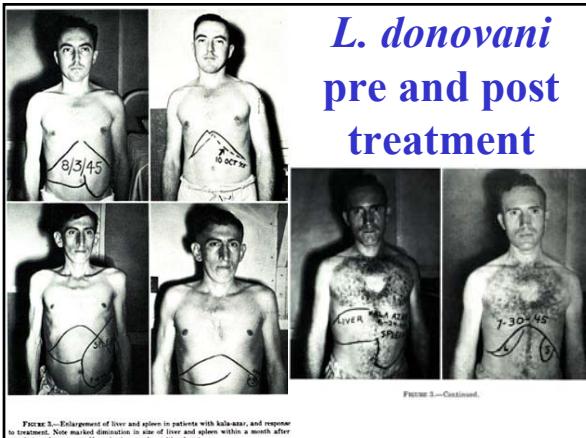
*L. donovani*  
pre and post  
treatment

FIGURE 3.—Continued.

FIGURE 3.—Enlargement of liver and spleen in patients with kala-azar, and response to treatment. Note marked diminution in size of liver and spleen within a month after completion of treatment. Note also improved nutritional state.

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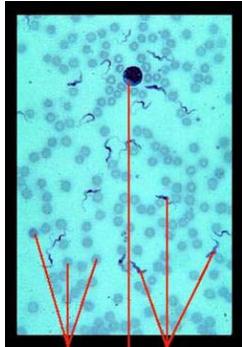


## Trypanosomes of Importance to Humans

### b. *Trypanosoma*

1. More recent (DNA based) classification systems have suggested that there may be several genera here, but current classification system is familiar and accepted.

## Trypanosomiasis



2. Usually known as "sleeping sickness" or other chronic malaises.
3. Tends to be intercellular - a blood parasite.

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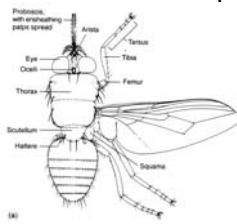
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## Trypanosomiasis

- c. Both are vectored by insects
- a. Probably is the original source of the parasite.



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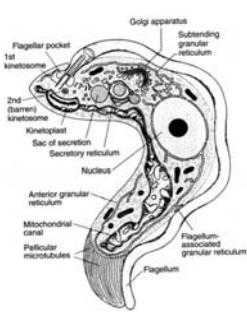
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## Trypanosomiasis

- a. Recognizable by **trypomastigote** form in blood of mammalian host.

1. But is capable of great transformation during life cycle.

2. Seems associated with different conditions of hosts bodies.



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## Trypanosome Transmission



Vectored by insects in two ways:

1. **Anterior station** - bite of insect.
2. **Posterior station** - feces of insect.

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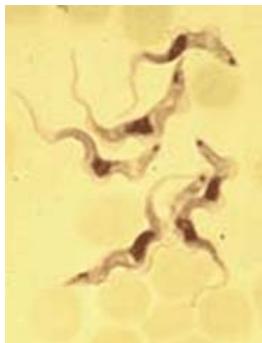
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## Trypanosome Transmission

This has generated two sections of trypanosomes that probably do reflect evolutionary relationships.



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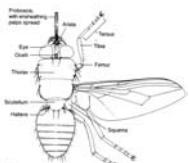
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## Trypanosome Systematics



1. **Section Salivaria** - metacyclic (infective) trypomastigotes in salivary glands.

2. **Section Stercoraria** - metacyclic trypomastigotes in hind gut.

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## Trypanosome Life Cycles

- a. *Trypomastigotes* ingested by insect with blood meal.
- b. Multiplication of *trypomastigotes* in insect midgut.
- c. Multiplication of *epimastigotes* in salivary glands or hind gut.
- 1. transformation to *metacyclic* stage (infective).

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## Trypanosome Life Cycles

- d. Transfer of *metacyclic trypomastigotes* to vertebrate host.
- e. Replication of *slender trypomastigote* in vertebrate.
- f. Transforms to *stumpy trypomastigote*  
- transferred to insect.
- g. These stages seem to be associated with different changes in metabolism of trypanosome - See fig. 5.6 in R&J.

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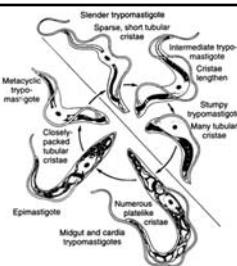
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## The Life Cycle of *T. brucei*

- a. Involves changes in Krebs cycle
  - 1. *Loss of mitochondrial function* in vertebrate host.
  - 2. Evidently occurs because glycolysis alone is possible.

FIGURE 5.6  
Diagram to show changes in form and structure of the mitochondria of *Trypanosomes* during their life cycle. The slender bloodstream forms lack a functional Krebs cycle and cytochrome chain. Stumpy forms have a partially functional Krebs cycle but still lack cytochrome. Trypomastigotes and epimastigotes have a functional Krebs cycle and cytochrome chain. The midgut and cardia forms have a fully functional mitochondrion with active Krebs cycle and cytochrome chain. Cytochrome may be associated with tubular cristae in the midgut and cardia forms. Return to tubular cristae in the salivary gland stages may therefore indicate loss of this electron transfer system.  
From K. Vickerman, "Morphological and physiological considerations of the trypanosomes", in *Advances in Parasitology*, Vol. 16, 1977, pp. 1-100.  
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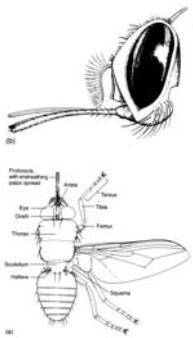
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## Important Species:

### 1. *T. brucei* (variants *gambiense* and *rhodiense*)

- a. Three species of African trypanosome cause different pathologies in cattle and man.

b. Vectored by the tsetse fly (*Glossina*).

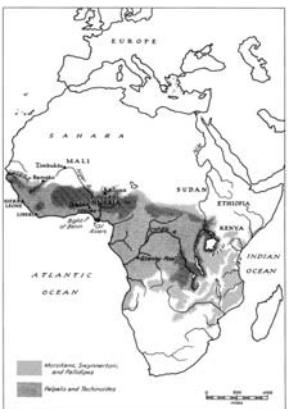


# Important Species

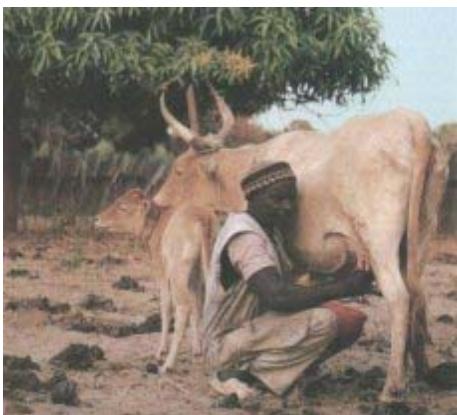
### *T. brucei brucei* -

nagana

1. Resident in native ungulates.
2. Infects introduced animals as well - often fatally.



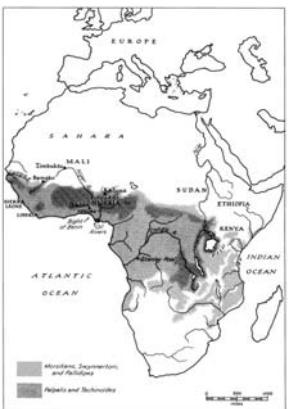
## Distribution of 5 *Glossina* species in Africa



## Important Species

*T. brucei gambiense* -  
West african sleeping sickness.

1. In humans only, no reservoir host known.
2. Causes chronic form of disease.
  - a. Often takes several years to form.



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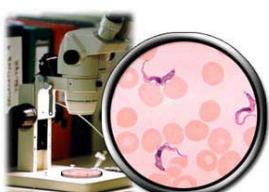
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### *T. brucei gambiense*



Trypanosoma brucei, in blood smear from patient with African sleeping sickness (CDC/Dr. Myron Schultz).

- b. Year one - localized in blood lymph.
- c. Year two - associated with nervous system.

1. Causes coma and death - often insanity too.

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## *T. brucei rhodensiense*



East african sleeping sickness

1. Onset is faster than *T. b. gambiense*.
2. Several reservoir hosts exist.
3. Involves lymph like b.b.g, but death comes sooner.

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## *T. brucei rhodensiense*



- a. Winterbottom's sign - swollen lymph nodes.
  1. Named after the British officer that noticed it.
  2. Was justification for throwing slaves overboard.

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From the movie, 'Amistad' 1997

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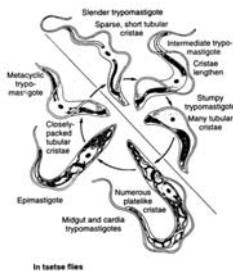
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## Immunological Notes

1. Successive dominance of VATs.

- a. Just as host's immune system becomes successful at overcoming the parasites **variant antigenic type** (VAT).

2. The parasite expresses another one - a new VAT.



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## Production of New VATs

1. Can go on for over 100 types!

- a. A good example of more rapid evolution of parasite than host due to more rapid generation time
- b. Some consider this the context not only for form of vertebrate immunity, but also the evolution of sex.

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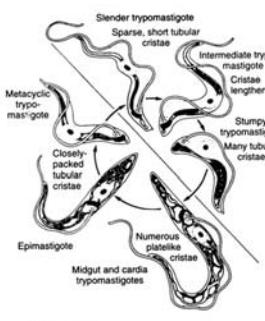
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## Production of New VATs



2. Three mechanisms of occurrence (see book for details).

- a. Result in a single dominant type, but with multiple circulating VATs.

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