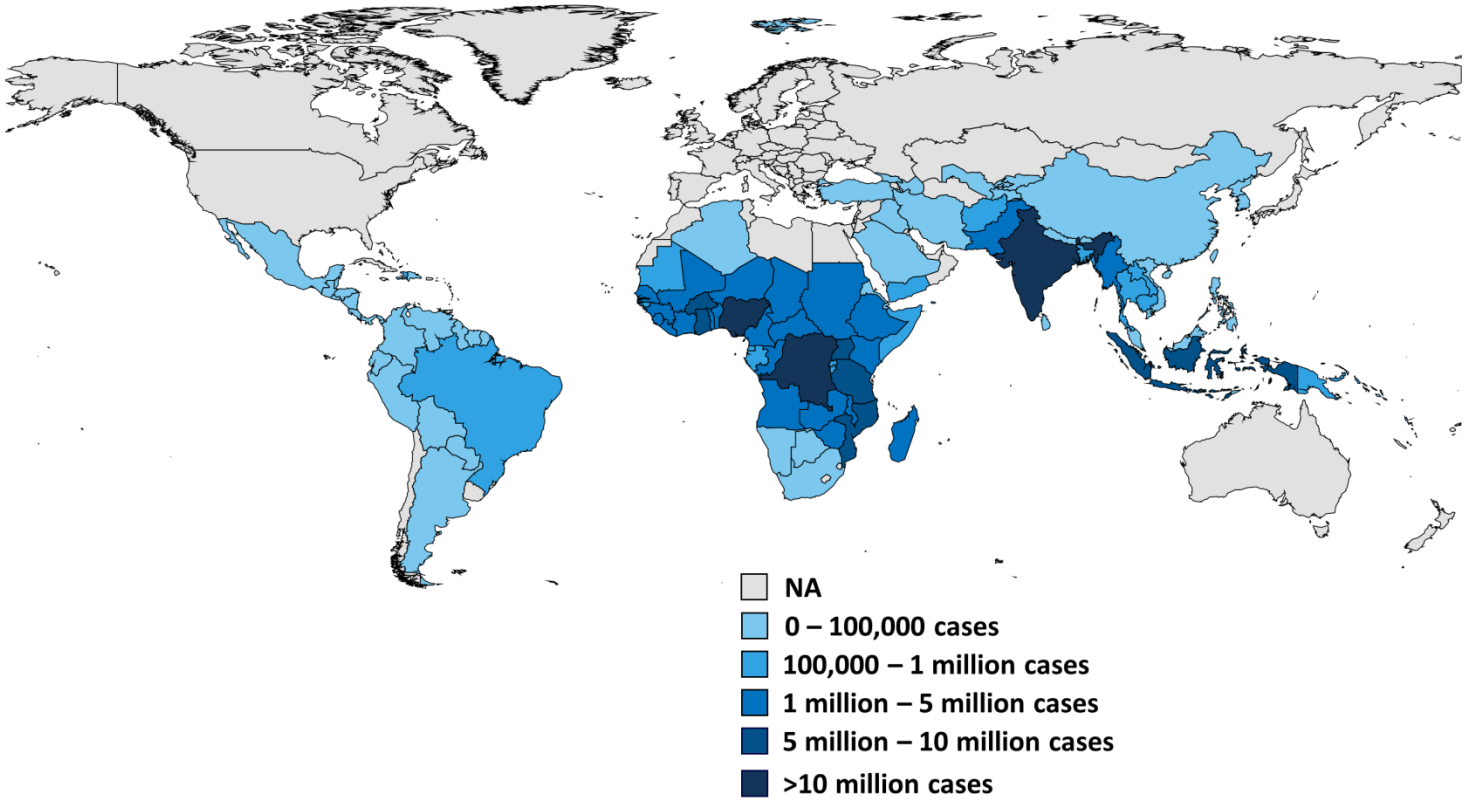
Malaria 1

Estimated Malaria Cases, 2012



SOURCE: Kaiser Family Foundation, http://kff.org/globaldata/, based on WHO, World Malaria Report 2013; December 2013.

THE HENRY J. KAISER FAMILY FOUNDATION

World Malaria Report 2013 from WHO

Impact on human health

- Malaria is endemic in 106 countries
- Caused about 198 million cases around the world in 2013
- Estimated 584 000 deaths in 2013, mostly in children <5 years(78%), especially in Africa
- 90% of deaths are estimated to have occurred in the WHO African Region
- Sri Lanka has had endemic malaria for many centuries, with occasional severe epidemics

Outline of classes on malaria

- Three lectures
  - Life cycles, morphology, transmission
  - Clinical features, pathogenesis and pathology, laboratory diagnosis Treatment, drug resistance
  - Epidemiology and immunology
- Three lab classes
  - Morphology of the 4 species
  - Examining stained thin blood films for Pv and Pf
  - Staining thin blood films with Leishman
- Tutorial

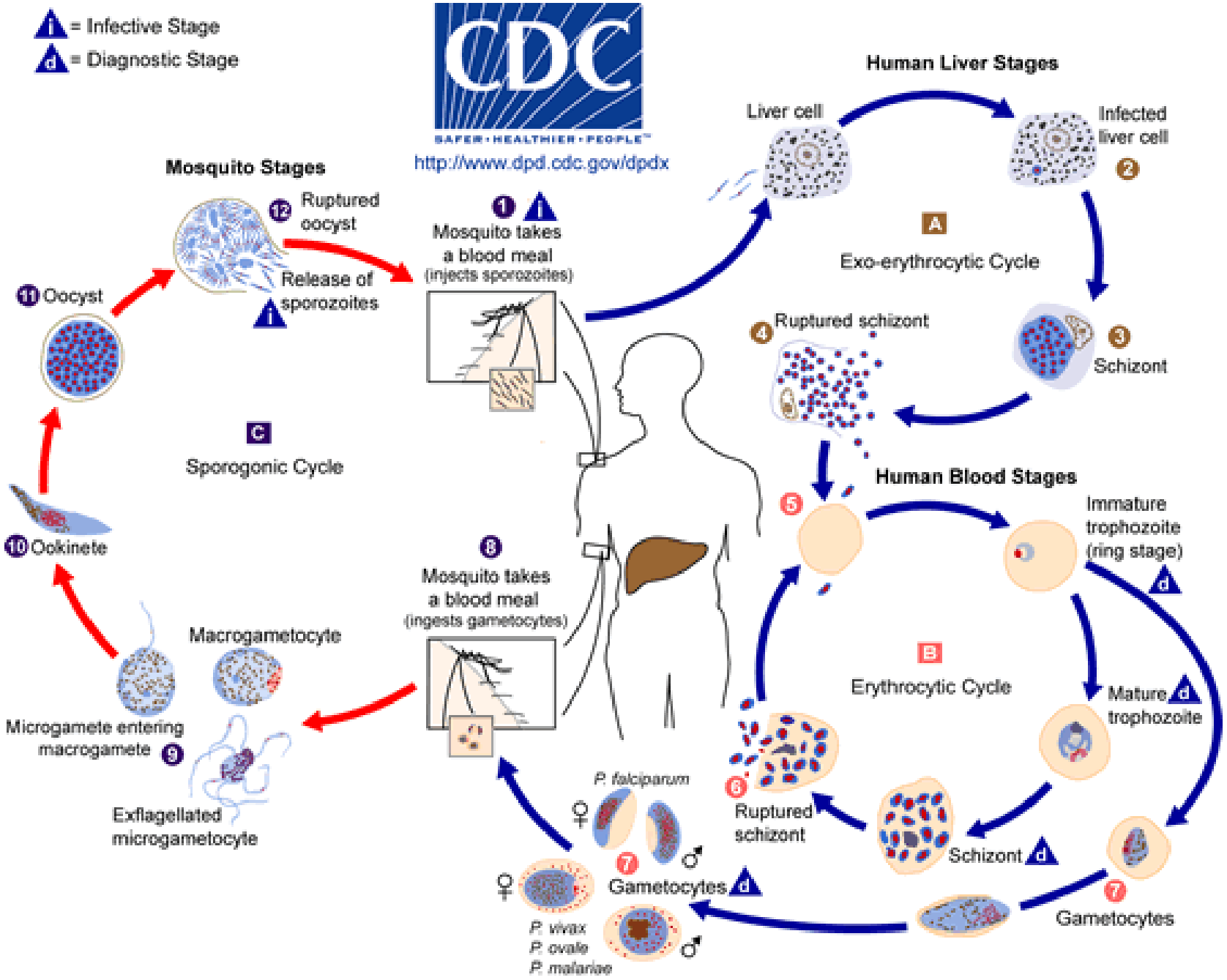
Malaria parasites that affect humans:

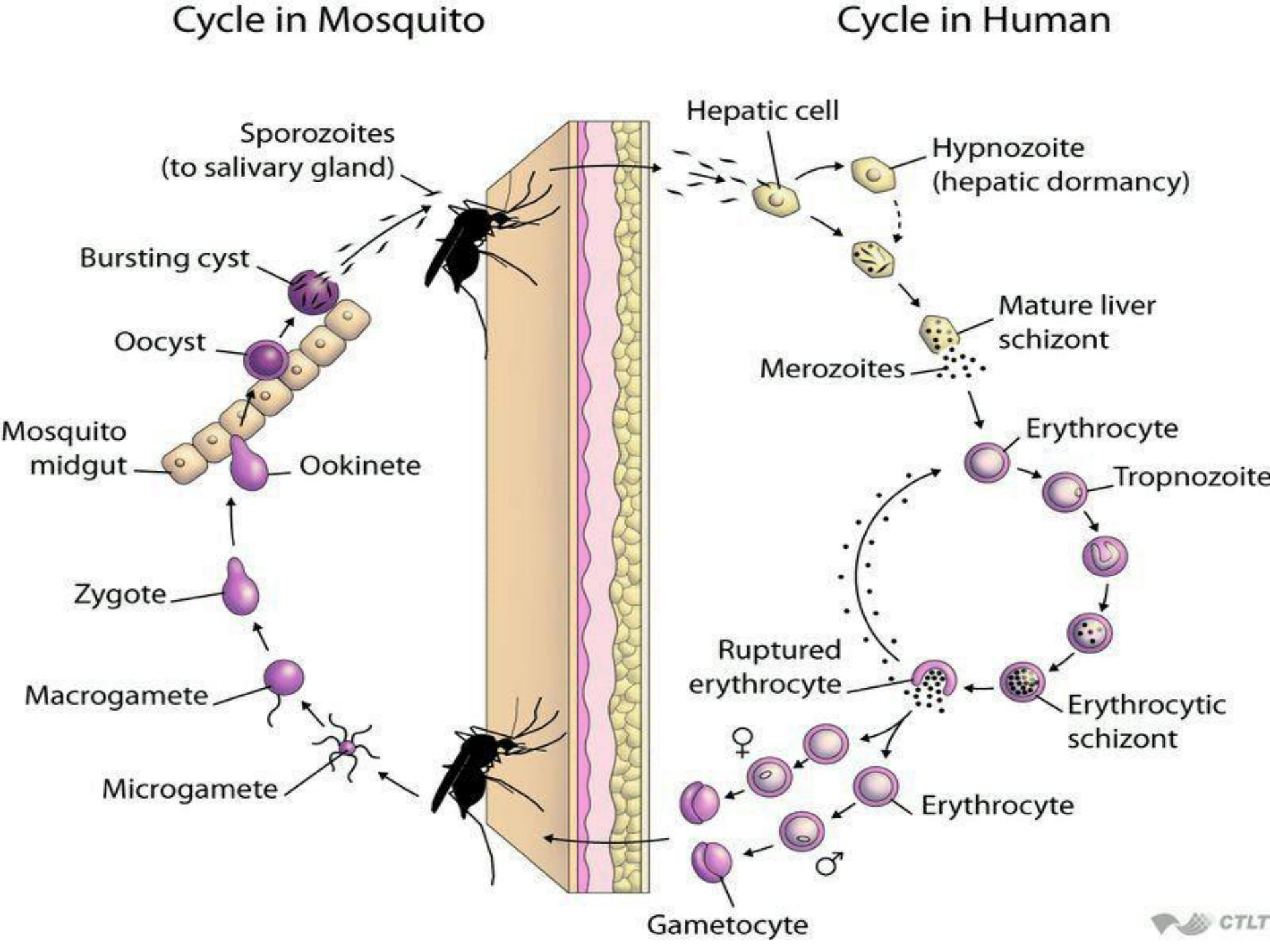
- *Plasmodium vivax*
- *Plasmodium falciparum*
- *Plasmodium malariae*
- *Plasmodium ovale*
- *Plasmodium knowlesi*

- Naturally acquired human infection with *P. knowlesi* was first described in Malaysian Borneo in 1965

Life cycle

- Two-host life cycle:
  - humans (asexual multiplication)
  - Anopheline mosquitoes (sexual multiplication)
- Rarely infects other vertebrate hosts


= Infective Stage
= Diagnostic Stage
CDC
SAFER · HEALTHIER · PEOPLE™
http://www.dpd.cdc.gov/dpdx
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
B
Erythrocytic Cycle
6
Ruptured schizont
Schizont
d
7
Gametocytes
P. falciparum
Gametocytes
d
P. vivax
P. ovale
P. malariae
1
Mosquito takes a blood meal (injects sporozoites)
Mosquito Stages
12
Ruptured oocyst
Release of sporozoites
11
Oocyst
C
Sporogonic Cycle
10
Ookinete
8
Mosquito takes a blood meal (ingests gametocytes)
Macrogametocyte
Microgamete entering macrogamete
9
Exflagellated microgametocyte

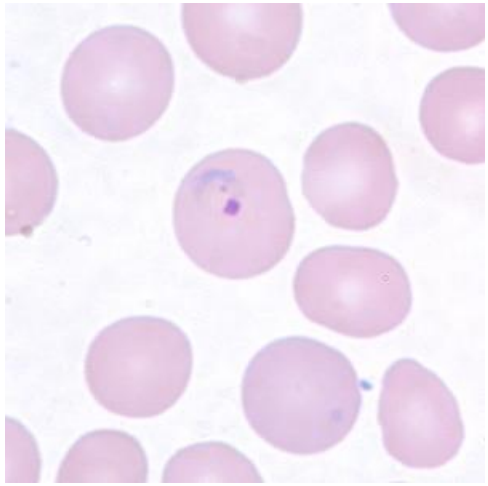

Cycle in Mosquito
Cycle in Human
Sporozoites
(to salivary gland)
Hepatic cell
Hypnozoite
(hepatic dormancy)
Bursting cyst
Oocyst
Mature liver
schizont
Merozoites
Mosquito
midgut
Ookinete
Erythrocyte
Tropnozoite
Zygote
Ruptured
erythrocyte
Erythrocytic
schizont
Macrogamete
Microgamete
Erythrocyte
Gametocyte

P. vivax life cycle

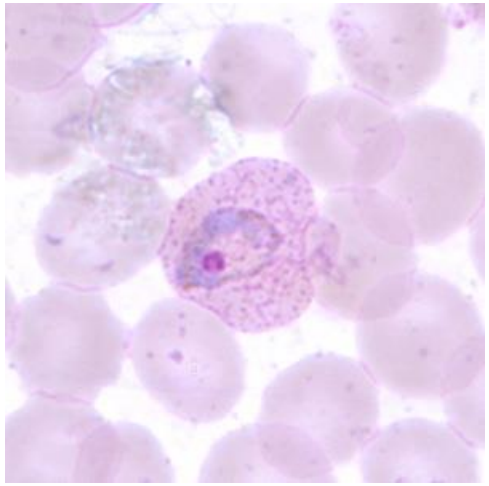
- Exo-erythrocytic schizogony takes 7 – 8 d
- 10 – 12,000 merozoites / hepatic schizont
- Hypnozoite formation
- Erythrocytic schizogony takes 48 h; 12 – 24 merozoites / schizont
- Gametocyte formation starts after 2-3 cycles of schizogony
- Sporogonic cycle takes 10 – 12 days at 30°C; longer in colder climates

Plasmodium vivax morphology

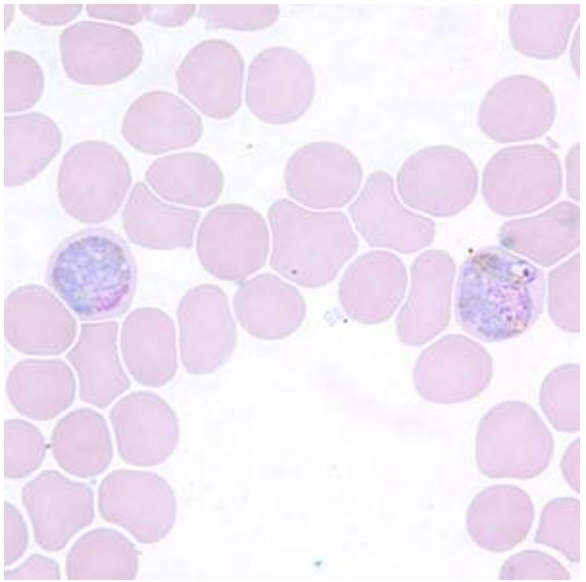
Ring trophozoite

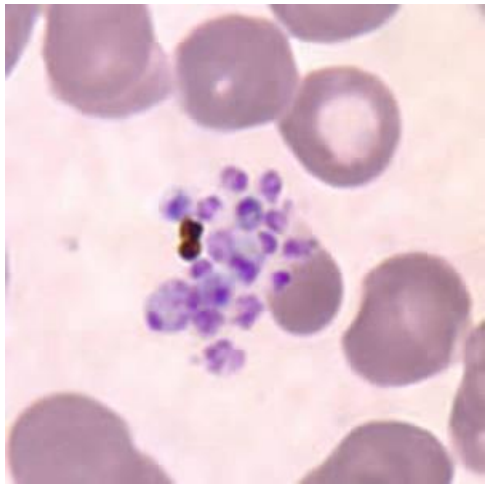


Amoeboid trophozoite

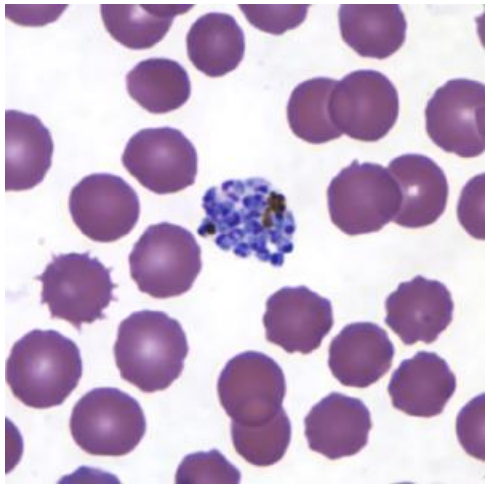


Gametocytes





Ruptured schizont



Mature schizont

P. falciparum life cycle

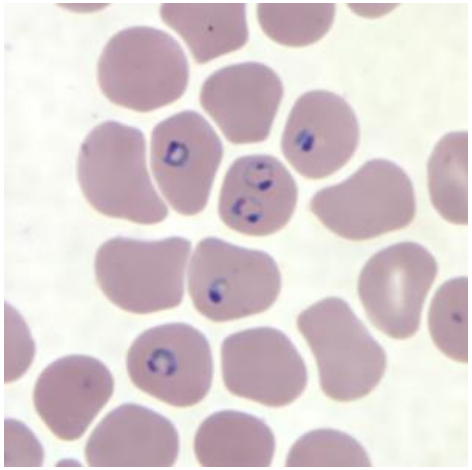
Main differences from *P. vivax* life cycle

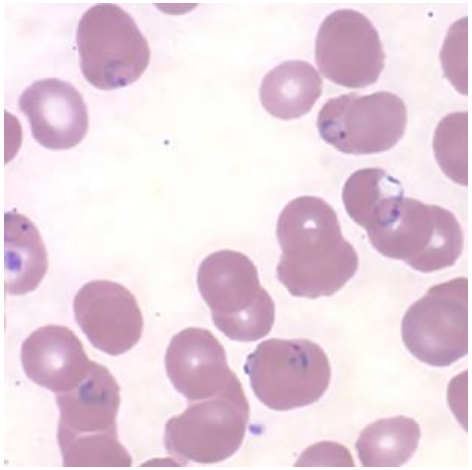
- Shorter exo-erythrocytic cycle (5-6 days)
- More merozoites per hepatic schizont (up to 30,000)
- No hypnozoite formation
- Merozoites can invade red cells of any age
- Multiple infection of red cells common
- Sequestration of erythrocytic schizonts

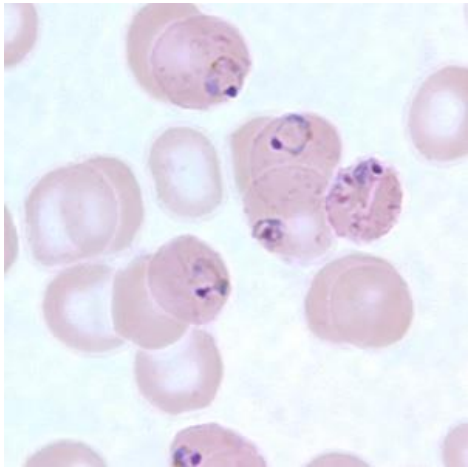
- Shorter sporogonic cycle

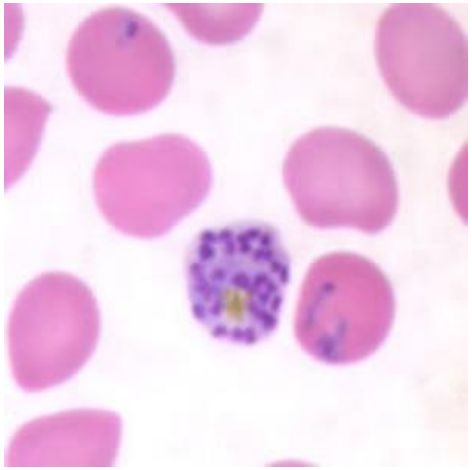
P. falciparum morphology

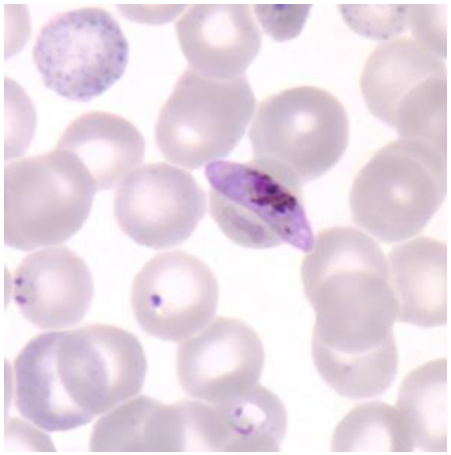
Ring trophozoites











Gametocyte

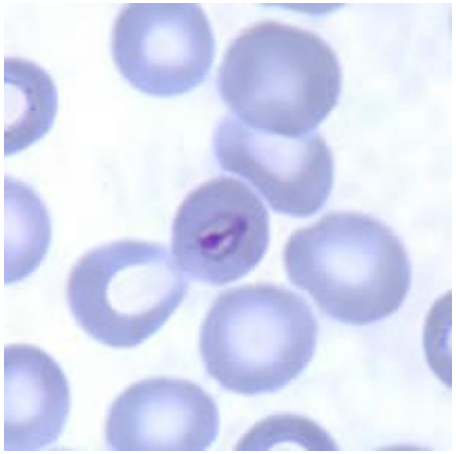
Maurer's clefts on red cells

schizont

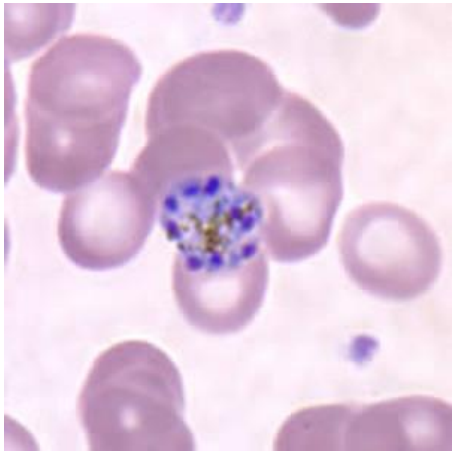
P. malariae life cycle and morphology

Differences from *P. vivax*

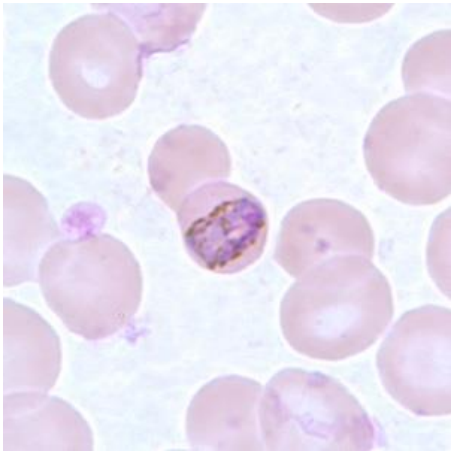
- Longer exo-erythrocytic cycle (15 days)
- Erythrocytic cycle takes longer (72 h)



Ring trophozoite – bird's eye form



Mature schizont – daisy head appearance

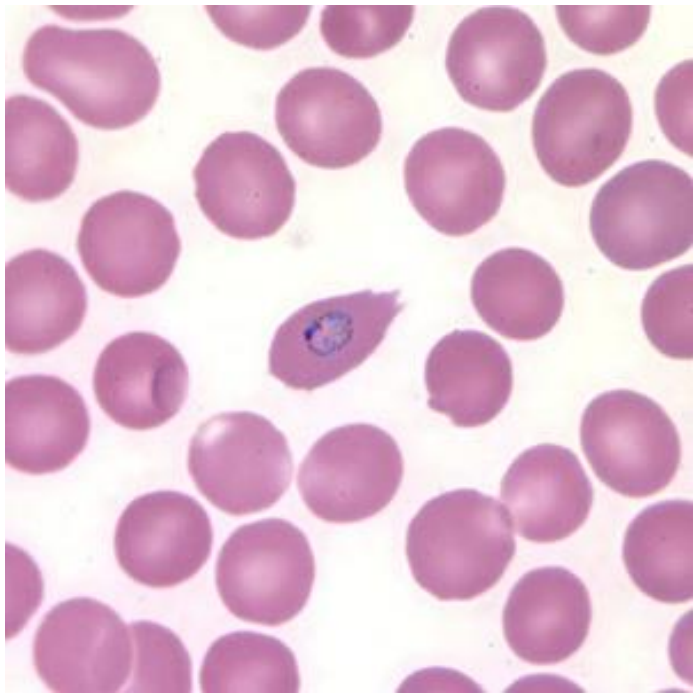


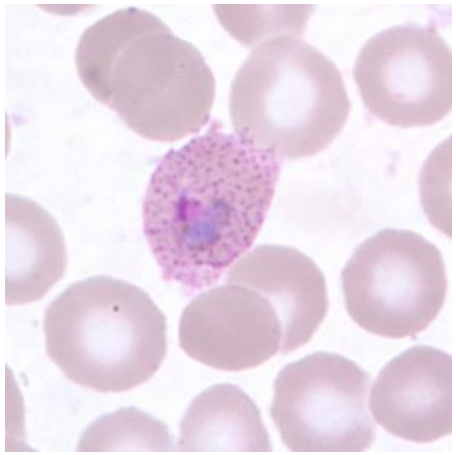
Band form

P. ovale life cycle and morphology

Life cycle virtually identical to *P. vivax*

Almost all infections seen in West Africa



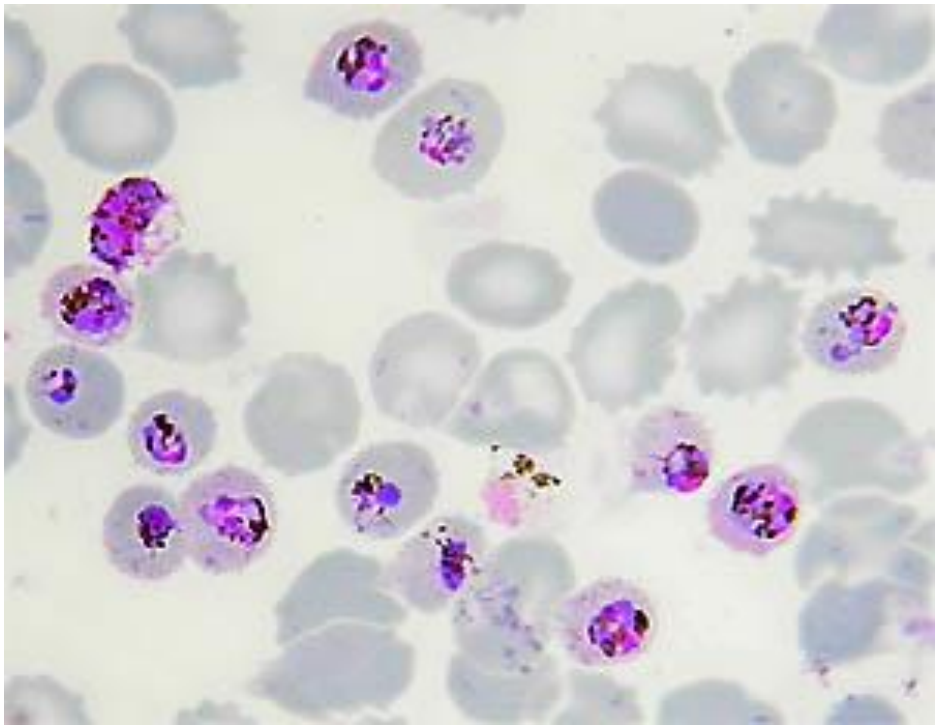


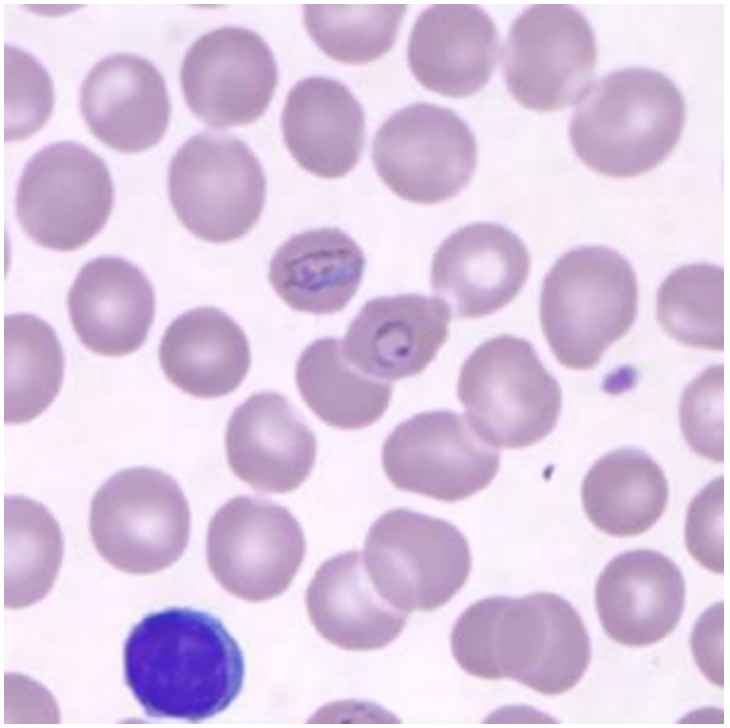
Trophoites with characteristic fimbriated edge

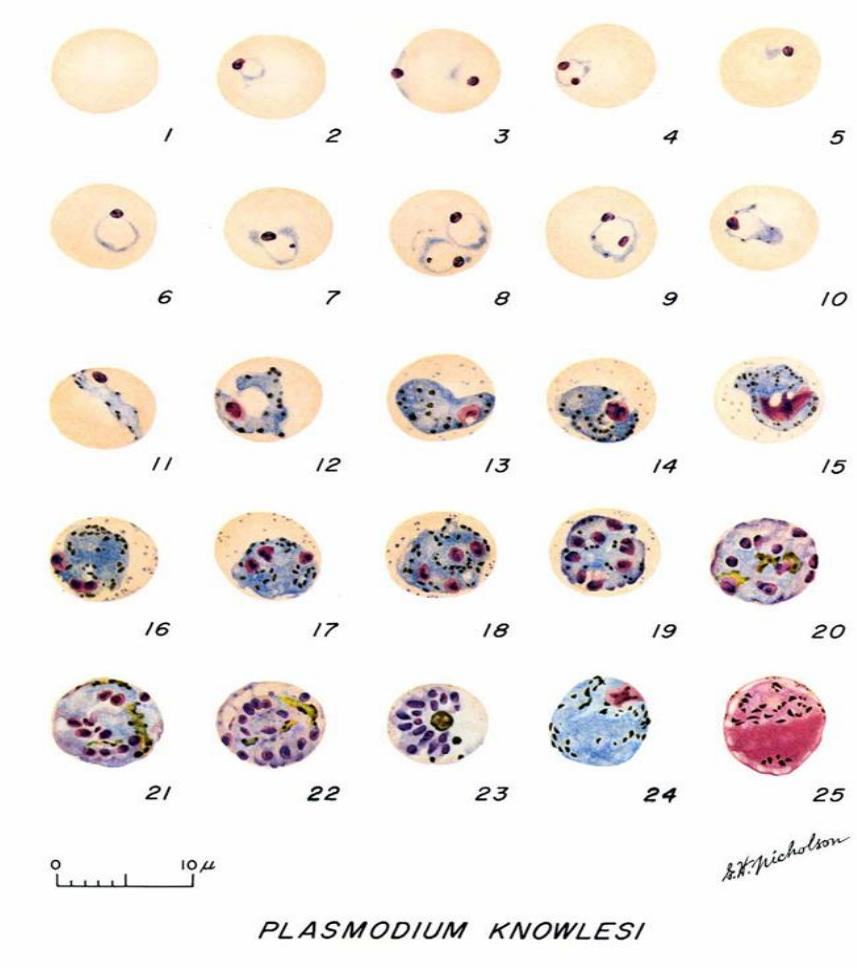
P. knowlesi

- *P. knowlesi* fifth and emerging human malaria parasite
- prevalent in South East Asia and can cause potentially life threatening malaria
- *P. knowlesi* is a zoonotic malaria parasite that is transmitted by mosquitoes of the *Anopheles leucosphyrus* group that feed on humans and monkeys
- Many *P. knowlesi* infections have been misdiagnosed by microscopy as *P. malariae*,

P. knowlesi






1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17
18
19
20
21
22
23
24
25
0
10 μ
S.K. Nicholson
PLASMODIUM KNOWLESI

Transmission of malaria

- Vast majority of cases are vector-borne - *Anopheles* mosquitoes
- May follow transfusion of unscreened, infected blood
- Congenital infections may also occur, especially if non-immune mother gets malaria just before delivery

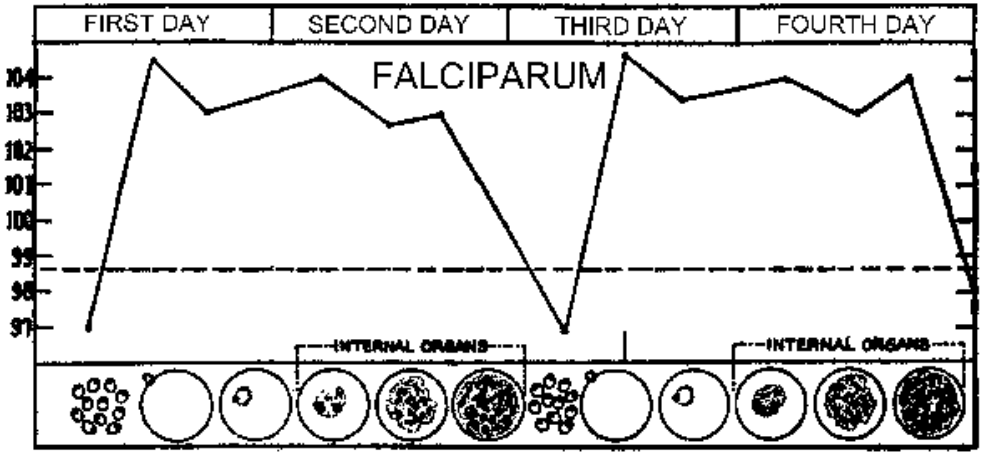
Clinical features and pathogenesis

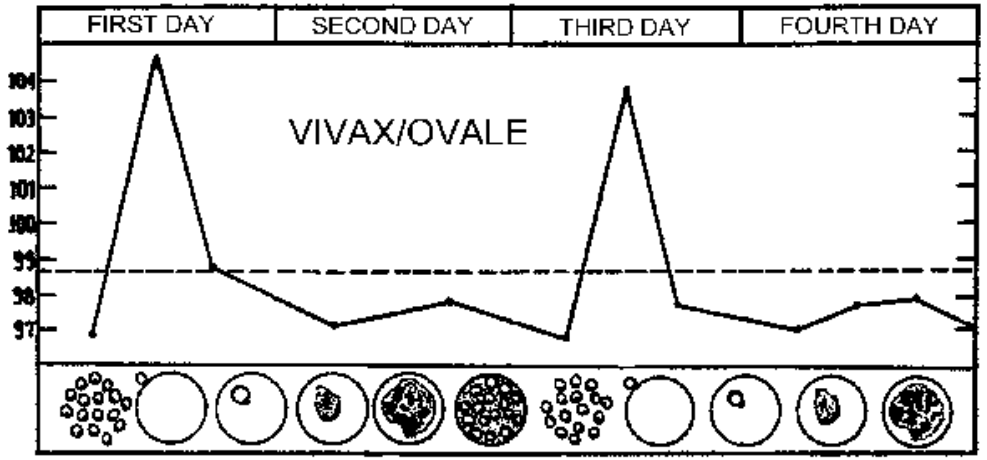
- Classical symptom is paroxysmal fever: high, intermittent fever with chills and rigors

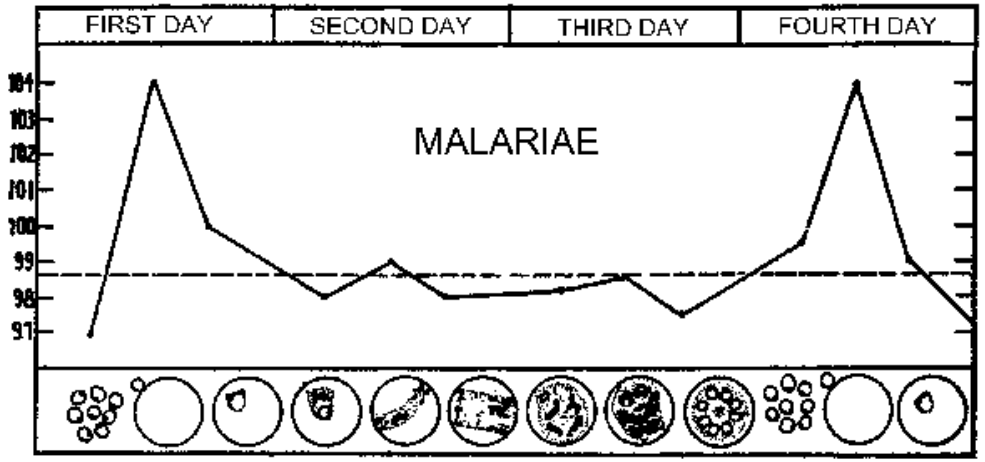
- Fever spike coincides with rupture of erythrocytic schizonts
  - Erythrocytic cycle synchronized so all parasites are at same stage of development at a given time
  - Rupture of infected red cells - release of malaria pigment and cellular debris leads to release of cytokines, esp TNF
  - TNF acts on thermoregulatory centre in hypothalamus

- Incubation period: minimum 7 days after exposure
- Pre-patent period is shorter

Typical fever charts in patients with malaria







Typical clinical features

- Prodromal features may occur before first bout of fever
- Febrile paroxysm lasts about 8 – 12 hours and has 3 stages
  - Cold stage
  - Hot stage
  - Sweating stage
- Patient may have mild anaemia and jaundice; also mild, tender, hepato-splenomegaly

Natural history of malaria

- Applicable to most cases of *P. falciparum* and all cases due to other species
- Patient has recurrent bouts of fever for several weeks, then recovers spontaneously
- Fever may recur after several months (even years) due to
  - Relapse - arising from hypnozoites, in *P. vivax* and *P. ovale*
  - Recrudescence – arising from surviving erythrocytic forms in *P. falciparum* and *P. malariae*

Severe and complicated malaria

- Occurs in a small but significant proportion of *P. falciparum* and knowlesi cases
- Does not occur with vivax , ovale and malariae
- Usually fatal, if left untreated
- Why only in falciparum and knowlesi malaria?
  - Parasitaemia is higher
  - Sequestration of maturing erythrocytic forms
- Pathogenesis involves
  - Cytoadherence, resulting in mechanical obstruction to blood flow and tissue hypoxia
  - Local release of cytokines and nitric oxide

Patients at risk of severe and complicated malaria

- In areas with high levels of transmission:
  - Young children
  - Pregnant women
  - Recent returnees
  - (other adults have developed protective immunity)
- In areas with lower levels of transmission
  - All age groups, but especially children
- In any region with malaria
  - Non-immune travellers
  - Migrant workers

Manifestations of severe and complicated malaria

- Cerebral malaria
- Severe anaemia
- Hyperpyrexia
- Hypoglycaemia
- Pulmonary oedema
- Fluid, electrolyte and acid-base disturbances
- Renal failure
- Hepatic dysfunction
- Circulatory collapse (algid malaria)
- Blackwater fever (massive intravascular haemolysis)

Cerebral malaria

- Commonest manifestation of SCM
- Any patient with malaria, who shows impairment of consciousness, should be considered as having cerebral malaria
- Onset usually after some days of fever; may be as little as 2 days in children
- May have generalized convulsions
- Show decerebrate rigidity in late stages

Cerebral malaria ctd

Decerebrate rigidity in a child with cerebral malaria

- Pathogenesis related to cerebral hypoxia and disturbances in neurotransmission
- May be aggravated by hypoglycaemia
- Many patients recover fully if treated, but children may have residual neurological deficits

Severe anaemia

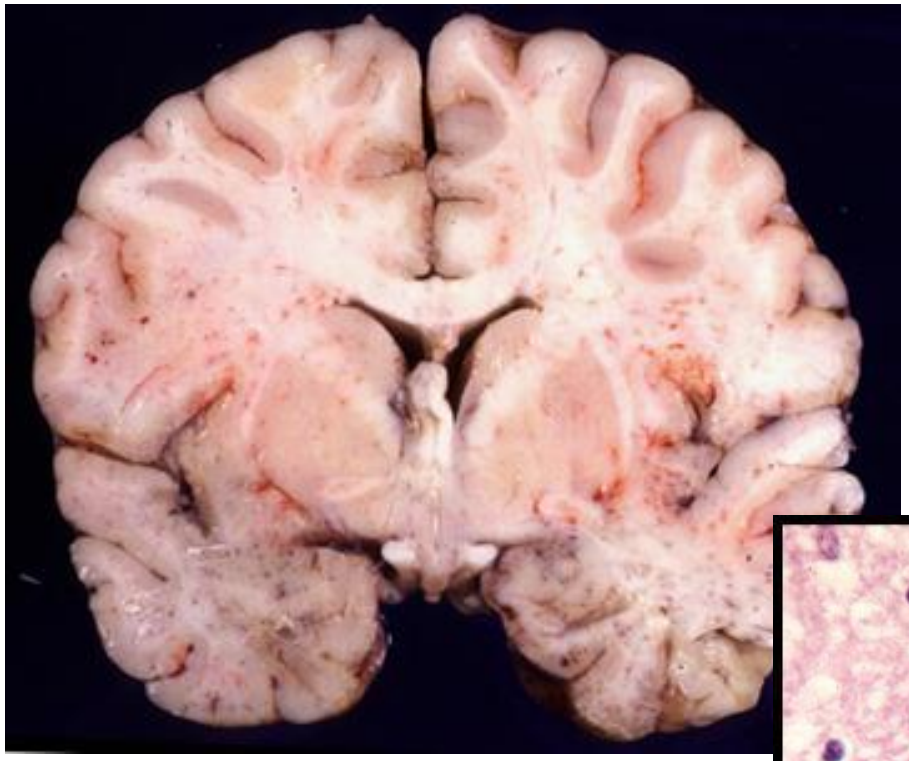
- Defined as Hb < 5 g / dl of blood
- Particularly common in children and pregnant women
- Several contributory factors:
  - Destruction of parasitized red cells (spleen)
  - Immune mediated destruction of un-infected red cells
  - Reduced production of red cells (dyserythropoiesis)

Falciparum malaria in pregnancy

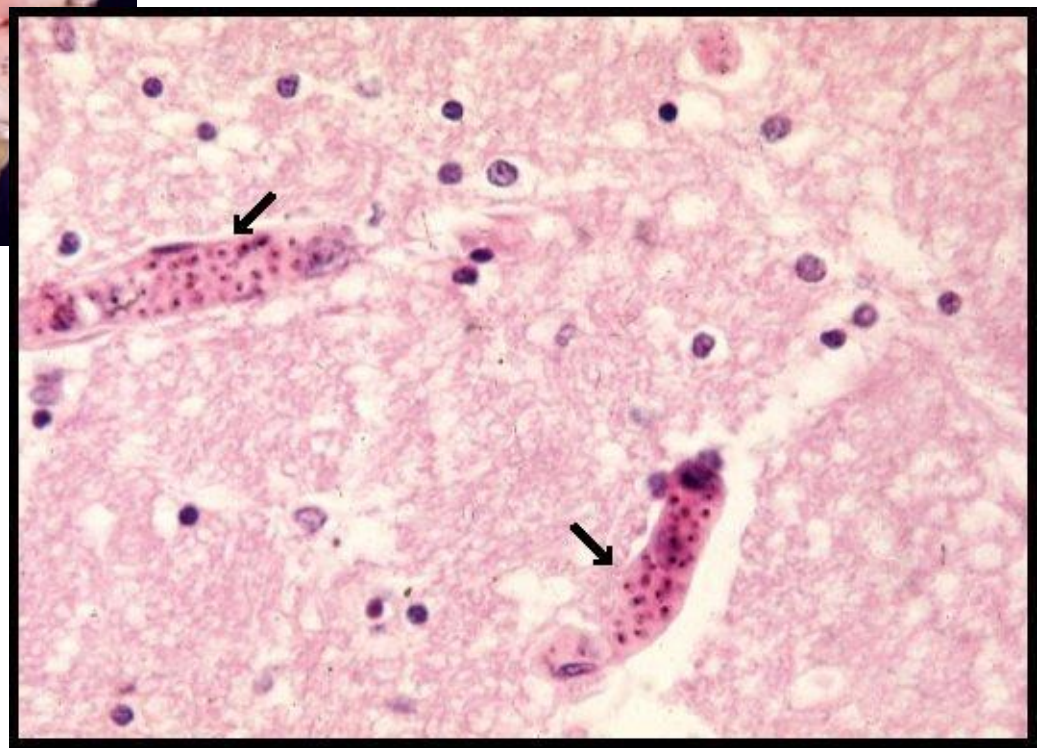
- Women in 1st or 2nd pregnancy particularly at risk of complications
- Parasites multiply in placenta
- Results in low birth weight and increased neonatal mortality; premature labour
- Causes several complications
  - Severe anaemia
  - Hypoglycaemia
  - Acute pulmonary oedema

Pathological changes in malaria

- In all cases:
  - Hyperplasia of lymphoreticular system, with active follicles in lymph nodes and spleen
  - Accumulation of malaria pigment in macrophages of liver and spleen
  - Marked vascular congestion of spleen
- In fatal cases
  - Spleen moderately enlarged, soft and red-grey
  - Liver also enlarged and grey; pigment in Kupffer cells
  - Brain is oedematous and markedly congested; petechial haemorrhages due to ruptured capillaries



Cross-section of eedematous brain with ring haemorrhages



H&E stained section showing congested capillaries with parasitized red cells and malaria pigment

Pathological changes ctd

- Kidney involvement may be
  - Acute renal failure in algid malaria and blackwater fever (acute tubular necrosis) in *P. falciparum*
  - Chronic – nephrotic syndrome in children with long-standing, untreated, *P. malariae* infections

Laboratory diagnosis of malaria

- **Parasitological diagnosis**: demonstration of parasites in red cells in stained blood films

- **Immunological diagnosis**: demonstration of malaria parasite antigens in blood or plasma

- **Molecular diagnosis**: demonstration of parasite DNA in blood

Parasitological diagnosis

- Microscopy is standard, classical diagnostic method
- Requires good microscope and skilled technician, but relatively inexpensive
- Thick blood films are more sensitive than thin films – enables confirmation of infection
- Species identification easier in thin films
- Smears stained with Giemsa or Leishman's stains
- Venous blood collected into EDTA may be used, but smears made directly from fingerprick blood are more sensitive

Immunological diagnosis

- Several Rapid Diagnostic Techniques (RDTs) now commercially available for diagnosis of malaria
- Based on detection of malaria antigens through an immune reaction that results in a colour change (immunochromatography)
- Antigens include
  - *P. falciparum* Histidine Rich Protein II
  - Parasite Lactate Dehydrogenase (LDH) isoenzymes
  - Parasite aldolase (isoenzymes)
- RDTs are easy to perform and don't require much technical expertise; but more expensive than microscopy
- Detection of antibodies NOT useful in diagnosis of infection

NEGATIVE

For all types of malaria spp.

POSITIVE

For Plasmodium sp.
(P. vivax / P. ovale / P. malariae)

POSITIVE

Plasmodium falciparum.
+/- (P. vivax / P. ovale / P. malariae)