Overview and Introduction to Malaria + Babesia

- Incidence
- Prevalence
- Transmission
- General lifecycle
- Treatment & Prevention

Definitions

- **accolé.** On the outer edge.
- ***Apicomplexa.*** A phylum containing protozoa whose life cycle includes feeding stages (trophozoites), asexual multiplication (schizogony), and sexual multiplication (gametogony and sporogony).
- **blackwater fever.** A complication of malaria infection in which red blood cells burst in the bloodstream (hemolysis), releasing hemoglobin directly into the blood vessels leading to hematuria, frequently causing kidney failure.
- **bradyzoites.** Slowly multiplying intracellular trophozoites of *Toxoplasma gondii;* intracellular tissue cysts in immune hosts contain bradyzoites that continue dividing within the cyst. Bradyzoites are also found in sarcocysts.
- **cryptozoite.** The stage of *Plasmodium* spp. that develops in liver cells from the inoculated sporozoites. Also called the *exoerythrocytic stage* or *tissue stage.*
- **gametes.** Mature sex cells.
- **gametocyte.** A sex cell that can produce gametes.
- **gametogony.** The phase of the development cycle of the malarial and coccidial parasite in the human in which male and female gametocytes are formed.

Definitions

- **hypnozoites.** Long-surviving modified liver schizonts of *P. vivax* and *P. ovale* that are the source of relapsing infections in these species.
- **Malarial pigment (Hemozoin).** An iron-containing product that accumulates as cytoplasmic granules when malaria parasites break down hemoglobin.
- **Maurer's dots.** Comma-shaped spots that are dark blue by Giemsa staining and found on the surface of red cells infected with *Plasmodium falciparum.* Maurer's dots consist of at least three groups of protein antigens.
- **merogony.** Asexual multiplication in coccidian life cycle. Usually occurs in intestinal epithelium.
- **merozoites.** The many trophozoites released from human red blood cells or liver cells at maturation of the asexual cycle of malaria.

Definitions

- **nosocomial.** An infection that originates in and is acquired from a medical facility. Infections present or incubating inside patients when they are admitted to the facility are *not* included in this definition.
- **oocyst.** The encysted form of the ookinete that occurs on the stomach wall of *Anopheles* spp. mosquitoes infected with malaria.
- **ookinete.** The motile zygote of *Plasmodium* spp.; formed by microgamete (male sex cell) fertilization of a macrogamete (female sex cell). The ookinete encysts (see **oocyst**).
- **paroxysm.** The fever-chills syndrome in malaria. Spiking fever corresponds to the release of merozoites and toxic materials from the rupturing parasitized red blood cells and shaking chills occur during subsequent schizont development. Occurs in malaria cyclically every 36 to 72 hours, depending on the species.
- **patent.** Apparent or evident.
- **pseudocysts.** A cystlike structure with a membrane covering formed by the host following an acute infection with *Toxoplasma gondii.* The cyst is filled with bradyzoites in immunocompetent hosts; it may occur in brain or other tissues. Latent source of infection that may become active if immunosuppression occurs.

Definitions

- **recrudescence.** A condition that may be seen in any malarial infection; infected red blood cells and accompanying symptoms reappear after a period of apparent “cure.” This situation reflects an inadequate immune response by the host or an inadequate response to treatment.
- **relapse (malaria).** A condition seen subsequent to apparent elimination of the parasite from red blood cells; caused by a reactivation of sequestered liver merozoites that begin a new cycle in red blood cells. True relapses occur only in *Plasmodium vivax* and *P. ovale* infections.
- **sarcocyst.** Infective cyst containing banana-shaped bradyzoites.
- **schizogony (merogony).** Asexual multiplication of ***Apicomplexa;*** multiple intracellular nuclear divisions precede cytoplasmic division.

Definitions

- **schizont.** The developed stage of asexual division of the *Sporozoa* (e.g., *Plasmodium* spp. in a human red blood cell; *Cystoisospora belli* in the intestinal wall).
- **sporocyst.** The fertilized oocyst in which the sporozoites of *Plasmodium* have developed.
- **sporogony.** Sexual reproduction of *Apicomplexa.* Production of spores and sporozoites.
- **sporoplasm.** Infectious protoplasm consisting of ions, lipids, monosaccharides, amino acids, nucleic acids and other proteins,
- **sporozoites.** Forms of *Plasmodium* that develop inside the **sporocyst,** invade the salivary glands of the mosquito, and are transmitted to humans.
- **tachyzoites.** Rapidly growing intracellular trophozoites of *Toxoplasma gondii.*
- **zygote.** The fertilized cell resulting from the union of male and female gametes.

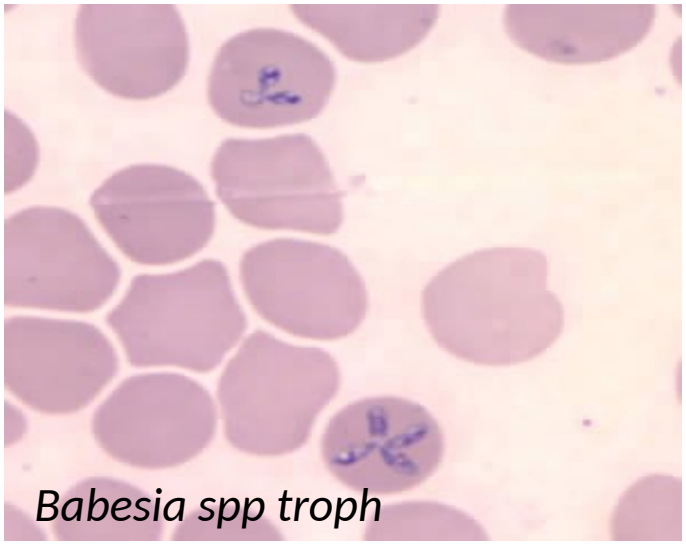
Sporozoa

Scientific Name	Common Name
Plasmodium vivax (plaz-mo'dee-um/vye'vacks)	Benign tertian malaria
Plasmodium falciparum (plaz-mo'dee-um/fal-sip'uh-rum)	Malignant tertian malaria
Plasmodium malariae (plaz-mo'dee-um/ma-lair'ee-ee)	Quartan malaria
Plasmodium ovale (plaz-mo'dee-um/ova'lee)	Ovale malaria
Plasmodium knowlesi (plaz-mo'dee-um/know-les'i)	None
Babesia spp. (bab-ee"zee'-uh)	None

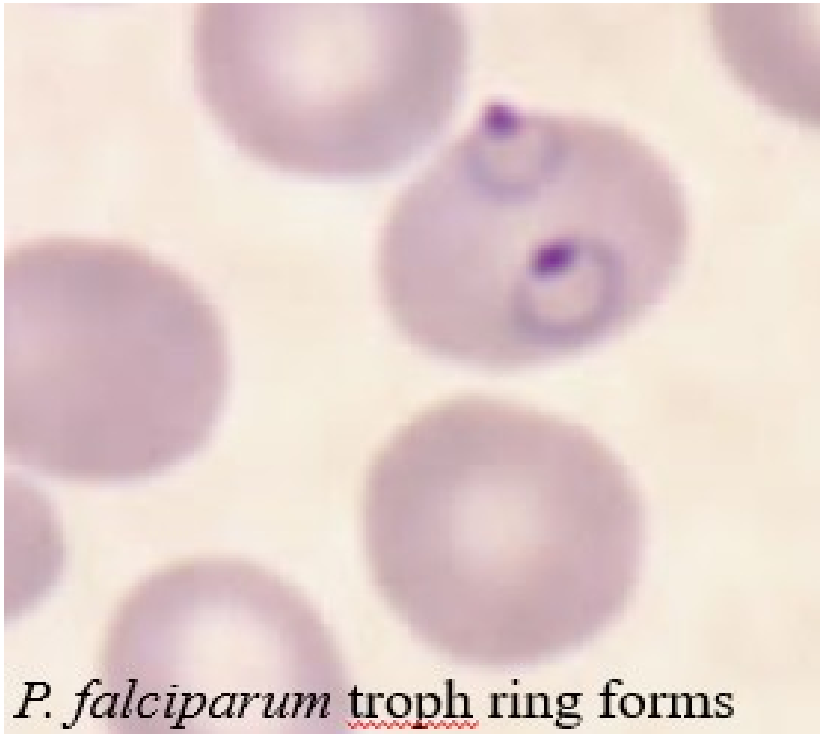
Classification of Blood and Tissue Protozoa

Part I: Sporozoa (malaria and babesiosis)

- ***Plasmodium falciparum***
- ***Plasmodium vivax***
- ***Plasmodium ovale***
- ***Plasmodium malariae***
- ***Babesia* spp.**



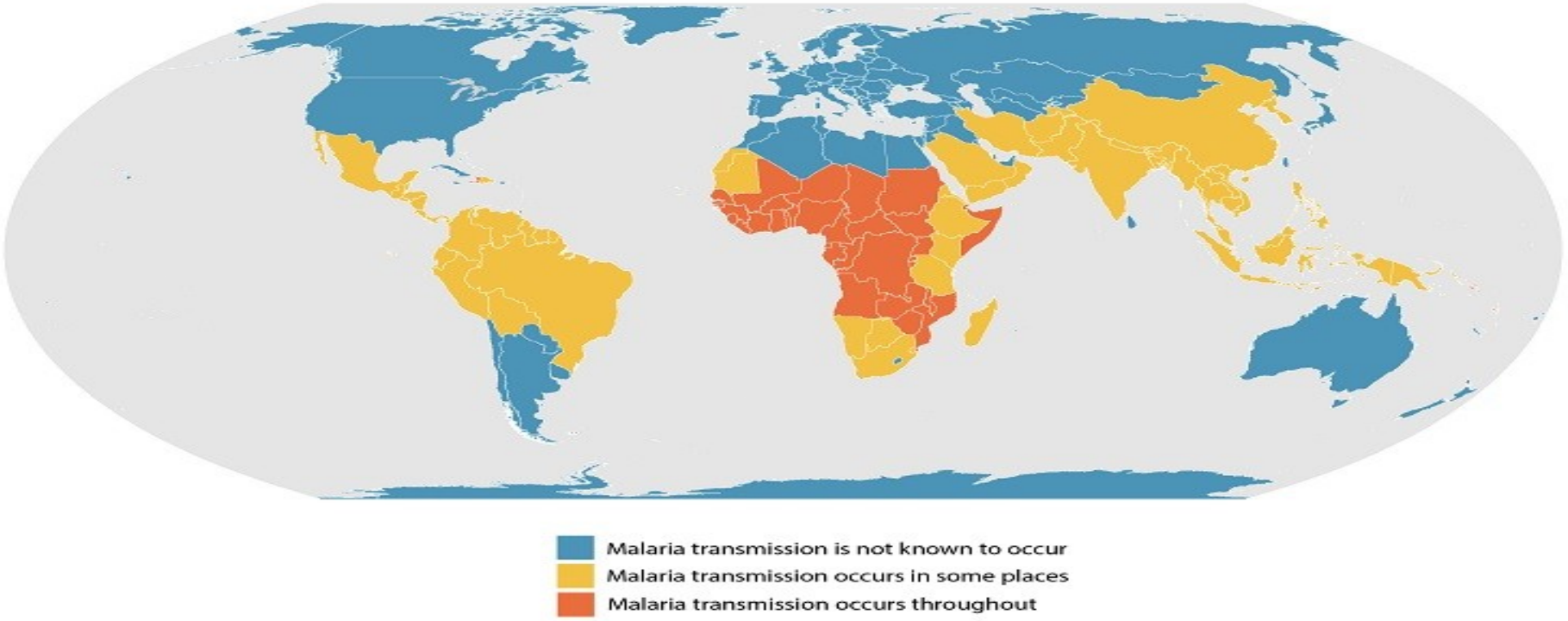
Babesia spp troph



P. falciparum troph ring forms

Malaria

- **Malaria:** number one killer of all parasitic diseases
  - Documented cases since 2700 BC
  - Estimated 500 million people worldwide are infected
  - Estimated 2.7 million people die worldwide
  - **95% of infections** are either **P. falciparum or vivax**



Epidemiology of Malaria

- Outside of US
  - **Endemic in tropics and subtropics**;
    - ½ of world's population lives in endemic areas
  - Tropical malaria: more stable and difficult to control
  - **Environmental conditions**: areas that allow multiplication of mosquito vectors; increased cases during **rainy season**
  - Malaria **decreases** at altitudes above 6000 ft
  - P. vivax and P. falciparum are **most** common

Epidemiology of Malaria

- **Inside of US:**
  - **Most cases** in travelers or immigrants from endemic areas
  - **Rare cases** of transmission in blood transfusion or blood contaminated needles
  - Malaria should be considered in a patient w/ **history of travel or residence in endemic areas**

Mosquito Disease Transmission

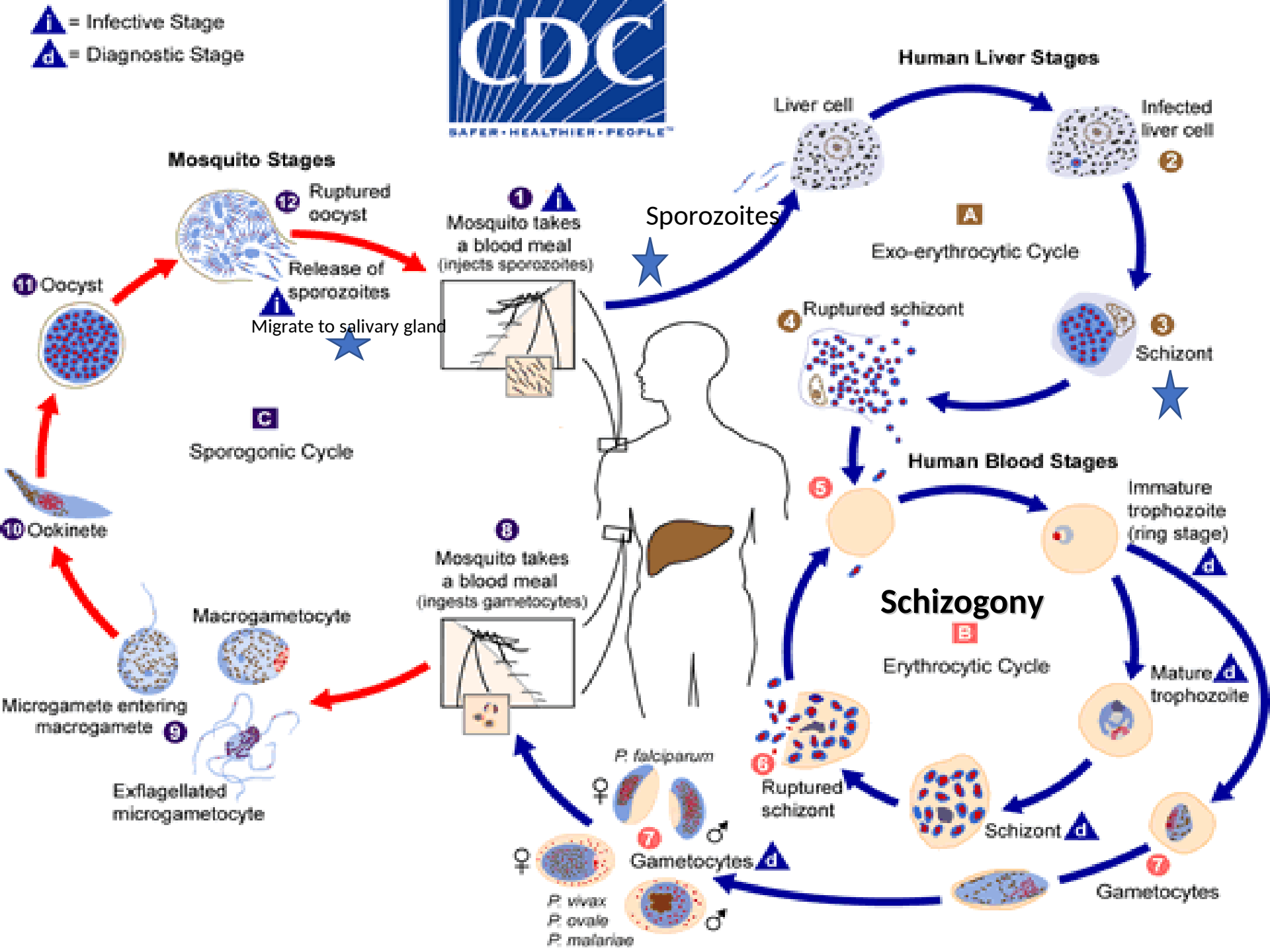
- ***Anopheles* spp (active 9pm-5am)**
  - ***Plasmodium* spp: Malaria**
- ***Aedes aegypti*, *Aedes albopictus* and other spp (active daytime)**
  - Flaviviruses: Dengue, Yellow Fever, Zika virus
  - Alphaviruses: Chikungunya virus
- ***Culex* spp (active 5pm-9pm)**
  - Flaviviruses: West Nile virus, Japanese encephalitis, St. Louis encephalitis virus

Life Cycle of Malarial Parasites

- **Vector:** **female *Anopheles*** mosquito
  - **Sporogony**: sexual cycle of *Plasmodium* spp
  - **Definitive** host
- **Humans = intermediate host**
  - **Schizogony**: asexual cycle
  - **Intermediate** host
  - Stages:
    - Pre-erythrocyte/primary exoerythrocytic
    - Erythrocytic
  - Cyclical illness of fever and chills

Life Cycle of Malarial Parasites

1. Mosquito takes a blood meal
   - Sporozoites from salivary gland enter human blood
   - Within 1 hour, sporozoites travel to liver and replicate in hepatocytes
     - **Schizogony** – massive reproduction results in merozoites in liver
2. Merozoites leave the liver into the blood and penetrate RBCs
   - Merozoites feed on hemoglobin and merozoite takes on mature trophozoite form
     - Trophozoites become schizonts
   - After several generations of infecting RBCs, some merozoites may male and female gametocytes
     - However, no sexual reproduction within humans
3. Mosquito takes a blood meal containing merozoites and gametocytes to complete the cycle


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

General Clinical Symptoms of Malaria

- Step 1 in humans:
- Exoerythrocytic cycle
  - Patient remains **asymptomatic** after mosquito bite and during liver stage

General Clinical Symptoms of Malaria

- Step 2
- Erythrocytic cycle (schizogony)
  - Paroxysmal: first clinical symptom; large numbers of RBCs rupture simultaneously; merozoites and toxic waste enter blood circulation
  - Cyclical Chills: lasting 10-15 min; followed by fever for 2-6 hr
  - After fever subsides, patient has profuse sweating
  - Additional symptoms: headache, lethargy, anorexia, nausea, vomiting, diarrhea, splenomegaly

General Clinical Symptoms of Malaria

- Recrudescence: P. vivax and P. ovale
  - Dormant forms may occur in liver (**hypnozoites**) during infections with P. vivax and P. ovale
  - May remain dormant for months to years after initial infection
  - Recurrence of malaria with rupture of hypnozoites

Epidemiology of *Plasmodium* spp

Plasmodium spp	Geographic Distribution
P. falciparum & P. vivax	Most common spp worldwide (95% of infections)
P. falciparum	Prevalent in Africa and S Pacific Southern & SE Asia Central and South America
P. vivax	Prevalent in southern & SE Asia South Pacific Islands Central and South America (wide distribution)
P. ovale	Occurs most often in West Africa
P. malariae	Less frequent occurrence sporadic worldwide distribution
P. knowlesi	Forested areas of Southeast Asia (early rings similar to P. falciparum, later troph stages similar to P. malariae)

Treatment of Malaria

- **No ideal/universal drug, vaccine, or treatment exists:**
- **Oral chemoprophylaxis**: traveling to endemic area?
  - **Chloroquine**
  - **Doxycycline**
  - **Mefloquine**

Treatment of Malaria

- **Chloroquine phosphate**
- **Atovaquone-proguanil** = selective inhibition of parasitic electron transport chain
- **Proguanil**: dihydrofolate reductase inhibitor
  - stops deoxythymidine synthesis
- **Quinine** sulphate (PO) + doxycycline
- Severe malaria (>5% parasitemia) = IV quinidine gluconate + PO doxycycline

Prevention of Malaria

- Mosquito **Avoidance Measures**
  - **Chemoprophylaxis** for endemic areas
  - Use mosquito **repellant**
  - Wear **protective clothing**; light weight long sleeves
  - Sleep under insecticide-treated (pyrethrin) **mosquito net**
- Mosquito **Control Measures**
  - Tropical malaria: stable, **difficult to control** & eradicate
  - Use of **insecticide**: e.g. aerial spraying
  - Elimination of mosquito breeding places
    - **Reduce areas of standing water**: e.g. old tires
- GMO mosquitoes

Diagnostic Lab Testing for Malaria

Diagnostic Lab Testing

- Vitally important to be accurate and fast
- P. falciparum or knowlesi can be rapidly **fatal**
- Required highly trained, competent staff
- Multiple samples are required (36 hour time period) via venipuncture
- Typical testing include:
  - Blood smear evaluation
  - Serological assays
  - Molecular assays

Lab Diagnosis of Malaria

- **Timing of Blood Collection ** very important **
  - Peak of fever **not** ideal for collection; infected RBCs rupture -> merozoites present in circulating blood -> difficult to identify species
  - In-between paroxysms: greatest number of parasites are present **inside** RBCs
    - **Look for schizonts and gametes and trophozoite rings**
  - subsequent specimens should be drawn at 6 to 12 hr interval because parasitemia may be intermittent; draw blood for 48 hr before considering patient free of malaria

Lab Diagnosis of Malaria

- Collection of Blood Films = the gold standard
  - Peripheral blood obtained by finger stick or EDTA tube
  - blood should be drawn immediately upon arrival at the hospital and tested as STAT
  - For EDTA tubes, smears must be made w/in one hour to retain true stippling in young RBC (P. vivax)
  - Giemsa stains preferred but Wright's stain can be used

Examination of Blood Films

- Prepare **thick** and **thin blood films** and stain with Wright-Giemsa stain
- **Thick film**: examination of larger blood volume than thin film
  - spread a few drops of blood (EDTA) over 2 cm area; stir 30 sec to prevent fibrin strands**; air dry**; do not fix in methanol; RBCs ruptured by stain; read under oil immersion (1000x)
    - Qualitative assessment
- **Thin film:** used for **specific identification of parasites**; fewer organisms per field compared to thick film
  - Prepare thin film; air dry; fix in methanol; stain and read 300 fields under oil immersion (1000x)
  - malarial parasites are seen within RBCs
  - Quantitation: <5% uncomplicated; >5% severe malaria (IV)

Blood Testing: Thick and Thin Smears

- Whole blood is collected in EDTA tube to detect parasites STAT
- Parasitemia and detection may vary by organism life cycle
- Freshly collected blood is stained with Wright Giemsa or Delafield's hematoxylin
- **Thin Smear:**
  - Examined under low and high power **looking for plasmodium**
  - Be sure to check the feathered edge and edges of coverslip (if used)
  - **300 fields** should be examined before calling a negative smear
- **Thick smear:**
  - Allows detailed examination under high power
  - RBCs are heavily concentrated and laked (lysed) during staining in the center of smear, intact RBCs may remain in periphery
  - **Examine 100 fields**

Serological, Automated, & Molecular Lab Testing for Malaria

- **Rapid Malarial Tests** using monoclonal antibodies directed against:
  - Histidine-rich protein 2 [HRP2]
  - Parasitic Lactate Dehydrogenase [pLDH]
- **BinaxNOW Rapid antigen** is FDA approved in US for P. falciparum and vivax currently
- **Molecular Testing:**
  - DNA probes directed against all 5 Plasmodium spp via PCR
    - Detects lower parasitemia loads than thick blood smears
- Automated instruments like Flow cytometry may be used but in parasitemia <0.5% the results may be unreliable

Clinically Significant Plasmodium Infections

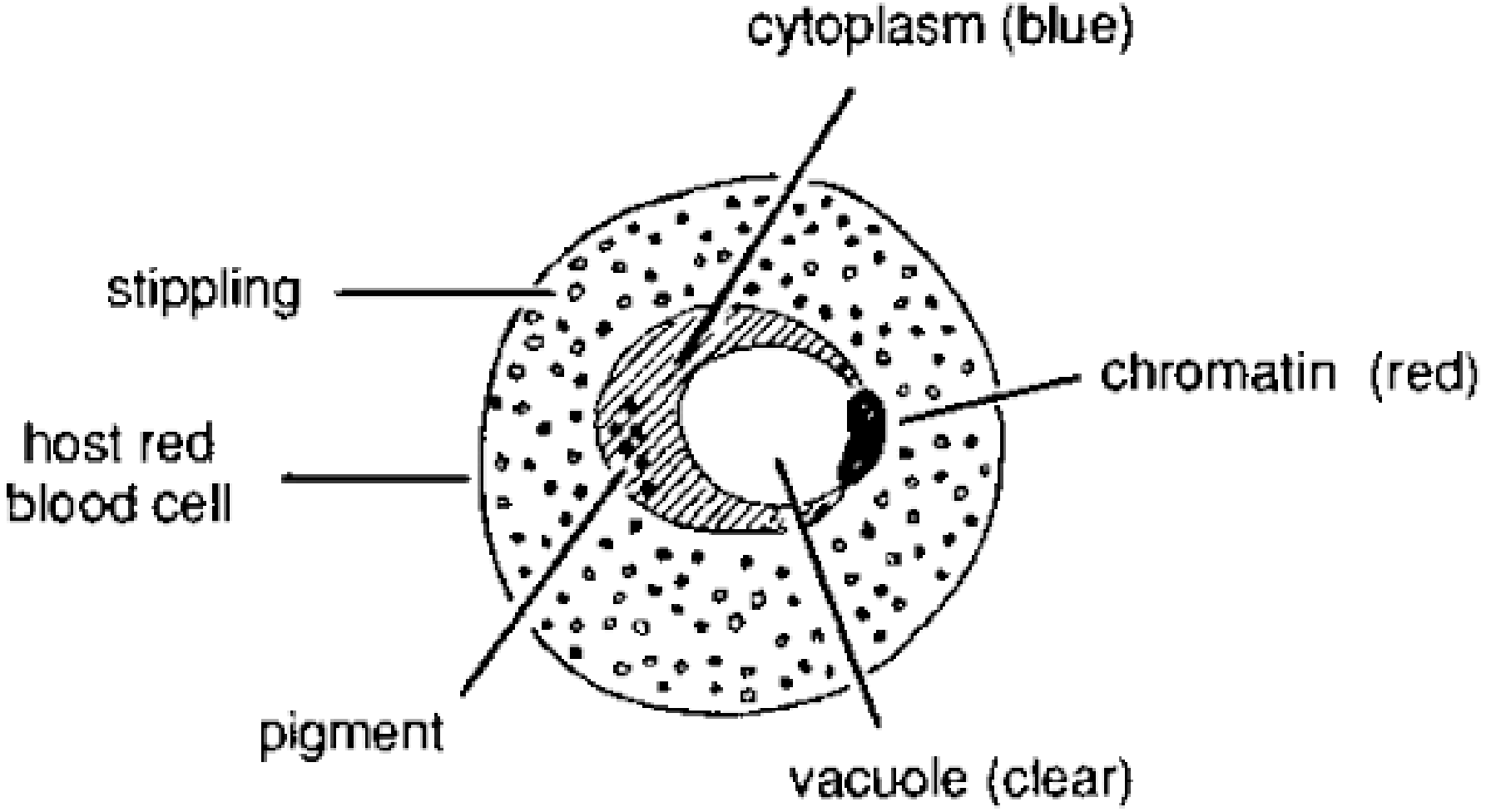
Differentiating Malarial Infections

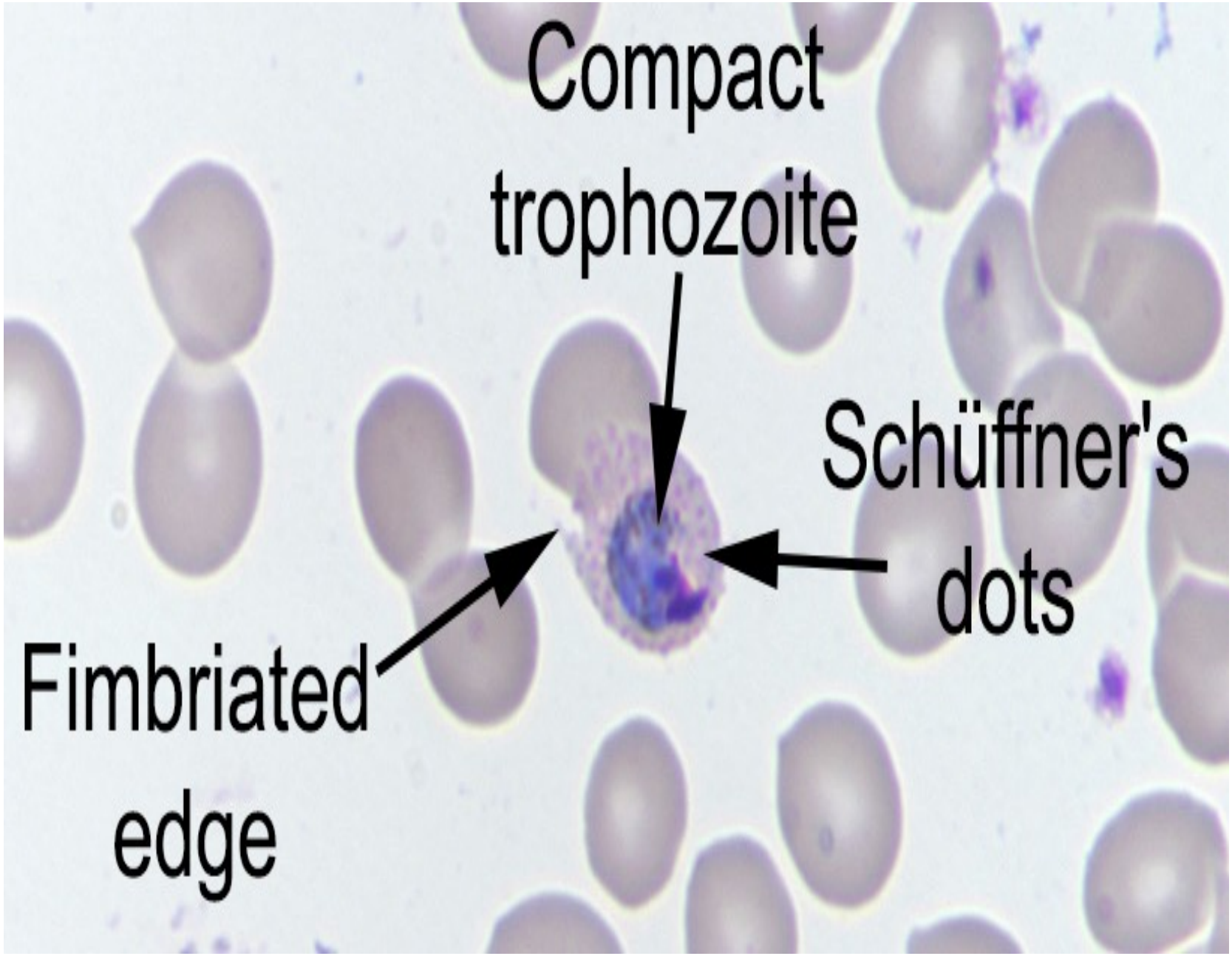
Scientific Name	Common Name	Cyclic Paroxysms	Relapses
P. falciparum	Malignant malaria	Every 36 to 48 hours	No
P. vivax	Tertian malaria	Every 48 hours	Yes
P. malariae	Quartan malaria	Every 72 hours	No
P. ovale	Ovale malaria	Every 48 hours	Yes
P. knowlesi	Quotidian malaria	Every 24 hours	No

Plasmodium Morphology of RBC Forms

- **Ring Form (early Trophozoite):** blue cytoplasmic circle connected to red chromatin dot; internal vacuole (space)
- **Developing Trophozoite:** varies among *Plasmodium* spp; amount of RBC space invaded is greater than early ring; pigment (brown) is often visible
- **Immature Schizont:** evidence of active chromatin replication w/ cytoplasmic material; pigment granules (brown); as multiplication continues more space is occupied within RBC
- **Mature Schizont:** emergence of merozoites; arrangements vary by species of *Plasmodium*
- **Microgametocyte:** male form; most are round in shape (*P. falciparum* is crescent shaped); large diffuse chromatin mass (pink)
- **Macrogametocyte:** female form; most are round in shape (*P. falciparum* is crescent shaped); compact chromatin mass (pink) surrounded by cytoplasmic material

Diagram of Young Trophozoite in Ring Form




Compact
trophozoite
Schüffner's
dots
Fimbriated
edge

Plasmodium falciparum

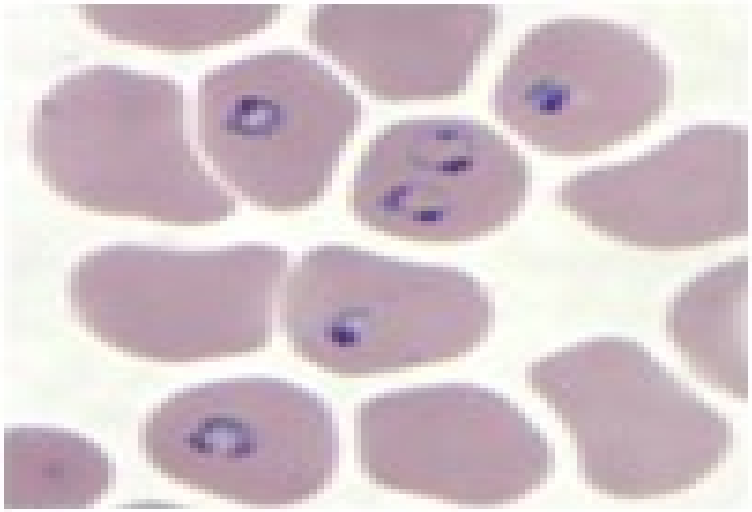
- **Malignant Tertian malaria**
- 36-48 hour cycle
- Infects **all** RBCs regardless of age
- High parasitemia % in peripheral blood
  - Usually 1-20%, but may be higher
- **Various** RBC size
- **No** schuffner dots, but may have **Maurer's dots**
- **Multiple** trophozoite ring in cells
  - **Delicate ring** with **one or two dots of chromatin**
- **Crescent shaped gametocytes "Banana"**

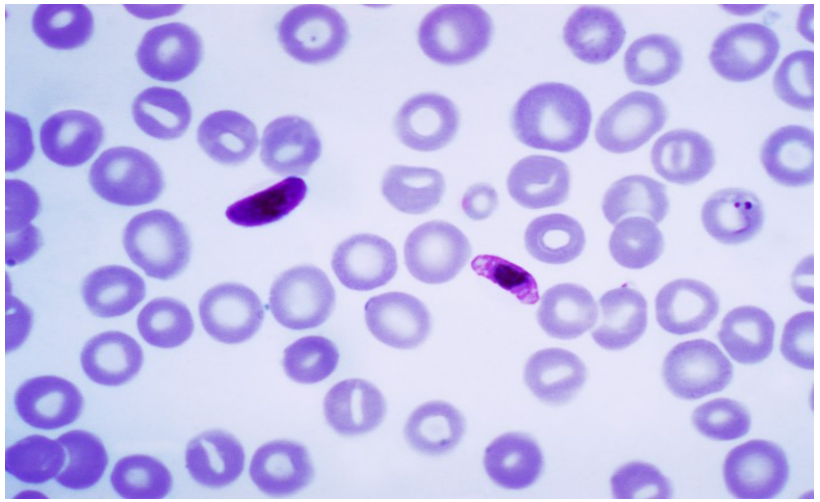
Plasmodium falciparum

- **Ring form: delicate, small ring that is 1/5 RBC diameter**
- Ring form often has **double**, **small chromatin dots**
- **Multiple** rings common in one RBC
- Developing trophozoite: heavy rings; only seen in severe infections
  - occur in capillaries of viscera, not seen in peripheral blood
- Mature schizont is **not** normally seen in peripheral blood
- Gametocytes: crescent-shaped
- **Stages found in peripheral blood**:
  - Young ring form trophozoites
  - Gametocytes
  - No schizonts

Plasmodium falciparum

- Predominates in sub-Saharan Africa
- Schizogony occurs in spleen, liver, and bone marrow in erythrocytic stages
  - **Schizonts can produce up to 24 merozoites**
- Unique **cytoadherence**
  - RBC membrane sticks to endothelial lining of capillaries in organs as parasite grows

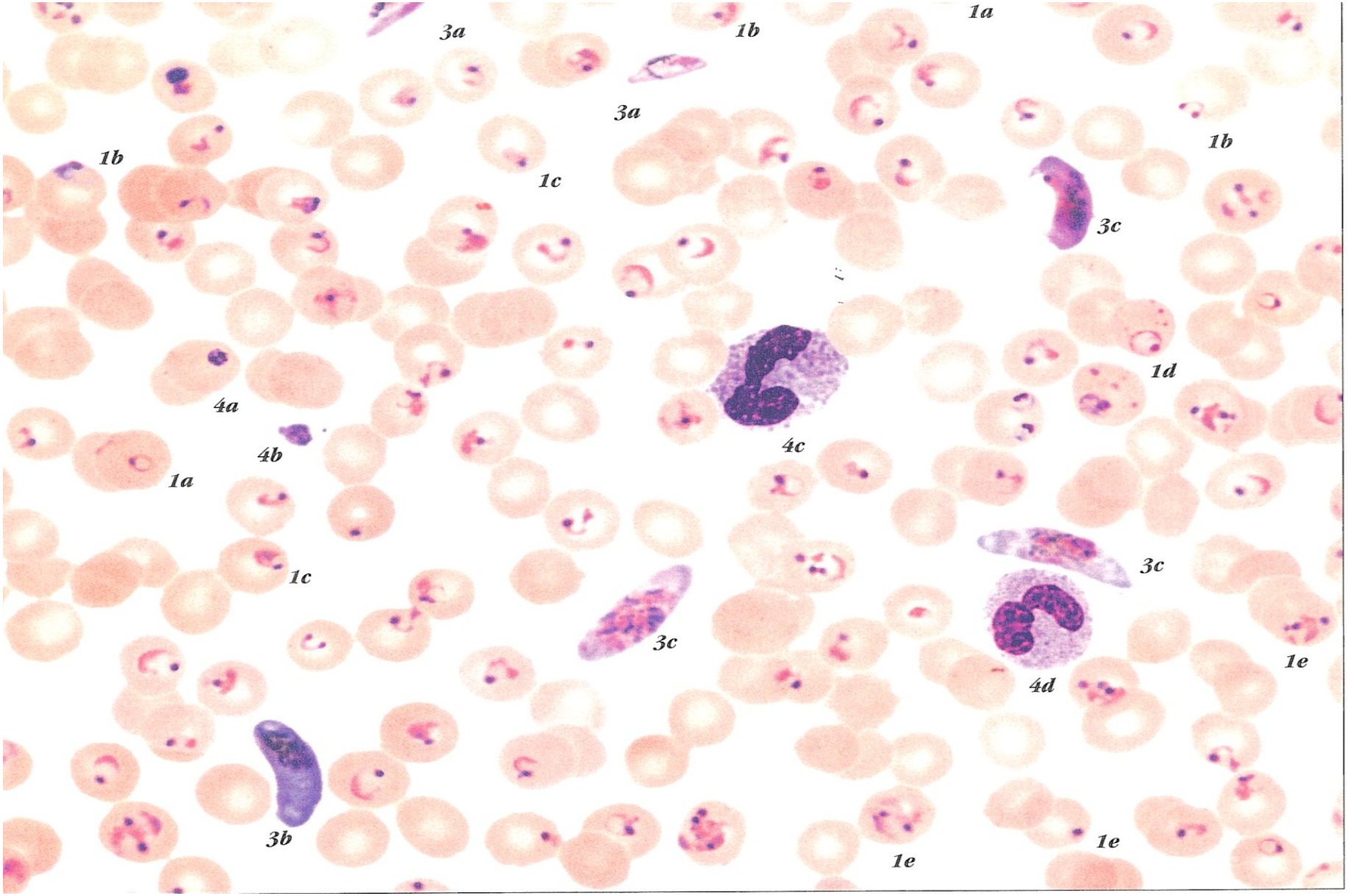




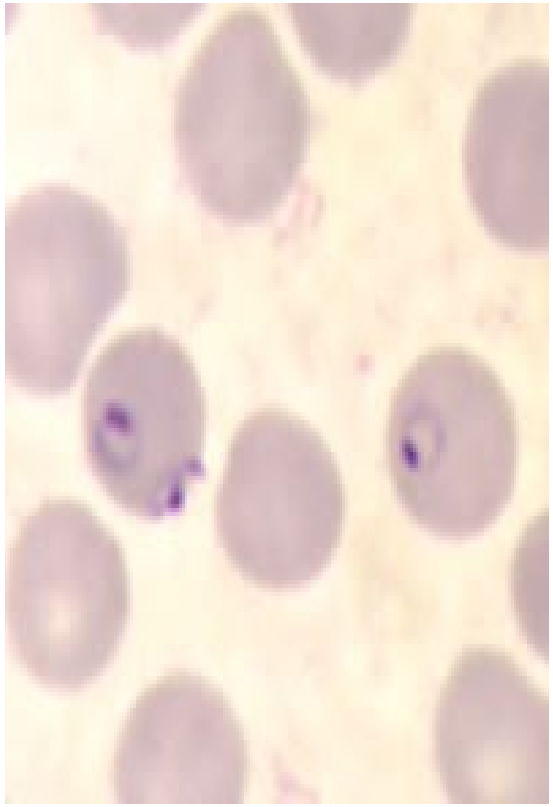
P. falciparum pathogenesis

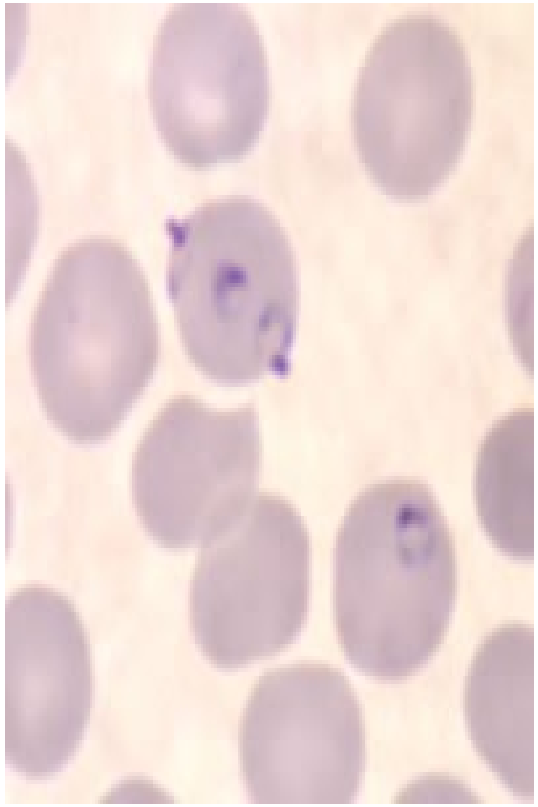
- Incubation period **is 8-12 days post infection** followed by 3-4 days of prodromal illness:
  - Aches, pains, headache, fatigue, anorexia, nausea, fever, and chills
- **Infections can be fatal** due to blood vessel obstructions in organs [liver GI, adrenal glands], intravascular hemolysis, and renal damage
  - **Blackwater fever**
- Highly likelihood of cerebral invasion
  - Fever of 107 F
  - Occurs in 10% of cases and makes up 80% mortality associated with this organism

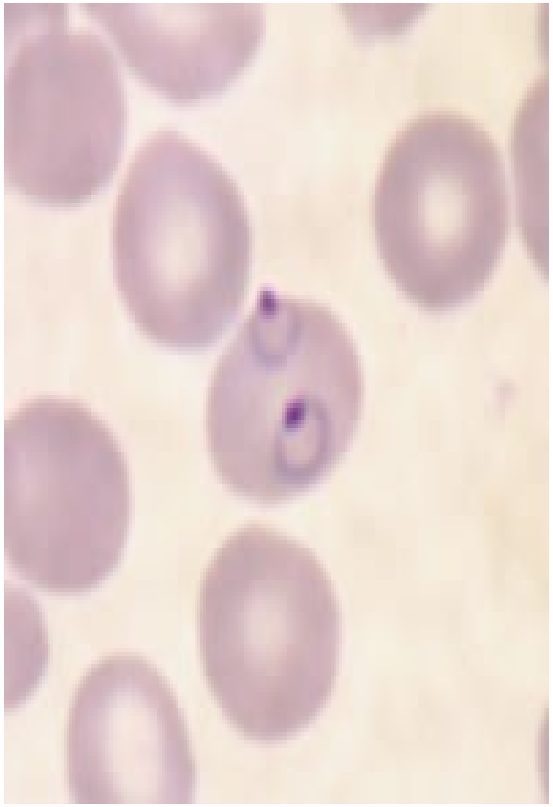
Plasmodium falciparum Stages



Plasmodium falciparum: Thin Blood Film Smear

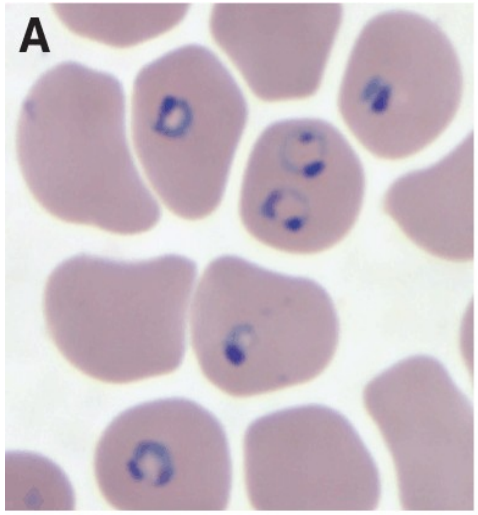


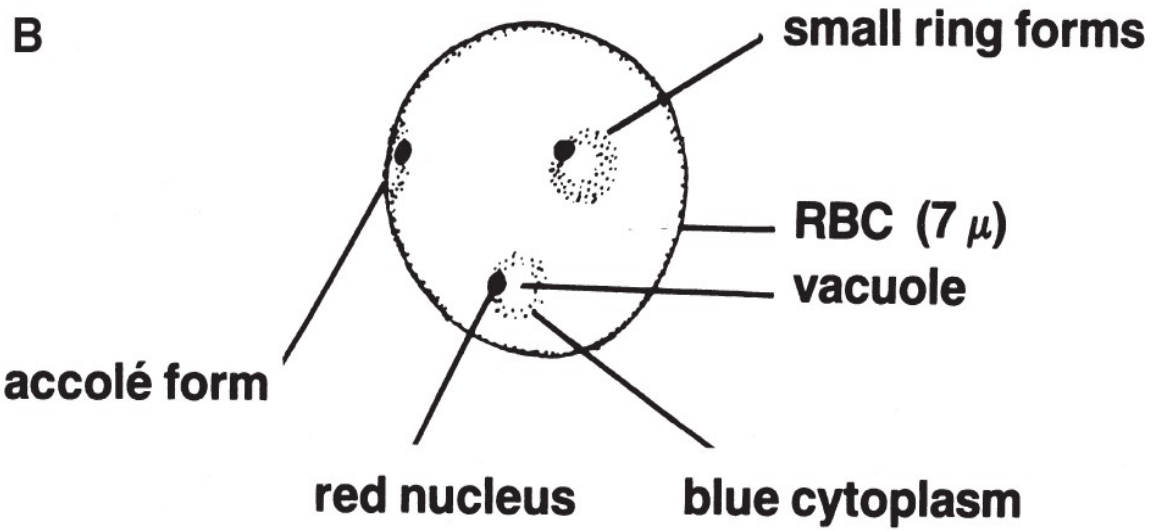


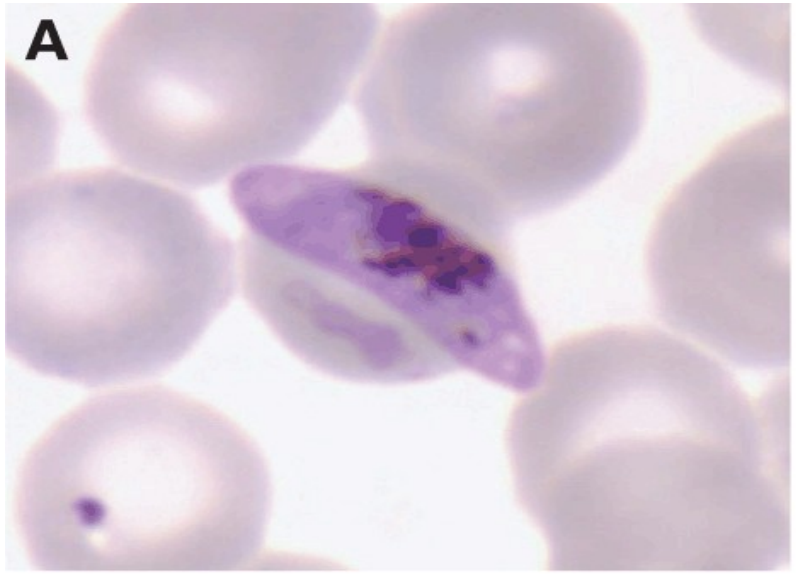


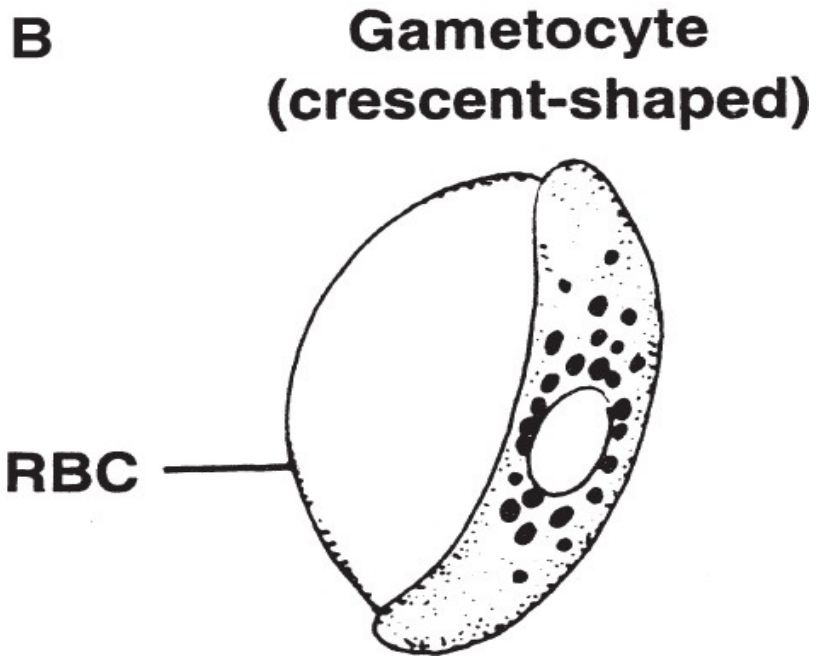
Type	P. falciparum
Incubation period	8-11 days
S&S severity	Mild
Initial fever pattern	Continuous
Cycling pattern	36-48 hours
Duration of untreated infection	1.5-2 years
Age/size of RBC	All RBCs ; varies
Anemia	Severe
CNS involvement	Very common
Nephrotic Syndrome	Rare

Plasmodium falciparum









Plasmodium vivax

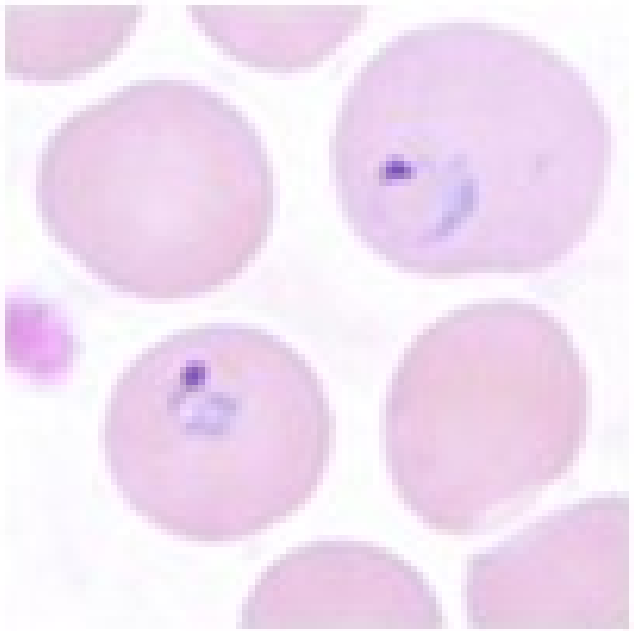
- **Benign tertian malaria**
- **48 hour cycle**
- Infects **young** RBCs + reticulocytes
- **Enlarged** RBCs
- **Schuffner dots**
- Delicate trophozoite ring form
  - **Ameboid / irregular**
- **Schizont contains 12-24 merozoites**

Plasmodium vivax “Benign Tertian Malaria”

- Mostly found in Asia, Latin America, and some parts of Africa
- Max parasitemia is 2 – 5%
- Splenomegaly from **increased** RBC turnover
- **Hypnozoites** in liver may cause relapse due to dormant merozoites
  - **Relapse** can be weeks, months, or years
- Infected **RBCs** are **larger** than normal
- **Some humans have natural resistance**
  - Duffy A and B negative people = resistant

Plasmodium vivax

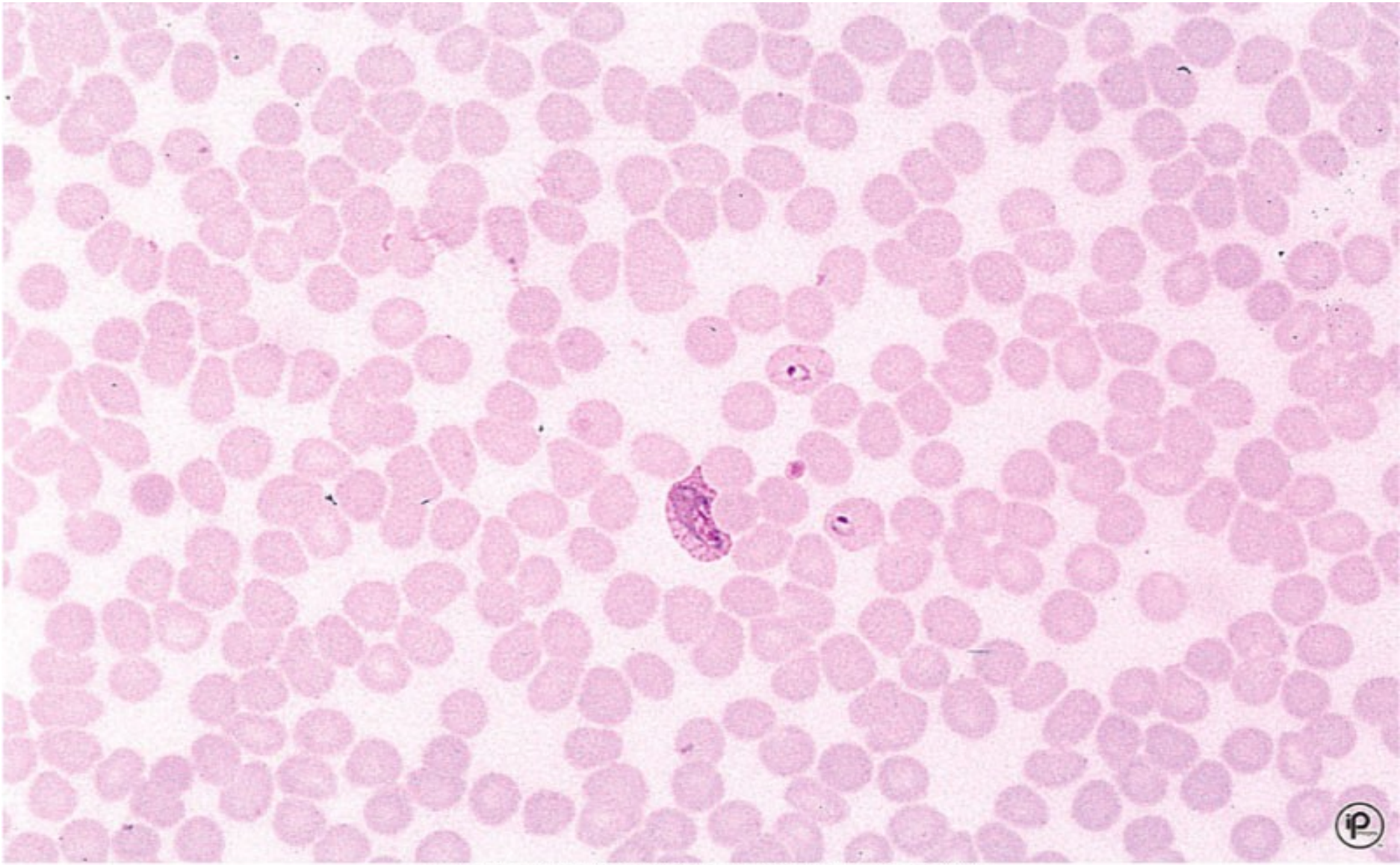
- **Ring form: delicate, large ring**
  - **Diameter is 1/3 – ½ of the RBC**
  - **Single, heavy chromatin dot**
- Developing trophozoite: irregular, ameboid; fine light brown pigment
- Schizont: 12-24 merozoites w/ chromatin & cytoplasm
  - merozoites fill entire RBC
- Gametocytes: round or oval (if seen)
- **Stages found: all stages in peripheral blood**



P. vivax pathogenesis

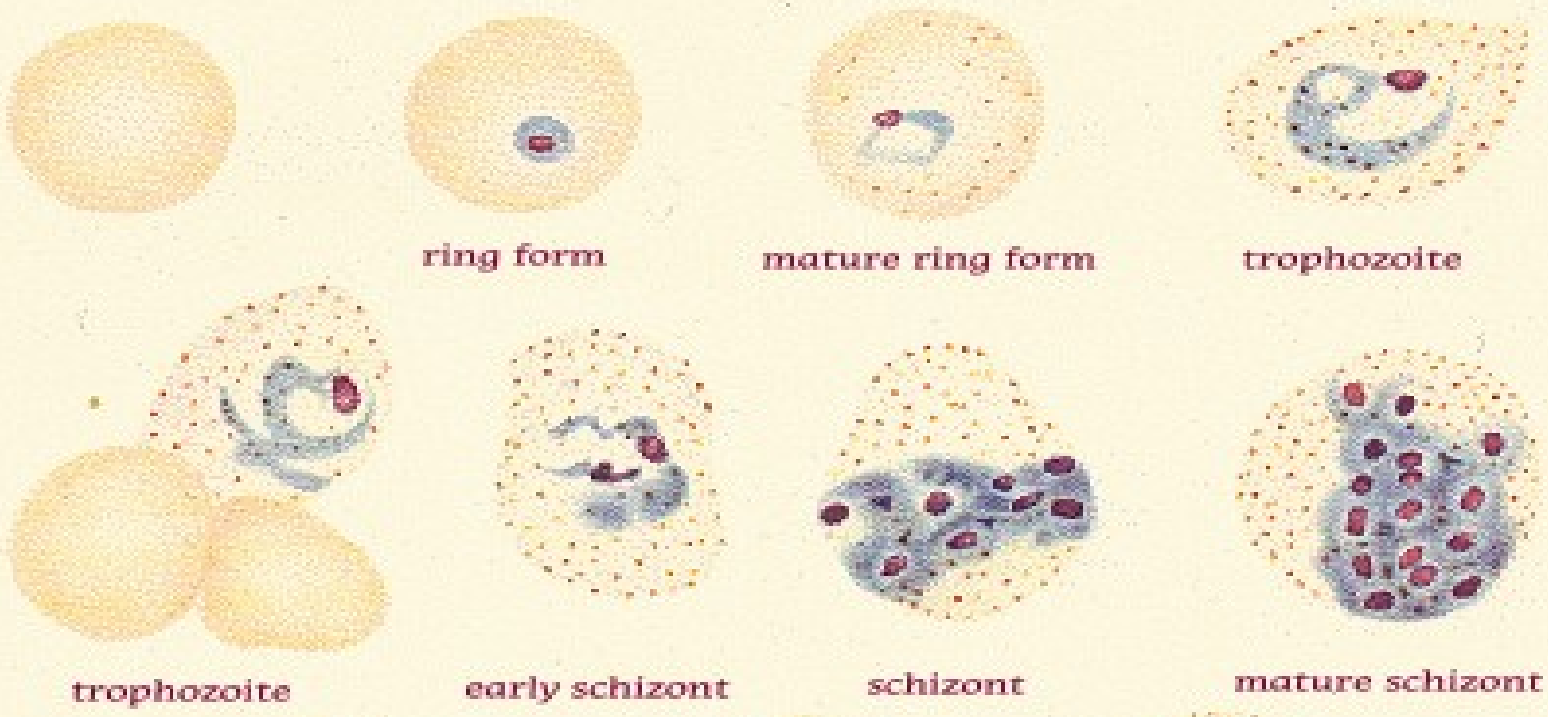
- S&S include headache, photophobia, muscle aches, anorexia, N/V
- **Rare** cases of CNS involvement, but are fatal
- Primaquine resistant strains are extra virulent
  - Renal failure, circulatory collapse, severe anemia, RDS, and jaundice

Plasmodium vivax rings



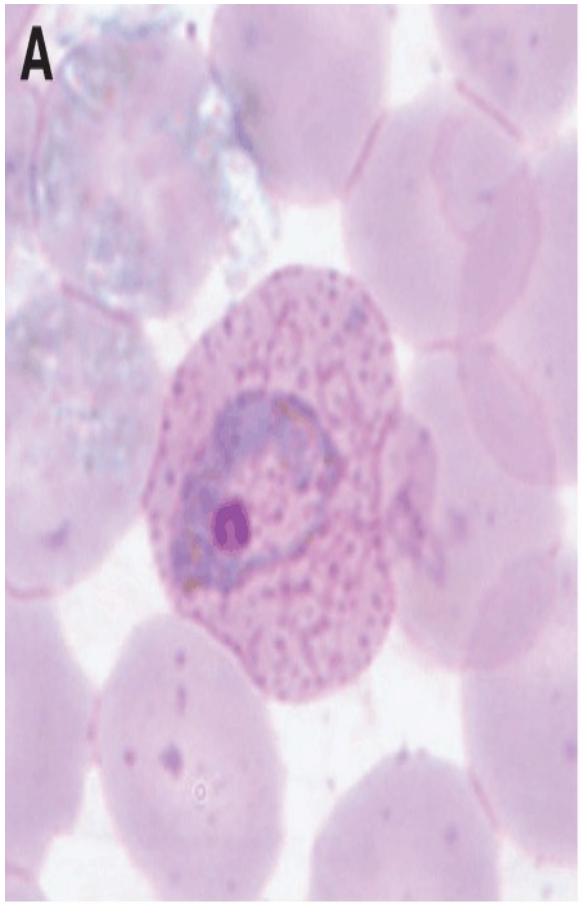
(From Marler LM et al: *Parasitology* CD-Rom, Indiana Pathology Images, 2003.)

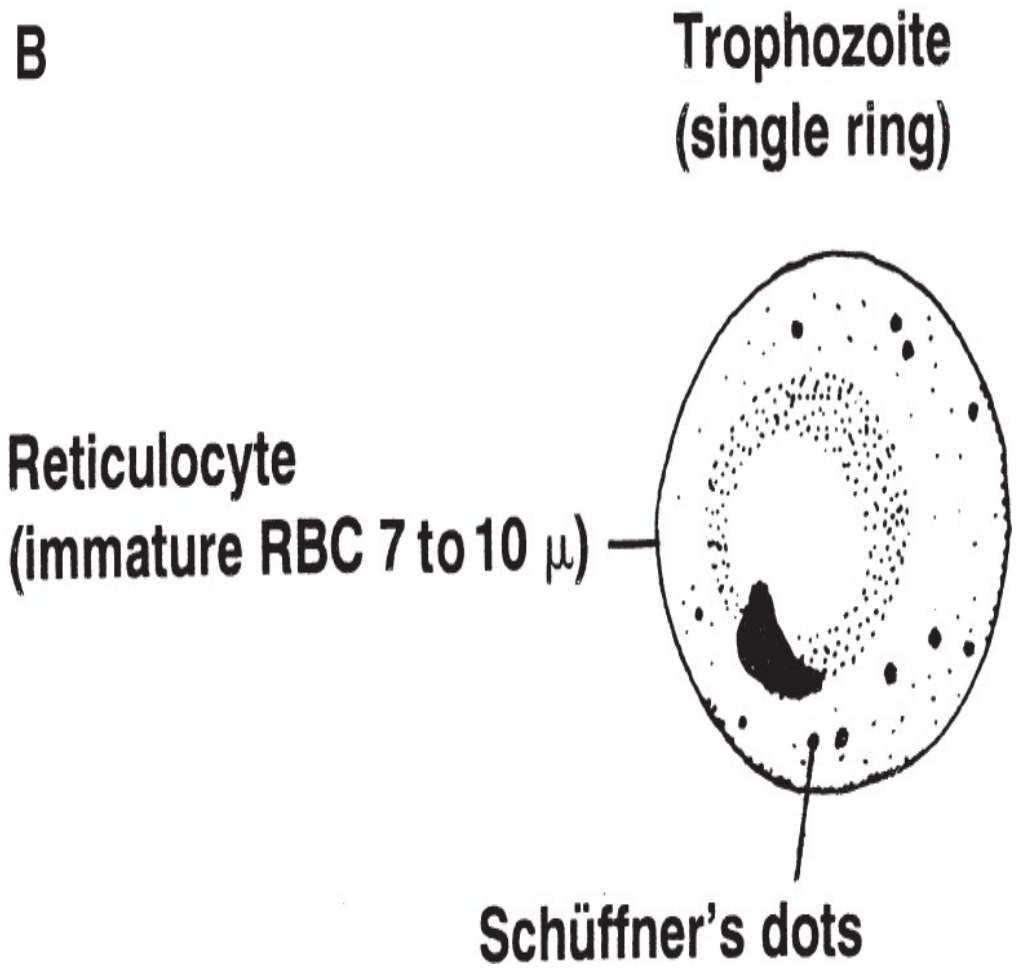
P. vivax


ring form
mature ring form
trophozoite
trophozoite
early schizont
schizont
mature schizont
developing gametocyte
female gametocyte
male gametocyte

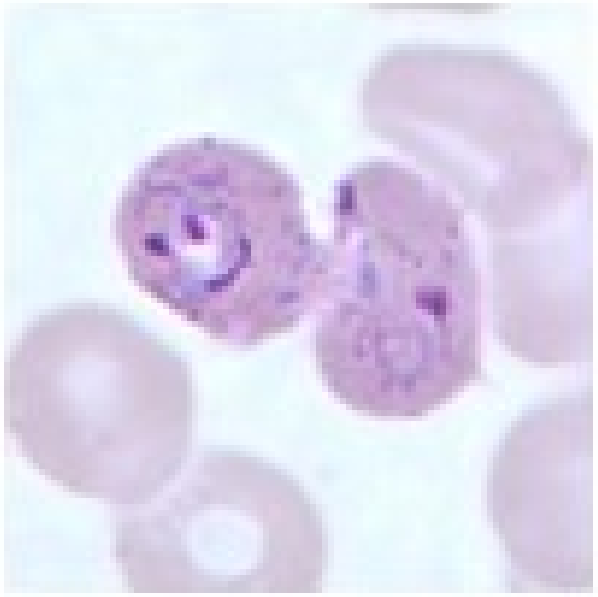
Type	P. vivax
Incubation period	8 – 17 days
S&S severity	Mild to moderate
Initial fever pattern	Irregular until 48 hours
Cycling pattern	48 hours
Duration of untreated infection	5-7 years
Age/size of RBC	Young RBCs/retics ; large
Anemia	Mild to moderate
CNS involvement	Rare
Nephrotic Syndrome	Possible

Plasmodium vivax





Plasmodium ovale



- **48 hour cycle**
- Tends to infect **young** RBCs and reticulocytes
- **Enlarged** RBCs with fimbriated [oval] edges
  - RBCs have “torn or ripped edging”
- **Schuffner dots**
- Small delicate trophozoite ring form
- Mature trophozoite is slightly amoeboid
- **Schizont contains an average of 8 merozoites**

Plasmodium ovale

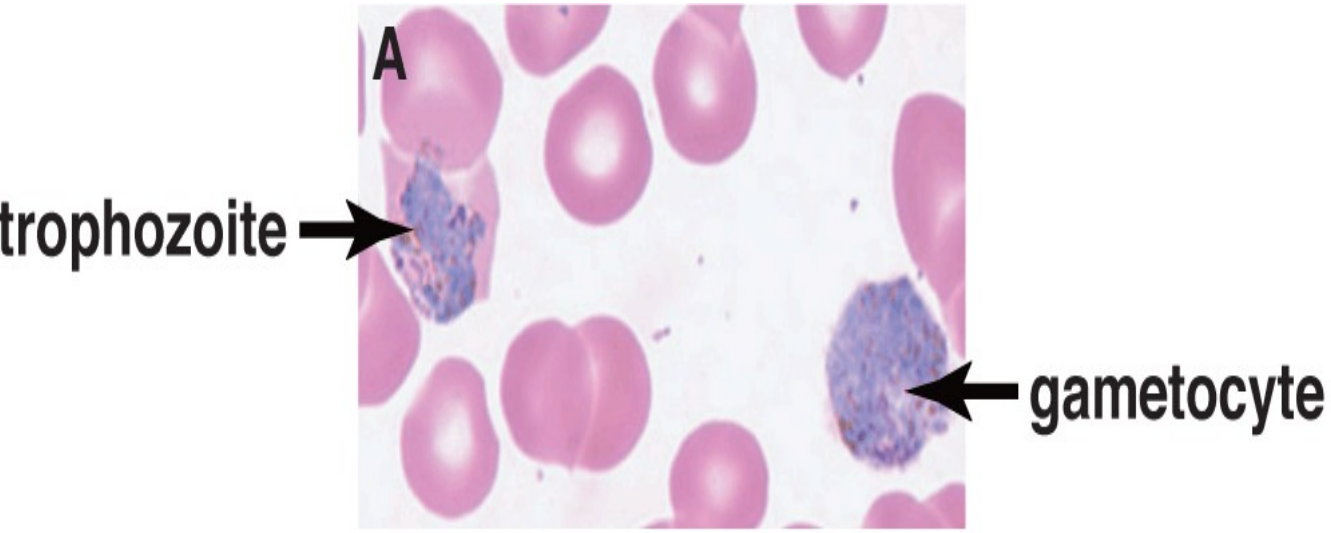
- Occurs most often in **West Africa**
- Similar in appear to P. vivax but less severe
- **Ring form: delicate, large ring, usually larger than P. vivax; 1/3 RBC**
  - **single chromatin dot; ring surrounds a vacuole**
  - **occasional multiple rings**
- Developing troph: ring shape maintained; dark yellow-brown pigment
- Mature schizont**: 6 -12 merozoites**, often in **rosettes**
  - merozoites fill entire RBC
- Gametocytes: round or oval
- Stages found: **all** stages in peripheral blood

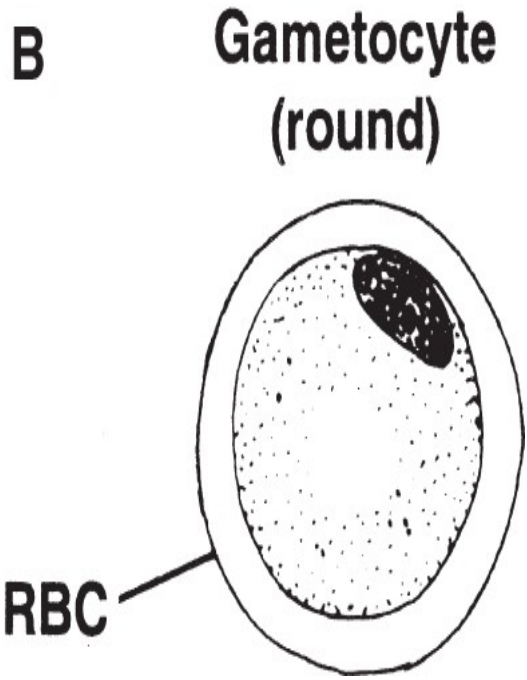
Plasmodium Ovale

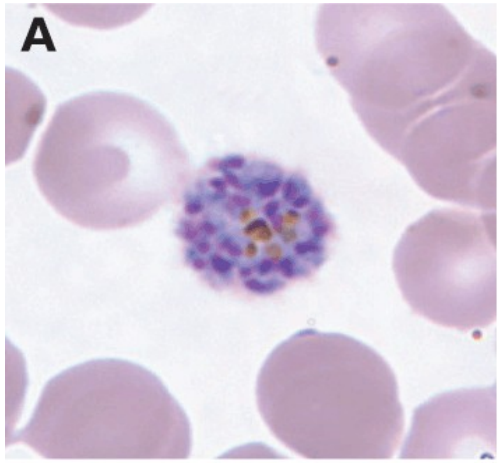
- Clinically less severe and resembles P. vivax
- Usually **spontaneous recovery** after 6-10 cycles
- Only infects reticulocytes (parasitemia 2-5%)
- Regular 48 hour paroxysmal of S&S
- **RBCs** tend to be **larger** than normal
- Have **Schuffners dots** (also known as James stippling)
- Ring trophozoites are less ameboid than P. vivax
- Schizont has up to 8 merozoites
- Usually only prevalent in West Africa

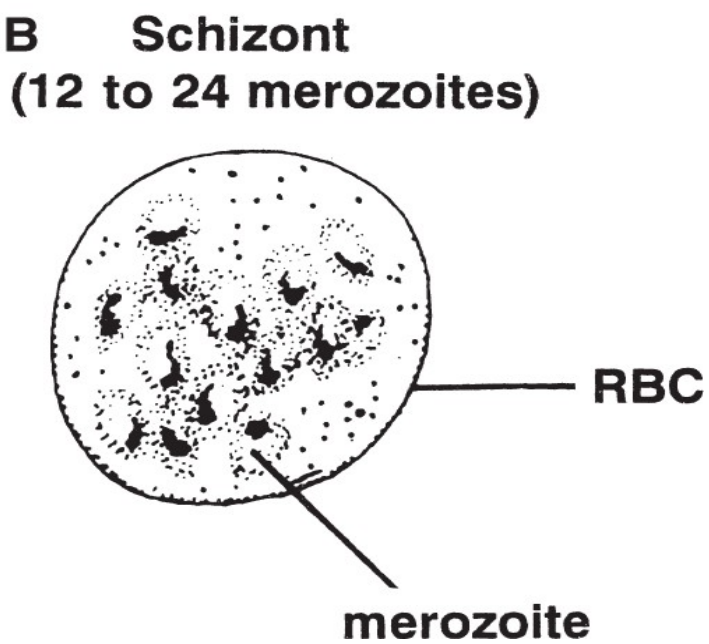
Type	P. ovale
Incubation period	10-17 days
S&S severity	Mild
Initial fever pattern	Irregular until 48 hours
Cycling pattern	48 hours
Duration of untreated infection	1 year
Age/size of RBC	Young RBCs/retics ; large
Anemia	Mild
CNS involvement	Possible
Nephrotic Syndrome	Rare

Plasmodium ovale









Plasmodium malariae

- **Quartan Malaria**
- 72 hour cycle
- Tends to infect old RBCs
- **RBCs are normal to small in size**
- **No stippling**
- Thick ring with large nuclear chromatin
- Trophozoite tends to form a "**band**" across RBC
- Schizont contains ***6-12 merozoites***

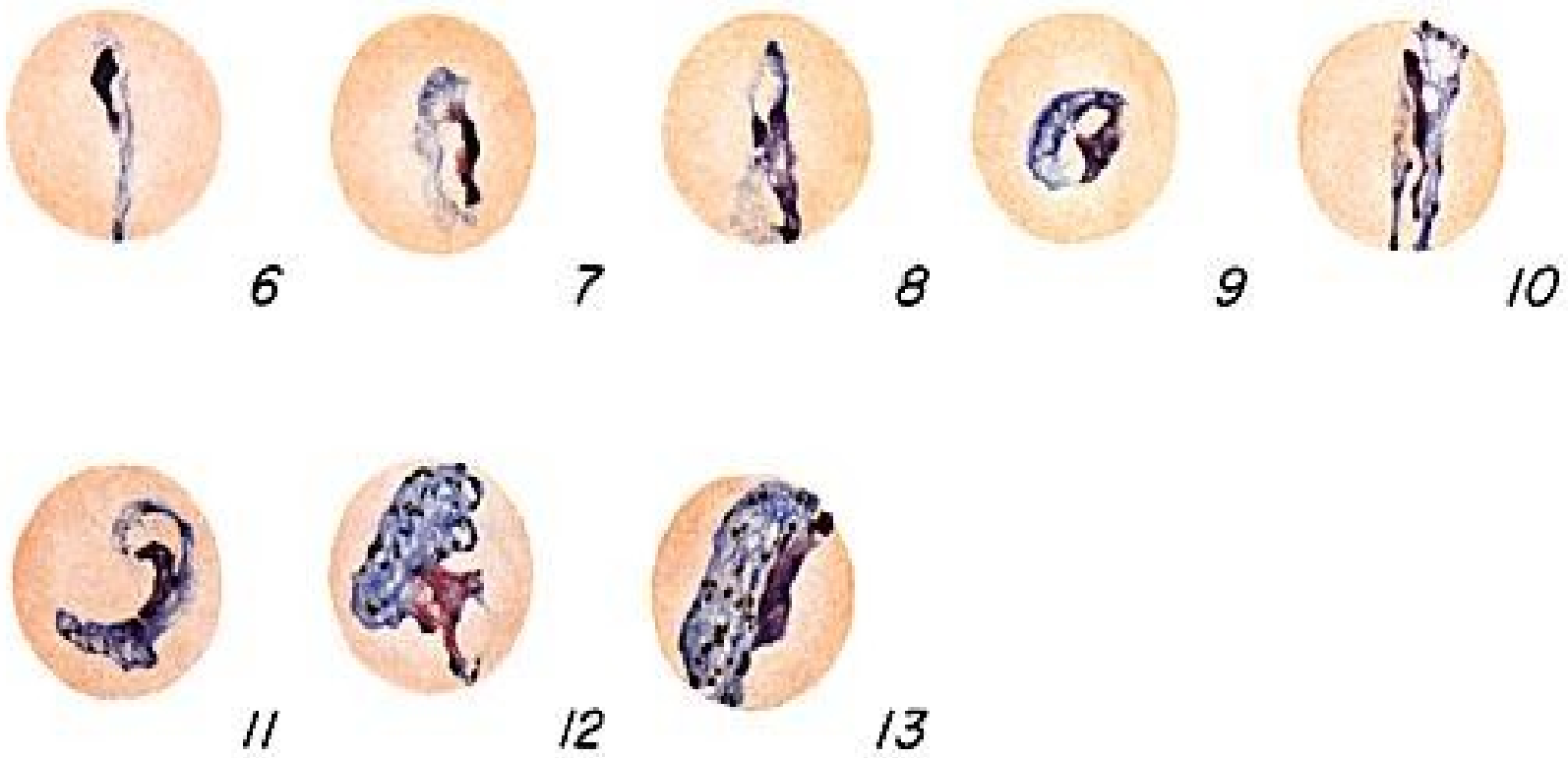
Plasmodium malariae

- Longer incubation period 18-40 days; flulike symptoms
- Spontaneous recovery or chronic repeated attacks may occur
- Mature schizont: 6-12 merozoites arranged in rosettes or clusters
  - central arrangement of brown-green pigment often seen
- Gametocytes: older forms have oval shape; smaller than P. vivax
- Stages found: **all** stages seen in peripheral blood
  - Few to rare ring form
  - Mature trophozoites
  - Schizonts

P. malariae pathogenesis

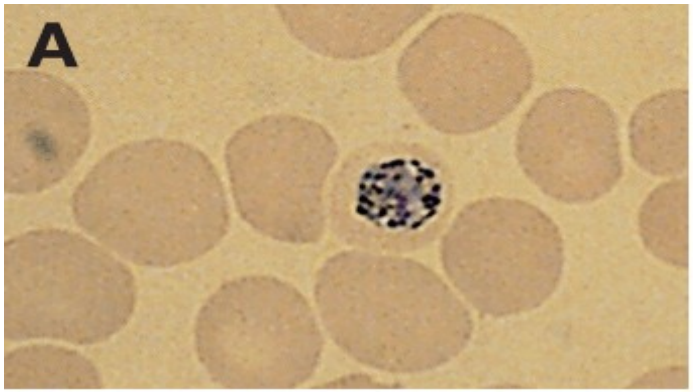
- **Damages the kidneys**
  - Proteinuria and nephrotic syndrome
  - Membranous glomerularnephritis due to accumulation of immune complexes
- May have reoccurring infections with low grade parasitemia for over 50 years
  - Chronic

Plasmodium malariae: Trophozoite Stages



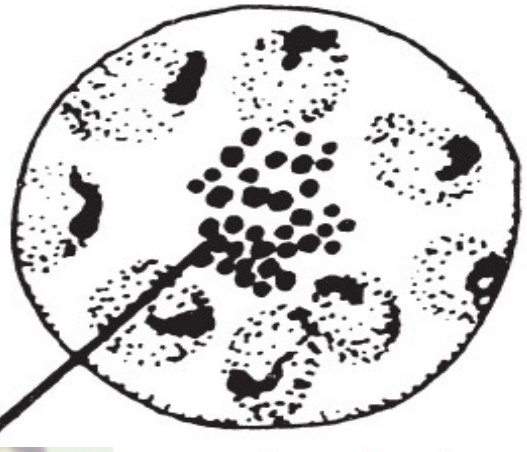
Type	P. malariae
Incubation period	18-40 days
S&S severity	Mild to moderate
Initial fever pattern	Regular until 72 hours
Cycling pattern	72 hours
Duration of untreated infection	20+ years
Age/size of RBC	Older RBCs ; smaller
Anemia	Mild to moderate
CNS involvement	Rare
Nephrotic Syndrome	Very common

Plasmodium malariae

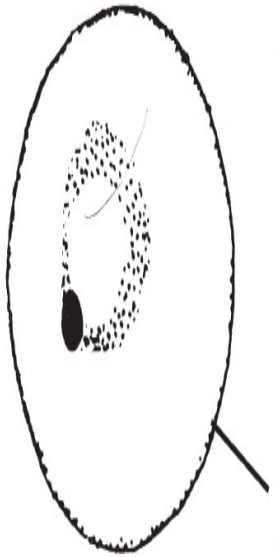


B

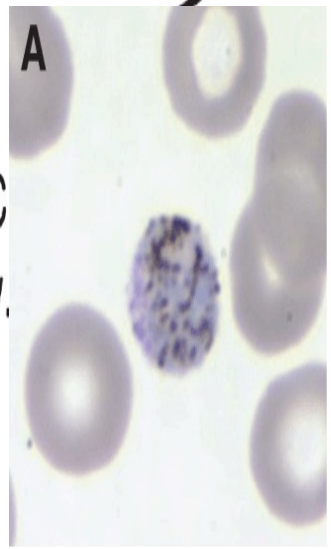
Schizont
(6 to 12 merozoites)



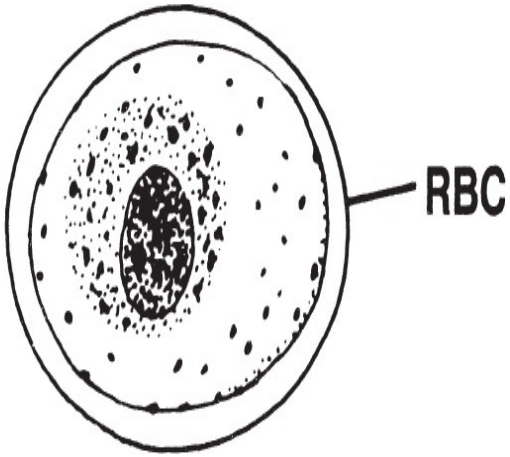
Trophozoite
(single ring)



B *Note*: Trophozoite forms band across RBC during early schizogony.



B **Gametocyte**
(ovoid)

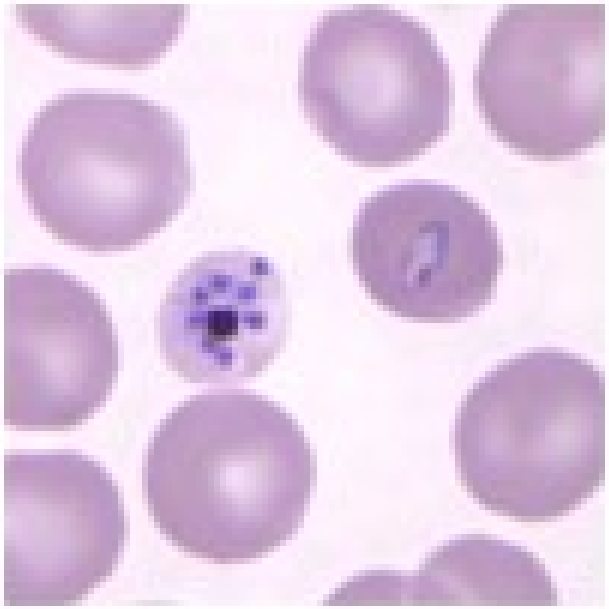


Plasmodium knowlesi

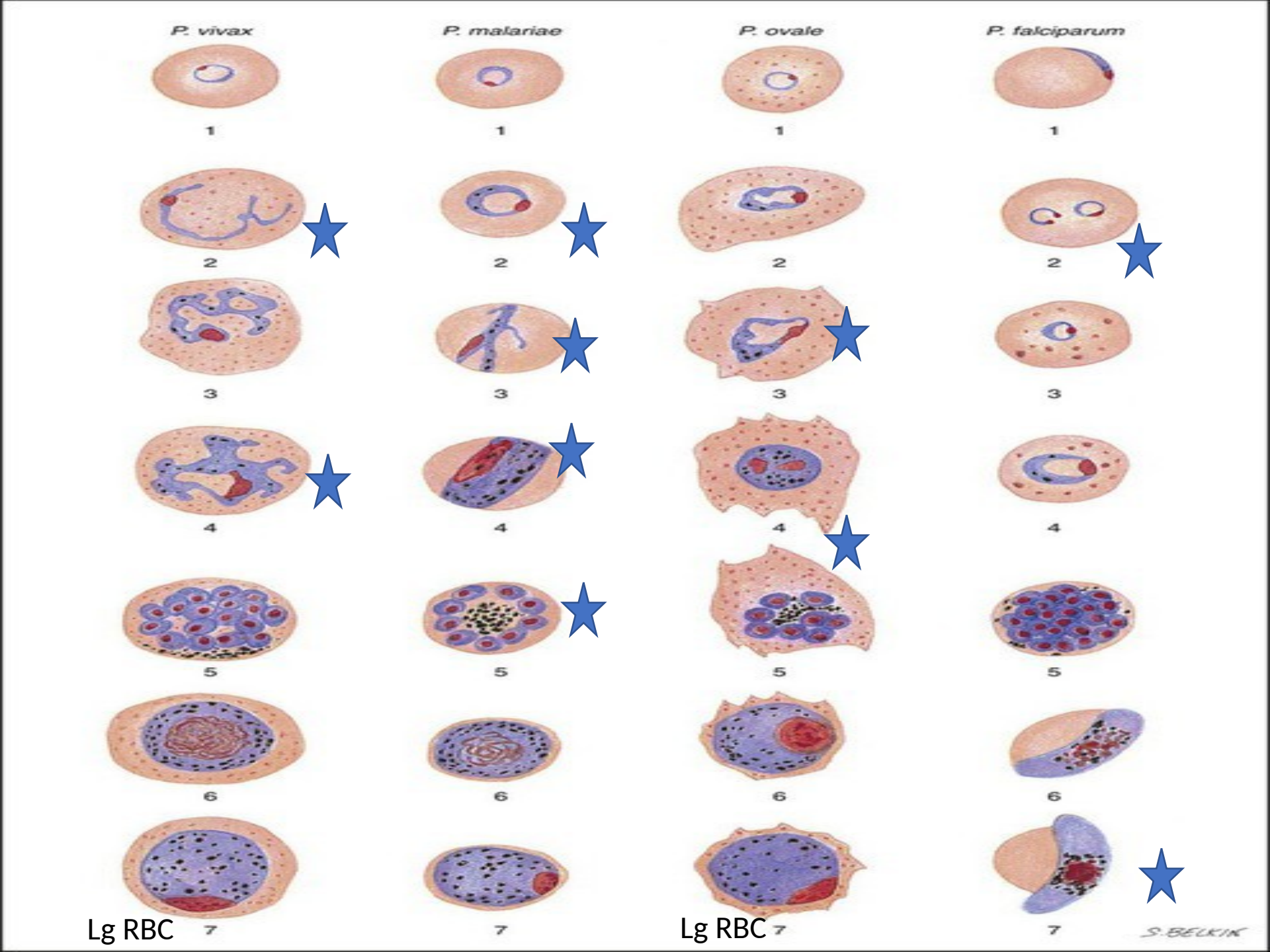
- 24 hour cycle
- Tends to infect **all** RBCs regardless of age
- **High** parasitemia load
- **RBC size variable** but typically normal
- **No** Schunner dots
- **Hard to diagnosis, but proper diagnosis is crucial!**
  - **Early** stages = resembles P. falciparum
  - **Late** stages = resembles P. malariae
- 2-3 trophozoite rings per RBC - early
  - Delicate rings with 2-3 chromatin rings
- Mature trophozoite is banded - late
  - Gametocytes are round
  - Schizonts contain up to 16 merozoites

Plasmodium knowlesi [**simian malaria**]

- **Some humans have natural resistance**
  - Duffy A and B negative people = resistant
- Possibly zoonotic in S.E Asia with humans living in proximity to macaques



Type	P. knowlesi
Incubation period	9-12 days
S&S severity	Mild to moderate
Initial fever pattern	Regular every 24 hours
Cycling pattern	24 - 27 hours
Duration of untreated infection	Unknown
Age/size of RBC	All RBCs ; varies
Anemia	Moderate to severe
CNS involvement	Possible
Nephrotic Syndrome	Possible


P. vivax
P. malariae
P. ovale
P. falciparum
1
2
3
4
5
6
7
Lg RBC
Lg RBC
S. BELKIN

Case Question 1

An 7 year-old female presents to the ED with chills and fever of 3 days duration along with diarrhea, nausea, and vomiting. She had been visiting relatives in Ghana, West Africa, for 3 weeks and had a history of mosquito bites during her visit.

Since malaria is part of the differential diagnosis, a blood sample is collected, blood films are prepared and stained with Wright-Giemsa.

Small double ring forms are observed in some RBCs and double chromatin dots are observed in some rings. Developing trophozoite forms are not observed. RBCs are not enlarged. Which of the following malarial parasites is most likely causing her infection?

a. *Plasmodium vivax*
b. *Plasmodium ovale*
c. *Plasmodium falciparum*
d. *Plasmodium malariae*

Review Question 2

A fatigued but alert 38 year-old female was seen in the outpatient clinic because of frequent headaches, occasional nausea and vomiting, and periodic fever. She recently returned from rural Thailand, where she had spent 6 months as a teacher. A blood smear is ordered for the suspected diagnosis of malaria.

Which of the following choices would fit *Plasmodium falciparum* based on the microscopic examination of the blood smear.?

a. Numerous large ovoid trophs in some of the red cells
b. Oval shaped, fimbriated red cells with dark-staining trophs
c. Dividing parasites (schizonts) in red cells with 8-12 merozoites
d. Double ring forms found in some of the red cells

Review Question 3

The seriousness of *P. falciparum* infection compared to the other three forms of malaria is due to which one of the following?

a. Destruction of lymphocytes compromises the immune response against malaria.
b. Extensive damage to the liver can occur during the pre-erythrocytic phase of the parasite cycle.
c. Infected RBCs with sticky surfaces adhere to the interior lining of blood vessels and block blood flow.
d. Bloodstream parasites reinvade the liver and induce a more severe disease state.

Babesia species

- **Arthropod** vector disease of parasitic infection
  - Transmitted by Ixodes tick
    - Definitive hosts are ticks
  - Ticks can live on wild, domestic, and farm animals
    - White-footed mouse is well known intermediate host
  - Humans are intermediate host
- **Worldwide** prevalence
- **In US** cases predominate in the **northeast**
  - Babesia microti – **most common** in US
  - Babesia divergens – most common in Europe
  - B. bovis
  - B. equis

Transmission and Symptoms of Babesiosis

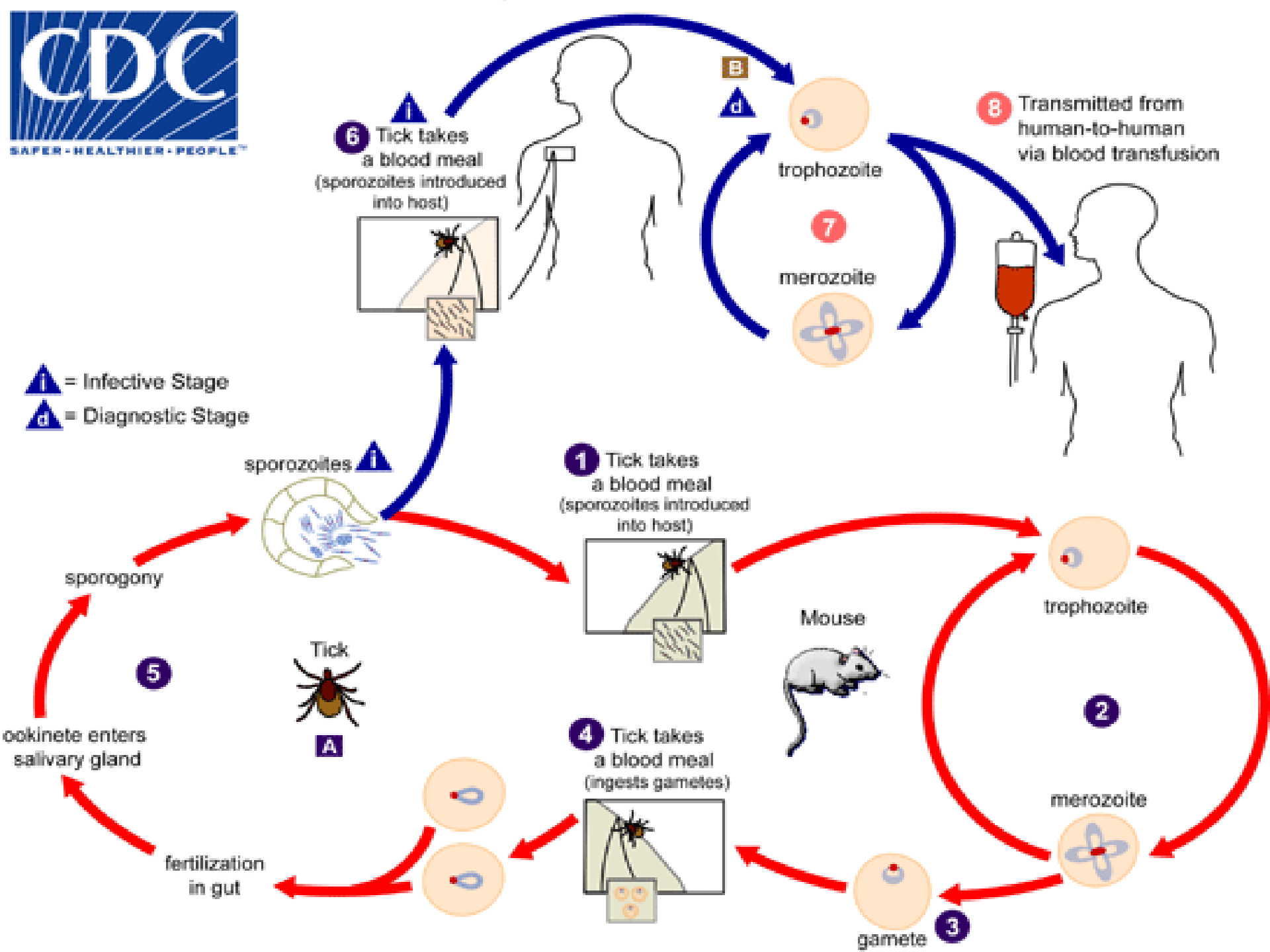
- **Female Ixodes tick bites human**
  - transfers Babesia sporozoites from salivary gland into human blood
- Malaria-like symptoms occur in 1-4 weeks
- Majority of cases are subclinical and self-limiting
- Severe cases occur in asplenic, immunocompromised
- Treatment: quinine + clindamycin 7 days *or* azithromycin + atovaquone

Symptoms of Babesiosis

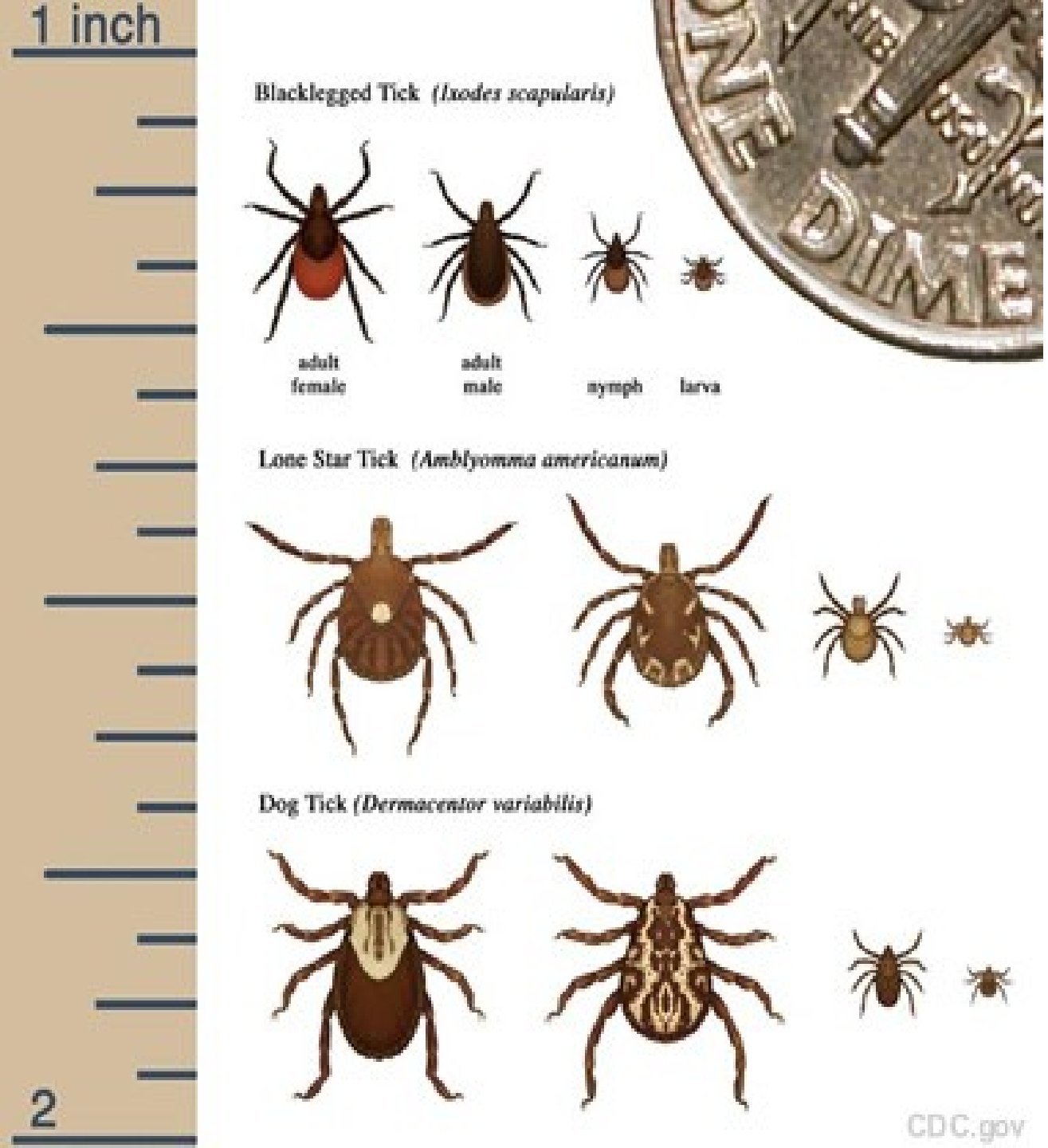
- Malaria-like symptoms follow bite of infected tick in about 1-4 weeks
- **Majority of cases are subclinical and self-limiting**
- **More severe cases may present as hemolytic anemia**
- Similar to malaria: fever and hemolytic anemia, malaise, anorexia, fatigue
- Followed by: chills, fever, sweats, myalgia, headache, nausea, vomiting, mild hepatosplenomegaly
- **No periodic fevers: differ from malaria**

Life Cycle of Babesiosis

- Ixodes tick bites human and sporozoites enter blood
- **Sporozoites enter RBCs directly** and asexually reproduce
  - **Only** ring trophozoites seen in blood film
- Humans are **dead end hosts**; little or no transmission back to ticks through blood meals have been documented
- Ticks feed on white-footed mouse (intermediate host) and acquire gametocytes
- Sporogony occurs in ticks (definitive host)
- Adult ticks also feed on deer (indirectly enhance tick population)


CDC
SAFER·HEALTHIER·PEOPLE™
i = Infective Stage
d = Diagnostic Stage
sporozoites
i
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
1 Tick takes a blood meal (sporozoites introduced into host)
trophozoite
Mouse
2
merozoite
3
gamete
4 Tick takes a blood meal (ingests gametes)
fertilization in gut
ookinete enters salivary gland
5
Tick
A
sporogony

Ticks and Disease Transmission



Ixodes

Borrelia (Lyme), *Babesia*, *Anaplasma* (HGA, human granulocytic anaplasmosis)

Amblyomma & Dermacentor

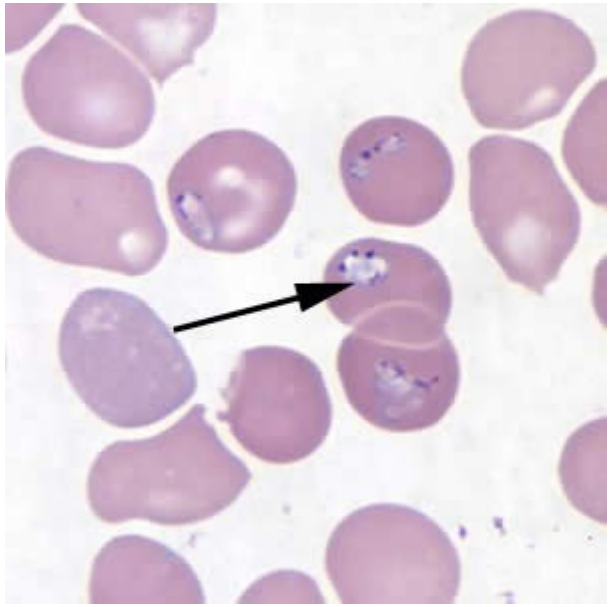
Erhlichia (HME, human monocytic erhlichiosis)

Dermacentor

Rickettsia rickettsii (RMSF, Rocky Mtn Spotted fever)

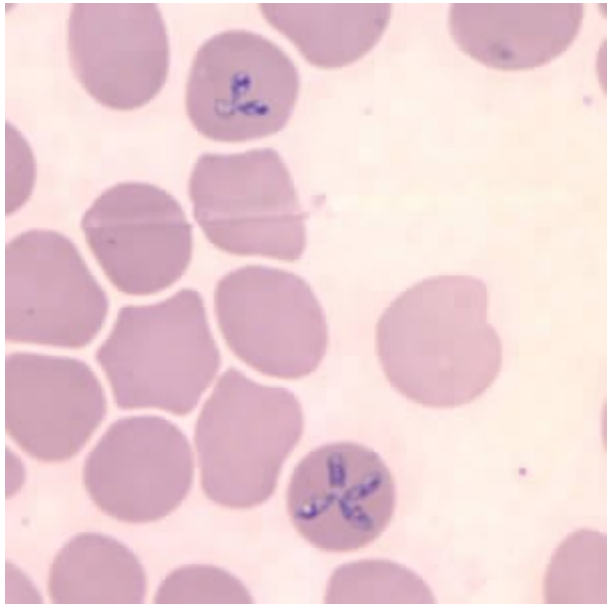
Lab Diagnosis of Babesia spp

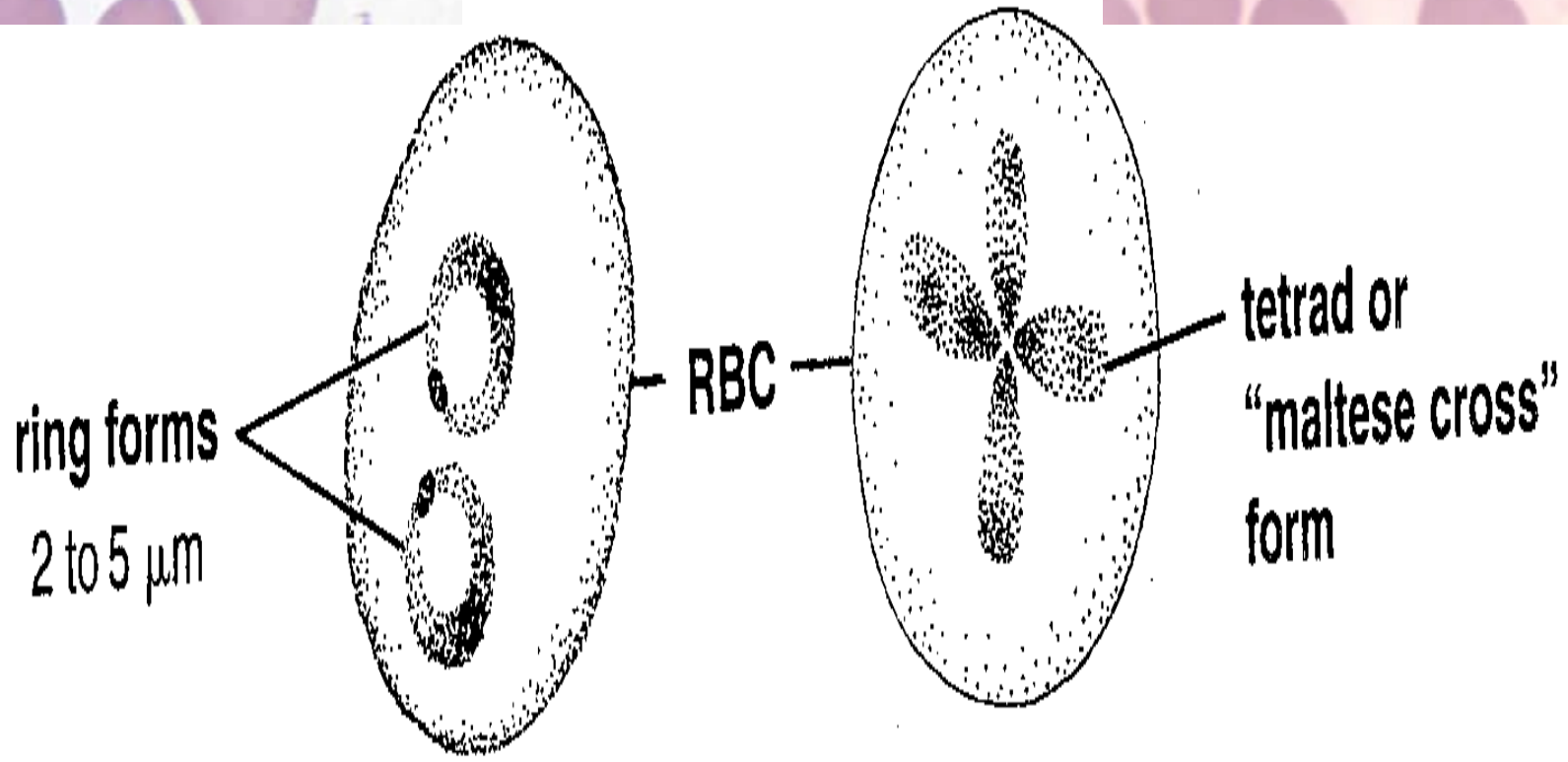
- Methods of Diagnosis
  - examination of multiple thin and thick blood films
  - **trophozoites** are pear shaped usually in pairs or tetrads in the **Maltese cross** formation
  - extracellular trophozoites may be observed
  - **4 to 5 ring forms per RBC may be seen**
  - often confused w/ P. falciparum
  - RBC's **not** enlarged
  - diagnosis is a problem due to the size (2to 4 µm) and low level parasitemia
- Molecular PCR are available in some laboratories



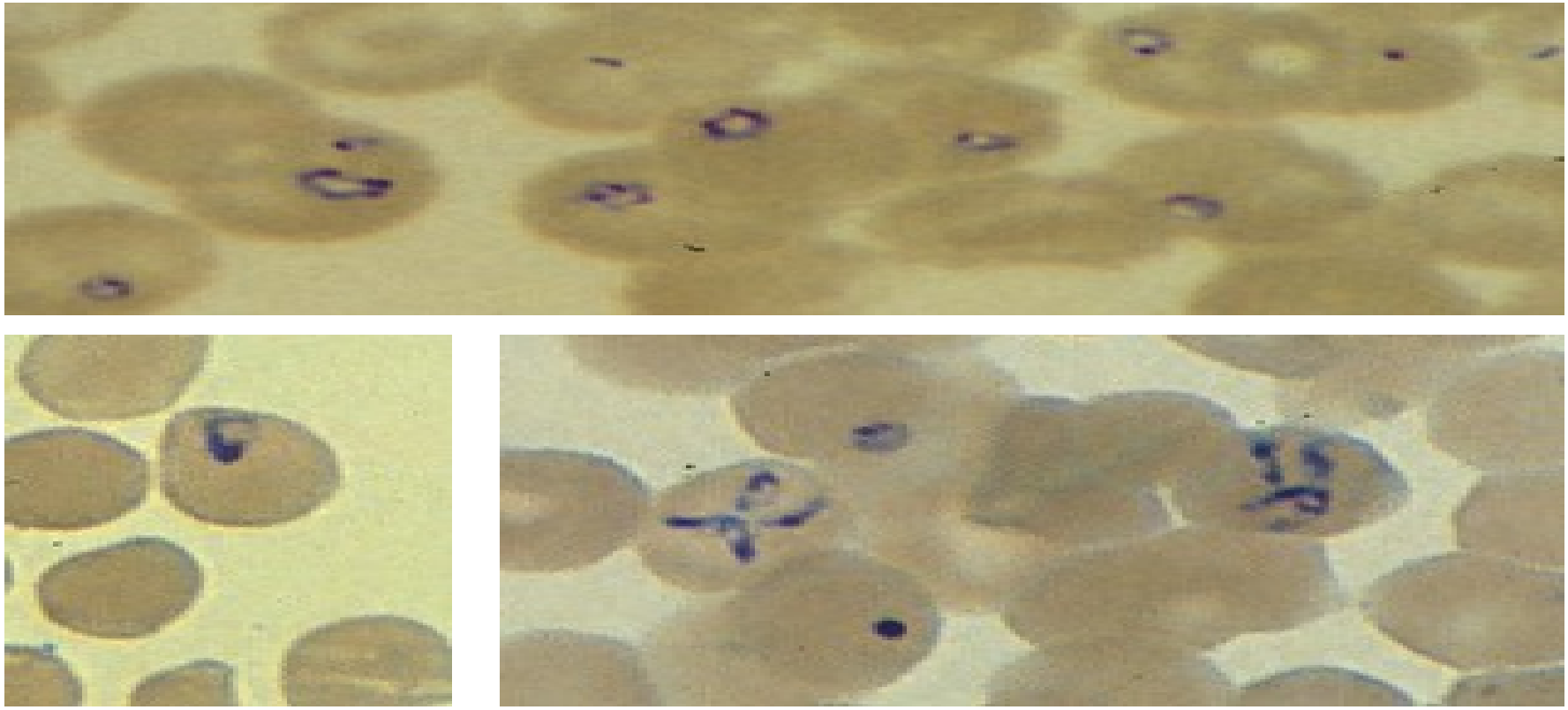
Babesia microti

Trophozoites



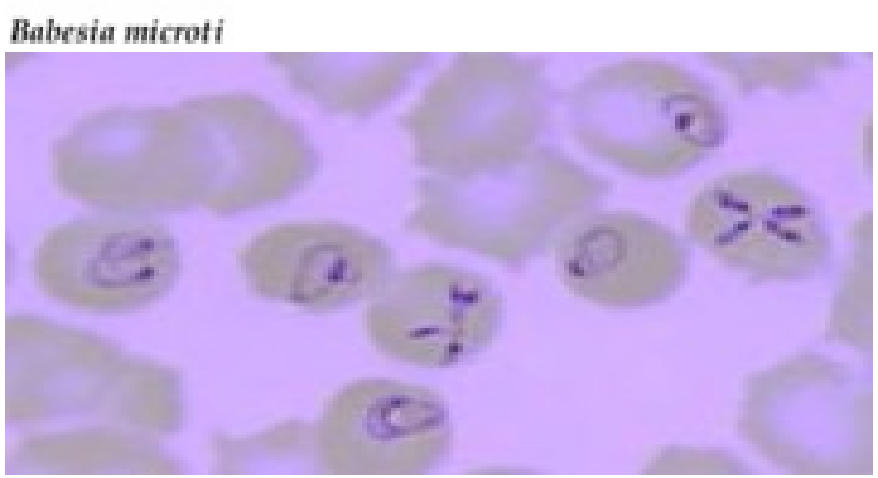


Babesia microti

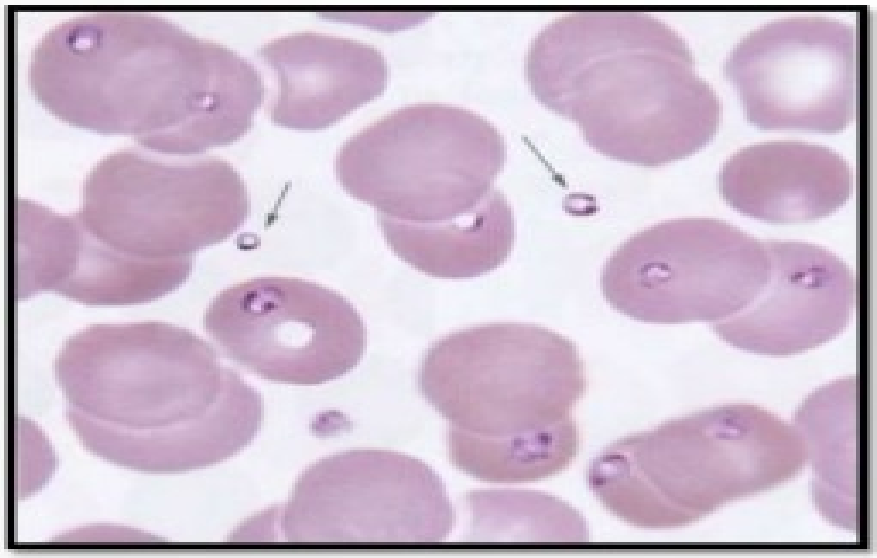


- *B. microti* in infected RBC, tiny rings that could be mistaken for *P. falciparum*
- Note the tetrad formation (Maltese cross)

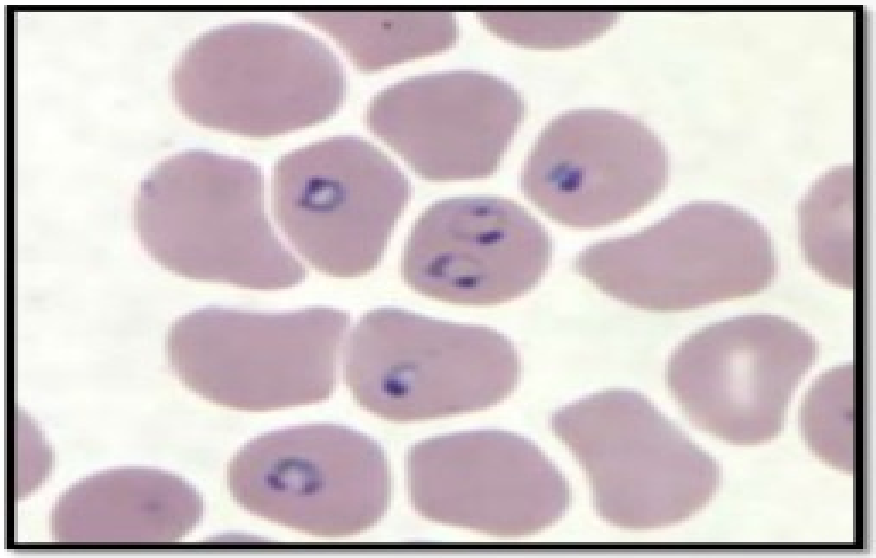
Babesia vs Malaria



- Babesia trophozoites rings are smaller [typically 1 – 5 um]
- Multiple rings per RBC are much more common
- Babesia rings may form tetrads, or a Maltese cross



Babesia microti



Plasmodium falciparum

Homework

- Blackwater fever is caused by _____ and the diagnostic features are _____.

- The most prevalent malaria parasite is _____ and the diagnostic features are _____.

- Relapses may be found in *P.* _____ infections and recrudescences may be found in *P.* _____ infections.

- “Band” form trophozoites are noted in *P.* _____ infections.

- Reticulocyte infections are seen in *P.* _____ infections.
- For each of the following, list two diagnostic morphologic features seen in peripheral blood films.
- *P. malariae:*
- *P. vivax:*
- *P. ovale:*
- *P. falciparum:*
- *P. knowlesi* is difficult to identify because parasitic forms seen at blood smear may resemble those seen in _____ and_____.
- Why are *P. falciparum* infections more severe than infections caused by the other *Plasmodium* spp.?

- The definitive host for *Babesia* spp. is a(n)

- The diagnostic forms seen in RBCs appear and may be confused with