Diagnosis of Infectious Agents in the Peripheral Blood Smear

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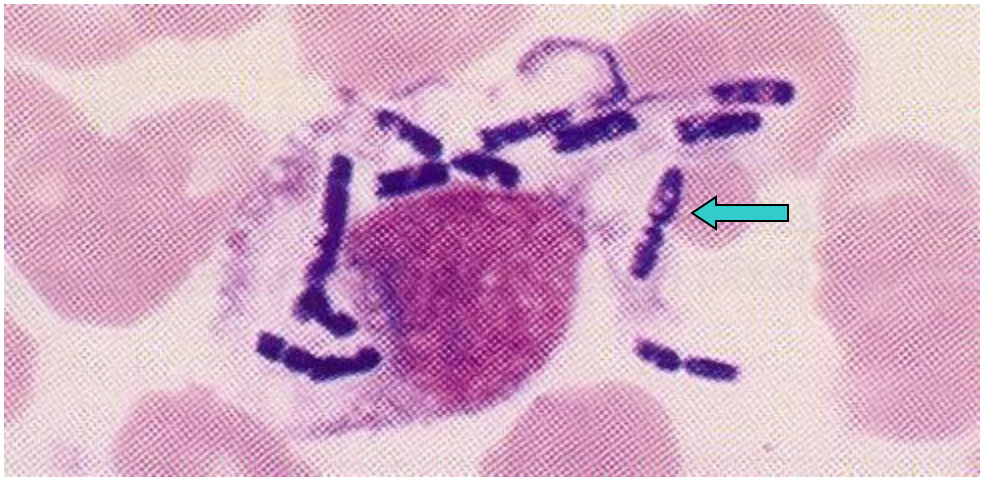
July 24, 2006

Pathogens detected in PS

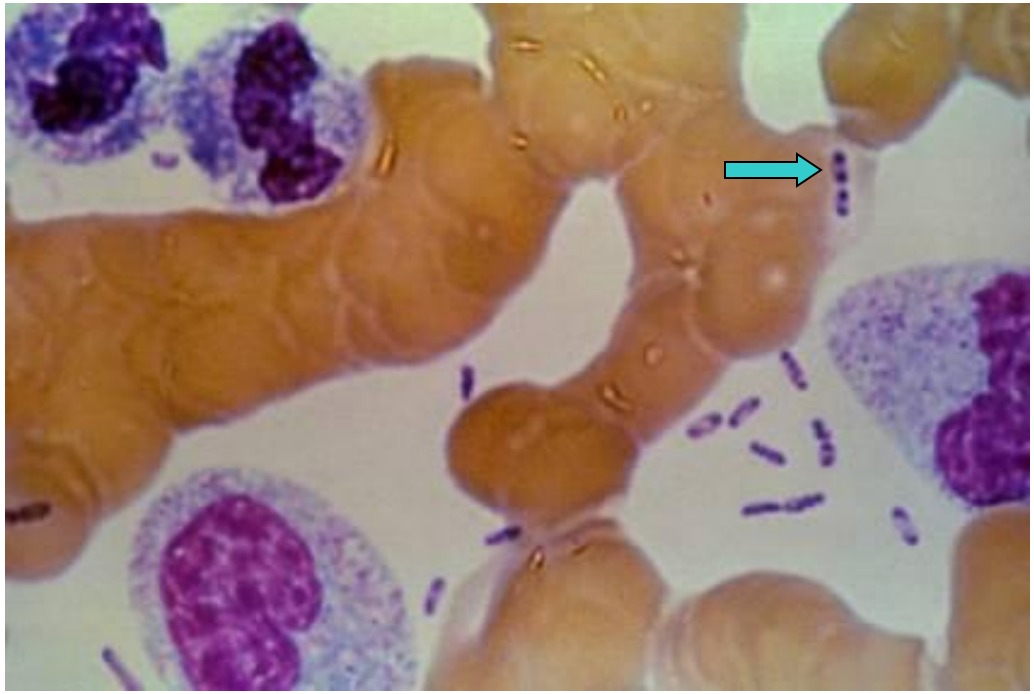
- Bacteria
  - Neisseria meningitidis
  - Streptococcus pneumoniae
  - Staphylococcus sp.
  - Bartonella bacilliformis
  - Yersinia pestis
  - Bacillus anthracis
  - Mycobacteria (buffy coat)
  - Ehrlichia sp.
- Spirochetes
  - Borrelia
- Fungi
  - Candida sp.
  - Histoplasma
  - Cryptococcus
- Protozoa
  - Plasmodium sp.
  - Babesia sp.
  - Trypanosoma sp.
- Filariae
  - Loa loa
  - Wuchereria bancrofti
  - Brugia malayi

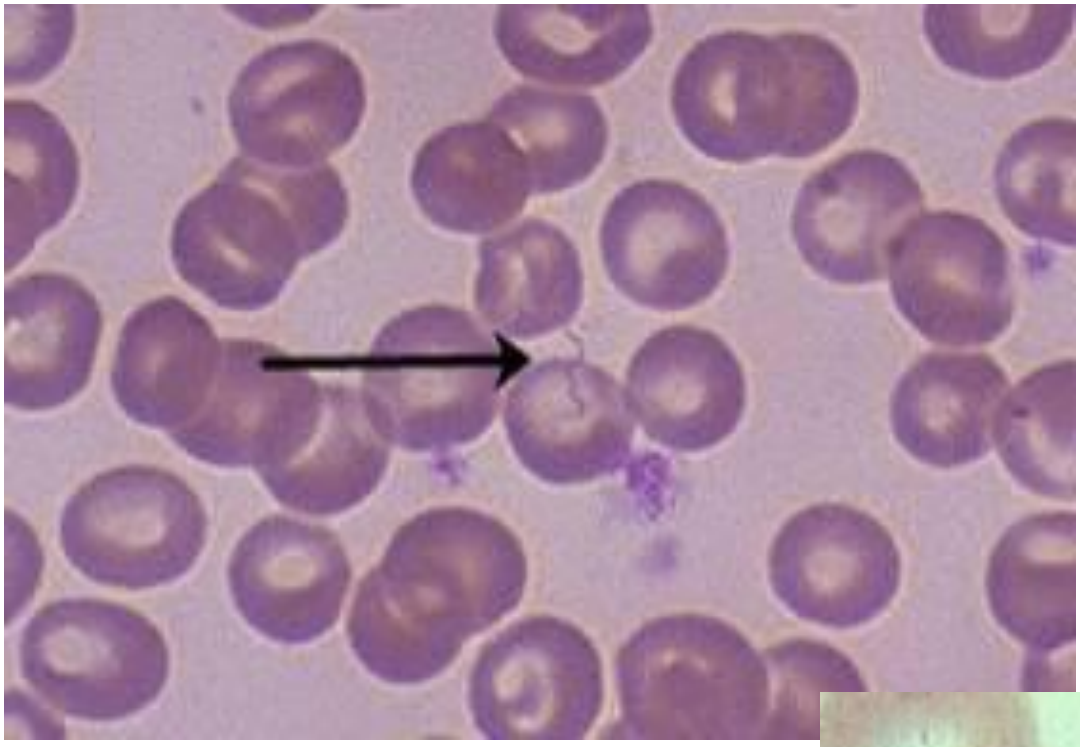
Bacteria



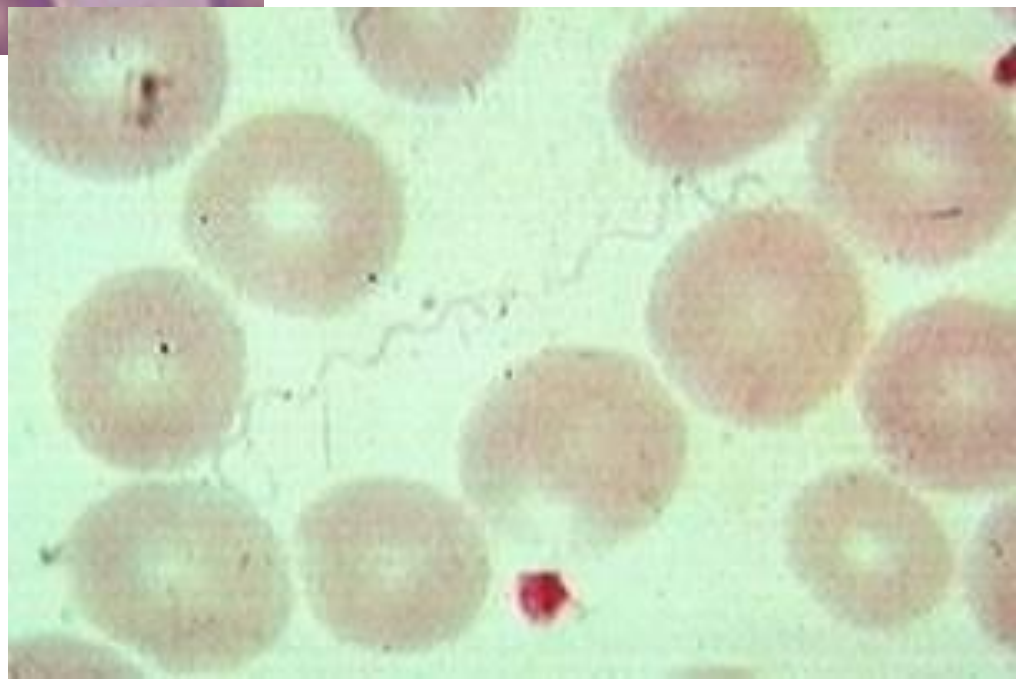
Streptococcus pneumoniae

Yersinia pestis





Borrelia recurrentis



- Epidemic (louse-borne) relapsing fever – Ethiopia, Rwanda
- Endemic (tick-borne) relapsing fever – worldwide & western US
- Antigenic variation cycles & endotoxin

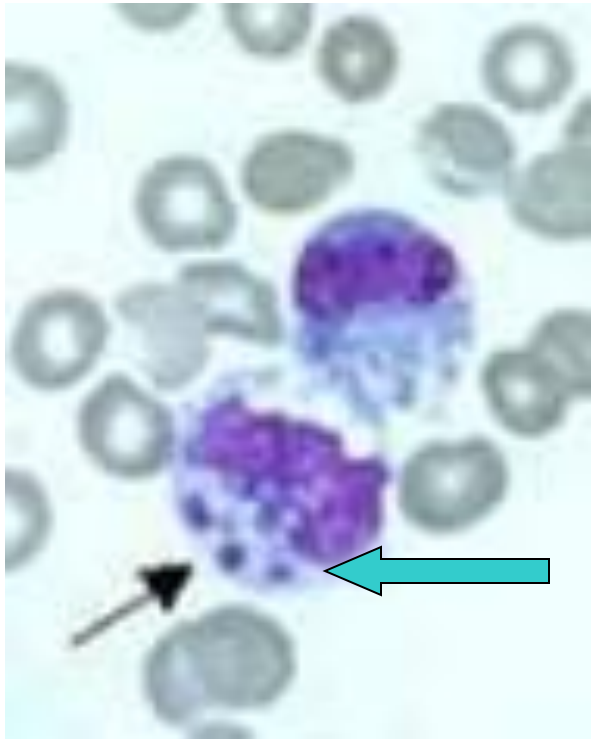
Ehrlichiosis

- *Ehrlichia sennetsu*
  - Sennetsu fever
  - Japan
  - Spread via consumption of raw fish
- **Ehrlichia chaffeensis*
  - Human monocytic ehrlichiosis
  - SE, Mid-Atlantic & South Central US
  - Spread via Lone Star Tick
  - Reservoir = white tailed deer

- *Ehrlichia ewingii & Ehrlichia phagocytophilia*
  - Human granulocytic ehrlichiosis
  - Midwestern & Atlantic US
  - Spread via Ixodes Tick
  - Reservoir = white-footed mouse, chipmunks

Ehrlichiosis Symptoms

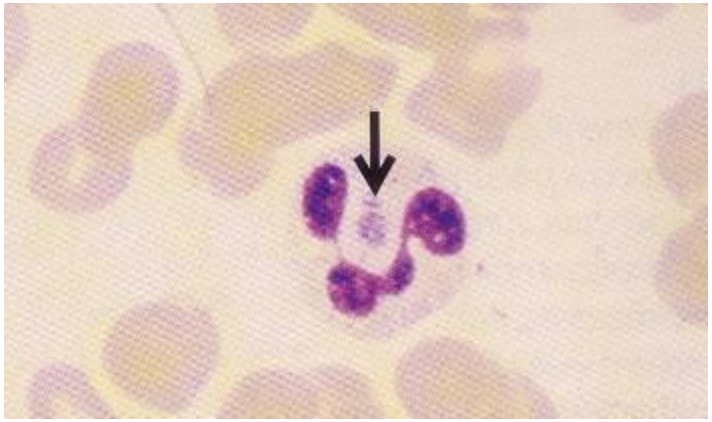
- 1-3 weeks after bite
- High fever, headache, malaise, myalgias
- Leukopenia & Thrombocytopenia
  - Destruction of leukocytes
- 20% develop rash (more often in children than in adults)
- Mortality <5%
  - Death in elderly, immunocompromised (AIDS)
- Most common April-October



Ehrlichia sp.

Morulae

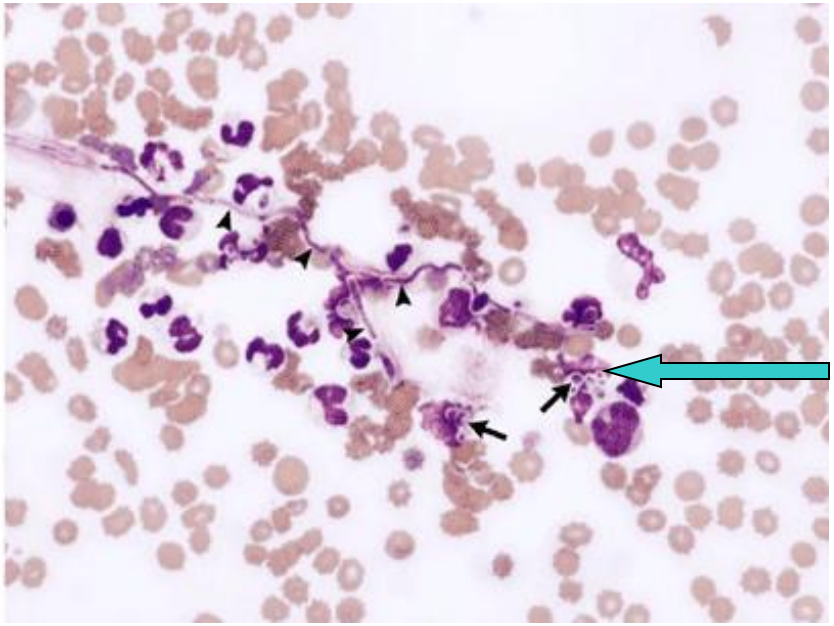
- <10% of HME
- 20-80% of HGE
- Better visualized with Giemsa stain



Diagnosis:

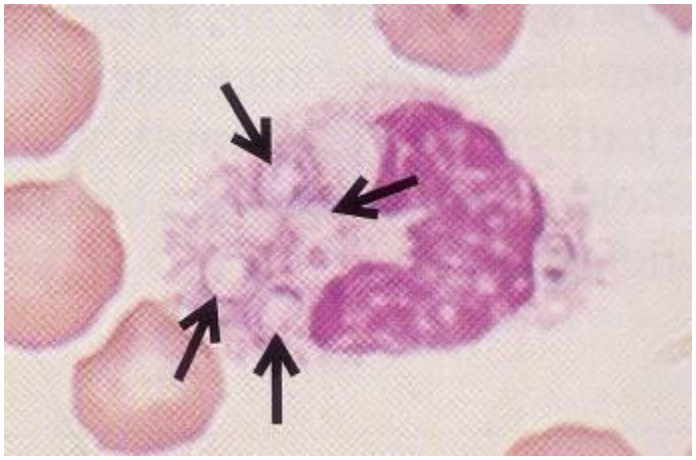
- Serology
- DNA probes

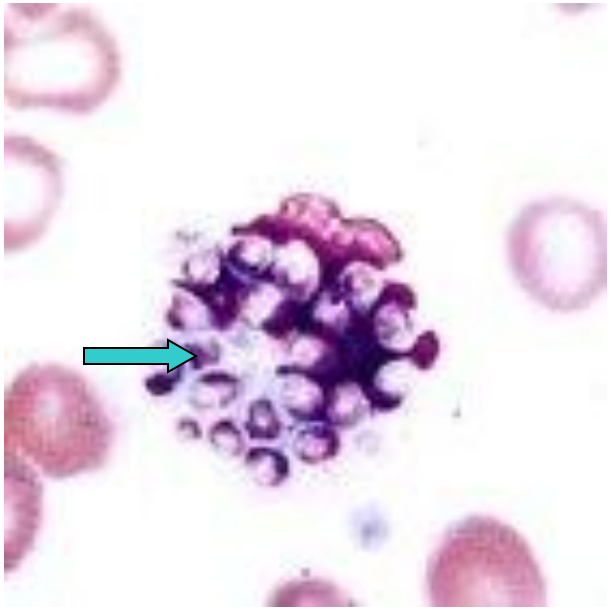
Fungi



Candida albicans

Histoplasma capsulatum



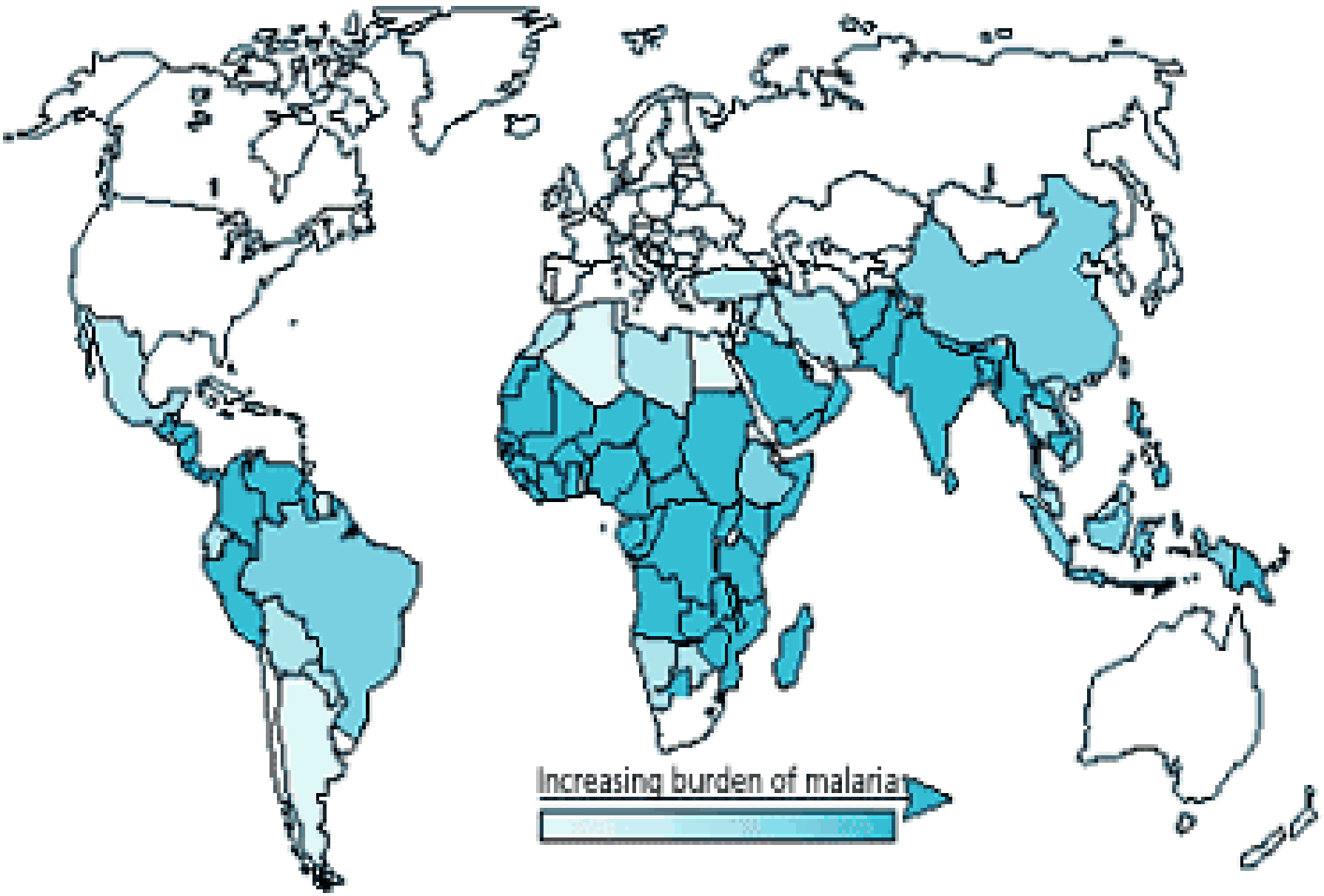


Protozoa

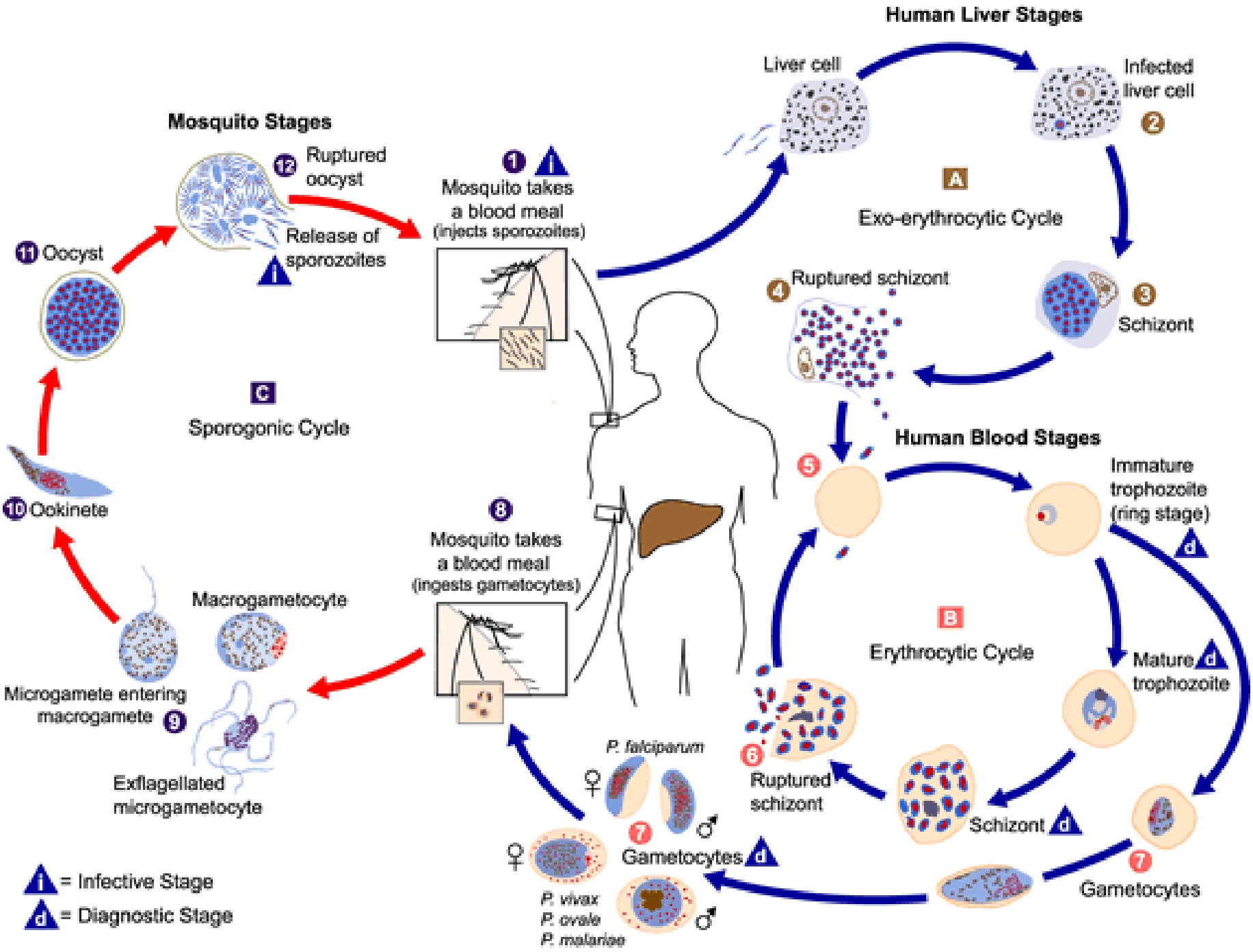
Malaria

- 4 species of *Plasmodium*
- 300-500 million cases/year
- 1-2 million deaths/year
- Spread via female anopheles mosquitoes, blood transfusion, IVDA
- Cyclic fevers, headache & malaise
- Thick & thin blood smears prepared
  - Collection – midway between fever cycles

Estimate of World Malaria Burden



Source: RBM data/J.Sachs 1999


Human Liver Stages
Liver cell
Infected liver cell
2
A
Exo-erythrocytic Cycle
3
Schizont
4
Ruptured schizont
Human Blood Stages
5
Immature trophozoite (ring stage)
d
Mature trophozoite
d
B
Erythrocytic Cycle
7
Gametocytes
Schizont
d
6
Ruptured schizont
P. falciparum
♀
♂
7
Gametocytes
d
P. vivax
P. ovale
P. malariae
♀
♂
1
i
Mosquito takes a blood meal (injects sporozoites)
Mosquito Stages
12
Ruptured oocyst
Release of sporozoites
i
11
Oocyst
C
Sporogonic Cycle
10
Ookinete
8
Mosquito takes a blood meal (ingests gametocytes)
Macrogametocyte
Microgamete entering macrogamete
9
Exflagellated microgametocyte
i = Infective Stage
d = Diagnostic Stage

Plasmodium falciparum

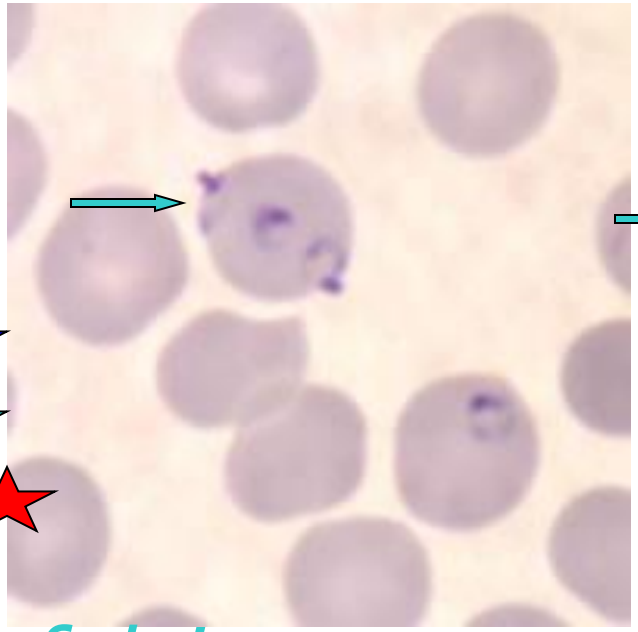
- Malignant tertian malaria
- Asia & Africa
- Most deadly and severe infections
- Shortest incubation period = 7-10 days
- Infects all ages of RBCs → higher parasitemia
- Mature trophozoites and schizonts sequestered in microvascular system → tissue ischemia
  - Rarely seen in peripheral smear
- GI symptoms
- Black water fever
  - Intravascular hemolysis → kidney damage
  - Capillary plugging due to RBC debris
- Can involve CNS
- Widespread drug resistance

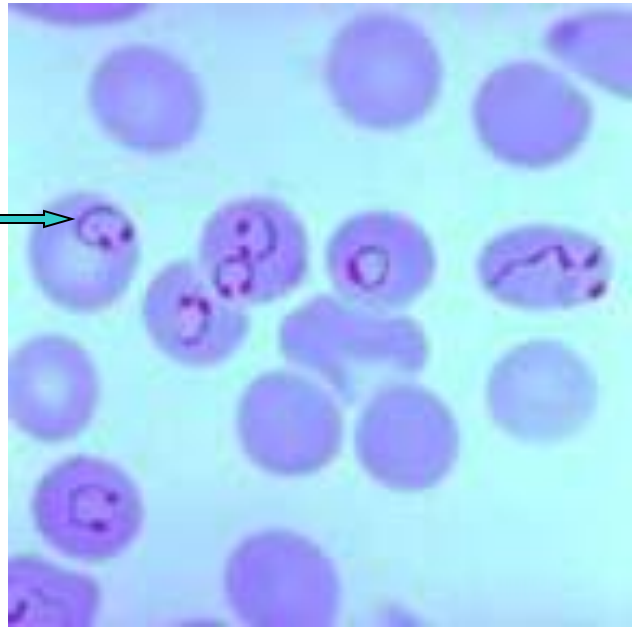
Trophozoites (rings)

Multiple rings/cell

Appliqué forms

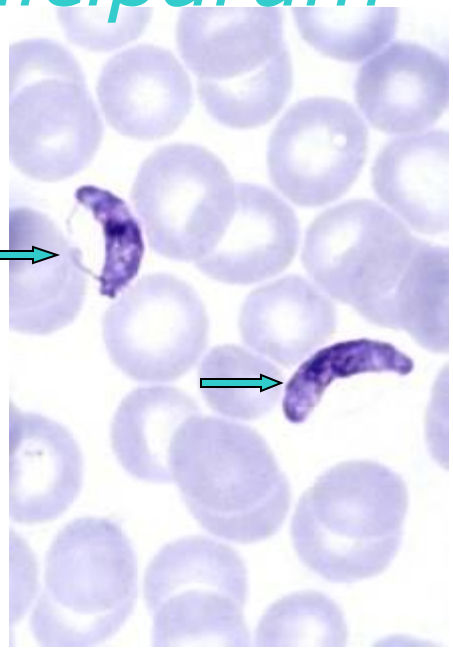
1-2 chromatin dots

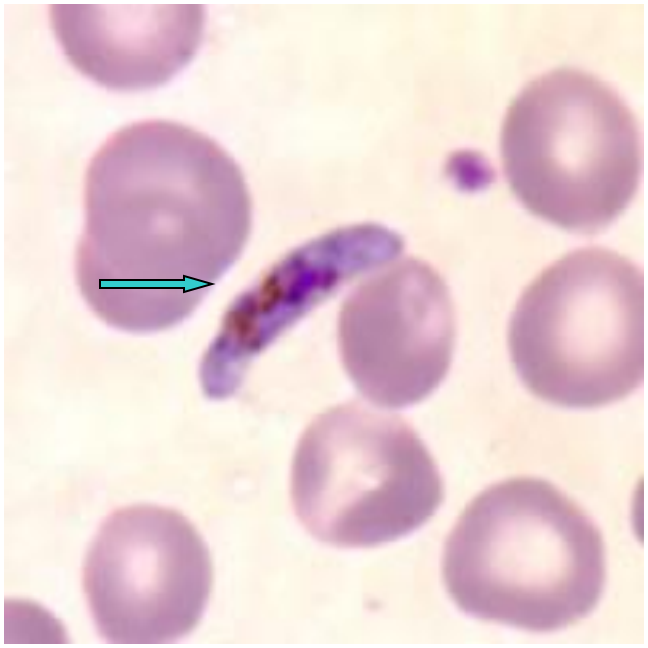




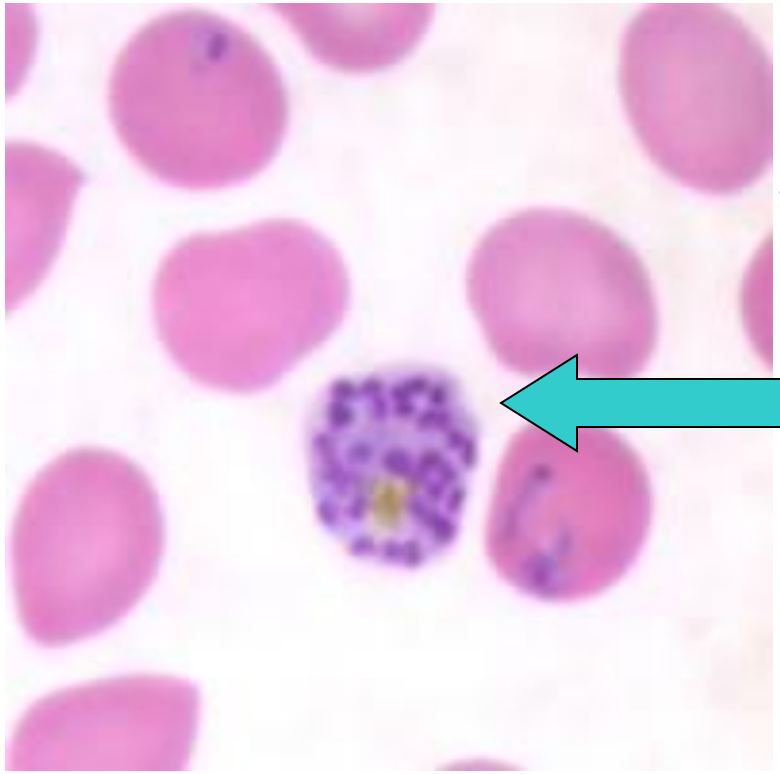
Plasmodium falciparum

Gametocytes



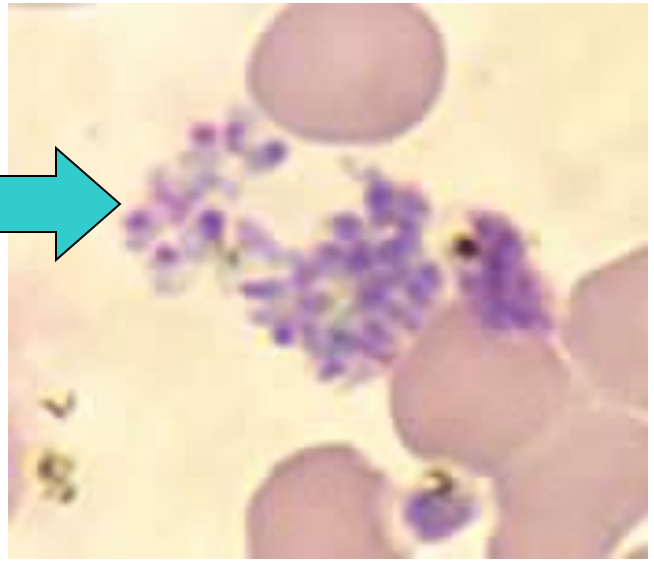


Plasmodium falciparum



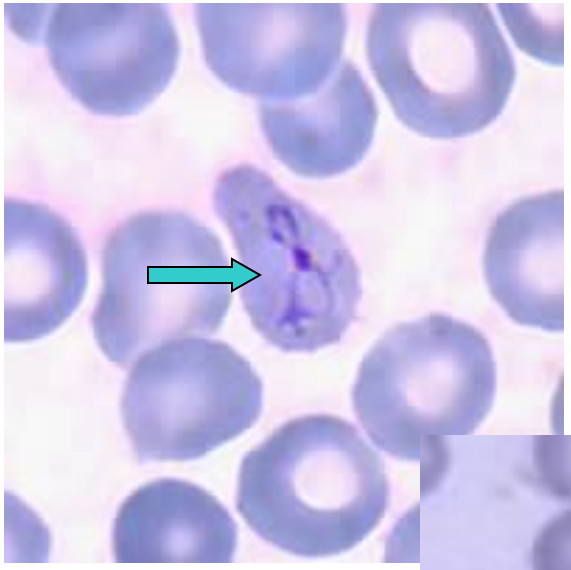
Schizont

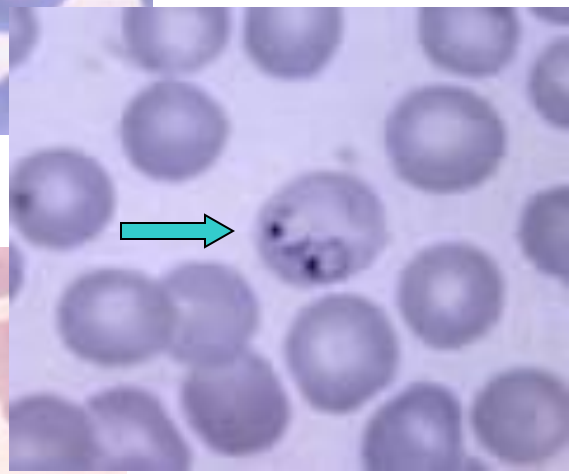
Ruptured Schizont

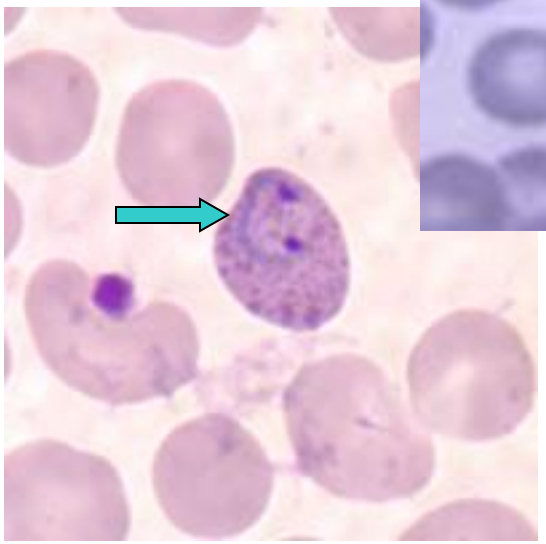


Plasmodium vivax

- Benign tertian malaria
- Latin America, India, Pakistan
- Only infects reticulocytes
- Produces HYPNOZOITES
- Relapses up to 5 years after infection
- Uses Duffy (Fy) antigen as receptor
- Treatment = Primaquine (for hypnozoite)







Plasmodium vivax

Early Trophozoites (rings)

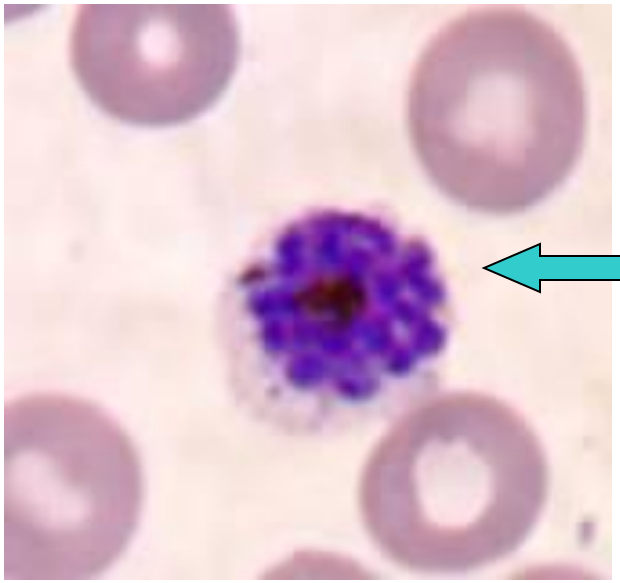
Ameoboid rings

Enlarged RBCs

Schuffner's Dots

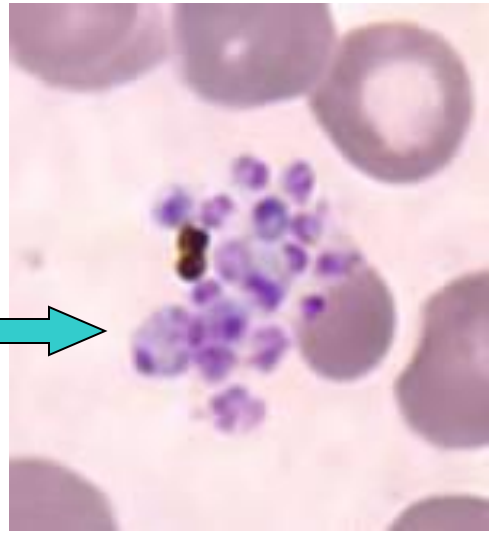
Mature trophozoite

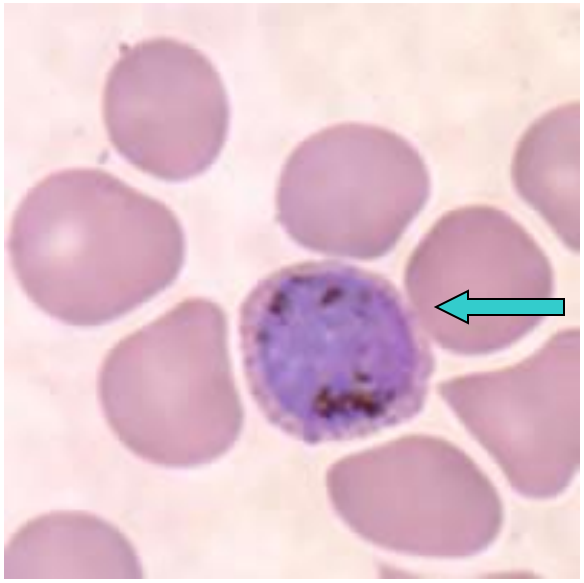
Plasmodium vivax



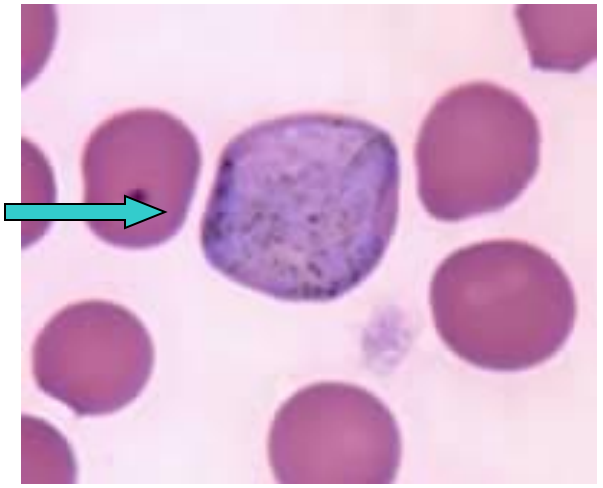
Schizont

Ruptured Schizont





Gametocytes



Plasmodium ovale

- Ovale/benign tertian malaria
- Africa, Asia, South America
- Only infects reticulocytes
- Produces HYPNOZOITES
- Relapses up to 5 years after infection
- Treatment = Primaquine (for hypnozoite)

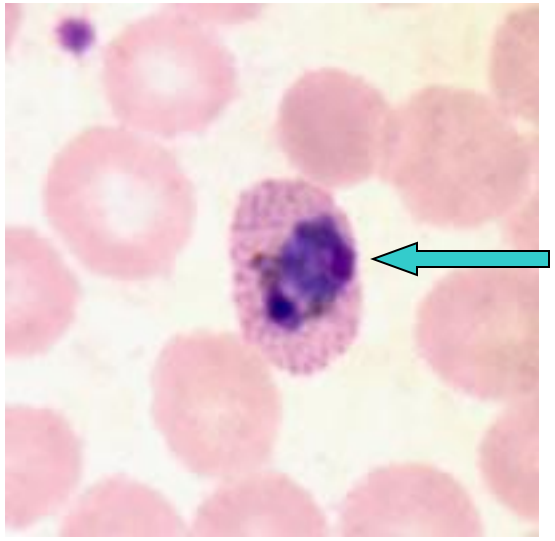
Plasmodium ovale

Early trophozoite (ring)

Enlarged RBCs

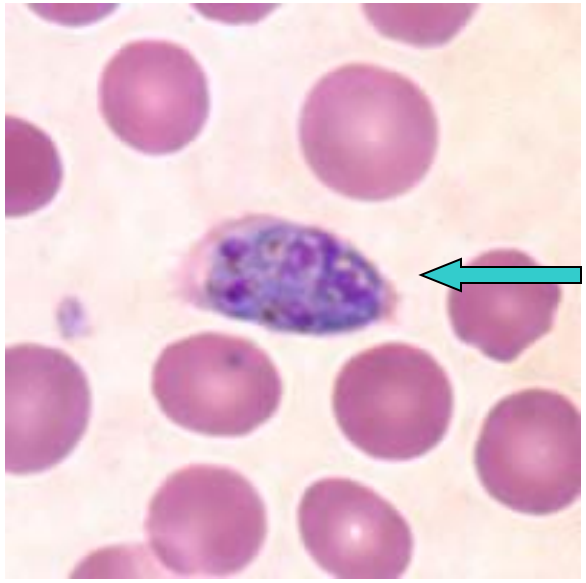
Fimbriated/Oval RBCs

Schuffner's Dots

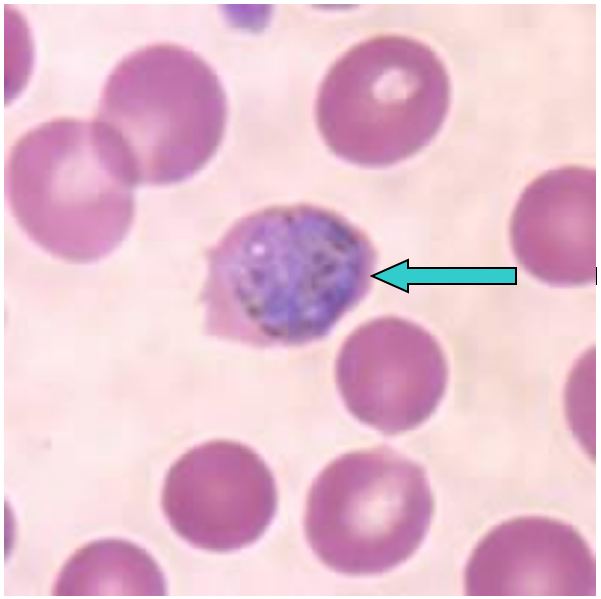


Mature trophozoite

Plasmodium ovale



Schizont

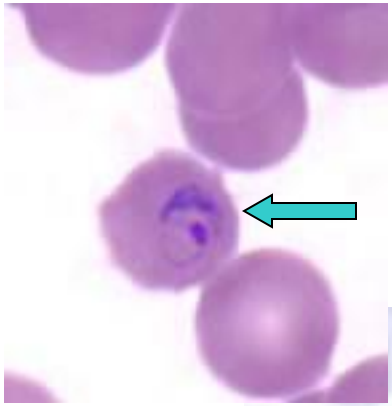


Gametocytes

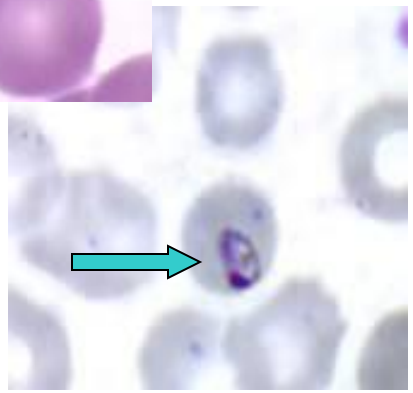
Plasmodium malariae

- Quartan malaria
- Asia & Africa
- Only infects mature RBCs
- Low-grade parasitemia that can persist >40 years
- Longest incubation period = 18-40 days, even years!

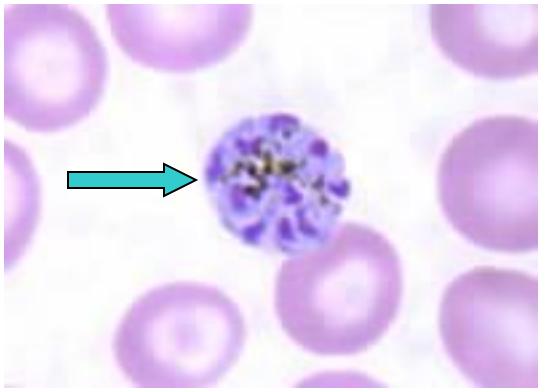
Plasmodium malariae



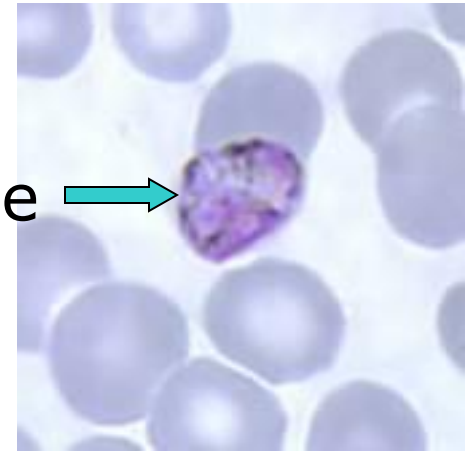
Early trophozoites
(rings)

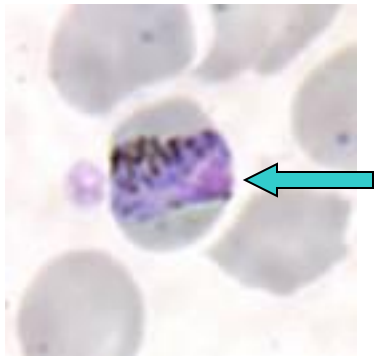


Schizont



Gametocyte

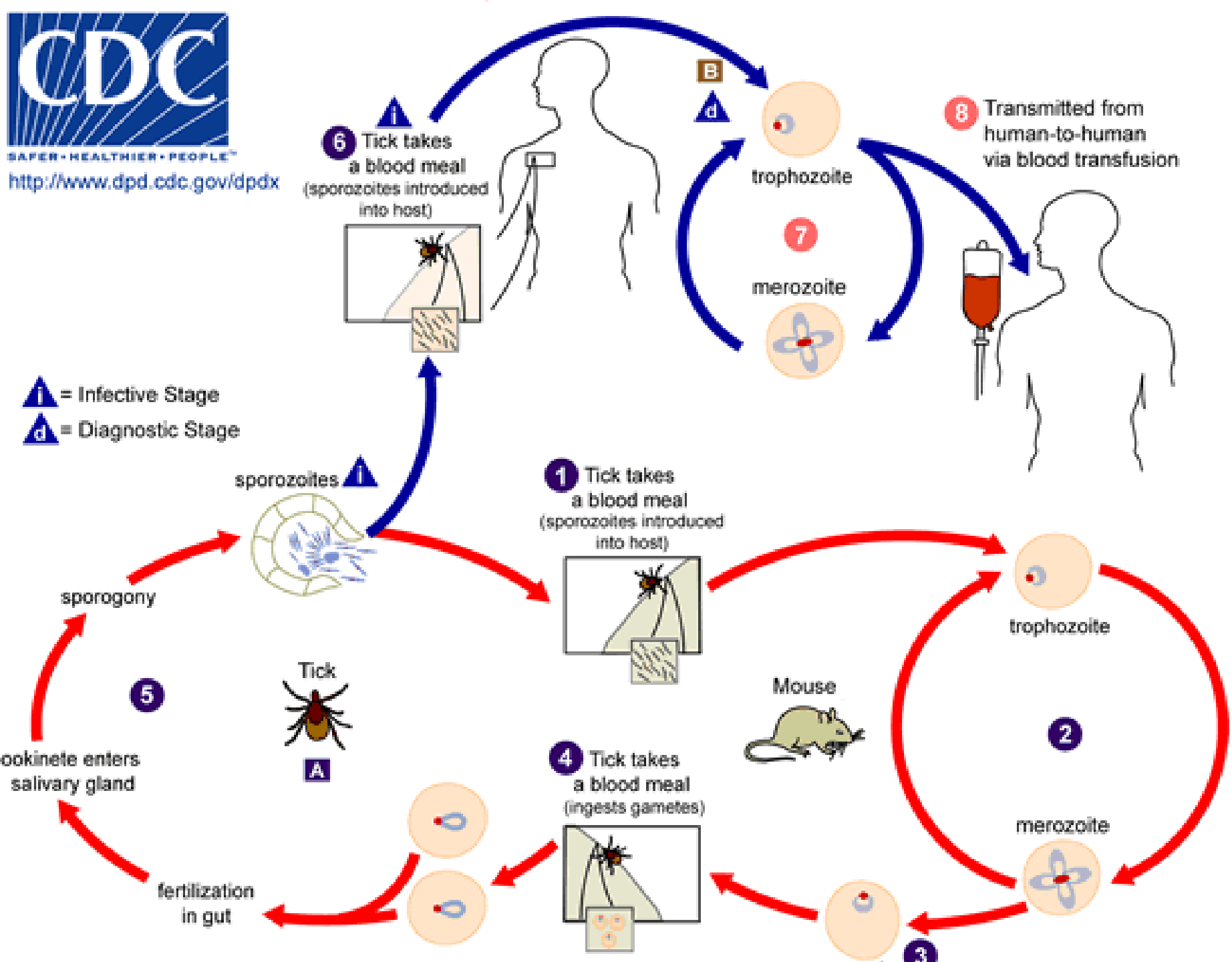




Mature trophozoite
(band form)

Babesiosis

- *Babesia microti*
- Reservoir = deer, cattle, rodents
- Humans = accidental hosts
- Spread via Ixodes tick and transfusion
- Symptoms
  - Most infections asymptomatic or mild
  - 1-4 weeks after bite → headache, fever
  - Later may → hemolytic anemia, hepatomegaly, renal failure


CDC
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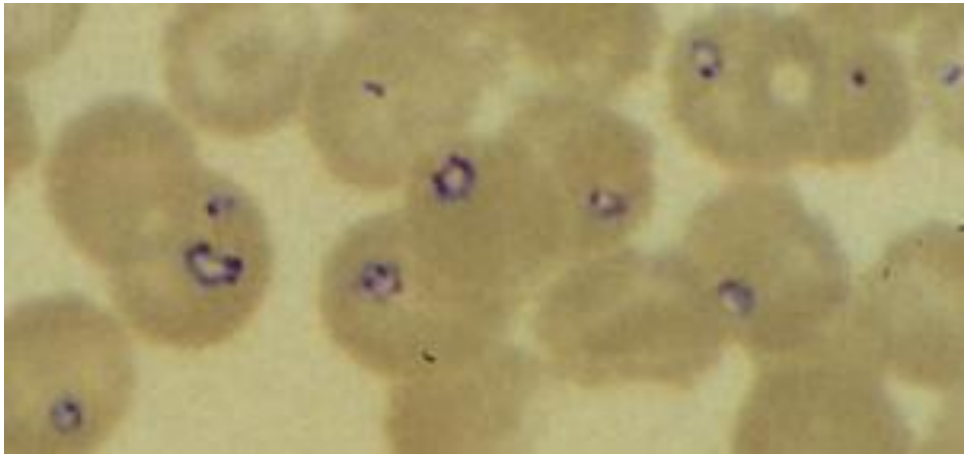
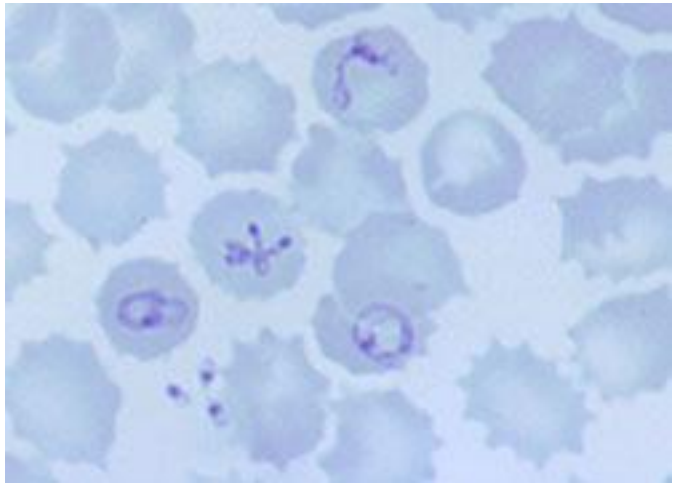
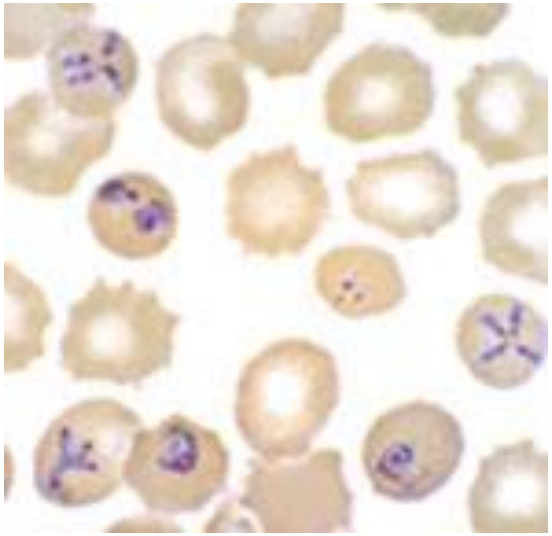
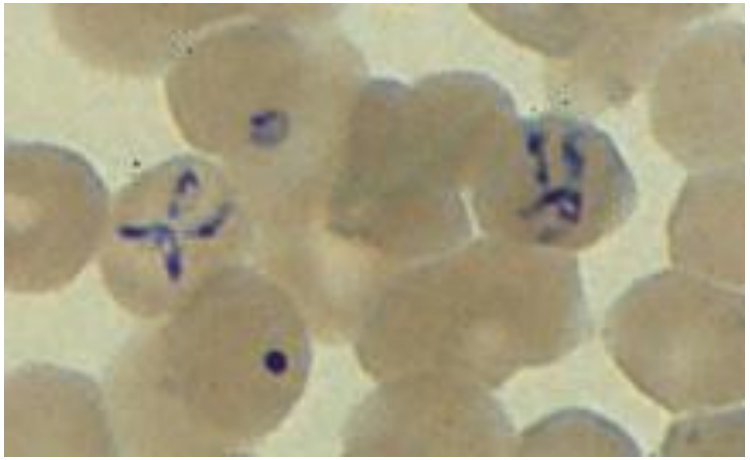
http://www.dpd.cdc.gov/dpdx
i = Infective Stage
d = Diagnostic Stage
6 Tick takes a blood meal (sporozoites introduced into host)
i
B
d
trophozoite
7
merozoite
8 Transmitted from human-to-human via blood transfusion
sporozoites
i
1 Tick takes a blood meal (sporozoites introduced into host)
trophozoite
sporogony
Tick
5
A
Mouse
2
ookinete enters salivary gland
4 Tick takes a blood meal (ingests gametes)
merozoite
fertilization in gut
3





Babesia microti



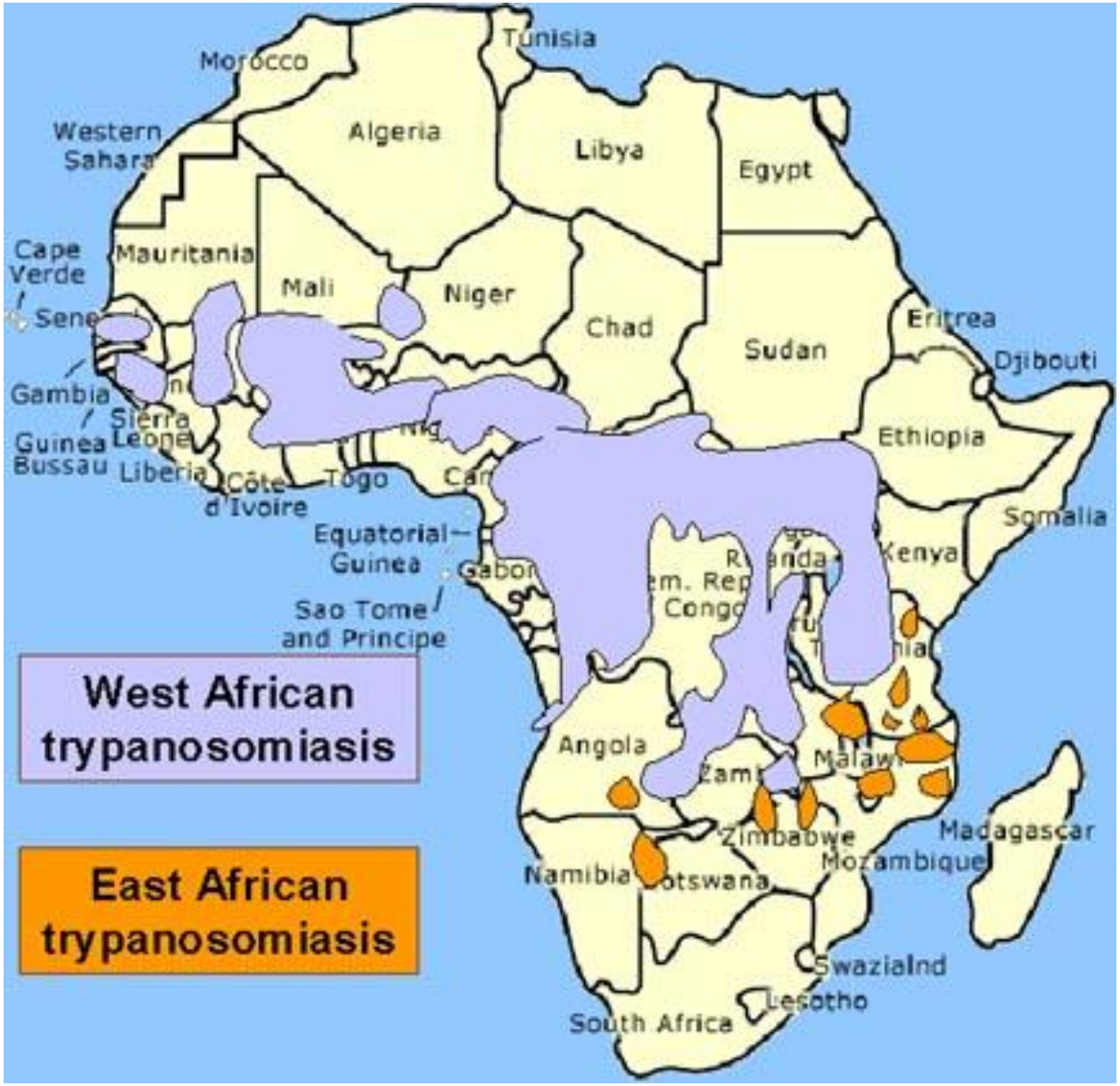


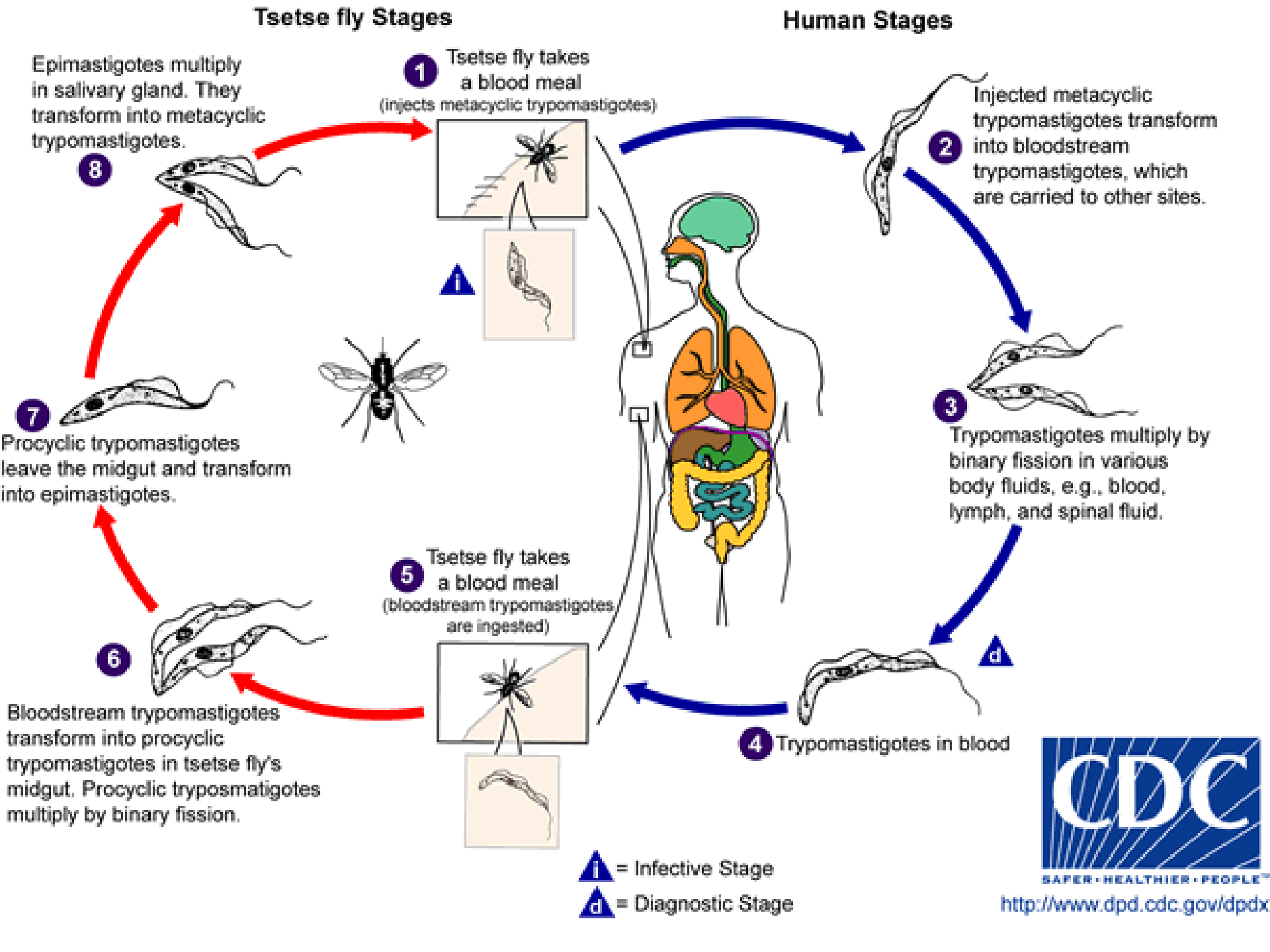
Trypanosomiasis

- African trypanosomiasis
  - Sleeping Sickness
- American trypanosomiasis
  - Chagas' Disease

African Sleeping Sickness

- *Trypanosoma brucei gambiense*
  - West African Sleeping Sickness
  - Reservoir = humans
- *Trypanosoma brucei rhodesiense*
  - East African Sleeping Sickness
  - Progresses more rapidly with more severe symptoms
  - Reservoir = wild game
- 6000-10000 human cases annually
- Spread via Tsetse fly & blood transfusion


Tunisia
Morocco
Western Sahara
Algeria
Libya
Egypt
Cape Verde
Mauritania
Mali
Niger
Chad
Sudan
Eritrea
Djibouti
Gambia
Sierra Leone
Guinea Bussau
Liberia
Côte d'Ivoire
Togo
Ethiopia
Somalia
Equatorial Guinea
Gabon
Kenya
Sao Tome and Principe
Congo
Angola
Malawi
Zimbabwe
Madagascar
Mozambique
Namibia
Botswana
Swaziland
Lesotho
South Africa
West African trypanosomiasis
East African trypanosomiasis


Tsetse fly Stages
Human Stages
Epimastigotes multiply in salivary gland. They transform into metacyclic trypomastigotes.
8
1 Tsetse fly takes a blood meal
(injects metacyclic trypomastigotes)
i
2 Injected metacyclic trypomastigotes transform into bloodstream trypomastigotes, which are carried to other sites.
3 Trypomastigotes multiply by binary fission in various body fluids, e.g., blood, lymph, and spinal fluid.
d
4 Trypomastigotes in blood
5 Tsetse fly takes a blood meal
(bloodstream trypomastigotes are ingested)
6 Bloodstream trypomastigotes transform into procyclic trypomastigotes in tsetse fly's midgut. Procyclic tryposmatigotes multiply by binary fission.
7 Procyclic trypomastigotes leave the midgut and transform into epimastigotes.
i = Infective Stage
d = Diagnostic Stage
CDC
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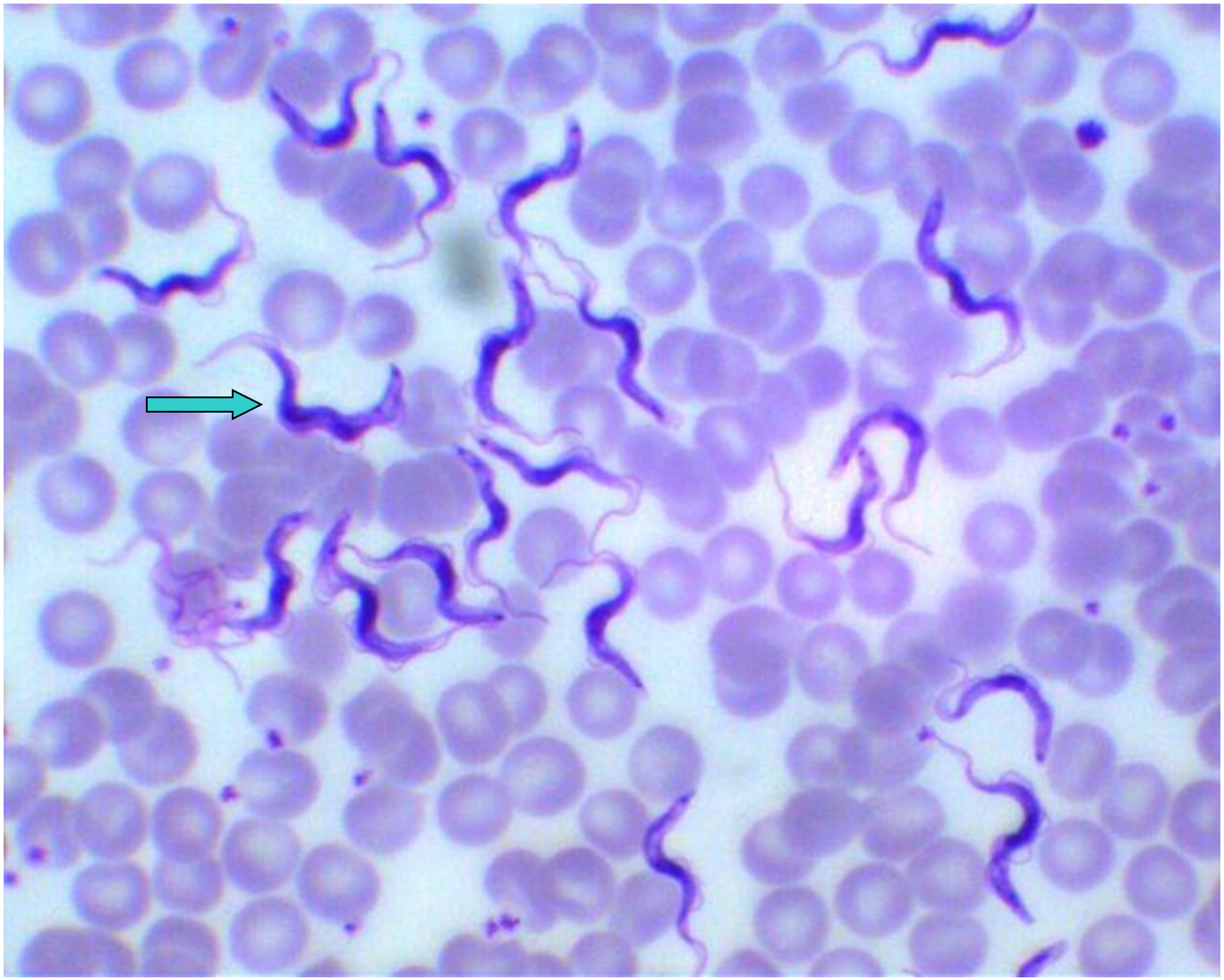
Symptoms

- Chancre (1-3 weeks) → parasitemia (2-3 weeks)
  - Fever, malaise, insomnia, headache
  - Winterbottom's sign = enlargement of posterior cervical glands
- CNS
  - Changes in character, tremors, sleepiness

Trypanosoma brucei

Trypomastigotes



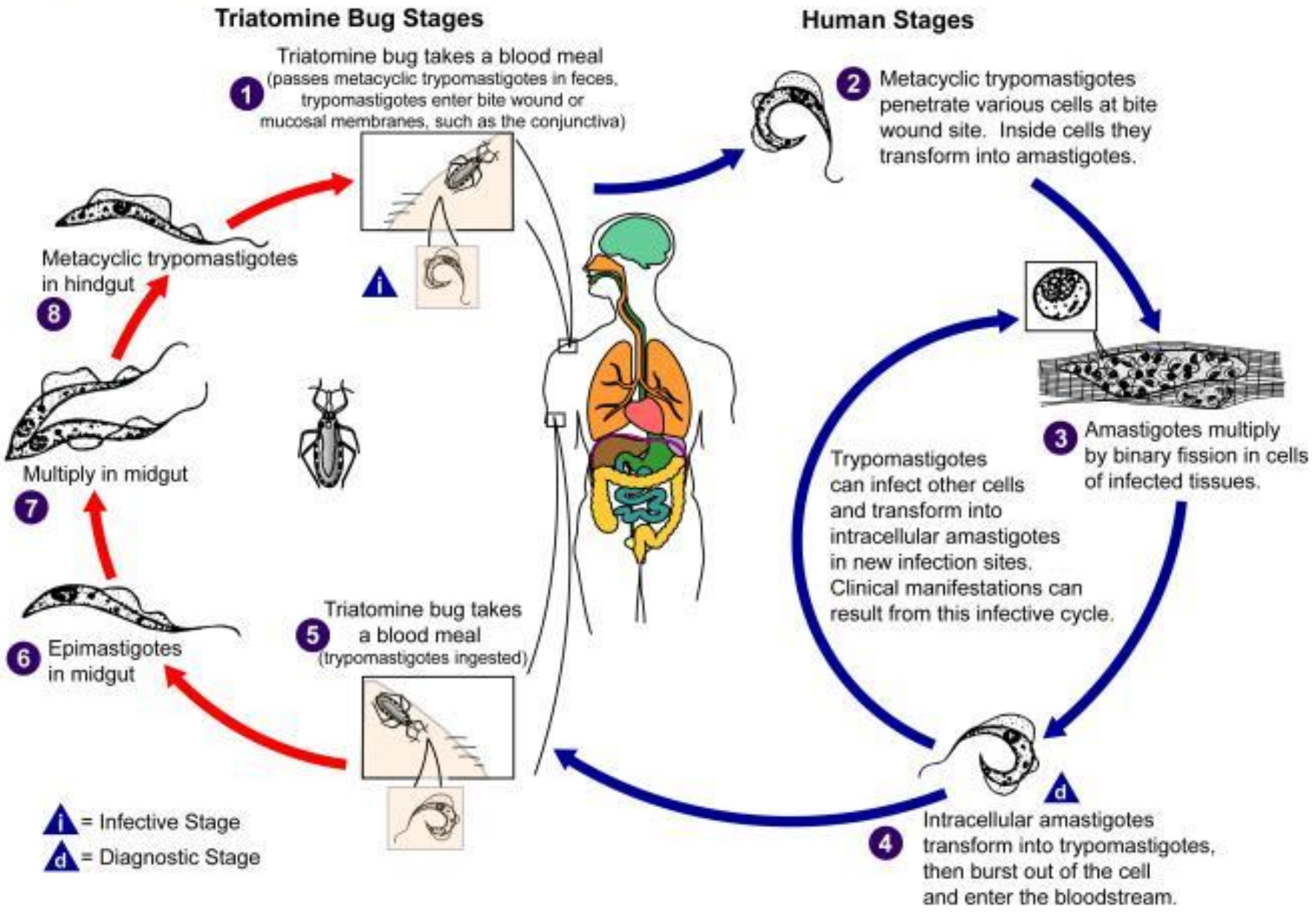
Chagas' Disease

- *Trypanosoma cruzi*
  - Central and South America
  - Rare cases in Texas, California & Maryland
  - Estimated 16-18 million people infected
  - 50,000 die annually from disease
  - Spread via Riduvid (kissing) bug
  - Also spread via blood transfusion and transplacentally
  - Many wild & domestic animals serve as reservoir


Trypanosoma cruzi
Chagas' disease
American trypanosomiasis
© CD-ROM ILLUSTRATED LECTURE NOTES ON TROPICAL MEDICINE

Trypanosomiasis, American (Chagas disease)

(Trypanosoma cruzi)

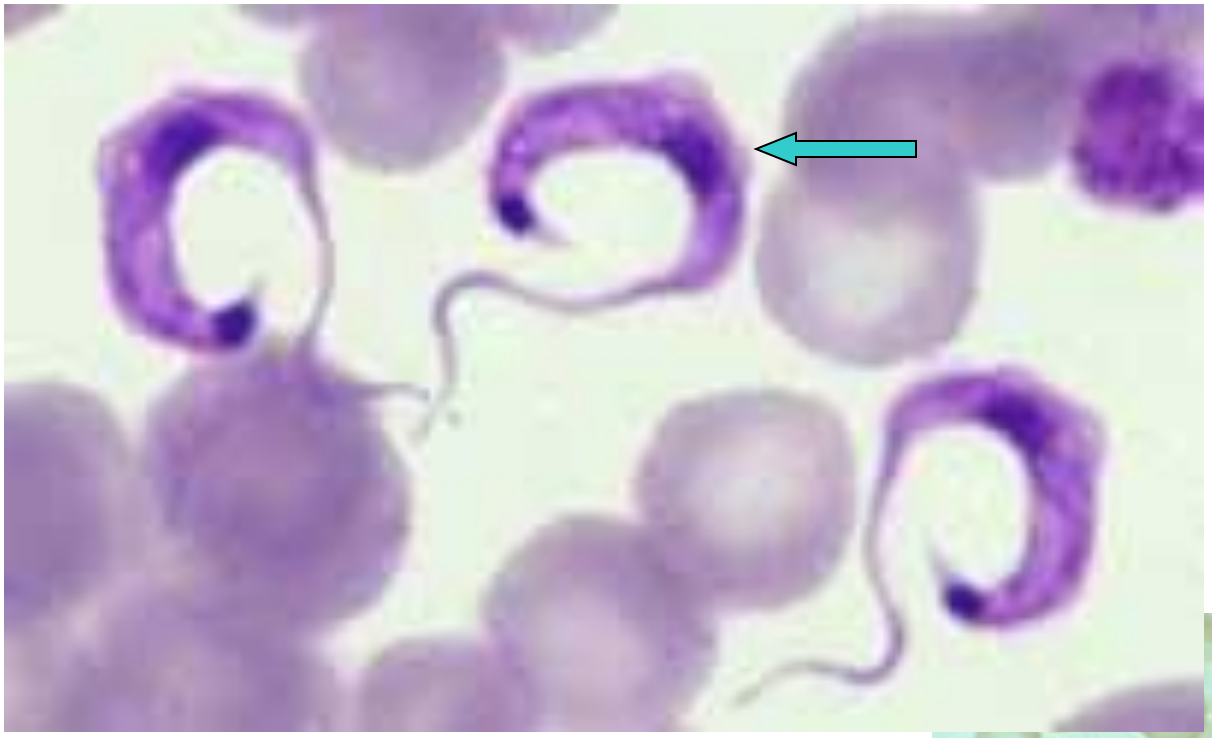


Symptoms

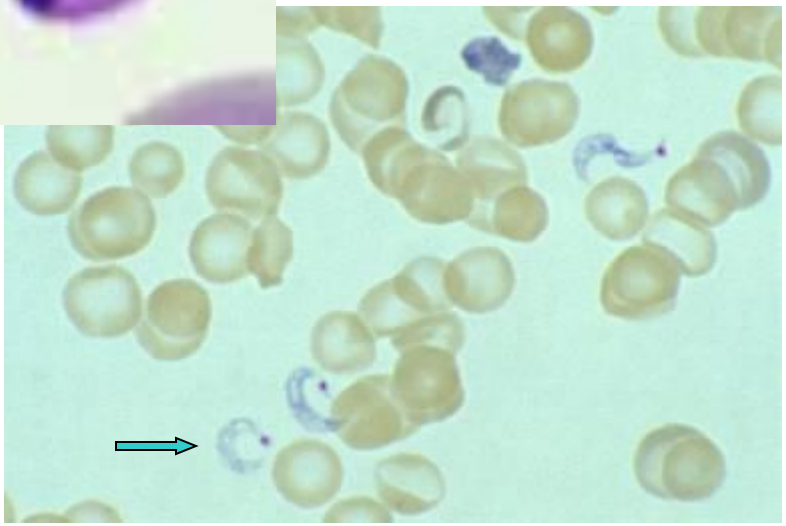
- Chagoma
  - Ramana's sign = unilateral conjunctivitis & orbital edema
- Acute Stage
  - 7-14 days after infection
  - Usually not recognized and resolves
  - Malaise, fever, hepatomegaly, rash
  - Acute myocarditis
  - Meningoencelphalitis in children

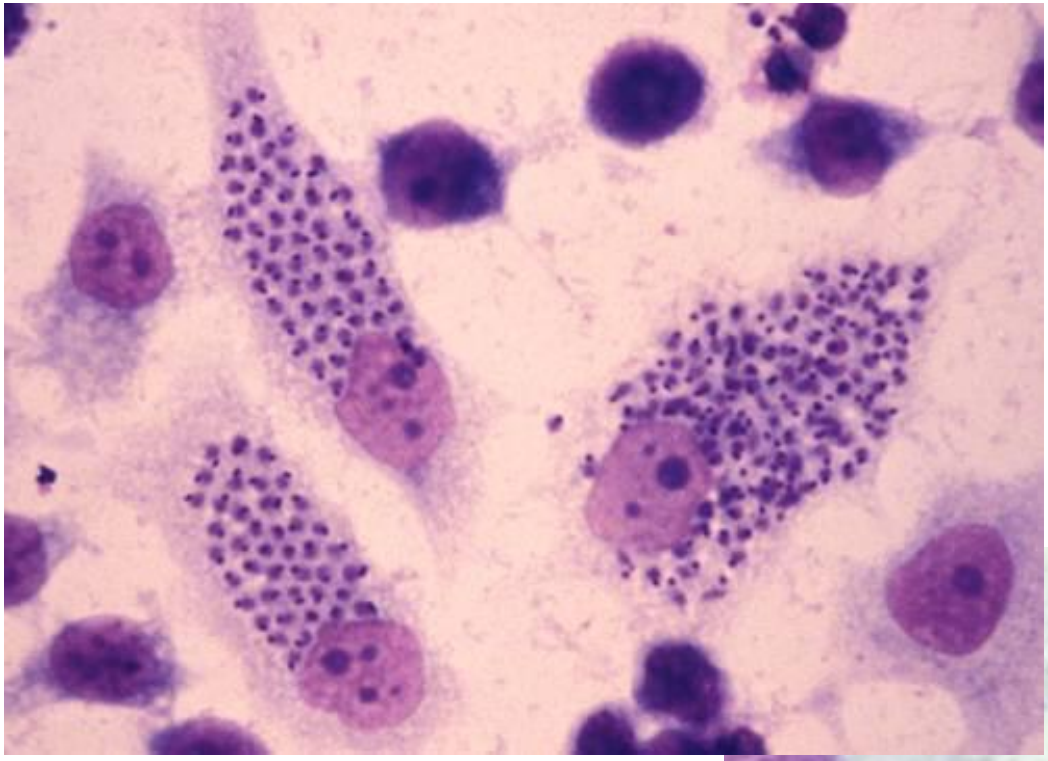
Chronic Stage
- Approximately 10-20% of infected
- Relapsing & remitting symptoms
- Myocardial insufficiency, cardiomegaly, arrhythmias
- Megaesophagus & megacolon

Trypanosoma cruzi

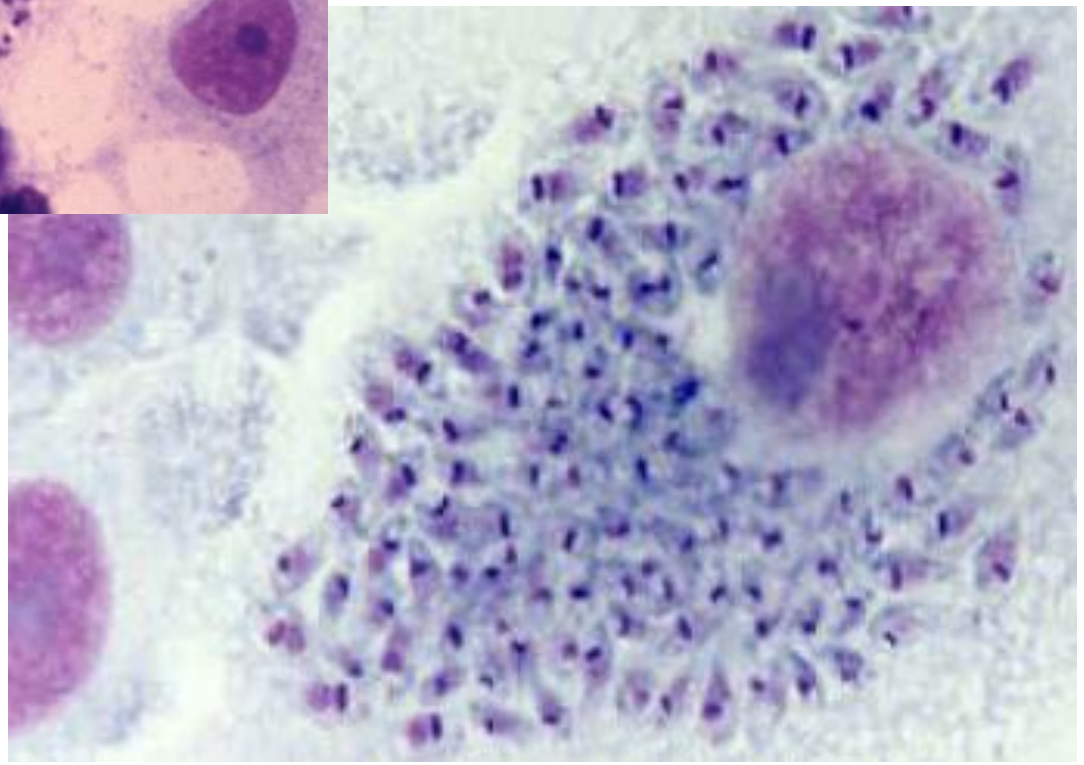


Trypomastigotes





Trypanosoma cruzi



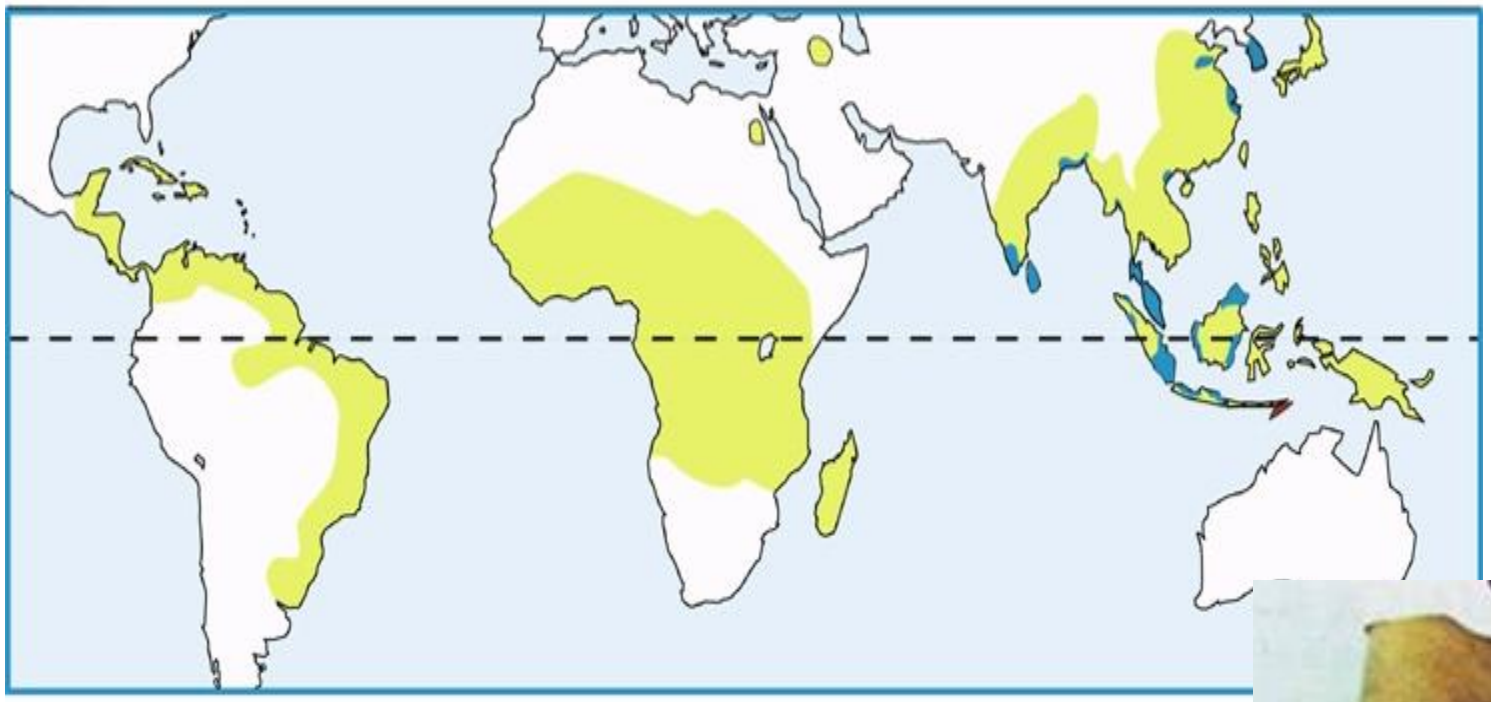
Amastigotes in tissue

Filariae

Elephantitis / Filariasis

- *Wuchereria bancrofti & Brugi malayi*
- Spread via mosquitos
- Can persist in humans up to 10 years
- May be asymptomatic
- Acute - Fever, lymphangitis
- Chronic – Filarial elephantitis
- Higher numbers of organisms in blood at night

Wucheria bancrofti, Brugia malayi

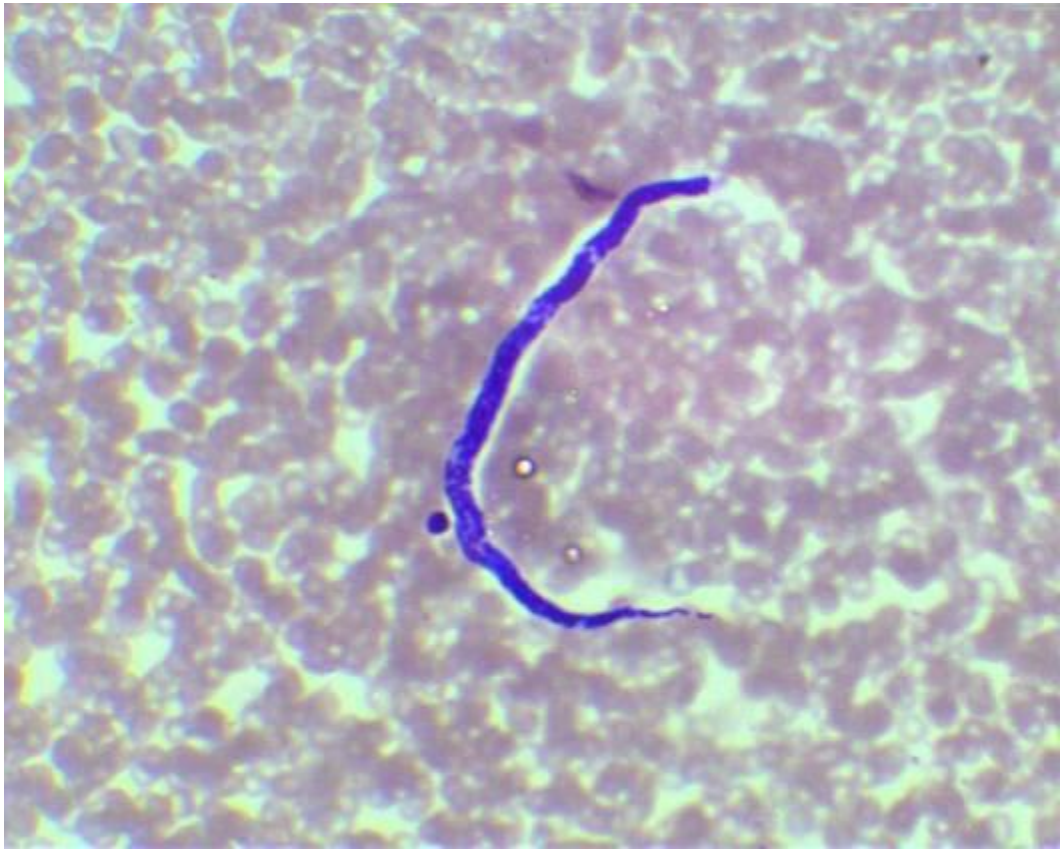


Brugia malayi

Wuchereria bancrofti

Loa Loa

- Endemic in West Africa, Nigeria
- Spread via mango fly
- Can persist up to 17 years
- Reservoir = monkey
- Symptoms 1 year after infection
  - Calabar swellings
  - Worm migration in conjunctiva
- Higher number of organisms in blood at daytime



Loa loa