Post-Graduate Degree Programme (CBCS)
in
ZOOLOGY
(M.Sc. Programme)

SEMESTER-III

Theory Paper-Minor Elective

Medical and Veterinary Parasitology
ZDSE(MN)T 305

Self-Learning Material

DIRECTORATE OF OPEN AND DISTANCE LEARNING
UNIVERSITY OF KALYANI
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Director's Message

Satisfying the varied needs of distance learners, overcoming the obstacle of distance and reaching the unreached students are the threefold functions catered by Open and Distance Learning (ODL) systems. The onus lies on writers, editors, production professionals and other personnel involved in the process to overcome the challenges inherent to curriculum design and production of relevant Self Learning Materials (SLMs). At the University of Kalyani a dedicated team under the able guidance of the Hon'ble Vice-Chancellor has invested its best efforts, professionally and in keeping with the demands of Post Graduate CBCS Programmes in Distance Mode to devise a self-sufficient curriculum for each course offered by the Directorate of Open and Distance Learning (DODL), University of Kalyani.

Development of printed SLMs for students admitted to the DODL within a limited time to cater to the academic requirements of the Course as per standards set by Distance Education Bureau of the University Grants Commission, New Delhi, India under Open and Distance Mode UGC Regulations, 2020 had been our endeavour. We are happy to have achieved our goal.

Utmost care and precision have been ensured in the development of the SLMs, making them useful to the learners, besides avoiding errors as far as practicable. Further suggestions from the stakeholders in this would be welcome.

During the production-process of the SLMs, the team continuously received positive stimulations and feedback from Professor (Dr.) Manas Kumar Sanyal, Hon'ble Vice-Chancellor, University of Kalyani, who kindly accorded directions, encouragements and suggestions, offered constructive criticism to develop it within proper requirements. We gracefully, acknowledge his inspiration and guidance.

Sincere gratitude is due to the respective chairpersons as well as each and every member of PGBOS (DODL), University of Kalyani. Heartfelt thanks are also due to the Course Writers-faculty members at the DODL, subject-experts serving at University Post Graduate departments and also to the authors and academicians whose academic contributions have enriched the SLMs. We humbly acknowledge their valuable academic contributions. I would especially like to convey gratitude to all other University dignitaries and personnel involved either at the conceptual or operational level of the DODL of University of Kalyani.

Their persistent and coordinated efforts have resulted in the compilation of comprehensive, learner-friendly, flexible texts that meet the curriculum requirements of the Post Graduate Programme through Distance Mode.

All the Self Learning Materials are self writing and collected from e-book, journals and websites.

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Theory (Discipline Specific Elective - Minor) - [ZDSE(MN)T-305]

Module	Unit	Content	Credit	Page No.
ZDSE(MN)T 305 Medical and Veterinary Parasitology	I	*Leishmania donovani* and Leishmaniasis	**2**	
	II	Structure, pathobiology prophylaxis and diagnosis of *Babesia* sp. *Anaplasma* sp. and *Theileria* sp.		
	III	i) *Trypanosoma cruzi* and Chagas disease ii) *Trichomonas foetus* in cattle.		
	IV	Outline structure and life cycle of *Plasmodium* spp.		
	V	*Trichuris trichiura*– biology, epidemiology, pathogenesis, diagnosis and treatment.		
	VI	Life cycle, biology, pathogenesis, epidemiology and control of *Loa loa*, *Dracunculus medinensis, Haemonchus contortus, Sarcoptes scabiei*.		
	Total counseling session 6hrs.			

UNIT I

Leishmania donovani and Leishmaniasis

Objective:

In this unit we will discuss about *Leishmania donovani* and Leishmaniasis.

Introduction:

Leishmania donovani is a haeomoflagellate parasite. In man it resides in Leishmanial form in lymphoid—macrophage (Reticuloendothelial) cells of the spleen, liver, bone- marrow, intestine and lymph glands.

Its vector host is Sand fly, *Phlebotomous sp.* It is found in Leptomonad form in the intestine. The disease caused by *Leishmania* is known as Leishmaniasis or Kala-azar. The disease is prevalent in the Eastern hemisphere specially in India, Southern U.S.S.R., Burma, Central China, Iraq etc.

Leishmania donovani

Morphology of *Leishmania donovani*:

- The parasite exists in two forms: **Amastigote** and **Promastigote**.

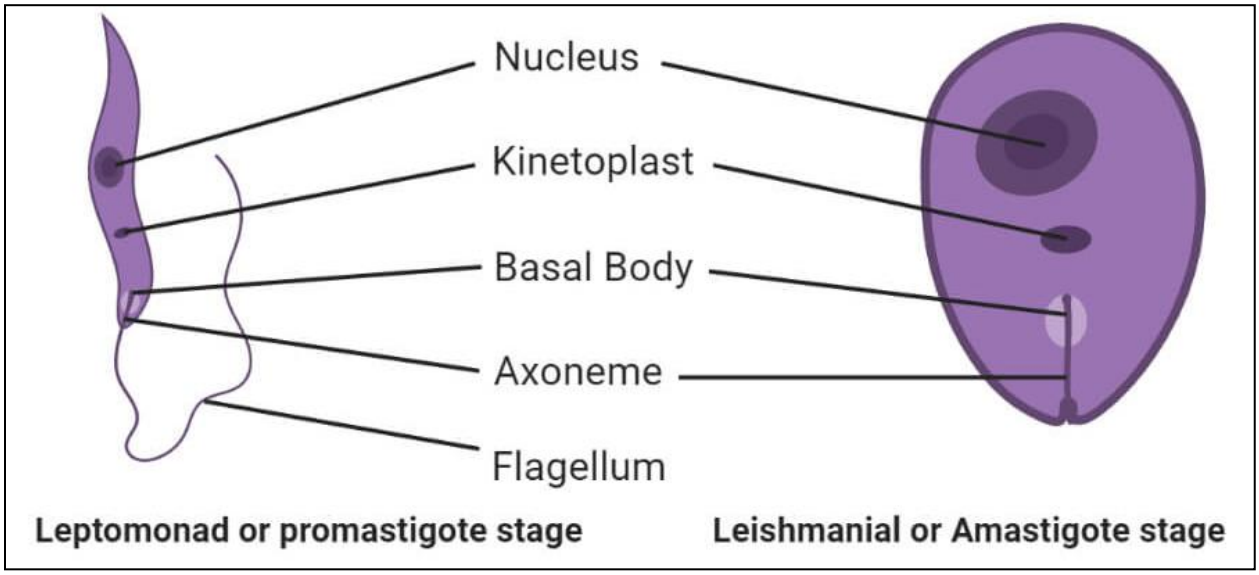

Fig : Morphological structure of *Leishmania Donovani*

1. Leishmanial or Amastigote stage:

- Amastogote is the aflagellar stage of the parasite.
- The parasites at Amastigote stage are found in man and other mammalian hosts.
- They are found inside monocytes, polymorphonuclear leucocytes or endothelial cells.
- Amastigotes are small, round to oval bodies measuring 2-3 μm in length.
- They are also known as LD (Leishman Donovan) bodies.
- Cell membrane is delicate and can be demonstrated only in fresh specimen.
- The nucleus is less than 1 μm in diameter, oval or round and is usually situated in the middle of cell.
- A rod –shaped kinetoplast lies at the right angles to the nucleus. It comprises of DNA containing body and a mitochondrial structure.
- Axoneme (rhizoplast) arises from the kinetoplast and extends to margin of the body. It represents the foot of the flagellum.
- Vacuole, which is clear unstained space, lies alongside the axoneme.
- They are stained well with Giemsa or Wright stain.
- In a Giemsa stained preparation the cytoplasm surrounded by a limiting membrane appear pale blue. The nucleus relatively is larger and stained red. The kinetoplast stained deep red.
- Amastigote divides by binary fission at 37°C.

2. Leptomonad or promastigote stage:

- Promastigotes are found in the digestive tract of sand fly (vector) and in the culture media.
- The fully developed promastigotes are long, slender and splindle-shaped. They measure 15 to 25 μm in length and 1.5 to 3.5 μm in breadth.
- A single nucleus is situated centrally.
- The kinetoplast lies transversely near the anterior end.
- The flagellum is single, delicate and measures 15- 28 μm and may be of same length as the body or even longer, projecting from front. The flagellum does not curve round the body of the parasite and therefore there is no undulating membrane.
- With Leishman stain, the cytoplasm appears blue, the nucleus pink or violet and the kinetoplast bright red.
- Promastigote multiplies by binary fission at 27°C.

Leishmaniasis

Kala-azar, also known as Dum-dum fever, is a serious oriental disease of man. It is found in India, China, Mediterranean countries and parts of Africa and South America. Its causative agent is a pathogenic flagellate, known as *Leishmania donovani* which is transmitted by the bite of small blood sucking sandflies called *Phlebotomus argentipes* (Fig 2). *Leishmania* species undergo multiplication as promastigotes *Phlebotomus argentipes*, but they are injected into a vertebrate host when the sand fly feeds, and they undergo additional multiplication, as amastigotes, in a variety of tissues.

The species concerned are as follows:

i. Indian vector: *P. argentipes*;
ii. Chinese vectors: *P. chinensis*, and *P. sergenti*;
iii. Mediterranean vectors: *P. perniciosus* (Italy and Sicily);
iv. Tropical American vector: *P. intermedius*;
v. East African vector: *P. martini*.

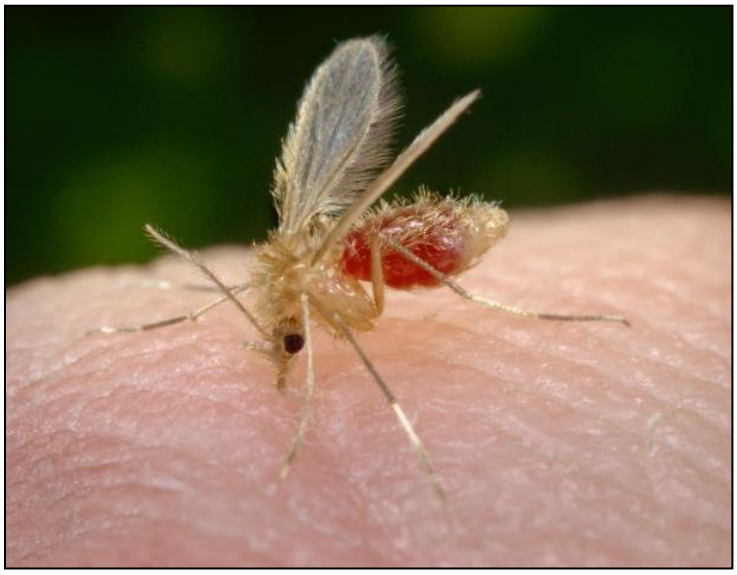

Fig 2: Diagram of *Phlebotomus argentipes*

Life cycle of *Leishmania donovani*:

Hosts

- *Leishmania* is also a digenetic parasite that requires 2 hosts for completion of its life cycle.

- The primary host is a vertebrate or man, in which the parasite feeds and multiplies asexually.
- The secondary host or vector is invertebrates or blood-sucking insects or sand-fly, belonging to the genus *Phlebotomus*.
- Some mammals like dogs, jackals, gerbils, and squirrels also serve as reservoir hosts in which the parasite does not undergo any change but simply waits for its introduction into the human host.

(I) <u>Life cycle in Man</u>

- The parasite has two stages in its life cycle:
- **Amastigote form** occurs in humans and mammals.
- **Promastigote form** occurs in sandfly.
- *L. donovani* is transmitted to humans or other vertebrates by the bite of blood-sucking sandfly *Phlebotomus argentipes*
- The parasites introduced by sandfly into the human body are in the promastigote form.
- Some of the promastigote entering the blood circulation directly become destroyed.
- while those entering the reticuloendothelial system(liver, spleen, bone marrow, and lymph nodes) change into amastigote or leishmanial forms.
- The amastigotes multiply by simple binary fusion inside the Reticuloendothelial system to form a large number of amastigotes.
- When the number of parasite reaches 50 to 200 or even more, the host cell rupture.
- The liberated parasites are taken up by new host cells and the multiplication cycle is repeated so that the reticuloendothelial system becomes progressively infected.
- Some of the free amastigotes are phagocytosed by the neutrophils and monocytes(macrophages) in the bloodstream.
- These heavily parasitized cells wander through the general blood circulation leading to a general infection.

(II) Life cycle in sandfly

- When the sandfly sucks the blood of an infected person, it obtains free amastigotes as well as the parasitized neutrophils and monocytes along with the blood-meal.

- The parasite begins a process of transformation and the amastigotes change to procyclic promastigotes and then to metacyclic promastigotes in the midgut of the sandfly.
- These promastigotes multiply by longitudinal binary fusion and produce large numbers of promastigotes completely filling the lumen of the gut.
- In 6 to 9 days, the number of parasites becomes enormous and heavily spread into the pharynx and buccal cavity. The salivary glands are not infected.
- Transmission into a new host occurs when such a heavily infected sandfly bites the host.

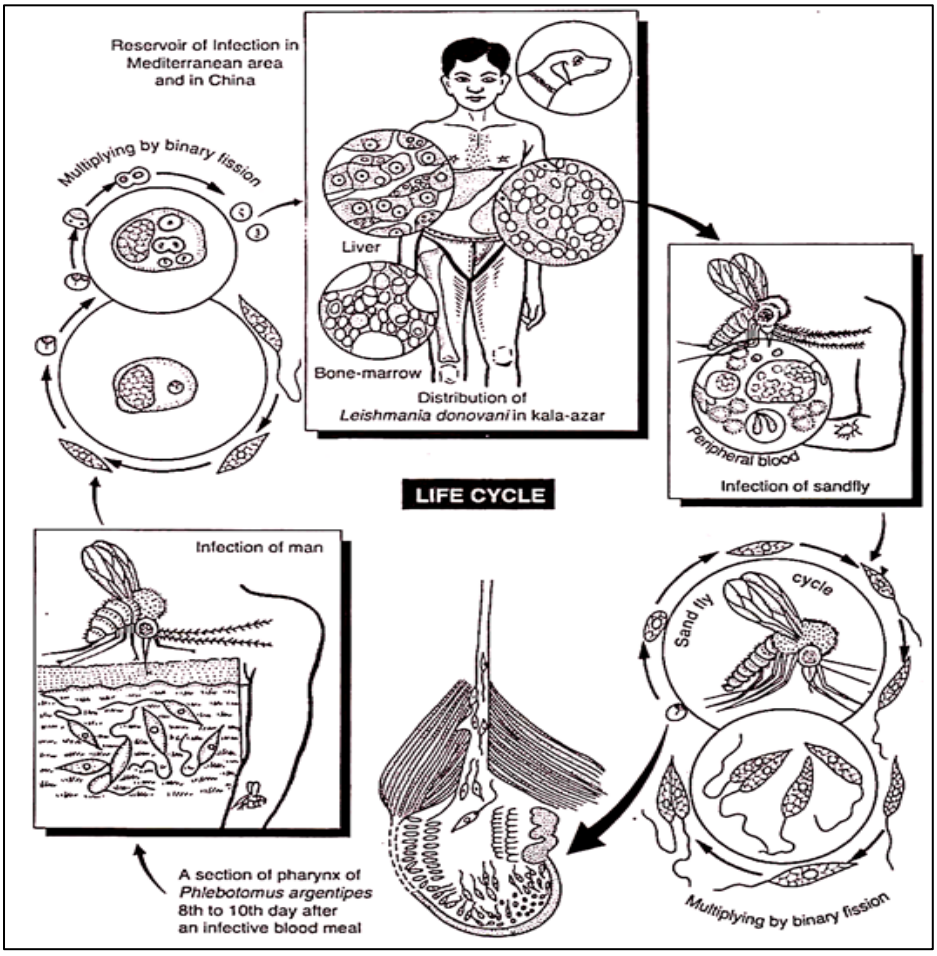

Fig 3: Life cycle of *Leishmania donovani* in man and sand fly

Mode of transmission:

- The infection is transmitted to Human mainly by the bite of vector sandfly of genus *Phlebotomus* and genus *Lutzomyia*.

- Less frequently the infection is transmitted by: Blood transfusion, congenital infection, accidental inoculation of cultured promastigotes in the lab workers and sexual intercourse.
- Males are affected more due to increase exposure through the occupation and leisure activities.

Pathogenesis of *Leishmania donovani*:

- After the inoculation of promastigotes by sand flies, they are deposited on the surface of skin and bind to macrophages in the skin.
- The sand fly, liberates biologically active substances, which promote infectivity of promastigotes by partially deactivating fixed macrophages in the skin.
- The outcome of leishmania infection appears to depend on the complex interaction between the parasite's virulence and the immune response of the host.
- Promastigotes activate complement through the alternative pathway and are opsonized.
- They produce activated products of complement such as C3b or C3bi. These activated products bind with two specific receptors present on the outer membrane of promastigotes.
- The receptors are- a 63kD mol. wt glycoprotien (gp63) and a lipophosphoglycan (LPG). These receptors play an important role in the parasites- macrophage interaction.
- These receptors bind with complement receptors (CR3 and CR1) present on surface of macrophages either directly or through bound C3b or C3bi receptors.
- The most important immunological feature is a marked suppression of the CMI to leishmanial antigens. In persons with asymptomatic self-resolving infection, T-helper cells predominate, although immune suppression years later can result in disease. An overproduction of both specific Ig and non-specific Ig also occurs. The increase gamma globulin leads to reversal of the albumin-globulin ratio commonly associated with this disease.

Leishmaniasis

Leishmaniasis is a disease that involves the Reticuloendothelial (RE) system. Parasitized macrophages disseminate the infection to all parts of body but more to the spleen, liver and bone marrow. The spleen is enlarged, with a thickening of the capsule and is soft and fragile; its vascular spaces are dilated and engorged with blood. The reticular cells are

markedly increased and packed with the amastigote forms of the parasite. In the liver, the kupffer cells are increased in size and number and infected with amastigote forms. Bone marrow turns hyper plastic and parasitized macrophages to replace the normal hemopoietic tissue.

- Proliferation and destruction of Reticuloendothelial cells of the internal organs and heavy parasitization of external organ by parasitized cells are the characteristic pathological changes seen in visceral leishmaniasis.

Types of leishmaniasis

Leishmaniasis comes in three forms: cutaneous, visceral, and mucocutaneous. Different species of the *Leishmania* parasite are associated with each form. Experts believe that there are about 20 *Leishmania* species that can transmit the disease to humans.

I. Cutaneous leishmaniasis

Cutaneous leishmaniasis causes ulcers on your skin. It's the most common form of leishmaniasis. Treatment may not always be necessary depending on the person, but it can speed healing and prevent complications.

II. Mucocutaneous leishmaniasis

A rare form of the disease, mucocutaneous leishmaniasis is caused by the cutaneous form of the parasite and can occur several months after skin ulcers heal. With this type of leishmaniasis, the parasites spread to your nose, throat, and mouth. This can lead to partial or complete destruction of the mucous membranes in those areas. Although mucocutaneous leishmaniasis is usually considered a subset of cutaneous leishmaniasis, it's more serious. It doesn't heal on its own and always requires treatment.

III. Visceral leishmaniasis

Visceral leishmaniasis is sometimes known as **systemic leishmaniasis or kala-azar**. Kala-azar also is known as **black fever or Dumdum fever** in Asia.

It usually occurs 3 to 6 months after being bitten by a sandfly. It primarily infects the reticuloendothelial system. It damages internal organs, such as the spleen and liver. It also affects bone marrow, as well as the immune system through damage to these organs. It is the most severe form of the disease and, left untreated, is usually fatal. Annually an estimated 50 000 to 90 000 new cases of VL occur worldwide with only between 25 to 45% reported to WHO. It remains one of the top parasitic diseases with outbreak and mortality potential. This disease is the second-largest parasitic killer in the world (after malaria), responsible for an estimated 20,000 to 30,000 deaths each year worldwide.

- **Post-Kala-Azar Dermal Leishmaniasis (PKDL)**

Post-kala-azar dermal leishmaniasis (PKDL) is usually a consequence of visceral leishmaniasis that may appear after 1 month or years after the treatment of visceral leishmaniasis. In this leishmaniasis, parasites invade the skin and develop macular, papular, or nodular rash usually on the face, upper arms, trunks, and other parts of the body. 5-10% of kala-azar patients are reported to develop this PkDL. It occurs mainly in East Africa and on the Indian subcontinent.PKDL is not-life threatening but still considered a potential source of *Leishmania* infection.

Symptoms of leishmaniasis

People can carry some species of *Leishmania* for long periods without becoming ill. Symptoms depend on the form of the disease.

I. Cutaneous leishmaniasis

The main symptom of this condition is painless skin ulcers. Cutaneous symptoms may appear a few weeks after being bitten by an infected sand fly. However, sometimes symptoms won't appear for months or years.

II. Mucocutaneous leishmaniasis

In people with the mucocutaneous form of the disease, symptoms usually appear one to five years after the skin lesions. These are primarily ulcers in their mouth and nose or on their lips. Other symptoms may include: runny or stuffy nose, nosebleeds, difficulty breathing

III. Visceral leishmaniasis

Symptoms often don't appear for months after the bite with this type of leishmaniasis. Most cases are apparent two to six months after the infection occurred. Common signs and symptoms include: weight loss, weakness, fever that lasts for weeks or months, enlarged spleen, enlarged liver, and decreased production of blood cells, other infections, swollen lymph nodes. It is followed by general weakness, emaciation, anaemia and a peculiar darkening of the skin.

Lab diagnosis of Visceral leishmaniasis (Kala-azar)

Lab diagnosis of Kala-azar depends upon direct and indirect evidence.

- **Direct evidence**

1. Microscopy

Samples:

- The demonstration of amastigotes in smears of tissue aspirates is the gold standard for the diagnosis of visceral leishmaniasis.
- Peripheral blood sample,
- a biopsy from the bone marrow: obtained by sternal or iliac crest puncture.
- splenic aspirates: biopsy material: obtained by a splenic puncture.
- enlarged lymph node
- The smears of aspirates are stained with Leishman, Giemsa, or Wright's stain for the detection of amastigote forms of the parasite through microscopy.
- For an accurate diagnosis of leishmaniasis, amastigotes should be visualized by light microscopy under oil immersion.
- Amastigote parasites can be seen within the macrophages often in large numbers. A few extracellular forms can also be seen.
- Buffy coat of **peripheral blood** can be utilized to look for the parasite microscopically. Buffy coat smears show a diurnal periodicity, more smears being positive when collected during the day than at night.
- **Splenic aspiration**, although incurring a risk of hemorrhage, is the most sensitive means (95%) for diagnosing leishmaniasis.
- **Bone marrow biopsy** demonstrates amastigotes in approximately two-thirds of patients.
- **Lymph node aspirates** are not useful in the diagnosis of Indian kala-azar, although it is employed in VL in some other countries.
- Although the specificity is high, the sensitivity of microscopy varies, being higher for **spleen** (93% to 99%) than for **bone marrow** (53-86%) or l**ymph nodes aspirates** (53-65%).
- It is the classic confirmatory test for visceral leishmaniasis.

2. Animal inoculation

- It is a very sensitive method.
- It is not used for routine diagnosis.
- In this method, the material is inoculated intraperitoneally or intradermally into the skin of the nose and feet of the Chinese golden hamster, and they are kept at 23- 26°C.
- In positive cases, the amastigote can be demonstrated in smears taken from ulcers or nodules developing at the sites of inoculation or from the spleen.

▪ Indirect evidence

1. Detection of antigen

- The concentration of antigen in the serum or other body fluids is very low.
- **ELISA** and **PCR** have been developed for the detection of leishmanial antigens.
- Two non-invasive antigen detection test in urine for VL are under evaluation.
- It showed good specificity but only low to moderate (48-87%) sensitivity.

2. Detection of antibodies

a. Complement fixation test was the first serological test used to detect serum antibodies in VL.

Specific leishmanial antigens prepared from cultures have been used in a number of tests to demonstrate specific antibodies. These tests include:

b. Indirect immunofluorescent antibody test (IFAT)

c. Counter immunoelectrophoresis (CIEP)

d. ELISA or **Western Blot** using whole parasite lysate**,** have shown high diagnostic accuracy in most studies but are poorly adapted for field settings. The ELISA assay is highly sensitive (80-100%) but the specificity varies with the antigen used, from 80-94% with whole parasite lysate.

3. Leishmanin skin test (Montenegro test)

- It is a Delayed hypersensitivity test (DHT).
- This was first discovered by Montenegro in South America and hence, named after him.

- 0.1 mL of killed promastigote suspension (10^6 washed promastigotes/ mL) is injected intradermally on the dorsoventral aspect of the forearm.
- A positive result is indicated by induration and erythema of 5 mm or more after 48-72 hours.
- A positive result indicates prior exposure to the leishmanial parasite.
- The test is positive in African kala-azar but not in India and Mediterranean kala-azar.
- In the active kala-azar, this test is negative and becomes positive usually 6-8 weeks after a cure from the disease.
- DTH testing is very useful in the diagnosis of cutaneous leishmaniasis.
- It is utilized primarily as an epidemiological tool and has little role in the establishment of a diagnosis of acute VL.

4. Blood count

- Complete blood count shows normocytic normochromic anemia and thrombocytopenia.
- Leukocyte count reveals leukopenia accompanied by a relative increase of lymphocytes and monocytes.
- Eosinophil granulocytes are absent. During the course of the disease, there is a progressive diminution of leukocyte count falling to 1,000/mm3 of blood or even below that.
- The ratio of leukocyte to erythrocyte is greatly altered and maybe about 1:200 to 1:100 (normal 1:750).
- Serum shows hypergammaglobulinemia and a reversal of the albumin: globulin ratio.
- **Liver function tests** show mild elevations of liver

Prevention and control

Preventing and controlling the spread of leishmaniasis is complex and requires many tools. Key strategies include:

- **Early diagnosis and effective prompt treatment** reduce the prevalence of the disease and prevents disabilities and death. It helps to reduce transmission and to monitor the spread and burden of disease. There are highly effective and safe anti-leishmanial medicines particularly for visceral leishmaniasis, although they can be

difficult to use. Access to medicines has significantly improved thanks to a WHO-negotiated price scheme and a medicine donation programme through WHO.

- **Vector control** helps to reduce or interrupt transmission of disease by decreasing the number of sand flies. Control methods include insecticide spray, use of insecticide-treated nets, environmental management and personal protection.
- **Effective disease surveillance** is important to promptly monitor and act during epidemics and situations with high case fatality rates under treatment.
- **Control of animal reservoir hosts** is complex and should be tailored to the local situation.
- **Social mobilization and strengthening partnerships** – mobilization and education of the community with effective behavioural change interventions must always be locally adapted. Partnership and collaboration with various stakeholders and other vector-borne disease control programmes is critical.

Probable questions:

1. What do you mean by Leishmaniasis?
2. What microorganisms cause leishmaniasis?
3. *Describe the morphology of Leishmania donovani*
4. Describe the amastigote stage of *Leishmania donovani.*
5. Describe the promastigote stage of *Leishmania donovani.*
6. What is the infective stage of leishmaniasis?
7. Name the vector of Leishmaniasis?
8. What is the most serious form of leishmaniasis?
9. Is there a vaccine for leishmaniasis?
10. Why is kala-azar called?
11. Discuss different types of Leishmaniasis.
12. How is leishmaniasis transmitted?
13. What are the complications of leishmaniasis?
14. What are the complications of cutaneous leishmaniasis?

15. What are the signs of Leishmaniasis?
16. How can you prevent leishmaniasis?
17. Elaborate the direct evidence of lab diagnosis of visceral leishmaniasis (Kala-azar).
18. What do you mean by montenegro test)

Suggested Readings:

1. Chatterjee K D. 2009. Parasitology: Protozoology and Helminthology. XIII Edition, CBS Publishers
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6. Bock R, Jackson L, de Vos A, Jorgensen W. Babesiosis of cattle. *Parasitology.* 2004;129 (Suppl): S247–69.

UNIT II

Structure, pathobiology, prophylaxis and diagnosis of *Babesia* sp., *Anaplasma* sp. and *Theileria* sp.

Objective:

In this unit we will discuss about structure, pathobiology prophylaxis and diagnosis of *Babesia* sp., *Anaplasma* sp. and *Theileria* sp.

Babesia sp.

Introduction

In the year 1888, Babes investigated outbreaks of diseases with symptoms of hemoglobinuria in cattle in Romania and was the first to discover piroplasm in the blood of cattle. Initially, he thought it to be a bacterium that had named as *Hematococeus bovis* and later it was changed to *Babesia bovis*. After 5 years, Smith and Kilborne demonstrated the causative organism of "Texas Fever" (babesiosis) as *Pyrosoma bigeminum* (*Babesia bigemina*). After that the first demonstration of transovarian transmission of *Babesia* through its tick vector – a finding of historic significance was done.

The first confirmed case of human fatal babesiosis caused by *Babesia divergens* was recorded in 1956. Since then, zoonotic significance of babesiosis that too as a potentially life threatening zoonotic infection in human came out. There are several species of *Babesia* which causes human infections throughout the world but major one is *Babesia microti* particularly in North America. Babesiosis is transmitted during blood feeding by infected ticks. The disease is considered as the most economically important tick-borne disease in tropical and subtropical areas.

If we take the cattle babesiosis in respect of economic impact then we could see losses occurs due to mortality, decreased milk or meat production, abortions, reduction of draft power, cost under the head of control measures, including increased cost of management to maintain ill animals.

Pathology is a branch of medicine that studies the causes and nature of disease. **Pathobiology** is a branch of biology that concentrates on the biological aspects of disease rather than the medical. Pathobiology is also interdisciplinary, combining with microbiology, genetics, anatomy and animal science.

Prophylaxis means treatment given or action taken to prevent disease.

Diagnosis means the identification of the nature of an illness or other problem by examination of the symptoms.

Morphology

On the basis of morphology, ***Babesia*** are divided into two groups – small ***Babesia*** (1.0–2.5 µm long) which included *Babesia bovis*, *Babesia gibsoni*, *Babesia microti*, *Babesia rodhaini*, etc., and large *Babesia* (2.5–5.0 µm long) which included *Babesia bigemina*, *Babesia caballi*, *Babesia Canis*, etc. The orientation of the parasite in the red blood cells (RBCs) depends on its size because large pyriform parasites meet at their pointed ends at an acute angle to each other and small forms make an obtuse angle to each other. More than 100 species of *Babesia* have been identified that are infecting many mammalian and some avian species.

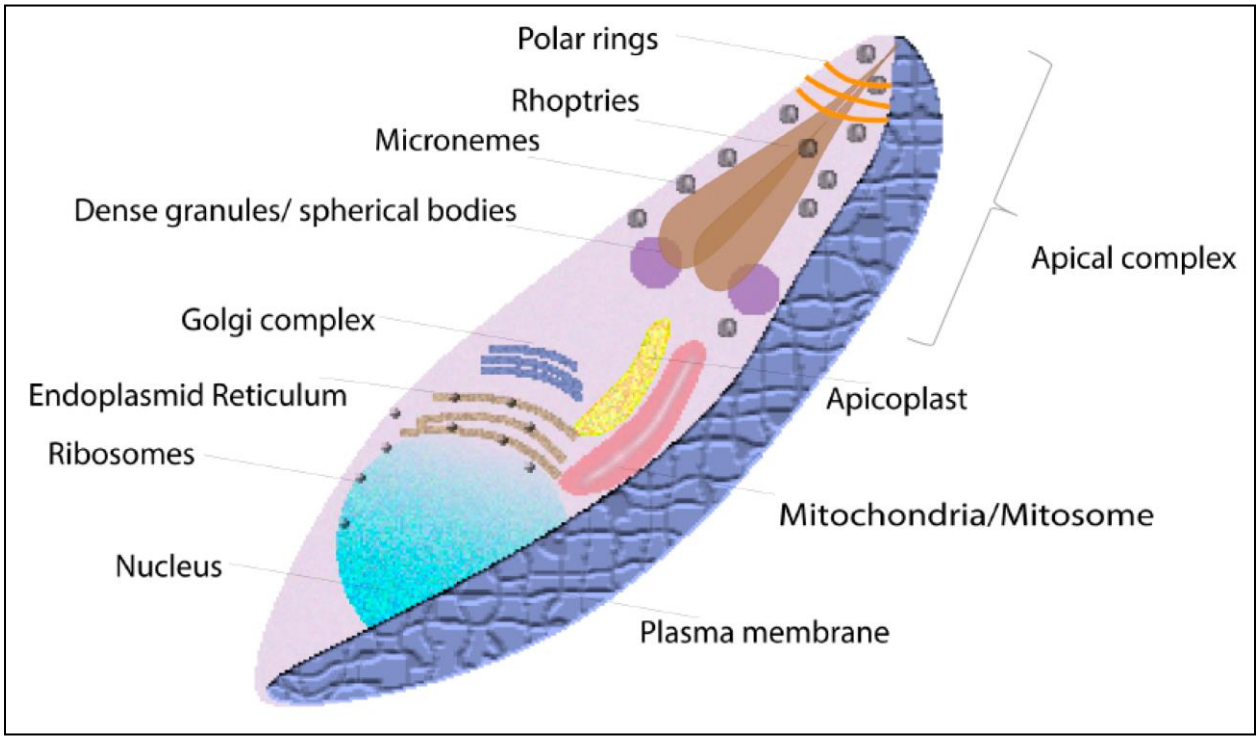

Fig: General structure of *Babesia* sp.

The following are the morphology of different *Babesia* spp. of animals according to their hosts.

I. <u>CATTLE AND BUFFALOES</u>

B. bigemina - large form (4.5 µm × 2.0 µm) of *Babesia*. The parasites are characteristically pear shaped. Round (2–3 µm in diameter) oval or irregularly shaped form may also be found.

B. bovis - small form (2.0 µm × 1.5 µm) of *Babesia*. Slightly larger than *B. divergens*, vacuolated signet ring forms are particularly common.

B. divergens - small form (1.5 µm × 0.4 µm) of *Babesia*. Generally remained as paired form, superficially lie on the RBC, stout and pyriform or circular forms may be found.

B. major - large form (3.2 µm × 1.5 µm) of *Babesia*. Pyriform bodies, the angle between the organism is <90°. Round forms with a diameter of about 1.8 µm are also available.

II. CANINE

Canine babesiosis is caused by two species of *Babesia* viz; *B. canis* and *B. gibsoni*, which are morphologically differentiated on the basis of their size.

B. canis - large form (4–5 µm long) of *Babesia*. Pyriform in shape, pointed one end, and round other. In a single RBC, multiple infection that is, more than one organism up to 16 may be found (Fig 1).

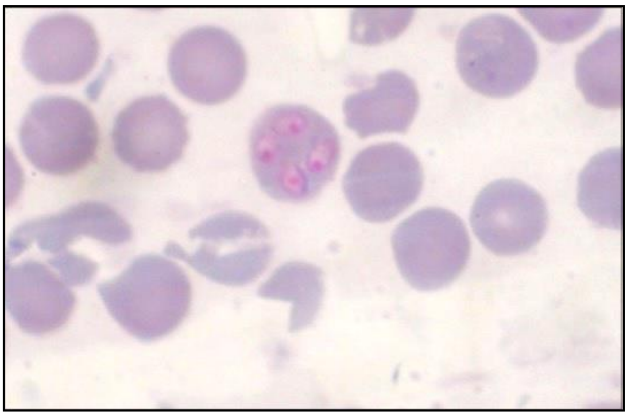

Fig 1: *Babesia canis* within RBC in Giemsa stained blood smear of a dog

B. gibsoni - small form (1.5–2.5 µm) of *Babesia*. Lack usual pyriform shapes, trophozoites are annular or oval; signet ring forms may occur (Fig 2).

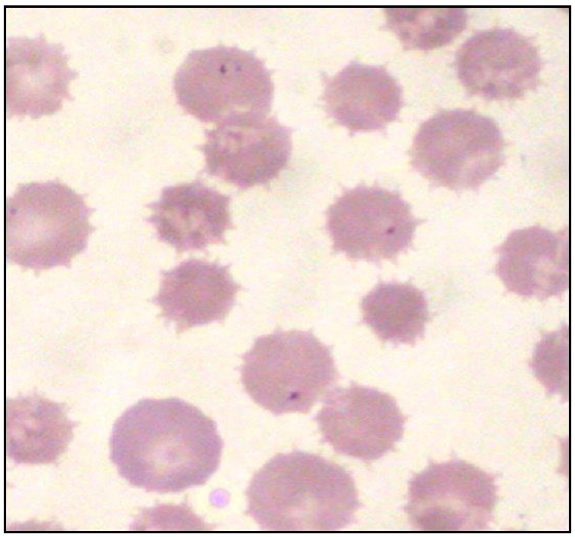

Fig 2: *Babesia gibsoni* within RBC in Giemsa stained blood smear of a dog

III. EQUINES

B. caballi - large form of *Babesia*. Commonly occur as pair. Pyriform and measures 2.5–4 µm long. Round or oval forms with 1.5–3.0 µm in diameter may be found.

Babesia equi - small form (2.0 µm) of *Babesia*. The piroplasms characteristically form a Maltese cross of four organisms.

Babesiosis

Epidemiology

Babesiosis is caused by infection with intra-erythrocytic parasites of the genus *Babesia*, is one of the most common infections of free-living animals worldwide and is gaining increasing interest as an emerging zoonosis in humans. Although capable of infecting a wide range of vertebrates, babesial parasites require both a competent vertebrate and nonvertebrate host to maintain transmission cycles. All babesial parasites described to date are transmitted by ixodid ticks to their vertebrate hosts. The parasites replicate in the vertebrate hosts' red blood cells and are called piroplasms due to their pear-shaped appearance when within the infected host cells. Most of what is known about the host response to babesial infections comes from observations of and studies on vertebrates other than humans. All mammalian hosts examined have been able to develop immunity to *Babesia* species, either after an episode of infection and recovery or after prophylactic immunization. Both humoral and cellular factors are involved in immunity to babesiosis.

Human babesiosis is caused by one of several babesial species that have distinct geographic distributions based on the presence of competent hosts. In North America, babesiosis is caused predominantly by *Babesia microti*,a rodent-borne piroplasm, and also occasionally by a newly recognized species, the so-called WA1 piroplasm. In Europe, babesiosis is considerably rarer but more lethal; it is caused by the bovine pathogen *Babesia divergens*. The spectrum of disease is broad, ranging from an apparently silent infection to a fulminant, malaria-like disease resulting occasionally in death.

- ***Invertebrate hosts***

Babesias can be found wherever certain species of ticks flourish. To date, only ixodid ticks have been identified as vectors for *Babesia* spp. except for one report that identified a nonixodes tick, *Ornithodoros erraticus*, as a reservoir for *Babesia meri*. Some *Babesia* species, such as*Babesia bigemina* and *Theileria equi* (*Babesia equi*) can infect more than one genus of ticks, whereas *B. microti* can only infect ticks from the genus *Ixodes* . Several tick vectors can carry more than one *Babesia* species. For instance, *Ixodes dammini* can harbor *B. microti*, usually but not exclusively in its nymphal stage.

The ecology and life cycle of *B. microti* and its interaction with *I. dammini* (also known as *Ixodes scapularis*) is the best understood of the *Babesia* species. The nymphal stage of *I. dammini* and its interaction with white-footed mouse (*Peromyscus leucopus*) is essential for the maintenance of *B. microti*. The adult stages of *I. dammini* feed primarily on deer (*Odocoileus virginianus*), which do not serve as reservoirs for *B. microti* .

They feed in the fall and again in the spring, after which they lay eggs. The eggs hatch in the summer (late July), and the larvae feed primarily on mice during August and

September. This is the point at which the tick can acquire *Babesia* organisms. These infected larvae overwinter and molt to become nymphs in the spring.

The nymphs feed on hosts from May through July.

Finally, nymphs that have fed moult into adults in the fall, completing the tick life cycle

It is believed that the tick responsible for transmission of *B. divergens* to humans is *Ixodes ricinus*. The life cycle of *I. ricinus* requires 3 years, as larva in the first year, nymphs in the second, and adults in the third.

- ***Vertebrate hosts***

More than 100 known *Babesia* species have been identified avian species. Almost any mammal that serves as a host for a*Babesia*-infected tick is a potential reservoir. The host ranges of *B. microti* and *B. divergens* vary from small terrestrial mammals to subhuman primates to humans for*B. microti* and from cattle to various rodent species and to humans for *B. divergens* (40, 119, 143). There are several examples of different and often more serious disease manifestations resulting from transmission of a *Babesia*species (e.g., *B. microti*) that is common in a wild vertebrate species (e.g., *P. leucopus*) to a poorly adapted vertebrate host (e.g., humans). As a natural reservoir for *B. microti*, most white-footed mice (*P. leucopus*) in babesiosis-enzootic areas are parasitemic; however, it is not unusual for less than 0.1% of the host erythrocytes to be infected.

Life Cycle

Apicomplexans/sporozoans (including the genera *Babesia*and its close relative *Theileria*) generally go through at least three stages of reproduction (Fig.3)): (i) gamogony—formation and fusion of gametes inside the tick gut, (ii) sporogony—asexual reproduction in salivary glands, and (iii) merogony—asexual reproduction in the vertebrate host

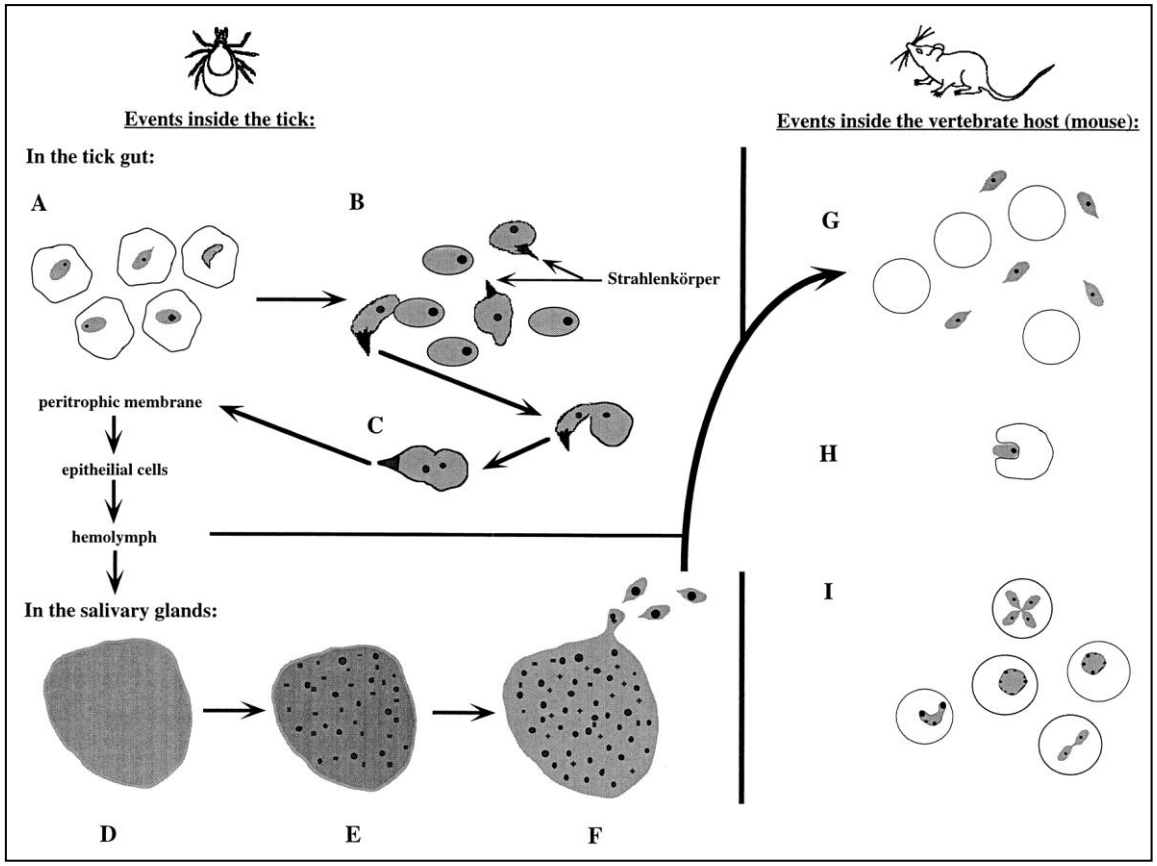

Fig. 3. Life cycle of *Babesia* spp. in the tick and vertebrate hosts. Events in the tick begin with the parasites still visible in consumed erythrocytes. Some are beginning to develop Strahlenkörper forms (A). The released gametes begin to fuse (note that only one of the proposed mechanisms is pictured; one gamete has a Strahlenkörper form, whereas the other does not) (B). The formed zygote then goes on to infect and move through other tissues within the tick (C) to the salivary glands. Once a parasite has infected the salivary acini, a multinucleate but undifferentiated sporoblast is formed (D). After the tick begins to feed, the specialized organelles of the future sporozoites form (E). Finally, mature sporozoites bud off of the sporoblast (F). As the tick feeds on a vertebrate host, these sporozoites are inoculated into the host (G). Not shown is the preerythrocytic phase seen in *Theileria*spp. and *T. equi* (*B. equi*). Sporozoites (or merozoites) contact a host erythrocytic and begin the process of infection by invagination (H). The parasites become trophozoites and can divide by binary fission within the host erythrocyte, creating the various ring forms and crosses seen on stained blood smears (I). Illustrations are not to scale.

Symptoms and Signs of Babesiosis

Asymptomatic *Babesia* infection may persist for months to years and remain subclinical throughout its course in otherwise healthy people, especially those < 40 years.

When symptomatic, babesiosis usually starts after a 1- to 2-week incubation period with nonspecific symptoms including malaise, fatigue, chills, fever, headache, myalgia,

and arthralgia. In healthy people, symptoms usually resolve after a week. In others, hepatosplenomegaly with jaundice, mild to moderately severe hemolytic anemia, mild neutropenia, and thrombocytopenia may occur. Noncardiac pulmonary edema can develop in severe disease.

Babesiosis is sometimes fatal, particularly in older patients, asplenic patients, and patients with AIDS. In such patients, babesiosis may resemble falciparum malaria, with high fever, hemolytic anemia, hemoglobinuria, jaundice, and renal failure. Splenectomy may cause previously acquired asymptomatic parasitemia to become symptomatic.

Babesiosis in neonates ranges from mild to severe febrile illnesses.

Diagnosis of Babesiosis

- Light microscopy of blood smears
- Serologic and polymerase chain reaction-based tests

Most patients with babesiosis do not remember a tick bite, but they may reside in or report a history of travel to an endemic region.

Babesiosis is usually diagnosed by finding *Babesia* in blood smears, but differentiation from *Plasmodium* species can be difficult. Tetrad forms (the so-called Maltese cross formation), although not common, are unique to *Babesia* and helpful diagnostically.

Serologic and polymerase chain reaction (PCR)–based tests are available. Antibody detection by indirect fluorescent antibody (IFA) testing using *B. microti* antigens can be helpful in patients with low-level parasitemia but may be falsely negative in those infected with other *Babesia* species. PCR-based assays can help differentiate *Babesia* from *Plasmodium falciparum* if blood smear findings are ambiguous, confirm infection in patients with low parasitemia, and identify the *Babesia* species.

Treatment of Babesiosis

- Atovaquone plus azithromycin
- Quinine plus clindamycin

Asymptomatic patients usually require no treatment, but therapy is indicated for patients with persistent high fever, rapidly increasing parasitemia, and falling hematocrit.

The combination of atovaquone and azithromycin given for 7 to 10 days has fewer adverse effects and is as effective as traditional therapy with quinine plus clindamycin in patients with mild to moderate babesiosis. Adult dosage is atovaquone 750 mg orally every 12 hours and azithromycin 500 to 1000 mg orally the first day followed by a daily dose of 250 to 1000 mg. In children > 5 kg, dosage

is atovaquone 20 mg/kg orally twice a day plus azithromycin 10 mg/kg orally once, then 5 mg/kg once a day for 7 to 10 days.

Quinine 650 mg orally 3 times a day plus clindamycin 600 mg orally 3 times a day or 300 to 600 mg IV 4 times a day for 7 to 10 days can also be used. Pediatric dosage is quinine 10 mg/kg orally 3 times a day plus clindamycin 7 to 14 mg/kg orally 3 times a day. Quinine plus clindamycin is considered the standard of care for severely ill patients. Recipients of quinine must be monitored closely for adverse effects.

Exchange transfusion has been used in severely ill patients with high (>10% of erythrocytes) parasitemia.

Prevention of Babesiosis

To prevent babesiosis, standard tick precautions should be taken by all people in endemic areas. Asplenic patients and patients with AIDS should be particularly cautious. People who have had babesiosis are deferred from donating blood and potentially organs to prevent transmission. Screening of blood and organ donors is now done in states with relatively high incidences of infection.

i. Tick bite prevention

Preventing tick access to skin includes

- Staying on paths and trails
- Tucking trousers into boots or socks
- Wearing long-sleeved shirts
- Applying repellents with diethyltoluamide (DEET) to skin surfaces

ii. DEET should be used cautiously in very young children because toxic reactions have been reported. Permethrin on clothing effectively kills ticks. Frequent searches for ticks, particularly in hairy areas and on children, are essential in endemic areas.

iii. Engorged ticks should be removed with care and not crushed between the fingers because crushing the tick may result in disease transmission. The tick's body should not be grasped or squeezed. Gradual traction on the head with a small forceps dislodges the tick. The point of attachment should be swabbed with alcohol. Petroleum jelly, alcohol, lit matches, and other irritants are not effective ways to remove ticks and should not be used.

iv. No practical means are available to rid entire areas of ticks, but tick populations may be reduced in endemic areas by controlling small-animal populations.

Anaplasma sp.

Anaplasmosis is a tickborne disease caused by the bacterium *Anaplasma phagocytophilum*. These bacteria are spread to people by tick bites primarily from the blacklegged tick (*Ixodes scapularis*) and the western blacklegged tick (*Ixodes pacificus*). People with anaplasmosis will often have fever, headache, chills, and muscle aches.

Anaplasmosis is most commonly reported in the Northeastern and upper Midwestern states.

Transmission of anaplasmosis

- ✓ *Tick Bites:* Primarily, through infected tick bites.
    - Blacklegged tick (*Ixodes scapularis*) widely distributed across the eastern United States. The blacklegged tick also transmits other pathogens (including *Borrelia burgdorferi*) in certain geographic areas, and coinfections have been reported.
    - Western blacklegged tick (*Ixodes pacificus*) along the West Coast.
- ✓ *Less commonly, through blood transfusion and organ transplant*
    - *A. phagocytophilum* can be transmitted through blood transfusions.
    - *A. phagocytophilum* has been shown to survive for more than a week in refrigerated blood.
    - Several cases of transfusion-transmitted anaplasmosis have been reported, including in cases of asymptomatic donors.
    - Patients who develop anaplasmosis within a month of receiving a blood transfusion or solid organ transplant should be reported to state health officials for prompt investigation.
    - Leukoreduction might reduce the risk of *Anaplasma* transmission; but it does not eliminate it. Cases associated with the use of leukoreduced blood products have been documented.
    - Blood products are not routinely screened for the presence of *A. phagocytophilum.*

Signs and Symptoms

- Signs and symptoms of anaplasmosis typically begin within 1–2 weeks after the bite of an infected tick.
    - Tick bites are usually painless, and many people do not remember being bitten.

I. *Early Illness*

Early signs and symptoms (days 1-5) are usually mild or moderate and may include:

- Fever, chills
- Severe headache
- Muscle aches
- Nausea, vomiting, diarrhea, loss of appetite

II. *Late Illness*

Rarely, if treatment is delayed or if there are other medical conditions present, anaplasmosis can cause severe illness. Prompt treatment can reduce your risk of developing severe illness.

- Respiratory failure
- Bleeding problems
- Organ failure
- Death

III. *Risk factors for severe illness:*

- **Delayed treatment**
- **Age:** being older puts you at risk
- **Weakened immune system:** People with weakened immune systems (such as those receiving some cancer treatments, individuals with advanced HIV infection, prior organ transplants, or people taking some medications) are at risk for severe illness

Diagnosis and Testing

- Your healthcare provider can order certain blood tests to look for evidence of anaplasmosis or other illnesses that cause similar symptoms.
- Test results may take several weeks.
- If your healthcare provider thinks you have anaplasmosis, or another tickborne infection, he or she may prescribe antibiotics while you wait for test results.

Treatment of Anaplasmosis

i. **Doxycycline** is the treatment of choice for anaplasmosis, and all other tickborne rickettsial diseases. Presumptive treatment with doxycycline is recommended in patients of all ages, including children <8 years.

ii. Doxycycline is most effective at preventing severe complications from developing if it is started early in the course of disease.

iii. When treated with doxycycline, fever generally subsides within 24–48 hours.

iv. Lack of a clinical response to doxycycline suggests that the patient's condition might not be due to anaplasmosis, or might be caused by other infections not responsive to doxycycline (see coinfections).

v. Resistance to doxycycline or relapses in symptoms after the completion of the recommended course has not been documented.

✓ **Treating Children and Pregnant Women**

i. The use of doxycycline to treat suspected rickettsial disease in children is standard practice recommended by both CDC and the American Academy of Pediatrics Committee on Infectious Diseases.

ii. A recent study found that short courses of doxycycline (5–10 days) did not result in staining of permanent teeth or enamel hypoplasia. Use doxycycline as the first-line treatment for suspected anaplasmosis in patients of all ages.

iii. Use of antibiotics other than doxycycline increases the risk of severe illness and patient death.

✓ **Other Treatments**

i. In cases of life-threatening allergies to doxycycline, severe doxycycline intolerance, and in some pregnant patients for whom the clinical course of anaplasmosis appears mild, physicians should consider alternate antibiotics.

ii. **Rifampin** has been used successfully in several pregnant women with anaplasmosis, and studies suggest that this drug appears effective against *A*. *phagocytophilum*.

iii. Small numbers of children <8 years of age have also been treated successfully for anaplasmosis with rifampin following a 7–10 day course.

iv. However, rifampin is not effective in treating RMSF, a disease that might be confused with anaplasmosis, nor is it an effective treatment for potential coinfection with Lyme disease.

v. Healthcare providers should be cautious when exploring treatments other than doxycycline, which is highly effective in treating multiple tickborne diseases including anaplasmosis, ehrlichiosis, Lyme disease, and RMSF.

✓ **Antibiotics as Prophylaxis**

i. Post-tick bite antibiotic prophylaxis is not recommended to prevent anaplasmosis.

ii. People who have been bitten by a tick should watch for signs and symptoms. They should see their healthcare provider if fever, rash, or other symptoms develop within two weeks of tick bite.

iii. Asymptomatic treatment for tick bites is not currently recommended.

Prevention

- There is no vaccine to prevent anaplasmosis. Prevent illness by preventing tick bites, preventing ticks on your pets, and preventing ticks in your yard.
- Ticks live in grassy, brushy, or wooded areas, or even on animals, so spending time outside camping, gardening, or hunting will bring you in close contact with ticks. Protect yourself, your family, and your pets. Here's how:
- Ticks can be active year-round, but ticks are most active during warmer months (April-September).

Before You Go Outdoors

- **Know where to expect ticks.** Ticks live in grassy, brushy, or wooded areas, or even on animals. Spending time outside walking your dog, camping, gardening, or hunting could bring you in close contact with ticks. Many people get ticks in their own yard or neighborhood.
- **Treat clothing and gear** with products containing 0.5% permethrin. Permethrin can be used to treat boots, clothing and camping gear and remain protective through several washings. Alternatively, you can buy permethrin-treated clothing and gear.
- **Use Environmental Protection Agency (EPA)-registered insect repellentsexternal icon** containing DEET, picaridin, IR3535, Oil of Lemon Eucalyptus (OLE), para-menthane-diol (PMD), or 2-undecanone. EPA's helpful search toolexternal icon can help you find the product that best suits your needs. Always follow product instructions. Do not use products containing OLE or PMD on children under 3 years old.
- **Avoid Contact with Ticks**
    - Avoid wooded and brushy areas with high grass and leaf litter.
    - Walk in the center of trails.

After You Come Indoors

Check your clothing for ticks. Ticks may be carried into the house on clothing. Any ticks that are found should be removed. Tumble dry clothes in a dryer on high heat for 10 minutes to kill ticks on dry clothing after you come indoors. If the clothes are damp, additional time may be needed. If the clothes require washing first, hot water is recommended. Cold and medium temperature water will not kill ticks.

Examine gear and pets. Ticks can ride into the home on clothing and pets, then attach to a person later, so carefully examine pets, coats, and daypacks.

Shower soon after being outdoors. Showering within two hours of coming indoors has been shown to reduce your risk of getting Lyme disease and may be effective in reducing the risk of other tickborne diseases. Showering may help wash off unattached ticks and it is a good opportunity to do a tick check.

Check your body for ticks after being outdoors. Conduct a full body check upon return from potentially tick-infested areas, including your own backyard. Use a hand-held or full-length mirror to view all parts of your body. Check these parts of your body and your child's body for ticks:

- Under the arms
- In and around the ears
- Inside belly button
- Back of the knees
- In and around the hair
- Between the legs
- Around the waist

Theileria sp.

Theilerioses are a group of tickborne diseases caused by protozoan parasites of the *Theileria* genus. A large number of *Theileria* spp are found in domestic and wild ungulates in tropical and subtropical regions of the world. The most important species affecting cattle are *T. parva* and *T. annulata*, which cause acute disease resulting in high levels of mortality. *T. lestoquardi, T. luwenshuni*, and *T. uilenbergi* are important causes of mortality in sheep, and *T. equi* sometimes causes clinical disease in horses. A therapeutic drug, buparvaquone, is available to treat the diseases, but it is expensive, and control of the diseases usually involves either prevention of tick infestation or, in some areas, vaccination.

Both *Theileria* and *Babesia* are members of the suborder Piroplasmorina. Although Babesia is primarily parasites of RBCs, *Theileria* use, successively, WBCs and RBCs for completion of their life cycle in mammalian hosts. The infective sporozoite stage of the parasite is transmitted in the saliva of infected ticks as they feed. Sporozoites invade leukocytes and, within a few days, develop to schizonts. In the most pathogenic species of *Theileria* (eg, *T. parva* and *T. annulata*), parasite multiplication occurs predominantly within the host WBCs, whereas less pathogenic species multiply mainly in RBCs.

Development of the schizont stage of pathogenic *Theileria* causes the host WBC to divide; at each cell division, the parasite also divides. Thus, the parasitized cell population expands and, through migration, becomes disseminated throughout the lymphoid system. Later in the infection, some of the schizonts undergo merogony, releasing merozoites that infect RBCs, giving rise to piroplasms. Uptake of piroplasm-infected RBCs by vector ticks feeding on infected animals is the prelude to a complex cycle of development, culminating in transmission of infection by ticks feeding in their next instar (trans-stadial transmission). There is no transovarial transmission as occurs in *Babesia*.

Occurrence of disease is limited to the geographic distribution of the appropriate tick vectors. In some endemic areas, indigenous cattle have a degree of innate resistance. Mortality in such stock is relatively low, but introduced cattle are particularly vulnerable.

East Coast Fever

East Coast fever, caused by *Theileria parva*, is an acute disease of cattle. It is usually characterized by high fever, swelling of the lymph nodes, dyspnea, and high mortality. It is a serious problem in east and southern Africa.

Etiology and Transmission of East Coast Fever

T. parva sporozoites are injected into cattle by infected vector ticks, Rhipicephalus appendiculatus, during feeding. Ticks acquire infection by feeding on infected cattle or African buffalo (*Syncerus caffer*), which carry the infection but do not show signs of disease. Both cattle- and buffalo-derived *T. parva* are highly pathogenic when transmitted to cattle, but the latter do not develop to the piroplasm stage and therefore are usually not transmitted by ticks from infected cattle.

Pathogenesis and Clinical Findings of East Coast Fever

An occult phase of 5–10 days follows before infected lymphocytes can be detected by microscopic examination of smears of cells aspirated from affected lymph nodes. Subsequently, the number of parasitized cells increases rapidly throughout the lymphoid system, and from about day 14 onward, cells undergoing merogony are observed and piroplasm-infected erythrocytes are detected. This coincides with progressively severe lymphocytolysis, marked lymphoid depletion, and leukopenia.

Clinical signs vary according to the level of challenge, and they range from inapparent or mild to severe and fatal. Typically, fever occurs 7–10 days after parasites are introduced by feeding ticks, continues throughout the course of infection, and may be >107°F (42°C). Lymph node swelling becomes pronounced and generalized as the number of infected lymphoblasts increases. Anorexia develops, and the animal rapidly loses condition; lacrimation and nasal discharge may occur. Terminally, dyspnea is common.

Just before death, a sharp decrease in body temperature is usual, and pulmonary exudate pours from the nostrils. Death usually occurs 18–24 days after infection.

The most striking postmortem lesions are generalized lymph node enlargement and massive pulmonary edema and hyperemia. Hemorrhages are common on the serosal and mucosal surfaces of many organs, sometimes together with obvious areas of necrosis in the lymph nodes and thymus. Anemia is not a major diagnostic sign (as it is in babesiosis).

Although the clinical and pathologic features and severity of disease caused by cattle- and buffalo-derived *T. parva* are broadly similar, the latter (sometimes referred to as Corridor disease) differ by exhibiting lower levels of schizont-infected lymphoblasts and no piroplasms.

Animals that recover are immune to subsequent challenge with the same strains but may be susceptible to some heterologous strains. Most recovered or immunized animals remain carriers of the infection.

Tropical Theileriosis

T. annulata is the causal agent of tropical theileriosis, which is widely distributed in north Africa, the Mediterranean coastal area, the Middle East, India, countries of the southern former USSR, and Asia. It is transmitted by several species of ticks of the genus *Hyalomma*. *T. annulata* can cause mortality of up to 90%, but strains vary in their pathogenicity.

The kinetics of infection and the main clinical features of the disease are similar to those produced by *T. parva*, but unlike East Coast fever, anemia is often a feature of the disease. Characteristic signs include fever and swollen superficial lymph nodes, and if the disease progresses, cattle rapidly lose condition. Animals that recover from infection are immune to subsequent challenge.

Diagnosis of Theileriosis in Cattle

- Diagnosis is based on clinical signs and detection of parasites in lymph node aspirates.
- Serology is only of value in detecting previous infection in recovered animals.

Confirmation of disease caused by *T. parva* and *T. annulata* relies on microscopic examination of Giemsa-stained smears of lymph node needle aspirates for the presence of schizonts in infected leukocytes. The intra-erythrocytic piroplasm stages are also readily detected in stained blood smears. Piroplasms assume various forms, but typically they are small and rod-shaped or oval. The schizonts and piroplasms of T parva and T annulata are morphologically similar.

Definitive diagnosis can also be confirmed using antigen-specific ELISAs or PCR on lymph node aspirates.

Treatment and Control of Theileriosis in Cattle

- Buparvaquone, often accompanied by anti-inflammatory drugs and antidiuretics, if there is evidence of pulmonary edema

Only a single compound, buparvaquone, is available for treatment of the diseases caused by Theileria parasites. Treatment is effective when applied in the early stages of clinical disease but may require more than one dose. Treatment is less effective in the advanced stages, when there is extensive destruction of lymphoid and hematopoietic tissues. Development of resistance to buparvaquone has also been reported for T annulata.

Prevention of Theileriosis in Cattle

Spraying or dipping of animals with acaracides is the most frequently used method for prevention of theileriosis, but this needs to be applied at regular intervals to be effective. Pyrethroid compounds are often used where animals are challenged with both tickborne diseases and trypanosomes.

Vaccination of cattle against T parva using an infection-and-treatment procedure is gaining acceptance in some regions. The components for this procedure are a cryopreserved sporozoite stabilate of homogenized ticks infected with the appropriate strain(s) of T parva and a single dose of long-acting oxytetracycline given simultaneously. Although oxytetracycline has little therapeutic effect when given after development of disease, it inhibits development of the parasite when given at the outset of infection.

Live vaccines using in vitro-cultivated parasitized bovine cells containing the schizont stage of T annulata are used in some countries to vaccinate cattle against T annulata. The infected cells are held as cryopreserved stock and, after thawing, approximately one million cells are administered subcutaneously. The parasitized cells need to be subjected to prolonged passage in vitro to ensure they are attenuated.

Cattle should be immunized 3–4 weeks before being allowed on infected pasture.

Probable questions:

1. Describe the general structure of *Babesia* sp. with diagram?
2. Describe the morphology of different *Babesia* spp. found in cattle and buffaloes?
3. Name the causative agent of Babesiosis.
4. Elaborate the epidemiology of babesiosis.
5. Name the invertebrate host of *Babesia* sp.
6. Describe the life cycle of Babesia sp. with diagram.
7. What are the sign and symptoms of babesiosis?
8. State the techniques of babesiosis prevention.
9. What are the treatments of Babesiosis?
10. How anaplasmosis is transmitted between different hosts?
11. What are the sign and symptoms of anaplasmosis?
12. What are the treatments of anaplasmosis?
13. How anaplasmosis can be prevented?
14. Describe the pathogenesis and clinical findings of East Coast Fever.
15. What is Tropical Theileriosis?
16. How the Theileriosis in cattle can be diagonosed?

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UNIT III

Trypanosoma cruzi and Chagas disease and *Trichomonas foetus* in cattle

Objective:

In this unit we will discuss about *Trypanosoma cruzi* and Chagas disease and *Trichomonas foetus* in cattle.

Introduction:

Trypanosoma cruzi is an obligatory parasite, meaning they require at least one host to complete their life cycle. Some species are heteroxenous that require more than one host to complete their life cycle. They are mostly transmitted by blood feeding invertebrates. *Trypanosoma cruzi* is the of human Chagas disease. Chagas disease is the highest impact infectious disease in Latin America and the most common cause of infectious myocarditis in the world. Although human Chagas disease is a huge public health problem, humans are in fact only incidental hosts for *T. cruzi*. *Trypanosoma cruzi* is very widely distributed in many wild and domestic mammals and thus will never be eradicated. However, there is one major benefit to zoonotic infections like *T. cruzi*: the many hosts that *T. cruzi* infects besides humans, including rodents, canines, and nonhuman primates, make outstanding models for studying the immunology of *T. cruzi* infection, providing highly reliable information relevant to the human infections.

Classification

Domain	Eukaryota
Phylum	Euglenozoa
Class	Kinetoplastea
Order	Trypanosomatida
Family	Trypanosomatidae
Genus	*Trypanosoma*

Morphology and Life Cycle

- They are unicellular, parasitic and flagellated protozoans.
- The mitochondrial genome of trypanosomes is known as kinetoplast DNA or kDNA. It is made up of catenated circles and minicircles and requires a different set of proteins during cell division.

- Trypanosoma exist in two different types and that have different types of life cycles – stercorarian and salivarian species.
- The stercoraria trypanosoma species first infects a triatomine kissing bug during a blood meal and develops in the posterior gut of the insect. It is then released in the faeces followed by deposition of the skin of the host. The trypanosoma then penetrates in the body of the host and causes infection.

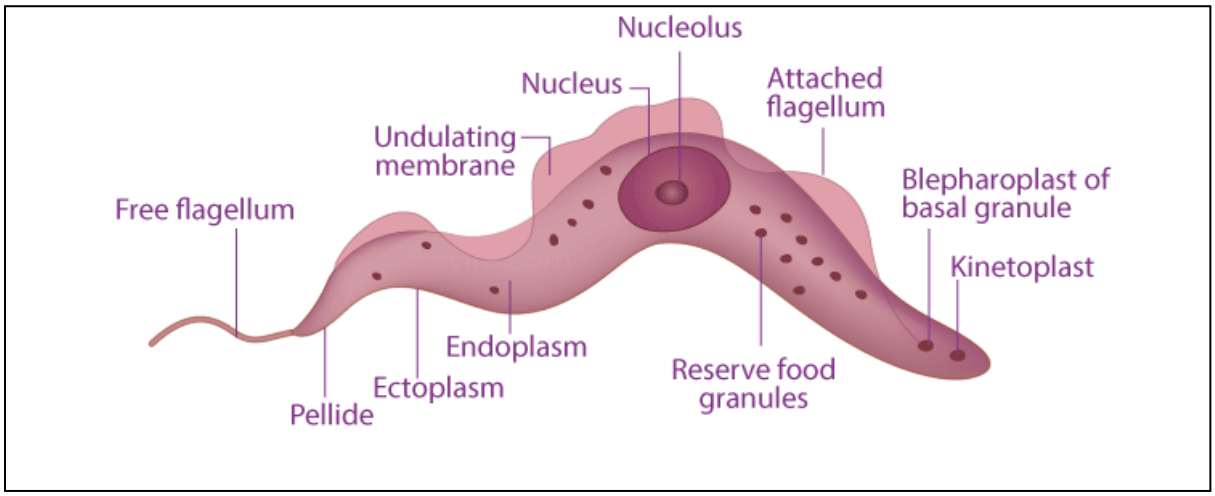


Fig: Morphological structure of *Trypanosoma* sp.

- The salivarian species develops in the anterior gut of Tsetse fly and is transmitted to the host by insect feed.
- The trypanosomes undergo a series of morphological changes during their transition from invertebrates to vertebrates.
- **Trypomastigote** is identified by their flagella that are attached to the body by an undulating membrane and the kinetoplast lies in the posterior portion of the body. This form of trypanosomes is found in the vertebrate host but is developed in the invertebrate host.
- **Amastigote** is another form of *Trypanosoma* that is identified by no visible external flagella or cilia. It is the intracellular form that is found during the replication phase of the organism.

Chagas disease

Chagas disease, also known as American trypanosomiasis, is a potentially life-threatening illness caused by the protozoan parasite *Trypanosoma cruzi.* About 6–7 million people worldwide are estimated to be infected with *T. cruzi*. The disease is found mainly in endemic areas of 21 continental Latin American countries, where it has been mostly transmitted to humans and other mammals by contact with faeces or urine

of triatomine bugs (vector-borne), known as kissing bugs, among many other popular names, depending on the geographical area.

Chagas disease is named after Carlos Ribeiro Justiniano Chagas, a Brazilian physician and researcher who discovered the disease in 1909.

Distribution

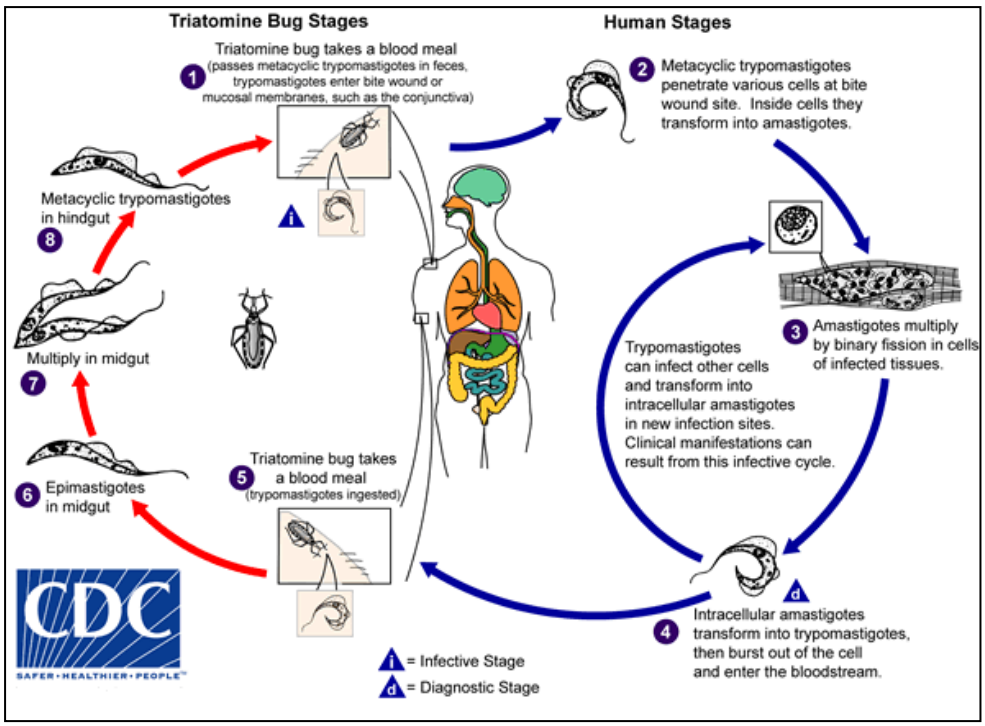
Chagas disease was once entirely confined to continental rural areas of the Region of the Americas (excluding the Caribbean islands). Due to increased population mobility over previous decades, most infected people now live in urban settings and the infection has been increasingly detected in the United States of America, Canada, and many European and some African, Eastern Mediterranean and Western Pacific countries.

Transmission

In Latin America, *T. cruzi* parasites are mainly transmitted by contact with faeces/urine of infected blood-sucking triatomine bugs. These bugs typically live in the wall or roof cracks of homes and peridomiciliary structures, such as chicken coops, pens and warehouses, in rural or suburban areas. Normally they hide during the day and become active at night when they feed on animal blood, including human blood. They usually bite an exposed area of skin such as the face (hence its common name, kissing bug), and the bug defecates or urinates close to the bite. The parasites enter the body when the person instinctively smears the bug's faeces or urine into the bite, other skin breaks, the eyes or the mouth.

T. cruzi can also be transmitted by:

- Consumption of food or beverages contaminated with *T. cruzi* through, for example, contact with faeces or urine of infected triatomine bugs or common opossums. This kind of transmission typically causes outbreaks with more severe case and mortality;
- passage from an infected mother to her newborn during pregnancy or childbirth;
- blood or blood product transfusion from infected donors;
- some organ transplants using organs from infected donors; and
- Laboratory accidents.



An infected triatomine insect vector (or "kissing bug") takes a blood meal and releases trypomastigotes in its feces near the site of the bite wound. Trypomastigotes enter the host through the wound or through intact mucosal membranes, such as the conjunctiva.

1. Common triatomine vector species for trypanosomiasis belong to the genera *Triatoma*, *Rhodnius*, and *Panstrongylus*. Inside the host, the trypomastigotes invade cells near the site of inoculation, where they differentiate into intracellular amastigotes.
2. The amastigotes multiply by binary fission and
3. Differentiate into trypomastigotes, and then are released into the circulation as bloodstream trypomastigotes.
4. Trypomastigotes infect cells from a variety of tissues and transform into intracellular amastigotes in new infection sites. Clinical manifestations can result from this infective cycle. The bloodstream trypomastigotes do not replicate (different from the African trypanosomes). Replication resumes only when the parasites enter another cell or are ingested by another vector. The "kissing bug" becomes infected by feeding on human or animal blood that contains circulating parasites.
5. The ingested trypomastigotes transform into epimastigotes in the vector's midgut.
6. The parasites multiply and differentiate in the midgut and
7. Differentiate into infective metacyclic trypomastigotes in the hindgut.
8. *Trypanosoma cruzi* can also be transmitted through blood transfusions, organ transplantation, transplacentally (from mother to unborn baby), and in laboratory accidents.

Signs and symptoms

Chagas disease presents in 2 phases. The initial acute phase lasts for about 2 months after infection. During the acute phase, a high number of parasites circulate in the blood, but in most cases symptoms are absent or mild and unspecific. In less than 50% of people bitten by a triatomine bug, characteristic first visible signs can be a skin lesion or a purplish swelling of the lids of one eye. Additionally, they can present fever, headache, enlarged lymph glands, pallor, muscle pain, difficulty in breathing, swelling, and abdominal or chest pain.

During the chronic phase, the parasites are hidden mainly in the heart and digestive muscles. One to 3 decades later, up to a third of patients suffer from cardiac disorders and up to 1 in 10 suffer from digestive (typically enlargement of the oesophagus or colon), neurological or mixed alterations. In later years the infection in those patients can cause the destruction of the nervous system and heart muscle, consequent cardiac arrhythmias or progressive heart failure and sudden death.

Treatment

To kill the parasite, Chagas disease can be treated with benznidazole or nifurtimox. Both medicines are fully effective in curing the disease if given soon after infection at the onset of the acute phase, including the cases of congenital transmission. The efficacy of both diminishes, however, the longer a person has been infected and the adverse reactions are more frequent at older age. Treatment is also indicated for those in whom infection has been reactivated (for example, due to immunosuppression), and for patients during the early chronic phase, including girls and women of childbearing age (before or after pregnancy) to prevent congenital transmission.

Infected adults, especially those with no symptoms, should be offered treatment because antiparasitic treatment can also prevent or curb disease progression. In other cases, the potential benefits of medication in preventing or delaying the development of Chagas disease should be weighed against the duration of treatment (up to 2 months) and possible adverse reactions (occurring in up to 40% of treated adult patients). Benznidazole and nifurtimox should not be taken by pregnant women or by people with kidney or liver failure. Nifurtimox is also contraindicated for people with a background of neurological or psychiatric disorders. Additionally, specific treatment for cardiac, or digestive or neurological manifestations may be required.

Control and prevention

The large reservoir of *T. cruzi* parasites in wild animals of the Americas means that the infection cannot be eradicated. Instead, the control targets are elimination of the transmission to humans and early health-care access of the infected people.

There is no vaccine to prevent Chagas disease. *T. cruzi* can infect many species of triatomine bugs, the majority of which are found in the Americas. Vector control has been the most effective method of prevention in Latin America. Blood screening is necessary to prevent infection through transfusion and organ transplantation and to increase detection and care of the affected population all over the world.

Depending on the geographical area, WHO recommends the following approaches to prevention and control

- spraying of dwellings and surrounding areas with residual insecticides;
- house improvements and house cleanliness to prevent vector infestation;
- personal preventive measures such as bednets, good hygiene practices in food preparation, transportation, storage and consumption;
- development of contextualized information, education and communication activities for different actors and scenarios about preventative measures and surveillance tools (one health approach);
- screening of blood donors;
- testing of organ, tissue or cell donors and receivers;
- access to diagnosis and treatment of people with medical indication or recommendation to do antiparasitic treatment, especially children and women of child-bearing age before pregnancy; and
- Screening of newborns and other children of infected mothers without previous antiparasitic treatment to do early diagnosis and provide treatment.

The medical care cost of patients with chronic cardiac, digestive, neurologic or mixed forms of the disease has been calculated to be >80% higher than the cost of spraying residual insecticide to control vectors and prevent infection.

First level of care (Primary health care), with its different health professionals, and in interaction of other health levels, has a key role in increasing current detection, treatment, follow up and notification.

Assessment of the available diagnostics (including rapid serologic or chemiluminescence tests, molecular biology tests) and most cost-effective algorithms, per territory, is fundamental to increase case detection.

Promotion of biomedical, psychosocial and environmental studies of the determinants and risk factors of Chagas disease is basic to propose more effective multidimensional approaches to prevent and control de disease.

National information systems to monitor the number of acute and chronic cases and the active transmission routes are essential. So far, 6 out of the 44 countries with disease cases have implemented it.

Trichomonas foetus in cattle

Trichomoniasis is a venereal disease of cattle characterized primarily by early fetal death and infertility, resulting in extended calving intervals. Distribution is likely worldwide. Diagnosis is confirmed by isolation of the organism. Imidazoles have been used to treat infected bulls, but none is both safe and effective. Control is by culling infected bulls.

Etiology and Epidemiology of Trichomoniasis in Cattle

The causative protozoan of trichomoniasis, *Tritrichomonas foetus*, is pyriform and ordinarily 10–15 × 5–10 mcm, but there is considerable pleomorphism. It may become spherical when cultured in artificial media. At its anterior end, there are three flagella approximately the same length as the body of the parasite. An undulating membrane extends the length of the body and is bordered by a marginal filament that continues beyond the membrane as a posterior flagellum. Although *T. foetus* can survive the process used for freezing semen, it is killed by drying or high temperatures.

T. foetus is found in the genital tracts of cattle. When cows are bred naturally by an infected bull, 30%–90% become infected, suggesting that strain differences exist. Variation in breed susceptibility to trichomoniasis may also exist. Bulls of all ages can remain infected indefinitely, but this is less likely in younger males. By contrast, most cows are free of infection within 3 months after breeding. However, immunity is not longlasting and reinfection does occur. Transmission can also occur when the semen from infected bulls is used for artificial insemination.

Clinical Findings of Trichomoniasis in Cattle

The most common sign of trichomoniasis is infertility caused by embryonic death. This results in repeat breeding, and attending stock persons often note cows in heat when they should be pregnant. This, along with poor pregnancy test results (eg, too many “nonpregnant normal” and late-bred cows) is usually the presenting complaint. In addition to a reduced number of cows estimated to calve during the regular calving season, an increased number of cows with a “nonpregnant abnormal” reproductive tract diagnosis is seen. These include cows with pyometra, endometritis, or a mummified fetus.

Fetal death and abortions can also occur but are not as common as losses earlier in gestation. *T. foetus* has been found in vaginal cultures taken as late as 8 months of gestation and, apparently, live calves can be born to infected dams. Pyometra occasionally develops after breeding.

Diagnosis of Trichomoniasis in Cattle

Diagnosis is confirmed by culture of *T. foetus*

History and clinical signs are useful but are essentially the same as those of bovine genital campylobacteriosis. Confirmation depends on isolation of *T. foetus*, which may be difficult to differentiate from other trichomonads resident in the digestive tract. Diagnostic efforts are directed at bulls, because they are the most likely carriers. Suction is applied to a pipette while it is used to vigorously scrape the epithelium in the preputial fornix. Alternatively, douching with saline or lactated Ringer's solution (without preservatives) can be used. Aspirates or douches, concentrated by centrifugation, are examined using darkfield contrast microscopy. This material is also transferred immediately to the surface of a liquid culture medium such as Diamond medium. Better success culturing the organism has been reported when using commercially available media-filled pouches. In addition, incubating the media beyond the standard 48 hours may also enhance the accuracy of the diagnosis. Sampling every 48 hours for 10 days from the bottom of the tube and examining at 100–400× may reveal the rolling jerky movements of *T. foetus*.

Studies have examined the possibility of using PCR assays to identify *T. foetus* directly from the preputial samples without an intervening culture.

Studies suggest that 90%–95% of infected bulls will be positive on culture, and that three successive cultures at weekly intervals will detect ~99.5% of infected bulls. A vaginal discharge (after treatment of pyometra) or vaginal mucus (obtained toward the end of a luteal phase) may also be of diagnostic value.

Treatment and Control of Trichomoniasis in Cattle

- Imidazoles have been used to treat bulls, but none are both safe and effective
- Control is by culling infected bulls

Various imidazoles have been used to treat bulls with trichomoniasis, but none is both safe and effective. Ipronidazole is probably most effective but, due to its low pH, frequently causes sterile abscesses at injection sites. In addition, bulls are probably susceptible to reinfection after successful treatment. Resistance to ipronidazole may also be a concern. The biggest problem, however, is that the success of treatment is measured by repeated sampling, which may mean the individual bull can never be definitively called negative. Therefore, an unqualified recommendation for the bull's use cannot be given.

Control consists of eliminating the infection from the bull battery by culling all bulls and replacing them with virgin bulls or by testing and culling positive bulls. Repeated testing in older bulls may be unsatisfactory, and it may be prudent to cull them all. Reinfection is prevented by exposing only the uninfected (clean) bulls to uninfected (clean) cows. Clean cows are assumed to be those with calves at foot (even though some infected cows may produce a live calf) and virgin heifers. In situations in which several herds are commingled on the same range, caution must be exercised to ensure that

cows and heifers are not exposed to potentially infected bulls at the home ranch before they are turned out on the common grazing pasture.

T. foetus can be safely eliminated from semen with dimetridazole.

Vaccines developed some time ago for use in cows and evaluated in the field were not highly effective, especially in the absence of other control measures. However, the efficacy of whole-cell *T. foetus* vaccines has recently been critically reviewed. Although there is some evidence to suggest that timely vaccination will improve reproductive performance in heifers, there is a distinct lack of evidence that vaccination will reduce "bull-associated outcomes."

Biosecurity of Trichomoniasis in Cattle

Once a herd diagnosis of trichomoniasis has been made, all efforts to eliminate or treat the organism will probably be unsuccessful if significant biosecurity measures are not in place. Although *T. foetus* can be safely eliminated from semen with dimetridazole, using artificial insemination as a reproductive strategy to circumvent the use of bulls will not sufficiently decrease the risk of reinfection without a significant biosecurity strategy. The commingling of breeding groups, even within one ranch, may serve to maintain carrier cows and bulls. Groups of infected and potentially infected cattle must be kept distinct from “clean” breeding groups. Also, the addition of leased mature bulls or the purchase of mature cows may be enough of a biosecurity break to maintain the organism in the herd and lead to an outbreak of reproductive failure later on. The herd veterinarian should be able to create a herd-specific biosecurity protocol for the herd in question.

Probable questions:

1. Write down the systematic position of *Trypanosoma cruzi*.
2. Describe the morphology of *Trypanosoma cruzi* with diagram?
3. What is Trypomastigote stage in *Trypanosoma cruzi*?
4. What is amastigote stage in *Trypanosoma cruzi*?
5. Name the causative agent of Chagas disease.
6. How *Trypanosoma cruzi* is transmitted from one host to another?
7. Write down the sign and symptoms of Chagas disease.
8. How Chagas disease can be controlled?
9. Discuss the Epidemiology of Trichomoniasis in Cattle.
10. What are the Clinical Findings of Trichomoniasis in Cattle?

11. Describe the procedure by which Trichomoniasis can be diagnosed in Cattle.
12. Discuss about the Treatment and Control of Trichomoniasis in Cattle

Suggested reading:

1. https://www.who.int/news-room/fact-sheets/detail/chagas-disease-(american-trypanosomiasis)
2. Cheng, T. C. (1986). General Parasitology. 2nd ed. Academic Press, Inc. Orlando.U.S.A.
3. Noble, E. R. and Noble G. A. (1989). Parasitology. The Biology of animal Parasites. 6th edn.
4. https://www.msdvetmanual.com/reproductive system/trichomoniasis/trichomoniasis-in-cattle

UNIT IV

Outline structure and life cycle of *Plasmodium* spp

Objective:

In this unit we will learn about Outline structure and life cycle of *Plasmodium* spp

Introduction:

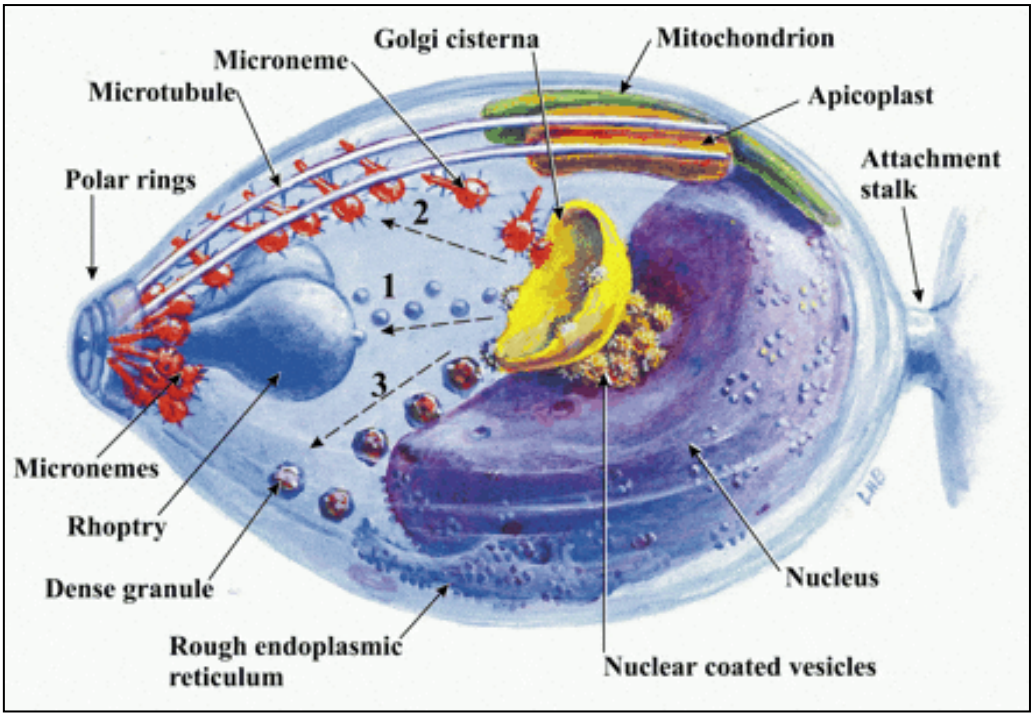
Plasmodium vivax is a protozoal parasite and a human pathogen. This parasite is the most frequent and widely distributed cause of recurring (Benign tertian) malaria; *P. vivax* is one of the five species of malaria parasites that commonly infect humans. Although it is less virulent than *Plasmodium falciparum*, the deadliest of the five human malaria parasites, *P. vivax* malaria infections can lead to severe disease and death, often due to splenomegaly (a pathologically enlarged spleen), *P. vivax* is carried by the female *Anopheles* mosquito, since it is only the female of the species that bites.

Structure of *Plasmodium* sp.

Plasmodium Falciparum is single-celled eukaryotes. Their adult stages are not motile, and their mode of movement is classified as Sporozoa. There are stages during its life cycle when it is motile and those stages include the sporozoite, trophozoite, and merozoite stages.

In *P. falciparum* infections, red blood cells (RBCs) are normal in size. Typically only rings and gametocytes are seen unless the blood sat before the smears were prepared.

1. **Rings:** *P. falciparum* rings have delicate cytoplasm and one or two small chromatin dots. RBCs that are infected are not enlarged; multiple infections of RBCs are more common in *P. falciparum* than in other species. Occasional appliqué forms (rings appearing on the periphery of the RBC) can be present.
2. **Gametocytes:** *P. falciparum* gametocytes are crescent or sausage shaped. The chromatin is in a single mass (macrogamete) or diffuse (microgamete).
3. **Trophozoites:** *P. falciparum* trophozoites are rarely seen in peripheral blood smears. Older, ring stage parasites are referred to as trophozoites. The cytoplasm of mature trophozoites tends to be denser than in younger rings. As *P. falciparum* trophozoites grow and mature, they tend to retain their ring-like shape and sometimes trace amounts of yellow pigment can be seen within the cytoplasm. Growing trophozoites in *P. falciparum* can appear slightly amoeboid in shape.
4. **Schizonts:** *P. falciparum* schizonts are seldom seen in peripheral blood. Mature schizonts have 8 to 24 small merozoites; dark pigment, clumped in one mass.



Structure of *Plasmodium* sp.

Epidemiology:

Plasmodium vivax is found mainly in Asia, Latin America, and in some parts of Africa. *P. vivax* is believed to have originated in Asia, but latest studies have shown that wild chimpanzees and gorillas throughout central Africa are endemically infected with parasites that are closely related to human *P. vivax.* These findings indicate that human *P. vivax* is of African origin. *Plasmodium vivax* accounts for 65% of malaria cases in Asia and South America. Unlike *Plasmodium falciparum*, *Plasmodium vivax* is capable of undergoing sporogonic development in the mosquito at lower temperatures. It has been estimated that 2.5 billion people are at risk of infection with this organism.

Although the Americas contribute 22% of the global area at risk, high endemic areas are generally sparsely populated and the region contributes only 6% to the total population at risk. In Africa, the widespread lack of the Duffy antigen in the population has ensured that stable transmission is constrained to Madagascar and parts of the Horn of Africa. It contributes 3.5% of global population at risk. Central Asia is responsible for 82% of globalpopulationatriskwithhighendemicareascoincidingwithdensepopulationsparticularlyin India and Myanmar. South East Asia has areas of high endemicity in Indonesia and Papua New Guinea and overall contributes 9% of global population at risk.

P. vivax is carried by at least 71 mosquito species. Many *vivax* vectors live happily in temperate climates—as far north as Finland. Some prefer to bite outdoors or during the day time, hampering the effectiveness of indoor insecticide and bed nets. Several key vector species have yet to be grown in the lab for closer study, and insecticide resistance is unquantified.

Systematic Position:

Phylum – Protozoa

Class – Sporozoa

Order – Haemosporidia

Genus – *Plasmodium*

Discovery:

Charles Laveran (1880) discovered *Plasmodium* in the blood of a malarial patient. In the year 1895 Ronald Ross, an Indian army doctor discovered the oocyte of *Plasmodium* in the stomach of the female *Anopheles* mosquito.

Mode of Infection:

When an infected female *Anopheles* mosquito bites a healthy person to suck blood, it injects the sporozoites (infective stage) into the human blood along with its saliva. Sporozoites are inoculated in thousands into the human blood. A sporozoite is microscopic, slender and sickle shaped animalcule.

Its body length is about 14 μ and breadth is 1μ Body is covered by a thin, elastic cuticle which gives a definite shape to its body. In the absence of any locomotory organelles the sporozoites show gliding movement. Two secretory glands are present at the anterior ends which are believed to help in penetration into the cell.

Causes

Malaria happens when a bite from the female *Anopheles* mosquito infects the body with *Plasmodium*. Only the *Anopheles* mosquito can transmit malaria. The successful development of the parasite within the mosquito depends on several factors, the most important being humidity and ambient temperatures. When an infected mosquito bites a human host, the parasite enters the blood stream and lays dormant with in the liver. The host will have no symptoms for an average of 10.5 days, but the malaria parasite will begin multiplying during this time.

The new malaria parasites are then released back into the bloodstream, where they infect red blood cells and multiply further. Some malaria parasites remain in the liver and are not released until later, resulting in recurrence. An unaffected mosquito becomes infected once it feeds on an infected individual. This restarts the cycle.

Life cycle and Transmission routes:

The life cycle of *Plasmodium* is completed inside the body of the two hosts such as male and female *Anopheles* mosquito. Hence, the life cycle is digenetic. Man is the primary or definitive host and female *Anopheles* mosquito is the secondary or intermediate host or vector.

The asexual life cycle in man is called schizogony. It is divided in to 3 phases

(a) Pre-erythrocytic schizogony,

(b) exo-erythrocytic schizogony,

(c) erythro-cytic schizogony.

The sporozoite is the infective stage. While sucking blood an infected mosquito injects the sporozoites into the body of man. The sporozoites enter the liver to carry on pre and exo-erthrocytic schizogony.

Through these two cycles large number of merozoites is produced. They enter R.B.C. to carry on erythrocytic schizogony. This erythrocytic cycle is repeated in every 48 hours which coincides with the appearance of the symptoms of malaria. Some of the merozoites produced by erythrocytic cycle never attack fresh R.B.C. They wait for the mosquito to be sucked. They are of two types: male and female gametocytes. They carry on sexual reproduction inside the stomach of mosquito.

The main mode of transmission of the disease is by bites from infected *Anopheles* mosquitoes that have previously had a blood meal from an individual with parasitemia. The human infection begins when an infected female anopheles mosquito bites a person and injects infected with sporozoites saliva into the blood circulation. That is the first life stage of *plasmodium* (stage of infection).

The next stage in malaria life cycle is the one of asexual reproduction that is divided into different phases: the pre-erythrocytic (or better, exo-erythrocytic) and the erythrocytic phase. Within only 30- 60 minutes after the parasites inoculation, sporozoites find their way through blood circulation to their first target, the liver. The sporozoites enter the liver cells and start dividing leading to schizonts creation in 6- 7days. Each schizont gives birth to thousands of merozoites (exoerythrocytic schizogony) that are then released into the bloodstream marking the end of the exoerythrocytic phase of the asexual reproductive stage. The liver phase occurs only once while the erythrocytic phase undergoes multiple cycles; the merozoites release after each cycle creates the febrile waves. A second scenario into the RBCs is the parasite differentiation into male and female gametocytes that is a non pathogenic form of parasite. When a female anopheles mosquito bites an infected person, it takes up these gametocytes with the blood meal (mosquitoes can be infected only if they have a meal during the period that gametocytes circulate in the human's blood).The gametocytes, then, mature and become microgametes (male) and macrogametes (female) during a process known as gametogenesis. The time needed for the gametocytes to mature differs for each

Plasmodium species: 3- 4 days for *P. vivax* and *P. ovale*, 6- 8 days for *P. malariae* and 8- 10 days for *P. falciparum*.

In the mosquito gut, the microgamete nucleus divides three times producing eight nuclei; each nucleus fertilizes a macro gamete forming a zygote. The zygote, after the fusion of nuclei and the fertilization, becomes the so- called ookinete. The ookinete, then, penetrates the mid gut wall of the mosquito, where it encysts into a formation called oocyst. Inside the oocyst, the ookinete nucleus divides to produce thousands of sporozoites (sporogony). That is the end of the third stage (stage of sexual reproduction/ sporogony). Sporogony lasts 8- 15days**.**

The oocyst ruptures and the sporozoites are released inside the mosquito cavity and find their way to its salivary glands but only few hundreds of sporozoites manage to enter. Thus, when the above mentioned infected mosquito takes a blood meal, it injects its infected saliva into the next victim marking the beginning of a new cycle.

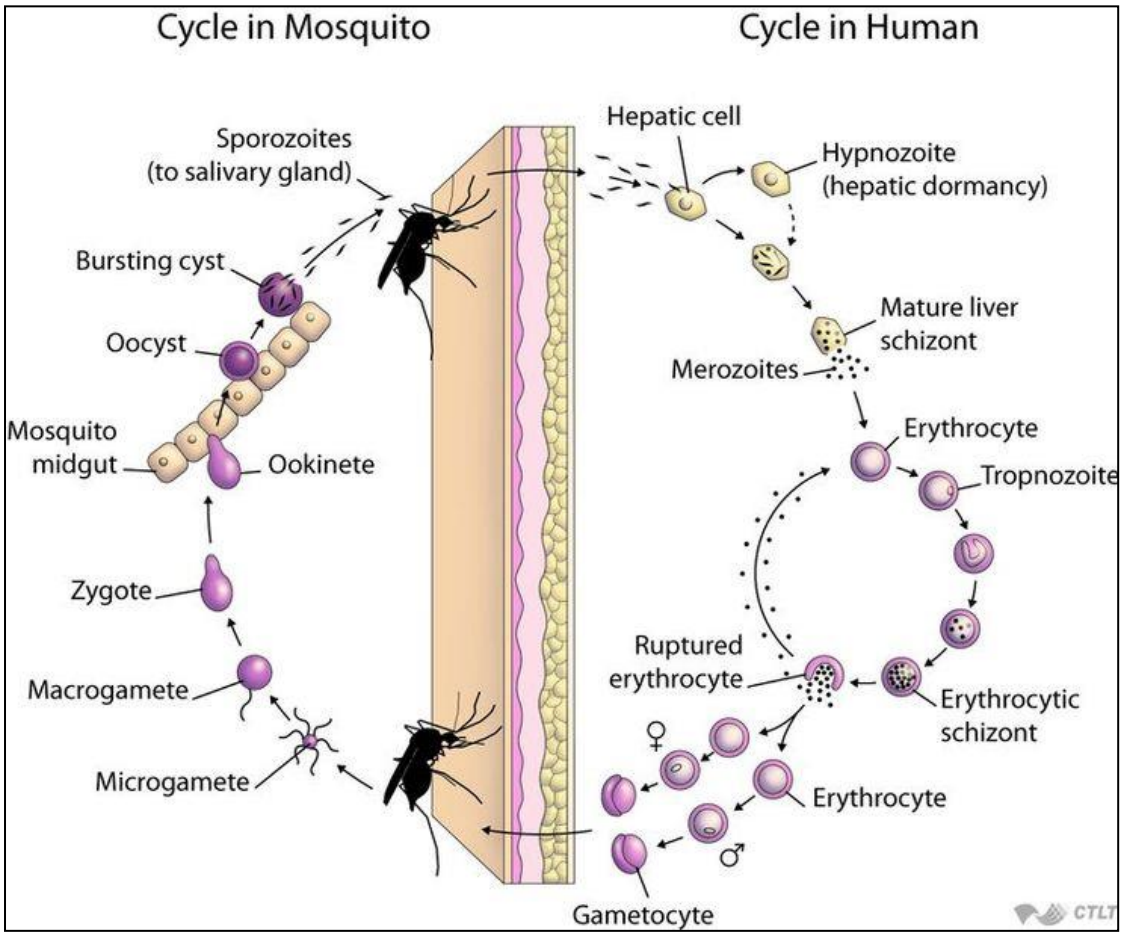


Fig: Life cycle of *Plasmodium* sp.

Clinical representation:

Pathogenesis results from rupture of infected red blood cells, leading to fever. Infected red blood cells may also stick to each other and to walls of capillaries. Vessels plug up and deprived issues of oxygen. Infection may also cause the spleen to enlarge.

Unlike *P. falciparum*, *P. vivax* can populate the bloodstream with sexual-stage parasites—the form picked up by mosquitoes on their way to the next victim—even before a patient shows symptoms. Consequently, prompt treatment of symptomatic patients doesn't necessarily help stop an outbreak, as it does with falciparum malaria, in which fevers occur as sexual stages develop. Even when symptoms appear, because they are usually not immediately fatal, the parasite continues to multiply.

The parasite can go dormant in the liver for days to years, causing no symptoms and remaining undetectable in blood tests. They form what are called hypnozoites (the name derives from "sleeping organisms"), a small form that nestles inside an individual liver cell. The hypnozoites allow the parasite to survive in more temperate zones, where mosquitoes bite only part of the year.

Sign and Symptoms:

A single infectious bite can trigger six or more relapses a year, leaving sufferers more vulnerable to other diseases. Other infectious diseases, including falciparum malaria, appear to trigger relapses.

- **Uncomplicated malaria**

This is diagnosed when symptoms are present, but there are no signs to indicate severe infection or dysfunction of the vital organs. This form can become severe malaria if left untreated, or if the host has poor or no immunity. Symptoms of uncomplicated malaria typically last 6 to 10 hours and recur every second day. Some strains of the parasite can have a longer cycle or cause mixed symptoms. As symptoms resemble those of flu, they may be undiagnosed or misdiagnosed in areas where malaria is less common. In uncomplicated malaria, symptoms progress as follows, through cold, hot, and sweating stages:

i. A sensation of cold with shivering
ii. Fever, headaches, and vomiting
iii. Seizures sometimes occur in younger people with the disease
iv. Sweats, followed by a return to normal temperature, with tiredness

In areas where malaria is common, many patients recognize the symptoms as malaria and treat themselves without visiting a doctor.

- **Severe malaria**

In severe malaria, clinical or laboratory evidence shows signs of vital organ dysfunction. Symptoms of severe malaria include:

i. Fever and chills
ii. Impaired consciousness
iii. prostration, or adopting a prone position
iv. multiple convulsions
v. deep breathing and respiratory distress
vi. abnormal bleeding and signs of anemia
vii. clinical jaundice and evidence of vital organ dysfunction

Prevention

The main way to prevent malaria is through vector control. There are mostly three main forms that the vector can be controlled:

(1) insecticide-treated mosquito nets,

(2) Indoor residual spraying and

(3) antimalarial drugs.

Long-lasting insecticidal nets (LLNs) are the preferred method of control because it is the most cost effective. The WHO is currently strategizing how to ensure that the net is properly maintained to protect people at risk. The second option is indoor residual spraying and has been proven effective if at least 80% of the homes are sprayed. However, such method is only effective for 3-6months. A drawback to these two methods, unfortunately, is that mosquito resistance against these insecticides has risen. National malaria control efforts are undergoing rapid changes to ensure the people are given the most effective method of vector control. Lastly, antimalarial drugs can also be used to prevent infection from developing into a clinical disease. However, there has also been an increase resistance to antimalarial medicine.

In 2015 the World Health Organization (WHO) drew up a plan to address vivax malaria, as part of their Global Technical Strategy for Malaria.

Probable questions:

1. Describe the general structure of Plasmodium *sp.*with diagram.
2. Elaborate the mode of infection of *Plasmodium*?
3. What is the symptom of malaria?
4. What is spleenomegaly?
5. Discuss the treatment of malaria.

Suggested reading:

1. Cheng, T. C. (1986). General Parasitology. 2nd ed. Academic Press, Inc. Orlando.U.S.A.
2. Noble, E. R. and Noble G. A. (1989). Parasitology. The Biology of animal Parasites. 6th edn.
3. Roberts, L. S., Janovy, J. and Nadler S. (2013) Gerald D. Schmidt &Lary S. Roberts'Foundation of Parasitology. 9th ed. McGraw-Hill International.

UNIT V

Trichuris trichiura– biology, epidemiology, pathogenesis, diagnosis and treatment

Objective:

In this unit we will discuss about biology, epidemiology, pathogenesis, diagnosis and treatment of *Trichuris trichiura*.

Introduction:

Trichuris trichiura commonly called the whipworm because of its characteristic whip-like shape. It causes trichosis in human which is an intestinal infection caused by invasion of the colon by the adult worm.

Habitat:

The adult worm lives in the large intestine (caecum) of human. It can also be present in the vermiform appendix and rectum.

Morphology

- The adult worms resemble the whip. The anterior $3/5^{th}$ of the end is very thin hair like and the posterior $2/5^{th}$ is thick and stout resembling the handle of a whip.
- The anterior ends penetrate the mucosa layer and remain deeply embedded.
- Adult worms are pinkish in color.
- The anterior end consists of a long oesophagus which is a minute channel while the posterior end contains the intestine and sex organs.

Male worm:

- The adult males are 3-4 cm in length and are recognized by their characteristic coiled posterior end.

Female worm:

- Female worms are longer than males measuring 4-5 cm in length.
- The caudal extremity is either comma shaped or an arc shaped

Eggs

- Egg of *Trichuris trichiura* has diagnostic value. Egg is barrel shaped with a mucous plug at each end.
- It is brown colored (bile-stained) and has a double shell.
- The egg measures 50-54 µm in length and 22-23 µm in breadth.
- Eggs contain an un-segmented ovum when it leaves the human hosts.
- The freshly passed eggs are non-infective to human.
- Eggs float in saturated NaCl solution.
-

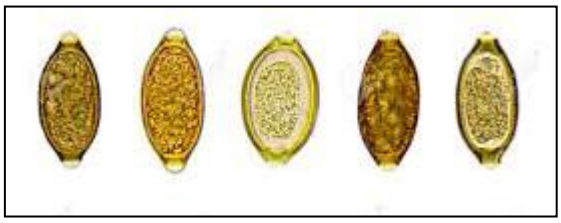


Fig: Egg structure of *Trichuris trichiura*

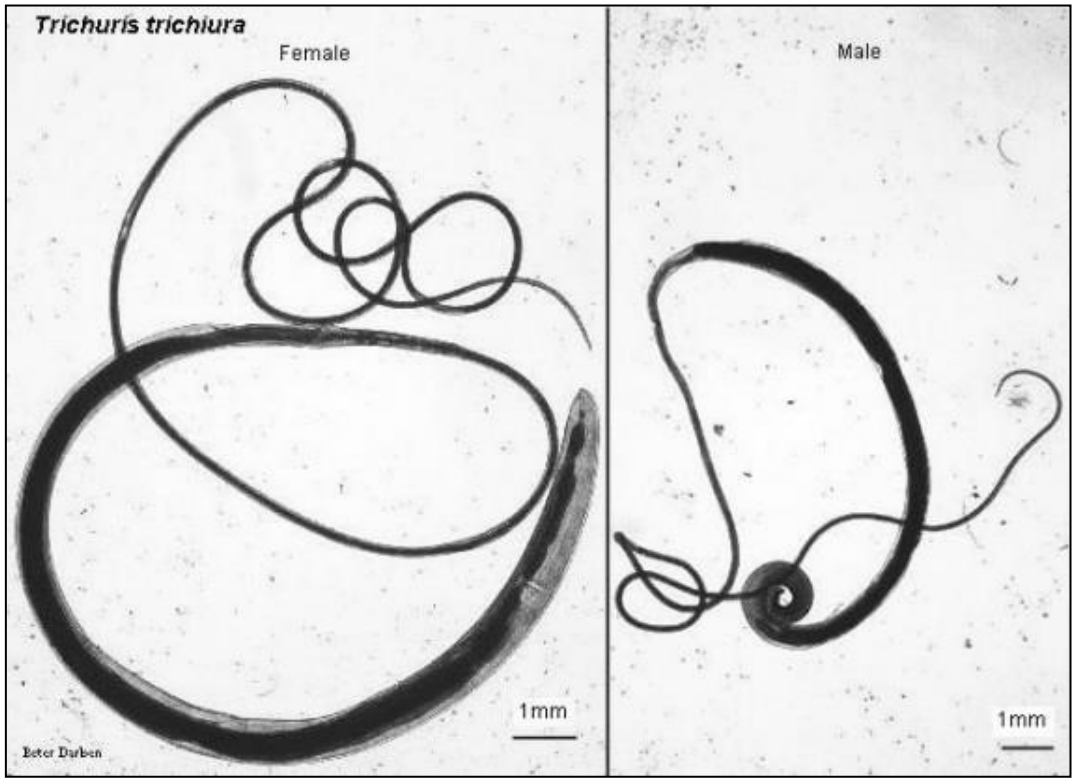


Fig: Male and female adult worm structure of *Trichuris trichiura*

Life cycle of *Trichuris trichiura*:

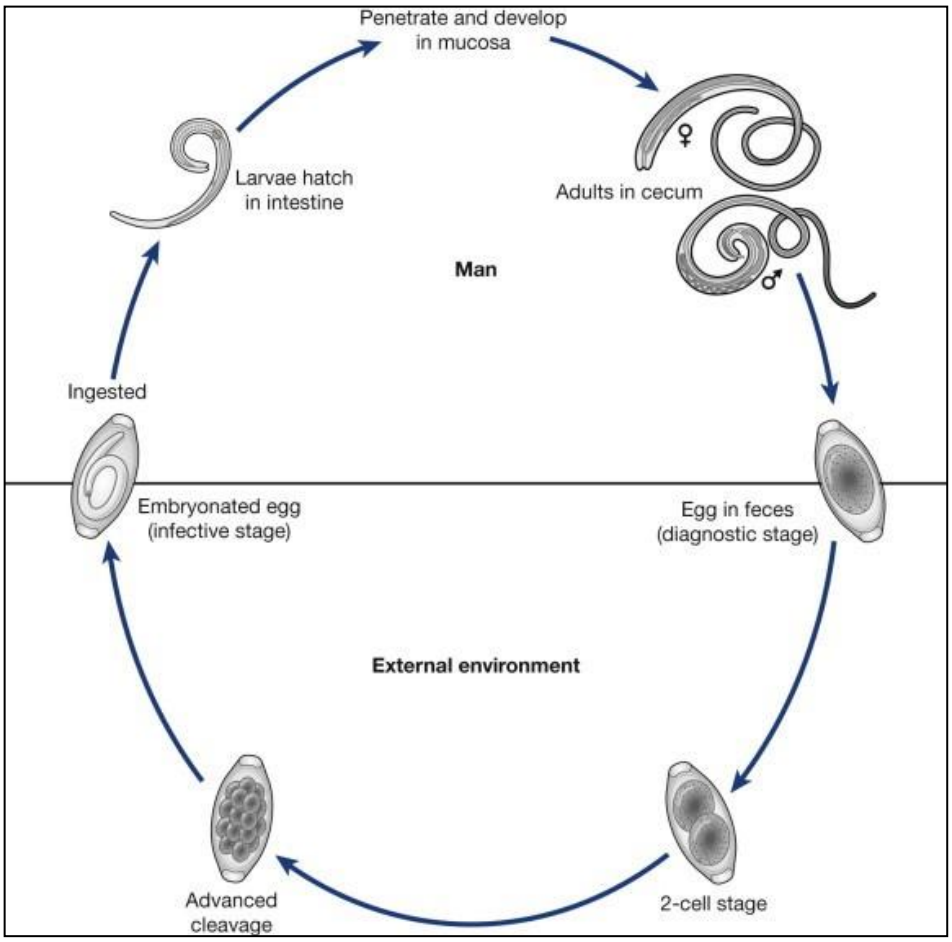


Fig: Life cycle of *Trichuris trichiura*

- The life cycle of *T. trichiura* is simple and is completed in single host, the man. However, change of host is needed for the continuation of species.
- No intermediate host is required.
- Human acquires infection by ingestion of food or water contaminated with embryonated eggs.
- The digestive enzymes dissolve the eggs shell and the larva emerges out through one of the poles of the eggs.
- The liberated larva then passes down into the caecum which is their site of the localization.
- In caecum the larvae develops into adult worm and become sexually mature within a month from the time of ingestion of eggs.
- The female worm after being fertilized by the male begins to lay the eggs, which is about 3 month after infection.

- The freshly laid eggs are un-embryonated and excreted out with the faeces.
- Each adult female can produce about 5000-7000 eggs per day for upto 5 years.
- Embryogenesis within eggs occurs in outside environment in the water or damp soil.
- In tropical climates larva develops within the egg in the course of 3-4 weeks. In temperate climates, the larva takes a long time (6-12 month) to complete its development.
- Once the egg is embryonated, it is infective to human.

Mode of transmission:

1. The food, water and soil contaminated with embryonated eggs are the chief sources of infection.
2. Ingestion of embroynated eggs in the contaminated food and water
3. Contaminated fingers during soil works

Pathogenesis

- The adult worm invades the intestinal mucosa by its thin, thread like anterior end and feeds on tissue secretions but not on blood.
- It causes petechial hemorrhage, inflammation, oedema and mucosal bleeding in the intestinal mucosa at their site of attachment.
- The lumen of appendix can be blocked in case of severe worm load.
- Presence of worms in the mucus membrane irritates the nervous plexus of mucosa causing diarrhea and cramps.
- Occasional eosinophilia can be present
- Approximately 0.005 ml of blood per worm per day is lost in the infected man.

Epidemiology

The egg of the whipworm is the infective stage, and favorable conditions for its maturation are a warm and humid climate. This is why most of the disease burden is seen in tropical climates, specifically in Asia and less often, in Africa and South America. It is also found in rural parts of the southeast United States. It is estimated that worldwide there are between 450 million to 1 billion active cases with most diagnosed in children. It is thought there is partial protective immunity that develops with age.

Worldwide, almost half of the 5 billion people that live in developing countries are infected with at least one soil-transmitted helminth species, and 10% with two or more helminth species. Young boys tend to be most affected as they are more likely to play outside and exhibit pica behavior.

Pathophysiology

The worm is acquired through fecal-oral transmission. A human host consumes infected eggs, typically while eating and drinking contaminated food or water. Once the embryonated eggs are ingested, the larvae hatch in the small intestine. From there they migrate to the large intestine, where the anterior ends lodge within the mucosa. This leads to cell destruction and activation of the host immune system, recruiting eosinophils, lymphocytes, and plasma cells. This causes the typical symptoms of rectal bleeding and abdominal pain. The parasite usually takes up residence in the terminal ileum and cecum. In some patients, the entire colon and rectum may be infested with the worm. The worm may live anywhere from 1-4 years without treatment. Eggs are expelled in the host feces unembryonated. The eggs will become embryonated in 2–4 weeks and are then infective.

Clinical manifestation

- The clinical manifestation of *Trichuris trichiura* depends upon the intestinal worm load of the person.
- Infection is asymptomatic in case of light infection with 100-200 worms.
- For moderate infection the number of worm should be more than 200 worms and this can manifest as vague abdominal discomfort and diarrhea (rarely bloody), vomiting, headache etc.
- **Trichuriasis**: In case of heavy infection with more than 800 worms, serious complications especially in children are observed.
- It causes bloody diarrhea with profuse mucus, abdominal pain and weight loss leading to the cachexia, severe anaemia.
- Distribution of a large number of worms throughout the colon and rectum may cause prolapse of the rectum.
- Migrating worms can occasionally cause appendicitis.

Diagnosis

- Specimen: stool, blood
- **Microscopy**:

- Finding of characteristic barrel-shaped eggs in the faeces on light microscopy.
- Stool concentration methods may be required to detect light infection
- Adult worms may occasionally be present in the stool.
- The degree of infection can be determined by egg count.
- In heavy infection stool is frequently mucoid and contains charcot-Leyden crystals.

- **Proctoscopy**: adult worm can be obtained from rectal mucosa sample.
- **Blood test**: shows eosinophilia

Treatment

- Mebendazole- drug of choice
- Albendazole
- Ivermectin

The suggested dose of mebendazole is 100 mg twice a day for 3 days or albendazole is 200 to 400 mg twice a day for 3 days. Mebendazole has been shown to be more effective and is considered first-line treatment. Albendazole and mebendazole have an inhibitory effect on tubulin polymerization which results in the loss of cytoplasmic microtubules. Ivermectin 200 mcg/kg daily can be used; however, it is not as effective as mebendazole and albendazole.

Probable questions:

1. State the scientific name of whipworm. Mention the habitat of whip worm.
2. Describe the egg structure of *Trichuris trichiura* .
3. Describe the morphological structure of *Trichuris trichiura* with diagram.
4. Describe the life cycle of *Trichuris trichiura* with proper diagram.
5. Which stage of Trichuris trichiura is infective?
6. What is the natural host for Trichuris trichiura?
7. State the mode of transmission of *Trichuris trichiura*.
8. State the pathogenesis of *Trichuris trichiura* .
9. What is Trichuriasis? Mention its symptoms.

10. Describe the Clinical manifestation of *Trichuris trichiura* infestation.
11. How trichuriasis can be diagonosed?

Suggested reading:

1. Izurieta, R., Reina-Ortiz, M. and Ochoa-Capello, T. (2018). Trichuris trichiura. In: J.B. Rose and B. Jiménez-Cisneros (eds), Water and Sanitation for the 21st Century: Health and Microbiological Aspects of Excreta and Wastewater Management (Global Water Pathogen Project). (L. Robertson (eds), Part 3: Specific Excreted Pathogens: Environmental and Epidemiology Aspects - Section 4: Helminths), Michigan State University, E. Lansing, MI, UNESCO.
2. Amoah, I.D., Singh, G., Stenström, T.A. and Reddy, P. (2017). Detection and quantification of soil-transmitted helminths in environmental samples: A review of current state-of-the-art and future perspectives. Acta Tropica. 169, pp. 187-201.
3. Global Health, Division of Parasitic Diseases and Malaria
4. Cheng, T. C. (1986). General Parasitology. 2nd ed. Academic Press, Inc. Orlando.U.S.A.
5. Noble, E. R. and Noble G. A. (1989). Parasitology. The Biology of animal Parasites. 6th edn.
6. Roberts, L. S., Janovy, J. and Nadler S. (2013) Gerald D. Schmidt &Lary S. Roberts' Foundation of Parasitology. 9th ed. McGraw-Hill International.

UNIT VI

Life cycle, biology, pathogenesis, epidemiology and control of *Loa loa, Dracunculus medinensis, Haemonchus contortus, Sarcoptes scabiei*

Objective:

In this unit we will discuss about Life cycle, biology, pathogenesis, epidemiology and control of *Loa loa, Dracunculus medinensis, Haemonchus contortus, Sarcoptes scabiei*.

Introduction:

Biology means the study of living organisms, divided into many specialized fields that cover their morphology, physiology, anatomy, behaviour, origin, and distribution. This field deals with all the physicochemical aspects of life.

Life cycle indicates**,** in biology, the series of changes that the members of a species undergo as they pass from the beginning of a given developmental stage to the inception of that same developmental stage in a subsequent generation.

Pathogenesis is defined as the origination and development of a disease. Insights into disease etiology and progression, the two major aspects of pathogenesis, are paramount in the prevention, management and treatment of various diseases.

Epidemiology is the study of how often diseases occur in different groups of people and why. Epidemiological information is used to plan and evaluate strategies to prevent illness and as a guide to the management of patients in whom disease has already developed. Like the clinical findings and pathology, the epidemiology of a disease is an integral part of its basic description. The subject has its special techniques of data collection and interpretation, and its necessary jargon for technical terms.

Loa loa

Loa loa filariasis

- Loa loa is a blood dwelling nematode that is parasitic in humans. The adult worm wanders through the subcutaneous tissue but is most obvious as it crosses the conjunctiva of the eye, hence leading to its common name, the African eye worm. It causes loa loa filariasis (loiasis). It is one of three parasitic filarial nematode that causes subcutaneous filariasis in humans.

Habitat

- The adult worm inhibits the subcutaneous tissue of man, often in the sub conjuctival tissue of the eye. The microfilariae are found in blood.

Morphology

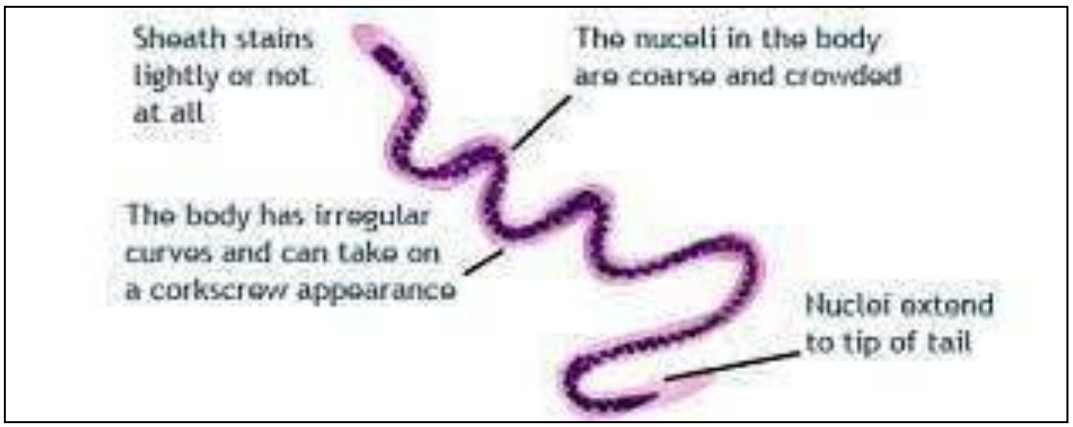


Fig: Morphology of *Loa loa*

Adult worm

- The adult worms are thin, whitish and thread like.
- The anterior and tapers to a narrow head. Surface of the body is covered with small knobs.
- Microscopically the cuticula is found to have numerous rounded protuberances (cuticular bosses) which vary in number and arrangement in two sexes.
- The female worm is 4-7 cm in length and 0.5mm in diameter.
- The life span of worm is 4-12 years.

Microfilaria

- They are found in peripheral blood during day time. Occasionally microfilaria have been demonstrated in the urine, sputum and even CSF.
- Microfilaria is sheathed and measures 250-300 mm in length and 6-8 mm in breadth.
- The column of nuclei extends upto the tail-tip.
- Sheath stains poorly with Geimsa stain but stains well with iron-haematoxylin.

Life cycle of *Loa loa*

- *Loa loa* completes its life cycle in two hosts:
- **Definitive host:** Human

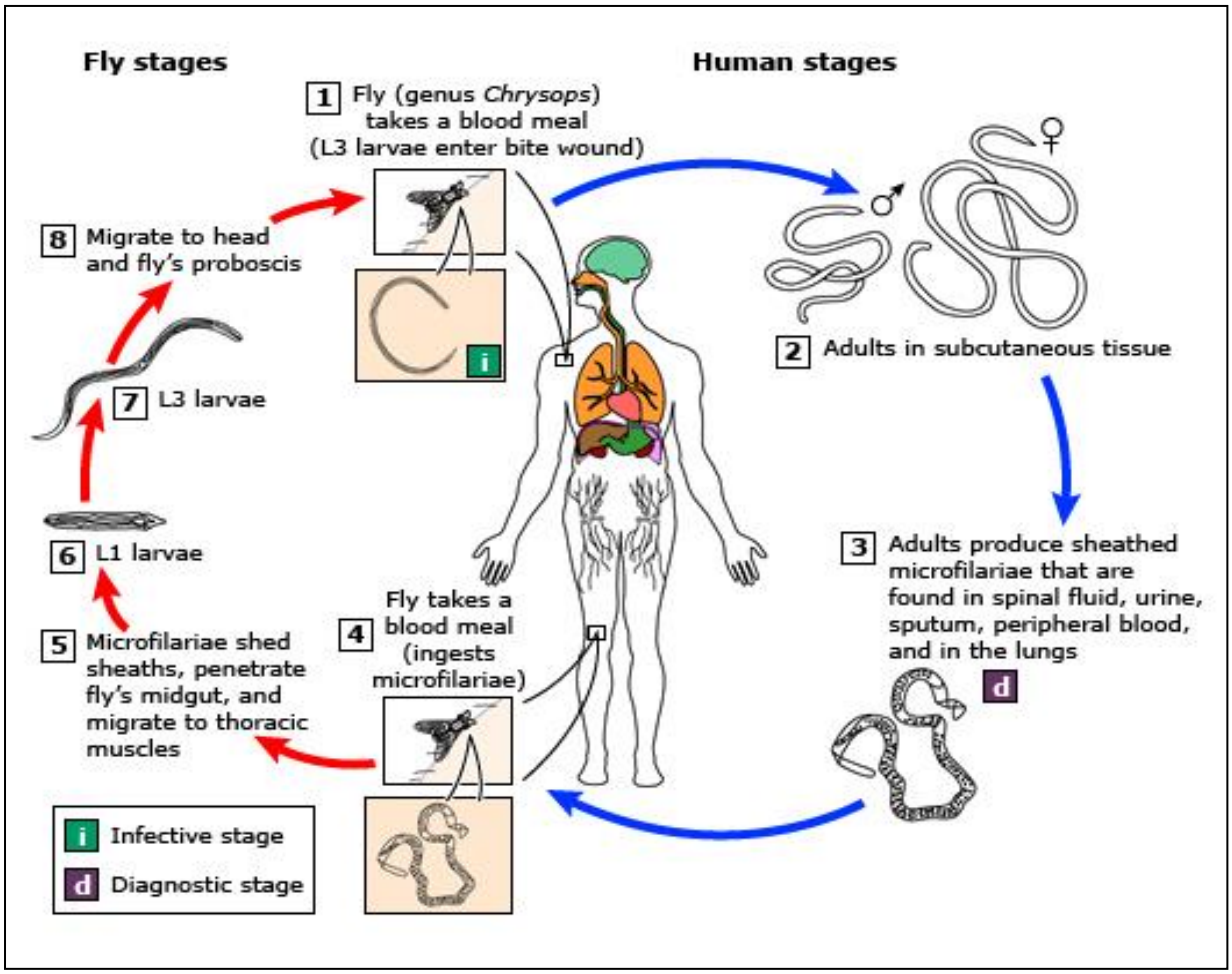


Fig: Life cycle of *Loa loa*

- Man acquires infection by the bite of infected female chrysops. During infection larva enter in large numbers through the punctured wound on the skin made by the fly, during the blood meal.
- The larva enters the subcutaneous tissue and mount to develop into adult worms within a period of 6-12 months.
- Adult worm occasionally migrate in the sub conjunctival tissue.
- The female worm after fertilize by males produce microfilarial larva that circulate in the peripheral blood during the day time and also found in the sub-cutaneous tissue.

- When a female chrysops bites the infected human to suck the blood, microfilaria is ingested that enter the fly's stomach.
- Microfilarial larvae lose their sheath, penetrate the wall of the stomach and invade thoracic muscles where they undergo changes to form the infective L3 larva.
- Development in fly is completed in about 10 days.
- The mature infective larva than migrate to the mouth parts of chrysops. When this fly bites a new host for blood meal, the cycle is again repeated.

Mode of transmission of *Loa loa*:

- Infected man is the only source and reservoir of infection for loa loa.
- Transmission is acquired by the bite of female chrysops species

Pathogenesis and pathology of *Loa loa*:

- Microfilaria is not pathogenic.
- Adult loa loa worms which live in the subcutaneous tissue are pathogenic.
- The migrating adult worms provoke an intense inflammatory reaction.
- **Calabar swelling** is the typical pathological feature of the loa loa filariasis. It is formed as a result of an allergic response to adult worms migrating in the subcutaneous tissue.

Clinical manifestation of *Loa loa* filariasis:

- Loa loa filariasis (loasis) is the disease produced by adult worm in human.
- The incubation period is on an average 3-4 years.
- Loiasis is asymptomatic in many pcases.

Skin lesions

- Skin lesions consist of colabar swelling or fugitive swelling.
- During migration of the adult worm, it causes oedema of subcutaneous tissues, known as calebur swelling. They disappear in course of 2-3 days and are regarded as allergic reaction of the tissues to filarial toxins.
- Localized pain and itching for several hours usually precede the onset of the swelling. Usually one swelling develops at a time. The swelling is non erythematous, measures 3-10 cm in diameter and last for few days to weeks. The wrist joints or knee joints are most frequently affected. Worms are not usually present in the swellings but are present below surface of the skin.

Ocular lesions

- These consists of;
- **Conjunctival granuloma:** It is caused by the migration of adult worms in the sub-conjunctival tissues. These granulomas are present as solitary or multiple small nodules measuring 2mm in diameter. These are found in the deeper layers of conjunctiva close to the sclera tissue.
- **Oedema of the eyelid:** It is painless condition frequently accompanied by itching but not fever or any other constitutional symptoms.
- **Proptosis:** This condition is known as 'bug eye' or 'bulge eye' caused by the edema of the orbital cellular tissue. It is a painless condition and of rapid onset frequently associated with itching.

Complications of loasis:

- These include frequent recurrence of fugitive swellings, endomyocardial fibrosis, retinopathy, encephalopathy, neuropathy and arthritis.
- Loa loa meingoencephalopathy is a severe and often fatal complication of infection.

Epidemiology of *Loa loa*

Loa loa parasites are found in West and Central Africa. Ten countries have areas where there are high rates of infection (i.e., where more than 40% of the people who live in that area report that they have had eye worm in the past). An estimated 14.4 million people live in these areas of high rates of infection. Another 15.2 live in areas where 20–40% of people report that they have had eye worm in the past.

Diagnosis of *Loa loa*:

- Clinical diagnosis is suggested in patients with typical fugitive swellings, high eosinophilia and history of residence in an area endemic for the disease
- Specific diagnosis is made by demonstration and identification of microfilaria in the peripheral blood
- Demonstration of the worm in the cornea or over the bridge of the nose.
- Identification of the adult worm surgically removed from the skin or conjunctiva.

Treatment of *Loa loa*:

- **Diethylcarbamazine(**DEC) is the drug of choice.
- Dose – 6mg/day, 3 times daily for 12 days

Prevention and control of *Loa loa*:

- Treatment of infected populations
- Using insect repellent
- Wearing protective clothing
- Avoiding visit to the places endemic for the disease

Dracunuculus medinensis

Introduction

- *Dracunuculus medinensis* is among the longest nematodes affecting humans.
- It is also known as Guinea worm, Medina worm, serpent worm or dragon worm.
- It causes dracunculiasis or dracontiasis, a nodular dermatosis produced by the development of *Dracunculus* parasite in the subcutaneous tissue.

Habitat:

- The adult females inhabit the subcutaneous tissue usually of the foot or lower limbs.
- Less frequently they also inhabit other parts of the body including the head and neck.

Morphology:

Adult worms

- **Male:**
  - Male worms are difficult to demonstrate as they are immediately after fertilizing the females. Hence, it has not yet been recovered from man.
  - The male measures 12-30 mm in length and 0.4 mm in breadth.
- **Female:**
  - It is slender long worm and is one of the longest nematodes known to cause infection in man.
  - It measures 60 cm to 1 mm or more in length and 1.5 to 1.7 mm in diameter, resembling a piece of long twine thread.
  - The body is cylindrical, smooth and milk white in color.

- The posterior end is extremely tapering and is bent to form a hook.
- A minute triangular mouth is present in an anterior end.
- An inner layer of 4 papillae-6 papillae and outer layer of 4 pairs of papillae surrounded the mouth.
- A pair of uteri, oviducts and tubules and a single unpaired vagina constitutes the female genital tract.
- The worm is ovoviviparous and discharge embryos in successive batches for a period of about 3 weeks until the gravid female completely implies its uterine contents.
- The body fluid is toxic and causes a bluster if female escapes into the tissue.The life span of female is about 1 year and that of the male is not more than 6 months.

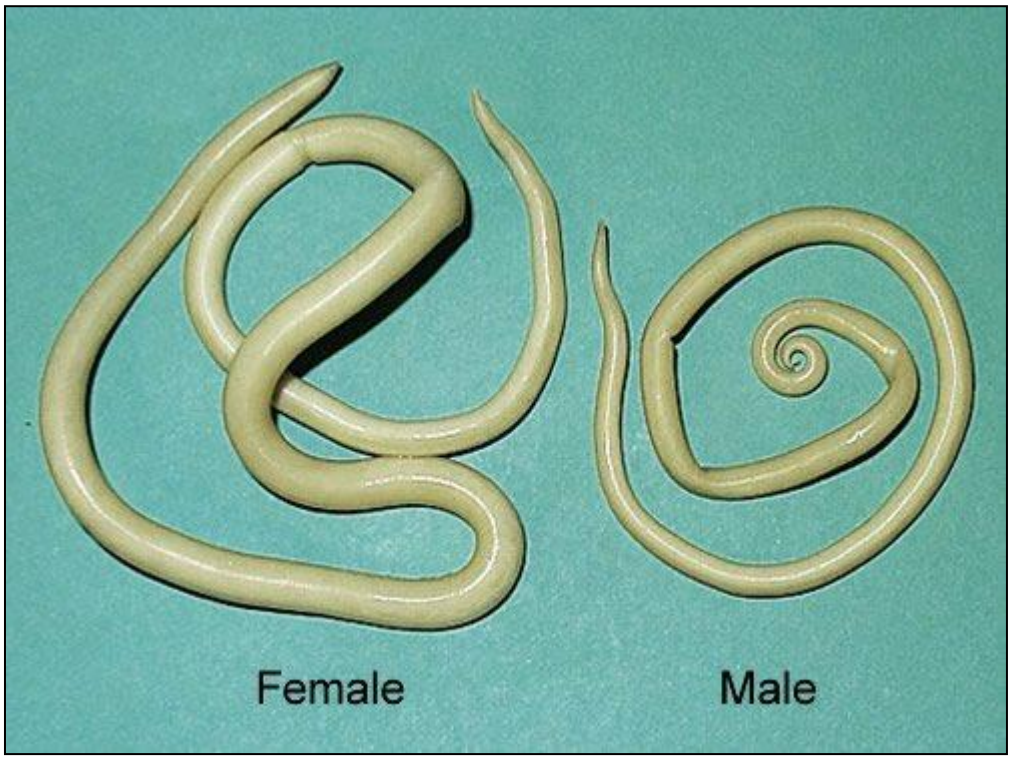


Fig: Morphology of *Dracunuculus medinensis*

larva:

- **First stage Larva**
  - It is unsheathed and coiled with a round anterior end and a long slender filariform tail.
  - It is large, measuring 650-750 mm in length and 17-20 mm in breadth.
  - The cuticle is conspicuously striated.

- It moves about with a shift motion, briskly coiling and uncoiling body.
- It shows a tad-pole like movement in water.
- These larva are set free only at the time of partition when the affected part is submerged in water.
- Further development proceeds in the body of a minute fresh water crustacean of the genus Cylops.

Fig: Larva of *Dracunuculus medinensis*

- **Third stage of larva:**
  - It is the infective stage of the parasite.
  - It is found in the body cavity of Cyclops.

Life cycle of *Dracunuculus medinensis* :

- Lifecycle is completed in two hosts:
  - Definitive host: Human
  - Intermediate host: Cyclops and other twelve species.
- **Entrance into man and development into adult worms:**
  - Man acquires infections by drinking unfiltered water containing infected Cyclops.
  - On reaching the stomach, the cyclopses are digested by the gastric juices and L3 larva is liberated.
  - The larva penetrated the gut wall and enters the retroperitoneal connective tissue where they grow and becomes sexually mature.

- The males die after fertilizing the females and disappear within 6 months of infection.
- The gravid female migrates and selects those parts of the skin labile to come in contact with water such as the backs of water carriers, anus and legs of washer men and legs of those who fill water in containers in 'step wells' and ponds.
- On reaching the skin surface, it secretes a toxin, producing a blister which later ruptures and forms an ulcer, contact with water stimulates the worm to protrude its head through the center of the ulcer and causes a reflex discharge of a milky fluid containing large number of first stage rhabditiform larva.

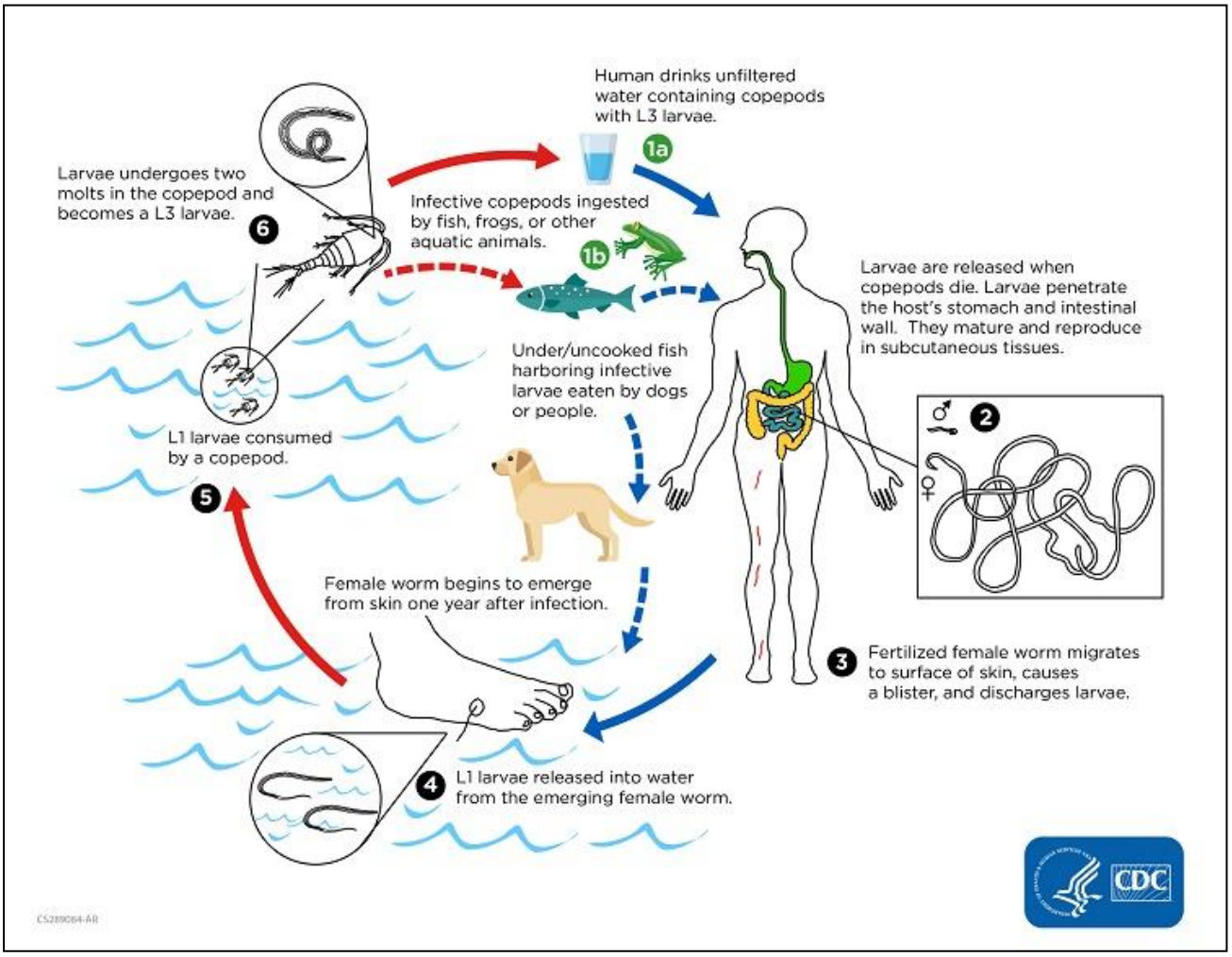


Fig: Life cycle of *Dracunuculus medinensis*

- **Development of larva in Cyclops:**
  - The larva discharged in water swims vigorously in water with tad-pole like movement.
  - These larvae further develop when ingested by suitable Cyclops species.

- In the body cavity of Cyclops, the first stage larva moults twice to develop into third stage larva, the infection from the parasite.
- These infected Cyclops have short life span than normal non infected ones.
- The infected Cyclops harboring third stage larva are infective to man and the cycle is repeated.

Mode of transmission:

- Water containing infected Cyclops is the main source of infection.
- Man acquires infections by drinking water contaminated with Cyclops harboring third stage larva.

Pathogenesis:

- The third stage larva are not pathogenesis and do not produce any pathological lesions.
- Only female adult worm is pathogenic.
- It produces a toxin, forming blister which is formed at the site at which the female worm comes out of the surface of the skin on coming contact with water.
- The blister is filled with the fluid which is bacteriologically sterile and contains numerous larvae and leucocytes.
- Diffusible toxins produced by the parasite are believed to cause urticaria, dyspnea, vomiting, mild fever and occasional fainting.

Clinical manifestation:

- The pre-patent period is 10-14 months.
- The symptoms are manifested during parturition of the female and are due to the liberation of a toxic substance causing allergic manifestation and blister formation.
- Septic infection can occur as a result of contamination by secondary organisms drawn in by the worm at the time of retraction.
- Blister formation appears wherever the female worms make an attempt to come to the surface of the body where it can readily discharge its larva.
- The blister is usually found on the lower extremities of the body especially between the metatarsal bones, sole of the feet or on the ankle and less frequently in arms, buttock, scrotum, head, neck and female breast.
- Blister formation is accompanied by intense burning pain, 'fiery serpent'.

- This may be accompanied by generalized reactions such as urticaria, nausea, vomiting, diarrhea and giddiness and marked burning sensation over the next few days the lesions vesiculates and blister ruptures producing a painful ulcer.
- The worm is often visible in the opening of ulcer.
- If the female worms break during the attempts of extractions the larva remain trapped in the subcutaneous tissue and may give rise to cellulitis and abscesses.
- In uncomplicated cases, lesions may only last for several weeks until the worm is completely expelled.
- However, many cases infection of the worm track with persistence of the lesions, chronic ulceration and possible sequelae, involving disseminated infection, phlegma of limbs, contractures of tendons, fibrous ankylosis or arthritis in the joints, die prematurely and calcify. The calcified worms can be trigger arthritis, locked joints or permanent clipping and deformations.

Laboratory diagnosis:

- **Detection of adult worms:**
  - This is possible when the female worm appears at the surface of the skin.
- **Detection of first stage larva:**
  - Specific diagnosis is made by the microscopic demonstration of the first stage larva in the discharge fluid.
- **Intradermal test:**
  - Infection of *Dracunculus* antigens intradermally causes a wheal to appear in the course of 24 hours in positive cases.
- **X-ray examination:**
  - Worms in deeper tissue after the death either become calcified or absorbed.
  - The position of calcified worm may be located by skiagraphy.
- **Blood examination**
- **Serodiagnosis:**
  - IFA, IHA, ELISA and western blot are the frequently used test for demonstration of circulating antibodies in the serum for diagnosis of dracunculiasis.

Treatment:

- There is no a specific drug or medicines to treat or prevent the disease.

- The mainstay of treatment is the extractions of the adult worm from the patient using a stick at the surface and wrapping the worm or few cm per day.
- Full extraction can take several days or weeks.
- Topical antibodies are applied to prevent secondary bacterial infections and the affected body part is bandaged with fresh gauze to protect the site.
- Surgical removal of worm using local anesthesia is another method of treatment
- Albendazole, Mebendazole, Niridazole, thiabendazole and metronidazole are used as anti-inflammatory agents.
- Analgesics such as aspirin or ibuprofen are given to help reduce the pain and inflammation.

Prevention and control:

- The disease can be transmitted only by drinking contaminated water and can be completely prevented through to relatively simple measures.
- Preventing people from drinking Cyclops contaminated water:
- Avoid drinking contaminated water
- Filtering water
- Boiling
- Treatment of water with larvicides to kill Cyclops
- Preventing people infected with the worm from entering water sources used for drinking.

Epidemiology

- *D. medinensis* infection is reported from 18 century of the world.
- The infection is particularly wide spread in Africa and Middle East.
- About 140 million people are estimated to suffer from dracunculiasis.

Haemonchus contortus

Small ruminants, such as sheep and goats, are extremely susceptible to the adverse effects of intestinal parasites. *Haemonchus contortus* is a major challenge throughout the world, particularly in regions with warm and wet conditions. Young and lactating sheep and goats are the most susceptible to *Haemonchus* infection. *Haemonchus* is incredibly costly to the Canadian sheep and goat industry due to significant performance losses, morbidity and mortality, drug costs, and labor associated with treatment and management. *Haemonchus* cannot be eradicated, but can be limited, through control, to decrease economic losses for producers.

What is *Haemonchus contortus*?

Haemonchus contortus is a blood-sucking nematode that feeds on blood from capillaries in the abomasum of ruminants. As a single worm ingests up to 50 µL of blood per day, high infection levels can cause severe blood loss (more than 100 ml daily), followed by anaemia and hypoproteinaemia. *H. contortus*-infected animals tend to have a reduced digestive capacity, which affects the uptake of nitrogen, organic matter and energy. In cases of heavy infection, animal death may occur.

Life cycle and Infectivity

The life cycle of *Haemonchus contortus* takes 17- 21 days to complete. It begins when larvae in the infective stage are ingested by sheep and goats during grazing. Once ingested, the larvae travel to the animal's abomasum where they continue to develop. Lastly, they molt in the adult form. Adult female worms produce thousands of eggs per day (5,000-10,000) which are secreted in the animal's feces onto pasture. Eggs will hatch into larvae under favorable conditions (e.g. warm and moist) and develop within fecal pellets through the immature stages in as short as 5 days. Infective larvae travel onto pasture where they are ingested by sheep and goats during grazing, restarting the life cycle. In Canada, infectivity is highest in late summer (mid-July to August) and early fall because *Haemonchus* prefers warm and humid conditions (>25°C). The two biggest sources of pasture contamination with *Haemonchus* eggs are 1) lambs and kids (late July/August) followed by 2) ewes and do in late gestation and lactation (usually spring). Larvae go into an inactive state inside the animal during winter to survive the Canadian climate. In this state, the worms do not lay eggs or cause damage to their hosts. In late April – early May as the worms resume activity, ewes or do develop severe infestations at the same time as late pregnancy or lactation when the animal's immune system is stressed. Ewes/does will contaminate the spring pasture with eggs and immediately expose vulnerable lambs or kids.

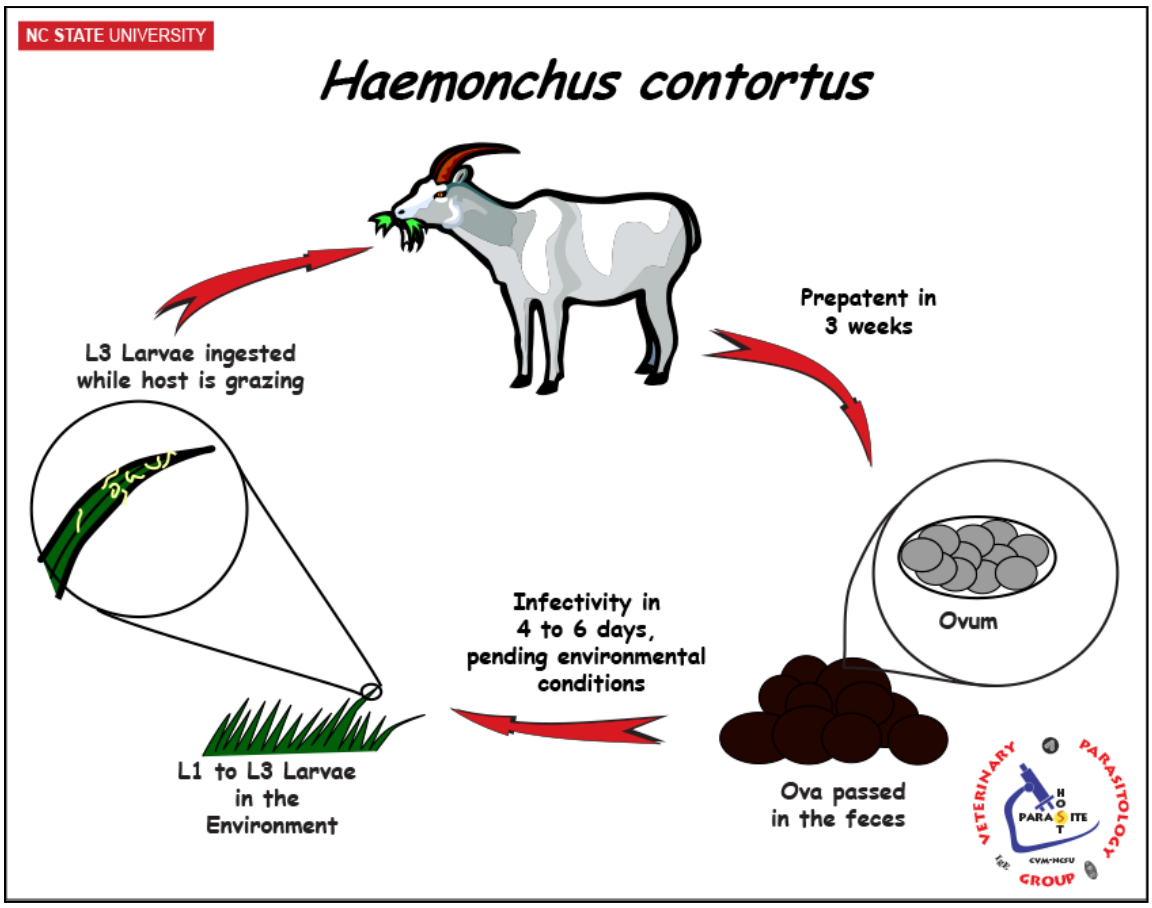


Fig: Life cycle of *Haemonchus contortus*

Signs of Haemonchosis

Signs of a Haemonchus contortus infestation include: anemia, dehydration, "bottle jaw" (accumulation of fluid in the lower jaw due to anemia), poor appetite, weight loss, and significantly reduced growth. Due to anemia, the conjunctival mucous membranes around the eyes appear pale pink to while color.

Gross lesions and the presence of characteristic nematodes in the abomasums often provide a diagnosis at necropsy. Animals with haemonchosis have marked pallor of mucous membranes and internal tissues. A characteristic gross lesion is widespread subcutaneous edema. This may be most striking in submandibular soft tissues, producing the so-called "bottle-jaw"

Control of *Haemonchus contortus*

The hematophagous nematode *Haemonchus contortus* is a common endoparasite that infects wild and domestic ruminants worldwide, especially in tropical and subtropical regions. To date, the most commonly applied control strategy is the administration of anthelminthic drugs.

Sarcoptes scabiei

Sarcoptes scabiei var. *hominis*, the human itch mite, is in the arthropod class Arachnida, subclass Acari, family Sarcoptidae.

It should be noted that races of mites found on other animals may cause a self-limited infestation in humans with temporary itching due to dermatitis; however they do not multiply on the human host.

EPIDEMIOLOGY

Scabies is a relatively common infestation that can affect individuals of any age and socioeconomic status. The worldwide prevalence is estimated to be 200 million people, with wide variation in prevalence among individual geographic regions. A systematic review of population-based studies from various regions of the world (excluding North America) found prevalence estimates ranging from 0.2 to 71 percent, with the highest prevalences in the Pacific region and Latin America. Scabies is particularly common in resource-limited regions.

Crowded conditions increase risk for scabies infestation. Epidemics can occur in institutional settings, such as long-term care facilities and prisons

Life Cycle:

Sarcoptes scabiei undergoes four stages in its life cycle: egg, larva, nymph and adult

Eggs: Females deposit 2-3 eggs per day as they burrow under the skin. Eggs are oval and 0.10 to 0.15 mm in length and hatch in 3 to 4 days. After the eggs hatch, the larvae migrate to the skin surface and burrow into the intact stratum corneum to construct almost invisible, short burrows called molting pouches.

Larva: The larval stage, which emerges from the eggs, has only 3 pairs of legs and lasts about 3 to 4 days. After the larvae molt, the resulting nymphs have 4 pairs of legs. This form molts into slightly larger nymphs before molting into adults.

Nymph: Larvae and nymphs may often be found in molting pouches or in hair follicles and look similar to adults, only smaller.

Adult: Adults are round, sac-like eyeless mites. Females are 0.30 to 0.45 mm long and 0.25 to 0.35 mm wide, and males are slightly more than half that size.

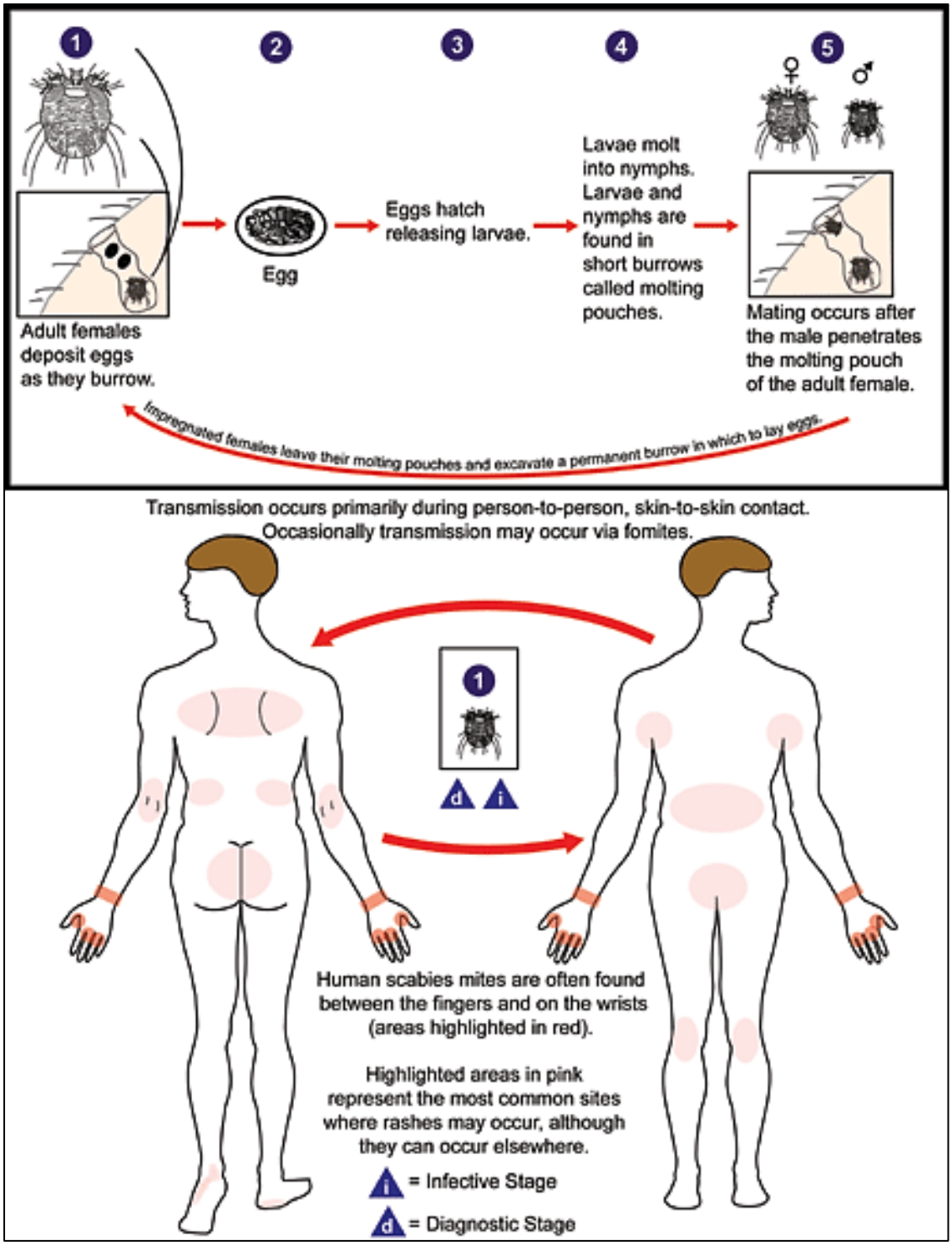


Fig : Life cycle of *Sarcoptes scabie*

Mating occurs after the active male penetrates the molting pouch of the adult female. Mating takes place only once and leaves the female fertile for the rest of her life. Impregnated females leave their molting pouches and wander on the surface of the skin until they find a suitable site for a permanent burrow. While on the skin's surface, mites hold onto the skin using sucker-like pulvilli attached to the two most anterior pairs of

legs. When the impregnated female mite finds a suitable location, it begins to make its characteristic serpentine burrow, laying eggs in the process. After the impregnated female burrows into the skin, she remains there and continues to lengthen her burrow and lay eggs for the rest of her life (1-2 months). Under the most favorable of conditions, about 10% of her eggs eventually give rise to adult mites. Males are rarely seen; they make temporary shallow pits in the skin to feed until they locate a female's burrow and mate.

Types of scabies

There's only one type of mite that causes scabies infestation. This mite is called *Sarcoptes scabiei.* However, these mites can cause several types of infestations.

1. *Typical scabies*

This infestation is the most common. It causes an itchy rash on the hands, wrists, and other common spots. However, it doesn't infest the scalp or face.

2. *Nodular scabies*

This type of scabies may develop as itchy, raised bumps or lumps, especially in the genital areas, armpits, or groin.

3. *Norwegian scabies*

Some people with scabies may develop another form of scabies known as Norwegian scabies, or crusted scabies. This is a more severe and extremely contagious type of scabies. People with crusted scabies develop thick crusts of skin that contain thousands of mites and eggs.

Crusted scabies usually develops in people with weakened immune systems. This includes people with HIV or AIDS, people who use steroids or certain medications (such as some for rheumatoid arthritis), or people who are undergoing chemotherapy.

The scabies mites can overpower the immune system more easily and multiply at a quicker rate. Crusted scabies spreads in the same way as normal scabies.

Mode of transmission:

Transmission occurs primarily by the transfer of the impregnated females during person-to-person, skin-to-skin contact. Occasionally transmission may occur via fomites (e.g., bedding or clothing). Human scabies mites often are found between the fingers and on the wrists.

Symptoms

- ✓ The most common symptoms of scabies, itching and a skin rash, are caused by sensitization (a type of "allergic" reaction) to the proteins and faeces of the parasite. Severe itching (pruritus), especially at night, is the earliest and most common symptom of scabies. A pimple-like itchy (pruritic) "scabies rash" is also common.
- ✓ Itching and rash may affect much of the body or be limited to common sites such as: between the fingers, wrist, elbow, armpit, penis, nipple, waist, buttocks, and shoulder blades. The head, face, neck, palms, and soles often are involved in infants and very young children, but usually not adults and older children.
- ✓ Tiny burrows sometimes are seen on the skin; these are caused by the female scabies mite tunneling just beneath the surface of the skin. These burrows appear as tiny raised and crooked (serpiginous) grayish-white or skin-colored lines on the skin surface. Because mites are often few in number (only 10-15 mites per person), these burrows may be difficult to find.

Possible Complications

The intense itching of scabies leads to scratching that can lead to skin sores. The sores sometimes become infected with bacteria on the skin, such as Staphylococcus *aureus* or beta-hemolytic streptococci. Sometimes the bacterial skin infection can lead an inflammation of the kidneys called post-streptococcal glomerulonephritis.

Treatment

Often consists of medications that kill scabies mites and their eggs. Since scabies is so contagious, doctors will usually recommend treatment for an entire group of people who are in frequent contact with a person who has scabies.

Recognizing scabies bites and the distinctive red rash can help you find treatment faster.

Application of ointments, creams, and lotions that can be applied directly to the skin. Oral medications are also available.

Your doctor will probably instruct you to apply the medicine at night when the mites are most active. You may need to treat all of your skin from the neck down. The medicine can be washed off the following morning.

Make sure you follow your doctor's instructions very carefully. You may need to repeat the topical treatment in seven days.

Some common medicines used to treat scabies include:

- 5 percent permethrin cream

- 25 percent benzyl benzoate lotion
- 10 percent sulfur ointment
- 10 percent crotamiton cream
- antihistamines, such as Benadryl (diphenhydramine) or pramoxine lotion to help control the itching
- An oral tablet called ivermectin (Stromectol) can be given to people who don't see an improvement in symptoms after initial treatment
- Sulfur is an ingredient used in several prescription scabies treatments. You can also purchase sulfur over the counter and use it as a soap, ointment, shampoo, or liquid to treat scabies.

Natural treatment of scabies

Common natural treatments for scabies include:Tea tree oil, Aloe vera, Capsaicin cream, Essential oils, Soaps

Prevention & Control

When a person is infested with scabies mites the first time, symptoms may not appear for up to two months after being infested. However, an infected person can transmit scabies, even if they do not have symptoms. Scabies usually is passed by direct, prolonged skin-to-skin contact with an infected person. However, a person with crusted (Norwegian) scabies can spread the infestation by brief skin-to-skin contact or by exposure to bedding, clothing, or even furniture that he/she has used.

Scabies is prevented by avoiding direct skin-to-skin contact with an infected person or with items such as clothing or bedding used by an infected person. Scabies treatment usually is recommended for members of the same household, particularly for those who have had prolonged skin-to-skin contact. All household members and other potentially exposed persons should be treated at the same time as the infested person to prevent possible re-exposure and reinfection. Bedding and clothing worn or used next to the skin anytime during the 3 days before treatment should be machine washed and dried using the hot water and hot dryer cycles or be dry-cleaned. Items that cannot be dry-cleaned or laundered can be disinfested by storing in a closed plastic bag for several days to a week. Scabies mites generally do not survive more than 2 to 3 days away from human skin. Children and adults usually can return to child care, school, or work the day after treatment.

Persons with crusted scabies and their close contacts, including household members, should be treated rapidly and aggressively to avoid outbreaks. Institutional outbreaks can be difficult to control and require a rapid, aggressive, and sustained response.

Rooms used by a patient with crusted scabies should be thoroughly cleaned and vacuumed after use. Environmental disinfestation using pesticide sprays or fogs generally is unnecessary and is discouraged.

Probable questions:

1. Describe the life cycle of *Loa loa*?
2. Describe the Clinical manifestation of *Loa loa* filariasis.
3. How *Loa loa* infection can be diagonosed?
4. State the habitat of *Dracunuculus medinensis*
5. Describe the morphological structure of *female Dracunuculus medinensis.*.
6. Describe the larval structure of *female Dracunuculus medinensis.*.
7. Describe the clinical manifestation of *Dracunuculus medinensis* infection.
8. What is the treatment of *Dracunuculus medinensis* infection?
9. How the *Dracunuculus medinensis* infection can be controlled?
10. Describe the life cycle of *Haemonchus contortus* with diagram.
11. What are the signs of Signs of Haemonchosis?
12. What is *Nodular scabies*?
13. What are the symptoms of scabies?
14. State the treatment of scabies.
15. How scabies can be controlled?

Suggested reading:

1. Cheng, T. C. (1986). General Parasitology. 2nd ed. Academic Press, Inc. Orlando.U.S.A.
2. Noble, E. R. and Noble G. A. (1989). Parasitology. The Biology of animal Parasites. 6th edn.
3. Roberts, L. S., Janovy, J. and Nadler S. (2013) Gerald D. Schmidt &Lary S. Roberts' Foundation of Parasitology. 9th ed. McGraw-Hill International.

Disclaimer:

The study materials of this book have been collected from books, various e- books, journals and other e-sources.