Wollo University

School of vet medicine

Vet. Protozoology Note

Year II VLT Students

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Veterinary Protozoology

Introduction

- The phylum Protozoa contains unicellular organisms from:
  - the kingdom Protista in the 5 kingdom classification of living things
  - a Phylum in the kingdom Animalia in the 2 kingdom classification
- Microscopic, except very few species
- Most protozoas are free-living organisms
- Of those that live in the body of animals, only a very small proportion are associated with disease
  - The etiologic significance of some is sometimes unclear
  - For example, certain intestinal flagellates tend to multiply when the host has diarrhea
- Protozoa of great pathogenic importance are:
  - Malaria and ameoba in man
  - Trypanosomes, piroplasms and coccidia in animals

Structure

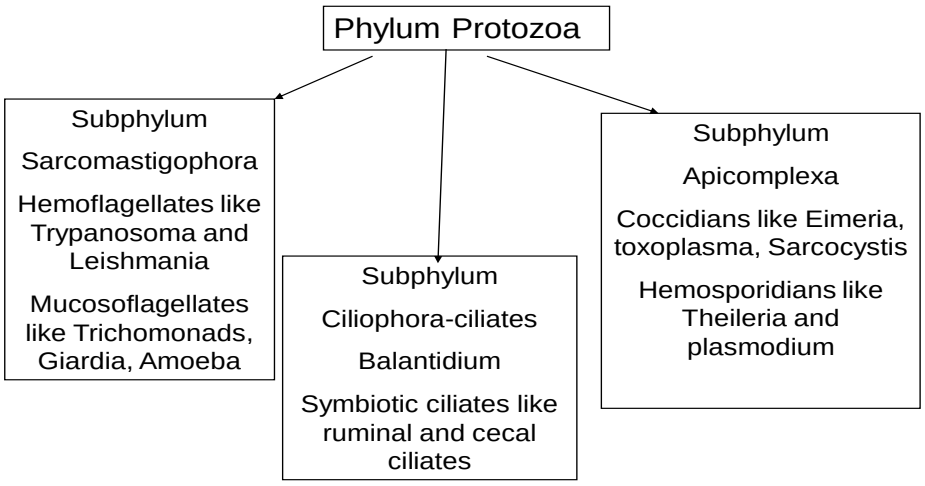
- ✓ Protozoa are eukaryotic: nucleus enclosed in a membrane
- ✓ Contain one or more nuclei
- ✓ Move by means of flagella, cilia, pseudopods or undulating ridges
  - Flagellum: a whiplike organelle composed of: A central axoneme and an outer sheath
    - May pass several points to form undulating membrane
  - Cilium: eyelashlike organelle, small flagellum
    - Found in the Ciliophora
  - Pseudopod: temporary locomotory organelle that can be formed and retracted as needed
- ✓ Organelles associated with nutrition
  - Particulate food material is ingested through a temporary or permanent mouth (cytostome)
  - Many protozoa ingest nutrient materials in solution-pinocytosis
  - In some no specialized organelles are necessary, nutrients being absorbed through the body wall
- ✓ Excretion: commonly through the body wall

Reproduction and lifecycle

- Asexual:
  - binary fission (each individual divides into two)
    - The plane of fission is longitudinal in flagellates and transverse in ciliates
  - Multiple fission (schizogony)
    - the nucleus divides several times before the cytoplasm divides
    - The dividing cell is known as a meront, schizont, agamont or segmenter
    - The daughter cells are merozoites or schizozoites
  - Budding: a small daughter individual separates from the side of the mother and grows to full size
- Sexual reproduction
  - Conjugation
    - Two individuals come together temporarily and fuse along part of their length
    - Their macronuclei degenerate, their micronuclei divide a number of times
    - One of the resultant haploid pronuclei passes from each conjugant into the other
    - The conjugants then separate, nuclear reorganization takes palce
  - Syngamy
    - Two gametes fuse to form a zygote
    - If the gametes are similar in appearance, they are called isogametes
    - If they are dissimilar, they are called anisogametes
    - Following syngamy, the zygote may or may not divide to form a number of sporozoites
    - Some protozoa form resistant cysts or spores. A cyst results from the formation by the protozoon of a heavy wall around itself.
    - Each spore or cyst may contain one or more sporozoites.

Classification

- Based on the structure and life cycle they can be grouped in to different sub phylum and classes. The common characteristics of each group are reflected by similarities in the disease they cause.



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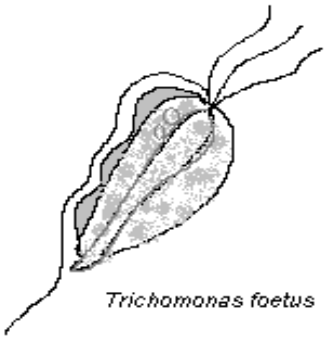
Trichomonads

- Pear-shaped, with rod-like axostyle that protrudes from the more pointed posterior
- 3-5 anterior flagella, and undulating membrane with a trailing flagellum running along its free edges



- Generally they are pyriform in shape rounded anteriorly and pointed posteriorly
- Reproduce by longitudinal binary fission
- Identification:
    - Host and site specificity
    - Number of anterior and trailing flagella
- Common spp

  T. foetus - venereal disease of cattle

  T. vaginalis- reproductive tract of women

  T. humanus – intestine of human

  T. suis – pig T. equi – equine

  T. galinarum - birds

  T. galine – birds

Trichomonas foetus

- ✓ Found in the vagina, uterus, macerated fetus, prepuce, penis, epididymis and vas deferens of cattle, pig, horse and deer but important in cattle.
- ✓ It is economically important in dairy farms. For example in USA it is 3rd important disease next to brucellosis and leptospirosis in causing abortion.
- ✓ Displays considerable pleomorphism
    - Varies from 10-25 µm in length
    - Three anterior flagella, and a long trailing flagellum that extends beyond the undulating membrane
- ✓ it is a venereal disease of bovine that causes infertility, abortion (up to 5 months) etc
- ✓ Transmission through infected bulls, AI, mechanical through gynecological examination.
- ✓ Semen if contaminated with preputial materials can be a source of infection

Pathogenesis

1. Cows – vaginitis, mucopurulent discharge, in the uterus invasion of the developing foetus causes death of the foetus (early abortion) which is usually unnoticed. If the fetal membrane completely removed there will be spontaneous recovery or sterile immunity will be developed. But if retention occurs purulent endometritis, 2ndery complication and permanent sterility is possible. The corpuslutium retained, the cervix closed and development of pyometra or false pregnancy is common.
2. Bulls – no lesions of diagnostic importance, pain on micturition, prepicial discharge which soon disappear may exist. No change in the libido and fertility. But it simply transmits the disease for a number of cows without being noticed.

Diagnosis

- ✓ History and clinical signs are very important.
- ✓ Parasite demonstration: preputial swabs for trophozoites in bulls, uterine or vaginal discharge flushing with physiological saline, amniotic/alantoic fluid, and aborted foetus after centrifugation examination under worm stage microscope motile organisms with distinct shape can be demonstrated.

Control:

- Cull infected bulls, slaughter policy
- Avoid use of communal bulls for breeding and use proper AI
- At least 4 months of sexual rest for infected cows so that the trophozoites diseapear
- Treatment: Metronidazole, Acriflavin

Other species

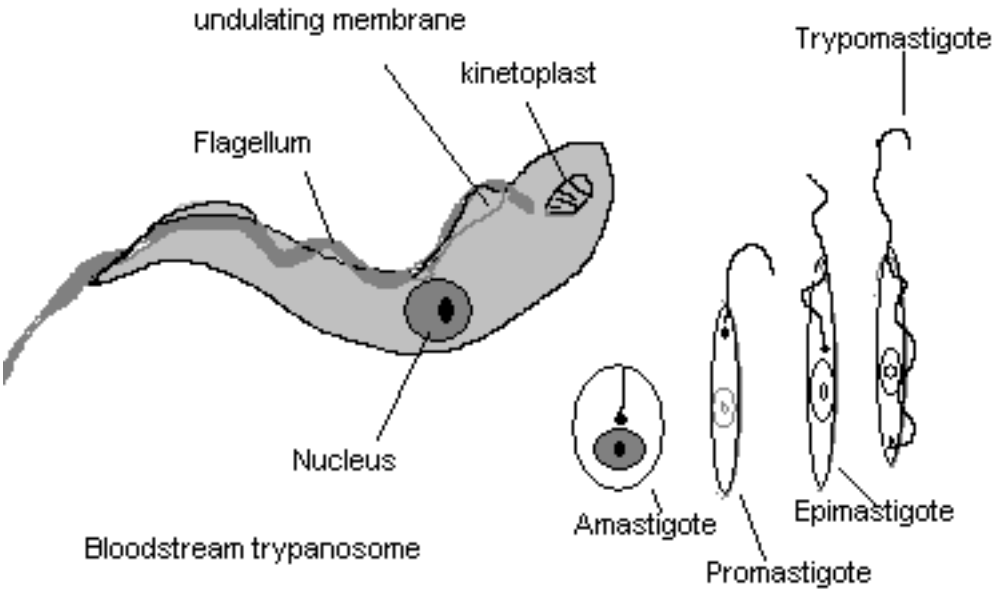
- ✓ *Trichomonas* spp can occur as oral parasites on various hosts
- ✓ *T. Vaginalis* causes vaginitis in women

- ✓ *T. gallinae* causes necrotic ulcerations in the esophagus, crop, proventriculus of pigeons, turkeys and chickens.

Trypanosomes and trypanosomoses

Main morphological features

- Trypanosomes are unicellular, flagellated organisms which live in the blood or tissue fluids of their vertebrate hosts.
- Have kinetoplast, nucleus and sometimes some granules.
- The locations of these structures are important in spp identification.
  - ✓ Position of kinetoplast (terminal, sub terminal or marginal).
  - ✓ Size of kinetoplast (large, medium or small).
  - ✓ Free flagellum (absent or present).
  - ✓ Position of the nucleus (usually central in adults but may not be central in developing stages).
  - ✓ Undulating membrane (distinct).
  - ✓ Posterior end (blunt or pointed).
- Trypanosomes moves by twitching its body in to S- shape. The direction of the movement is towards the free end of the flagellum (anterior end).
- The speed of the movement is variable and helpful in spp identification.
  - ✓ *T. vivax* – moves quickly across the field.
  - ✓ *T. bruci* – moves considerably distance but is less lively.
  - ✓ *T.* congolense – is active but confined to the same area.



- ■ VSG, variable surface glycoprotein
  - ✓ Covers the entire cell surface on top of the cell membrane

- ✓ Through this VSG trypanosomes escape the host immune system.
- ✓ It is the basis for antigenic variation of trypanosomes.

- Mechanism – trypanosomes are exposed to the host immune system. Because they are extracellular parasites. VSG are homodimers and each chain of the dimer is composed of about 500 amino acids.
- ❖ The trypanosomes of veterinary and medical importance are sub divided in to two sections. The **stercoraria** and the **salivaria** based on mode of development in the insect vectors and vertebrates hosts.
- ❖ **stercoraria** – development in the vector (bed bugs) ends with the formation of infective metatrypanosomes in the posterior part of the digestive tract. (Contamination with the feces of the vector).
  - ✓ Megatrypanum, Herpestosoma and Shizotrypanum *(T. cruzi).*
  - ✓ *T. theleri, and T. melophagum* transmited by Tabanid flies and sheep ked respectively are found in cattle and sheep respectively. But they are not pathogenic.
- ❖ **Salivaria** – the usual mode of transmission is inoculative (via saliva) through the biting mouth parts of the vector except dourine.
  - ✓ Duttonella, (*T. vivax and T. uniforme*).
  - ✓ Nanomonas (*T. congolense and T. sime*).
  - ✓ Picnomonas (*T. Suis*).
  - ✓ Trypanozoon (*T. bruci, T. rhodensiense, T. gambianse, T. evansi, T. equperdum*).

Trypanosomoses- is the disease condition in man and animals caused by parasitic trypanosomes.

- ❖ All spp of livestock are affected by the trypanosomes and the disease is characterized by relapsing parasitemia, anemia, loss of condition, abortion infertility, and if left untreated, high mortality.
- ❖ Transmission – bite of the infected Tsetse fly. The flies become infected when they feed on infected animal host. Once infested tsetse flies usually capable of transmitting Trypanosomes for the rest of its life.

Clinical features

- The clinical features are reflections of pathogenesis
  - ✓ Chancre
  - ✓ Lymphadenopathy
  - ✓ Anemia
  - ✓ Tissue damage
- Cattle exposed to infection by tsetse flies develop the disease after a prepatent period of 8-10 days (*T. vivax*), 12-16 days (*T. congolonse*), and 5-20 days (*T. bruci*).

Acute form

- Fever - parasitemia
- Enlargement of superficial lymph node
- Anaemia

Chronic form

- Intermittent fever - Anemia
- Progressive loss of condition -Prominent lymph nodes

- Infected animals are dull; they have a staring coat, lose weight and are easily exhausted, lagging behind the herd.
- As the anemia becomes more severe cattle invariably die of conjestive heart failure which results from anemia myocardial damage and increased permeability.

Pathology

The pathology of Trypanosomosis differs according to the species of trypanosomes causing the infection and the stage of the disease.

- Species of Trypanosomes

  -*T. congolonse* and *T. vivax* are parasites of the blood plasma and produce tissue injury primarily by the anemia.

  - *T. bruci* is more of tissue parasites infecting the intracellular fluids of connective tissue of various organs and extracellular fluids of the body cavities.

- Stage of the disease

  - In the acute stage of the disease post mortem findings may show extensive small hemorrhage on mucous and serous surfaces, emphysema in the lungs and mild gastro-enteritis. The spleen and lymphatic glands are grossly enlarged. The red bone marrow is present throughout the shaft of long bones.
  - In chronic stages the carcass may be anemic, and emaciated. The spleen is small and atrophic but the lymphatic glands have the same appearance as in the acute stage. Atrophy of body fat especially around the heart and kidneys. Hypertrophy of the heart, enlarged liver and chronic liver and venous congestion may be visible. The red bone marrow disappears from the long bones but not from the long bones but not from ribs and vertebrae.

Trypanosomal diseases

- ***Nagana***- given to tsetse transmitted group of disease of cattle (*T. vivax, T. bruci, T.congolonse).*
- ***Sura*** – disease of camel caused by *T. evansi* transmitted by biting flies.

- ***Dourine***- disease of equine mainly horses and donkey caused by *T. equperdum* transmitted by coitus and characterized by a slow progressive course, genital edema, dermal plaques and terminal paralysis. It is the only Trypanosome of vet importance that does not require an arthropod vector.
- ***Chagas disease***- *T. cruzi* transmitted by bed bugs.
- ***Sleeping sickness***- chronic disease caused by *T. gambianse* and *T. rhodesiensi*.

Diagnosis

- ✓ History, geographical distribution of the disease and clinical sign.
- ✓ Direct microscopy- wet blood film
  -thin smear
  -thick blood film
- ✓ Animal inoculation (laboratory rodents) except *T. vivax*.
- ✓ Serological tests (IFA, ELISA), difficult in interpretation because of cross-reactivity and persistency of antibody after treatment.

Treatment

- ❖ Trypanocidal drugs play a role in the control and treatment of trypanosomosis. However the range of drugs suitable for use is small and no new drugs have been marketed for more than 30 years.

Drugs include-Diminazine aceturate
-Homidium chloride
- Isometamidium
- suramin
-Quinapiramin prosalt

- ❖ Sanative pairs of drugs are pairs of drugs that do not induce resistance to each other.

Eg. Isometamidium and diminazin
Homidium and diminazin

Patern of drug resistance in trypanocidal drugs

Resistance	Quinapyramine	Homidium	Isometamidium	diminazin
Quinapyramine	**direct**	cross	Cross	cross
Homidium	Cross	**direct**	Cross	Protective
Isometamidium	Cross	cross	**Direct**	Protective
diminazin	cross	protective	protective	**direct**

LEISHMANIOSIS

- Intracellular parasites of macrophages in man, dog and wild animals.
- Vector: blood sucking ***Phlebotomine*** sand flies where the parasite undergoes morphological transformation and multiplication.
- Has both cutaneous and visceral forms and is of major importance as a disease of man.

SPECIES

- Difficult to distinguish morphologically

Species of dogs: *L. tropica* that cause cutaneous form '' oriental sore'' at the site of insect bite; *L. donovani* induce visceral form ''kala-azar'' i.e systemic infection; *L. braziliensis* cause lesions similar to *L. tropica.*

- *Leishmania* are related to *Trypanosoma* in so far as ovoid organisms within macrophages with rod-shaped kinetoplast and rudimentary flagellum.

LIFE CYCLE

- Multiply within macrophages, destroying them the parasite enter the macrophages.
- The Leishmanial/ amstigote form, after ingestion by flies transform into promastigote form in the insect gut, divide by binary fusion then migrate to the proboscis and when insects feed injected into a new host. Once within the macrophages the promastigote reverts to the amastigote and again divide.

PATHOGENESIS

- Foci type lesions surrounded by plasma cells and lymphocytes will be formed
- The infected macrophages will be destructed and the animal becomes immune to re infection
- Enlarged spleen, liver and lymph nodes as well as persistent skin lesions
- Haemolytic anaemia in case of *L. donovani*

CLINICAL SIGNS

- Only become apparent long after dogs have left the endemic area
- Cutaneous form: a shallow skin ulcer on the lip and eyelids
- Visceral form loss of hairs round eyes and latter all over the body, eczema (the parasites could be isolated from infected skin)
- Intermittent fever and generalized lymphadenopathy are typical signs

DIAGNOSIS

- Demonstration of the parasite in smear or scrapings from affected skin, lymph node and marrow biopsy.

TREATMENT AND CONTROL

- In man antimonial compounds and allopurinol are used
- No effective drug for canine host so animals with visceral form must be destroyed
- Reduction of sand flies as of mosquito control

BABESIOSES

- Caused by Babesia spp
- Class Piroplasmidia, intraerythrocytic parasite of domestic animals
- Cause anaemia and haemoglobinuria
- Transmitted by hard ticks and pass **transoversally**, via the egg, from one tick generation to the other and then *Babesia* are transmitted by any of the three stage soft tick (larval, nymphal or adult)
- The disease is sever in naive animals introduced into endemic areas.
- When infection persists from one stage to the next (two-or three-host ticks feeding on different hosts) transmission is said to be transtadial.

Babesia spp	Hosts	Vectors
B. bigemina, B. bovis, B. divergenes	Cattle	*Boophilus*
B. motasi, B. Ovis	Sheep and goats	*Rhipicephalus, Ixodes, Dermacentor Haemaphysalis,*
B. cabali, B. equi	Equines	*Hyaloma, Rhipicephalus, Dermacentor*
B. perroncitoi, B. trautmanni	Pigs	Rhipicephalus, Boophilus Dermacentor
B. canis, B. Gibsoni	Dogs	Rhipicephalus
B. Felis	Cats	?
B. microti, B. divergens	Man	Unknown

MORPHOLOGY AND IDENTIFICATION

- They are pyriform, may be rounded, elongated or cigar-shaped : small babesia with pyriform 1-2.5 µm long and large ones with 2.5-5.0µm long
- Blood film stained by Gimsa or Romanowsky dyes show the parasite to be within red cells, the cytoplasm appears blue while nucleus red with latter stain
- In the blood almost always singly or paired arranged with their characteristic angle with their narrow end opposed
- Electron microscopy show at their blunt end 'apical complex' which is concerned with assisting penetration of the erythrocyte

LIFE CYCLE

- Reproduction is asexually by binary fission to form two or sometimes four individuals within the red cell
- When cells rapture the parasites can penetrate new red cells
- When the parasitaemic blood is ingested by ticks, a sexual phase takes place in the tick gut followed by schizogony which results in elongated motile, club-shaped bodies, known as **vermicules.**
- Vermicule migrate in to the ovary of ticks and multiply and produce more vermicules
- In the ovary of the ticks vermicules invade the eggs, then multiply in the tissue of the hatched larvae, when larvae first feed, vermicules inter the salivary acini and within a few days, form the infective sporozoites which are inoculated into the new host before feeding of the tick ceases.
- Some species of *Babesia* may be transmitted via the ovary of two or more generations of female ticks (Vertical transmission).

PATHOGENESIS AND CLINICAL SIGNS

- Rapid destruction of red blood cells
- Haemoglobinuria ('red water), fever, increased heart and respiratory rate, jaundiced mucous membrane, diarrhoea
- Loss of weight, milk production and abortion
- In acute case PCV falls by 20% and death in few days

- Depending on the spp of *Babesia*, once the clinical sign appear, parasitaemia may involve 0.2% to 45% of the red blood cells
- In *B. bovis* and *B. canis* clumping of erythrocytes may also occur in the capillaries of the brain, producing nervous signs of hyperxcitability and incoordination
- Mild form is associated with less pathogenic spp or with relatively resistant hosts, characterized by fever anorexia, and slight jaundice for several days. At necropsy pale and jaundiced carcass, with thick and granular bile.

DIAGNOSIS AND TREATMENT

- ❖ History and clinical signs are usually sufficient
- ❖ To confirm: blood film examination stained with Giemsa will reveal the parasite
- ❖ If fever is subsided, impossible to find the parasites since they are rapidly removed from the circulation
- ❖ Treatment: Quinuronium sulphate, Pentamidine, Diminazen aceturate

CONTROL

- Vet. importance of Babesiosis is it constraints to the introduction of improved livestock from other area
- Reducing of ticks by regular dipping of animals with acaricides.
- Introducing tick resistant breed of cattle. Cattle immunization using blood from carrier animals in tropical regions.

THEILEROSIS

- Major constraint on livestock development in Africa, Asia, and Middle East
- Tick transmitted protozoan disease that undergo repeated schizogony in the lymphocytes releasing small merozoites which invade the red cells to become piroplasms.
- Highly prevalent in cattle and sheep in Africa, Asia, Europe and Australia

HOST SPECTRUM, VECTOR AND DISTRIBUTION

Species	Host	Vector	Disease	Distribution
T. parva	Cattle	*Rhipicephalus*	East coast Fever	East and central Africa
T. annulata	Cattle	*Hyalomma*	Mediterranean/tropical theileriosis	North Africa, South Europe, Asia
T. hirci	Sheep/goats	*Hyalomma*	Malignant theileriosis	North Africa, South Europe, Asia

IDENTIFICATION

- In red blood cells the piroplasms are rod-shaped (round, oval and ring-shaped can occur) and up to 2 μm and 1μm wide.
- With Giemsa stains, the cytoplasm is blue with a red chromatin dot at the end
- Commonly more than one parasite in each erythrocyte, in the cytoplasm of the lymphocytes in the lymph nodes and spleen the schizonts called Koch's blue bodies (macro/ microschizonts)

LIFE CYCLE

- The sporozoites are inoculated into cattle by ticks (*Rhipicephalus appendiculatus* (the brown ear tick for *T. Parva*); enter lymphocytes, usually via parotid lymph gland.
- The lymphocyte transforms to a lymphoblast which divides rapidly, the parasite also divides to form two infected cells
- 12 days after infection macroschizonts develop into microschizonts, which change into micromerozoits, liberated by rupture of the microschizonts, then invade the red cells to become piroplasms [never multiply in the RBC].
- The piroplasms require to be ingested by the larvae or nymphal stage of the tick vector where a sexual phase occurs in the tick gut then formation of sporoblasts in salivary glands
- The sporoblasts produce infective sporozoites, female ticks infect the host. Transmission is transtadial: by next stage of tick but not transovarian

PATHOGENESIS AND CLINICAL SIGNS

- ❖ Incubation period is one week, with no lesion and symptom.
- ❖ On the 2nd week, marked hyperplasia and expansion of infected lymphoblasts, initially in the regional lymph node at the site of tick bite

- ❖ Third week: lymphoid depletion and disorganization due to massive lymphocytolysis and depressed leucopoesis, may be due to activation of natural killer cells like macrophages.
- ❖ Terminal phase: necropsy shows atrophy of the cellular content of the lymph nodes and spleen, lung oedema, emphysema and petechial haemorrhages on the GIT mocosa.
- ❖ Occasionally nervous signs, `**turning sickness**` is observed due to presence of schizonts in cerebral capillaries.
- ❖ Enlarged regional lymph nodes, within few days generalized fever, loss of condition, dyspnoea, blood stained diarrhoea and petechial haemorrhages under the tongue. Death in 3 weeks.

- ***Indigenous cattle reared in endemic area are highly resistant but may remain carrier, newly introduced cattle may suffer from high mortality. An outbreak of East Coast Fever is enhanced by prolonged rainy season which is favorable for proliferation of tick vector.***

DIAGNOSIS

- Detection of macroschizonts in biopsy smear from lymph nodes and spleen of dead animals.
- Giemsa-stained blood smears show piroplasms in the RBC
- The indirect fluorescent antibody test is used to detect cattle which have recovered from East Coast Fever
- Tetracycline if effective at the time of infection but has no value to treat clinical cases, effective drugs are: anti-coccidial such as halofuginone and others as naphthaquinone compounds such as parvaquone and buparvaquone
- ***Control***: restriction of cattle movement during known outbreak of East Coast Fever, prevent access of nomadic cattle and buffalo, treating animals against ticks.
- Vaccination with *T. parva* strain and at the same time treating with long acting TTC had shown to be effective preventive measure in endemic areas.

ANAPLASMOSIS

- RBC invading protozoan disease (some says riketician).
- Transmitted by ticks or mechanically by biting insects or even contaminated hypodermal needles or surgical instruments
- Hosts: Cattle, wild ruminants, and perhaps sheep, may act as reservoir of infection
- Intermediate hosts: some 20 species of ticks, including the one-host *Boophilus*,

- Transovarian infection is known to take place but there is a little information on the development of the parasite in the ticks

IDENTIFICATION

- Main species: *Anaplasma marginale* and *A. centrale*, distributed in tropical and subtropical countries
- In Giemsa stained blood films the parasites are seen as small dark red inclusion bodies within the red cell
- Often only one organism in the cell localized marginally for *Anaplasma marginale* and centrally for *A. centrale* (mild pathogenic).

LIFE CYCLE

- Once in the blood, the organism inters the red cells by invaginating the cell membrane forming a vacuole, thereafter they divide to form an inclusion body containing up eight 'initial bodies' packed together
- The inclusion bodies are numerous during acute infection

CLINICAL SIGN

- Cattle aged 2-3 years develop typical often fatal anaplasmosis
- After incubation period of around four weeks, fever and parasitaemia appear
- During parasitaemia, the anaemia becomes more sever, within a week up to 70% of erythrocytes are destroyed
- The clinical sign include pyrexia, haemolytic anaemia, often jaundice, anorexia, laboured breathing, in cow sever drop of milk yield and abortion
- Death within a day in peracute case

PATHOGENESIS

- During parasitaemia, the anaemia becomes more sever, within a week up to 70% of erythrocytes are destroyed
- Necropsy: jaundiced carcass, a gross enlarged gall bladder, on section liver is suffused with bile, the spleen and lymph nodes are enlarged, unlike that of Babesiosis the urine is normal in colour

DIAGNOSIS

- Clinical sign: anaemia becomes sever within a week , laboured breathing, pyrexia, abortion
    - In cattle under one year and naive signs are mild, while aged of 2-3 years develop typical and peracute/fatal anaplasmosis
- Laboratory:
    - haematocrit estimation
    - demonstration of Anaplasma inclusions in the RBC
    - Detection of immune carriers by complement fixation and serum agglutination tests are used
- **Postmortem examination:** jaundiced carcass, a gross enlarged gall bladder, on section liver is suffused with bile, the spleen and lymph nodes are enlarged

✓ NB: unlike that of babesiosis the urine is normal in colour

TREATMENT AND CONTROL

- Tetracycline compounds are effective given before the parasitaemia
- Vaccination of stock with small quantities of blood containing the mildly pathogenic *A. centrale* or avirulent strain of *A. marginale* , Reduction of ticks and biting flies.

Coccidiosis

- ➲ Cocidia contains parasites which occur mainly in vertebrates. They are intracellular parasites and reproduce sexually and asexually. Sporozoites are the infective stages of these parasites.
- ➲ Differentiation b/n different groups is based on
  i. Sporulation time – some have very short up to 24 hrs.
  ii. Predilection site – SI, cecum, etc.
  iii. Number of sporocycst and sporozoite.
  iv. Duration of the life cycle – 2-3 days, 7days.
  v. Response to treatment – easy, difficult.
  vi. Pathogenesity – mild, pathogenic.
  vii. Type of cells affected – epithelial cells of GIT, kidney cells, liver cells.

Oocyst – a cyst which contains a zygote.

Sporocyst – a cyst within the oocyst it may or may not contain sporozoite.

Sporozoite – found in the sporocyst which is the infective stage of coccidian parasites.

Gamete – is the sexual stage it can be either a micro (male) or macro (female) gametes.

Zygote – developmental stage of the parasite after the fusion of the two gametes.

Types of life cycle

- ✓ **Homoxenous** – a life cycle where it has both sexual and asexual stage of development takes place within the host cell in one animal. It is a direct type of the life cycle and requires only a single host.
- ✓ **Hetroxenous** – a life cycle where by asexual stage of development takes place in one host and the sexual stage in another host.

Different groups of coccidian parasites

- Eimeria [*142*] – a single oocyst has 4 sporocysts and each sporocyst contains 2 sporozoites.
- Isospora [124] - a single oocyst has 2 sporocysts and each sporocyst contains 4 sporozoites.
  - ❖ Toxoplasma and sarcocyst are also similar to isospora.
- Cryptosporidium [104] – has no sporocyst but 4 sporozoites are located within the oocyst.
  - ❖ Within the family cryptosporididae there are other groups.

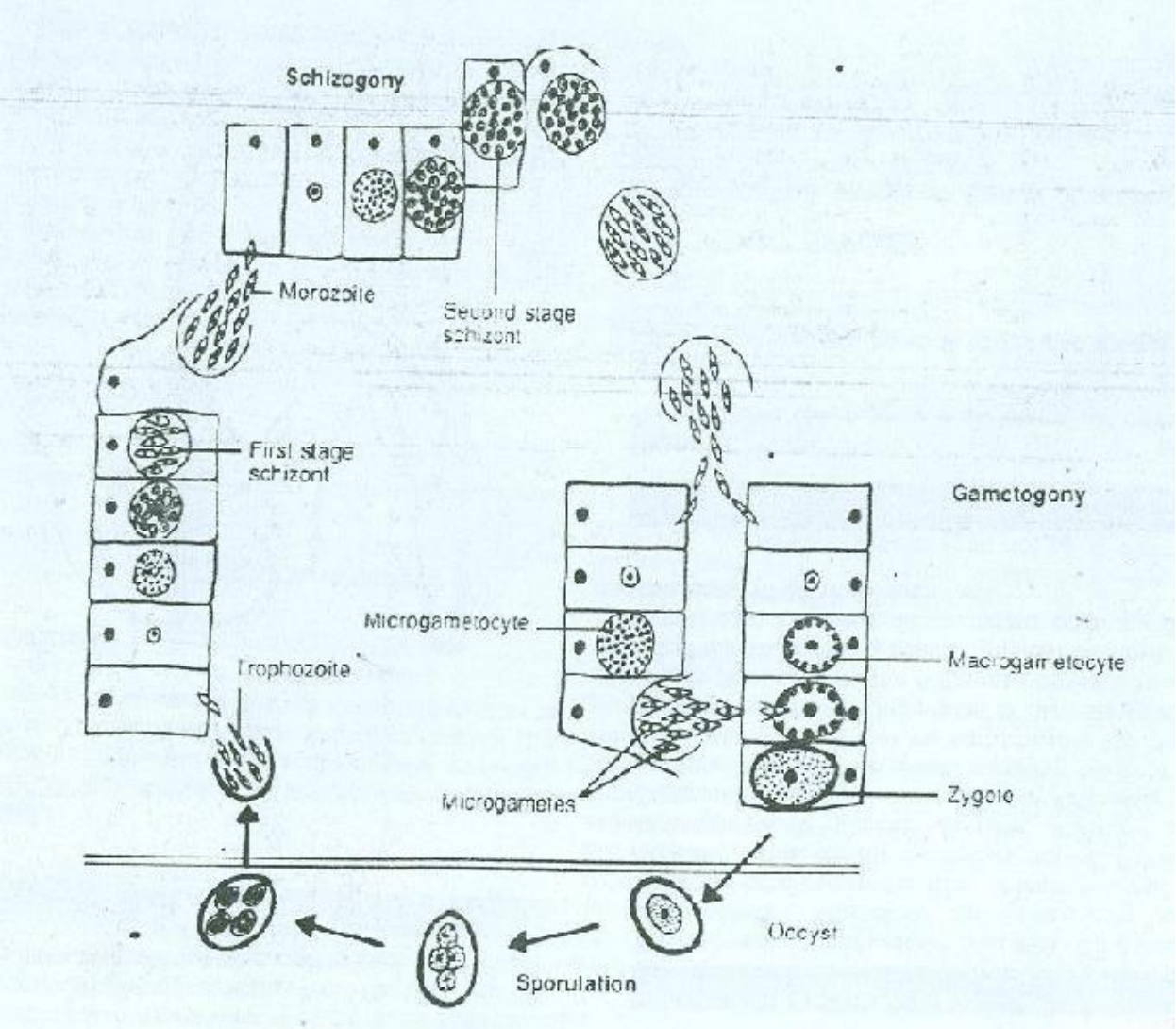
✓ Tyzzaria [118] – a single oocyst has 1 sporocysts and each sporocyst contains 8 sporozoites.

✓ Wenyonella - single oocyst has 4 sporocysts and each sporocyst contains 4 sporozoites.

Basic lifecycle of Eimeria

— Infected hosts pass oocysts to the environment and sporulation takes place in the presence of humidity temperature and oxygen (optimal conditions for each).

— Another host can be infected by ingestion of sporulated oocysts and within the host body sporozoites are released and enter the host cell (invasion of the host cell takes place) and undergo a series of asexual reproduction (schizogoney).
— Each sporozoite penetrate and enlongate and multiply to be trophozoit. Each trophozoit divide my multiple fission to form schizont containing enlongated and nucleated organism known as merozoite.
— The schizogony terminates when these merozoite give rise the male and female ametocytes.
— The macro gametocyte and micro gametocyte join together to form the zygote which after the destruction of a cell released in to the GIT (in the lumen). This sexual stage of development is called gametogony. This is a homoxenous type of lifecycle.



Eimeria

- ✓ Affects a wide range of hosts. It causes major disease condition primarily in poultry and cattle. It is a disease of young animals (chicken and calves). Older animals are usually carriers of the disease.
- ✓ The disease in poultry causes enormous financial losses which can be direct loss (loss from mortality and expense of treatment) and indirect loss (associated with production loss which is reduction in egg and meat production).
- ✓ The disease is widely distributed throughout the world especially in the tropics and it occurs where ever chicken present. Day old chickens even can be infected even though they are protected from natural protection.

Transmition-

- Fecal oral contamination i.e ingestion of sporulated oocysts with feed or water.
- Oocysts can be transported by people working in poultry house, and equipments mechanically.
- Older birds that carry infection are the main sources for young birds.
- There is no risk from hatchery transmition because the incubator temperature is high that destroy the oocysts.

Important species

In poultry there are many species of Eimeria

E. tenella – most pathogenic *E. maxima* – moderately pathogenic

E. accervulina- pathogenic *E. necatris* – lesspathogenic

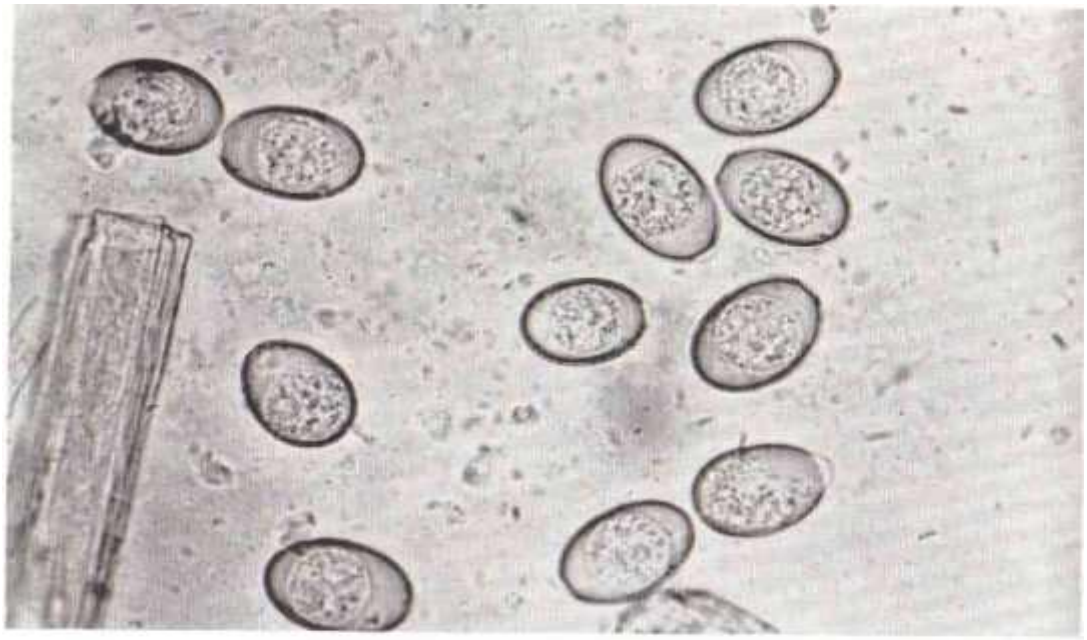
Pathogenesity depends on many factors. These include

- No of oocysts
- Site of infection
- Nutritional status of the host
- Age of the animal – sever in young individuals.
- Breed of the animal – exotic breeds are susceptible

Immunity – most birds develop a self limiting immunity i.e birds that survive infection develop strong immunity that protect from subsequent infection to that spp. but there is no cross protection in coccidiosis in birds. Immunity is species specific. Birds that have developed immunity cause a risk which they shade oocysts even though they do not die. This is a risk for the rest of the flock. So they should be culled out from the farm.

Diagnosis

- ✓ Demonstrations of oocysts from fecal examination using floatation fluid.
- ✓ serology



x 410

FIG. 35—Oocysts of **Eimeria arloingi**. A polar cap is present at one end of the cyst. x 410.

Treatment

Amprolium- 0.02- 0.03% with feed and water

Moensen -0.1- 0.2% with feed and water

Oxy TTC – the 1^0 target is to avoid 2^0 bacterial complications because there is extensive damage on the intestine.

- ♣ It is possible to use these drugs as prophilaxis. The problem is its drug resistance. So use in 'switch over' phenomena.

Vaccination – it is possible to prepare cocktel vaccine by attenuation of sporulated oocysts to be administered orally (Pos). But first the prevalent spp and pathogenic one must be determined in specific area because of spp specificity.

Other spp of Eimeria

There are some important spp

E. bovis – most frequent spp (cattle)

E. zurnii – less frequent (cattle)

E. adenoids (small rts)

E. ahsata (E. ovis) (Sheep)

Isospora

The disease affects a wide range of animals including humans and birds. *Isospora belli* is recognized as opportunistic infection in HIV infected individuals. It affects immune compromised individuals. Infection is through oral anal contamination and the lifecycle is basically the same with Eimeria. No intermediate host is involved. The predilection site is small intestine sometimes the large intestine.

Sarcocystis

- ✓ The disease is called sarcocystosis. Previously it was named as sarcosporidiosis.
- ✓ The life cycle is hetrixenous type in which asexual phase is in one host and the sexual phase is in another host. It requires two hosts.
    - Carnivores and omnivores (human being) act as a definitive hosts where as herbivores act as intermediate hosts.
- ✓ Sporozoites are released to the environment i.e sporulation takes place in the host. But occasionally sporulated oocysts canbe released and ingested by the intermediate hosts (herbivores). The sporozoites penetrate the intestinal mucosa (epithelial cells) and undergo asexual reproduction and carried to the circulation. It resides in the tissue as zoitocysts (bradizoites).
- ✓ The sites are skeletal muscles, cardiac muscles liver and occasionally the brain and remain as a cyst for long period.
- ✓ Definitive hosts are infected by ingestion of the cyst which contains the sexual stage.
- ✓ In the GIT they join to form Zygote and sporulation takes place and excreted with feces. Definitive hosts are not infected by ingestion of sporozoite and sporulated oocysts.

Important Species

Nomenclature is based on their biology which incorporates the intermediate and final host in the order. Although unaccepted to the systematic, this practice is for simplicity. It is proposed by many workers. At present the most important spp recognized with

— Dog as a final host	— cat as a final host	— Man as final host
Sarcocystis bovicanis	*S. bovifelis*	*S. bovihuminis*
S. ovicanis	*S. ovifelis*	
S. capricanis	*S. porcifelis*	
S. porcicanis		
S. equecanis		

Pathogenesis

- ✓ In the final host there is no visible clinical sign (non pathogenic) except mild diarrhea.
- ✓ The clinical manifestation in the intermediate host varies according to the location of the cyst.

  Cardiac muscle – interfears with the function of the heart, tachycardia, petecheal haemorage etc.

  Skeletal muscle – degeneration, lamness, problem in respiration etc.

  Cysts in the nervous system – encephalitis, nervous signs, continous head ache, blind ness.
    - The cysts in the muscles are visible with the naked eye during meat inspection and forms Meischer's tubule mostly 1 cm in diameter. In pigs it can confuse with *Trichinella spiralis* and *Cytsticercus cellulose*. So it can be confirmed after squash smear preparation.

Diagnosis

- In the final host it is like Eimeria i.e fecal examinationand identification of sporozoites/ sporulated oocysts. But we do not undergo fecal examination routinely because there is no clinical sign but simply accidentally during other purposes we may appreciate the oocysts.
- Most cases of sarcocystis infection revealed at meat inspection.
- In heavy infection in intermediate hosts clinical sign, histopathology, and serology (ELISA) are used for diagnosis.

Treatment

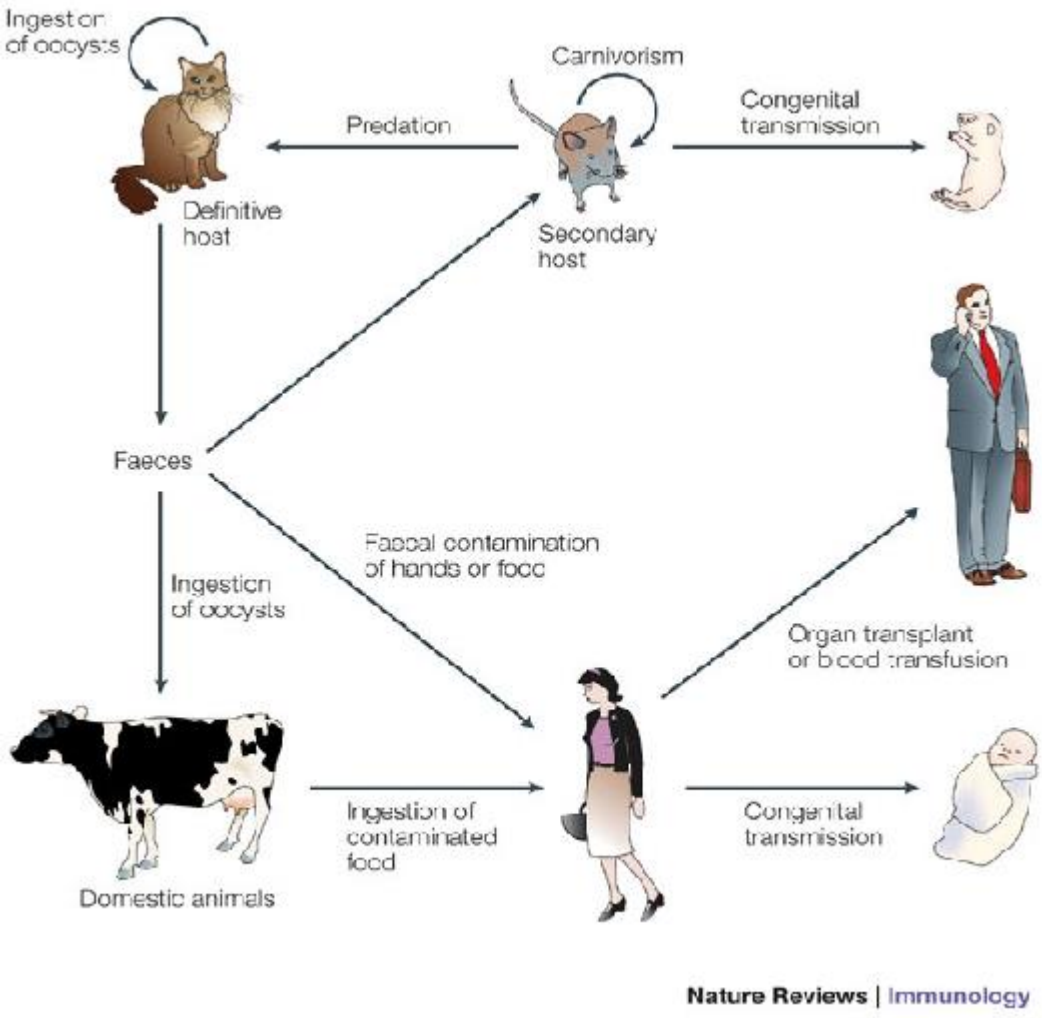
Treatment is not that much successful. In the definitive host anticoccidial drugs like Amprolium can be used. In the intermediate host treatment has a very little practical use since cysts are localized to be protected from the drug.

Control and prevention

- ✓ Avoid dogs and cats from contamination with animal feed stores.
- ✓ Avoid dogs to deficate in pens of animal houses.
- ✓ Carnivores should not be fed with non cooked meat.

Toxoplasma

- ❖ It has a very important spp called *T. gondii* which is an intestinal coccidian of cat (definitive Host). The life cycle involves facultative systemic phase which is an important cause of abortion in sheep and is also zoonosis.
- ❖ Life cycle is direct and indirect. The definitive host (cat) passes the oocyst to the outside. Intermediate hosts (herbivores, omnivores birds and rodents) can acquire oocystswhich results on the formation of Thachyzoites and Bradyzoites. Thachyzoites are highly invasive where as bradyzoites are less invasive due to antigen antibody reaction and they relay on the tissue through haematoginous root. The final hosts (cats) become infected by ingesting animals with the cysts in case of indirect type and ingestion of sporulated oocystes in case of direct type of life cycle. The cysts will be liberated or digested in the cats stomach and undergo schyzogony and gametogony to produce oocysts.



Clinical sign

No clinical sign in the definitive host even in heavy infection. Oocysts are simply observed at accidental routine fecal examination. In the intermediate hosts it depends on the site/tissue. Organs frequently affected are heart, brain, eye and other vital organs. So blindness, circulatory problem, and head ache are signs frequently observed specially in HIV infected individuals.

Diagnosis

- ✓ In the final host - fecal examination and observation of oocysts.
- ✓ Serology for both final host and intermediate hosts. MDAT modified direct agglutination test is common serological test.

Prevalence in our country

Up to 30% in cat was recorded. Serological surveys in human being indicate 37% in Debrebrahan 32% in Nazreth. From these 52% of the cases are associated with HIV.

Treatment

In the final host anticoccidial drugs like sulfonamides can be administered. In the intermediate hosts the treatment is not that much effective because of the localization of the cyst. Pentamidine is effective in human being especially in HIV infected personels.

Prevention and control

- ✓ Sanitation, personal Hygiene
- ✓ Avoid cat contamination
- ✓ Cats have habit that berried there feces this minimizes the dissemination of infection

Cryptosporidium

- ✓ *C. parvum* is the spp which has been responsible for clinical disease in domestic animals. But less important spp include *C. baileyi* and *C. melagridis* in birds. It has been associated with out breaks of diarrhea in calves, lambs, kids and piglets (young animals) dogs and cats also.
- ✓ The life cycle is basically the same with *Eimeria* and sporulation takes place in the host like sarcocystis.
- ✓ The predilection site is the surface epithelium of GIT (brush border) which interfears with absorption and digestion. This is the cause of diarrhea. Unlike that of *Eimeria* it lacks host specificity.
- ✓ Is a disease of animals and humans caused by protozoa known as *cryptosporidium.*
- ✓ *Cryptosporidium* was first described by Tyzzer in 1907 after isolating it from the gastric glands of mice.

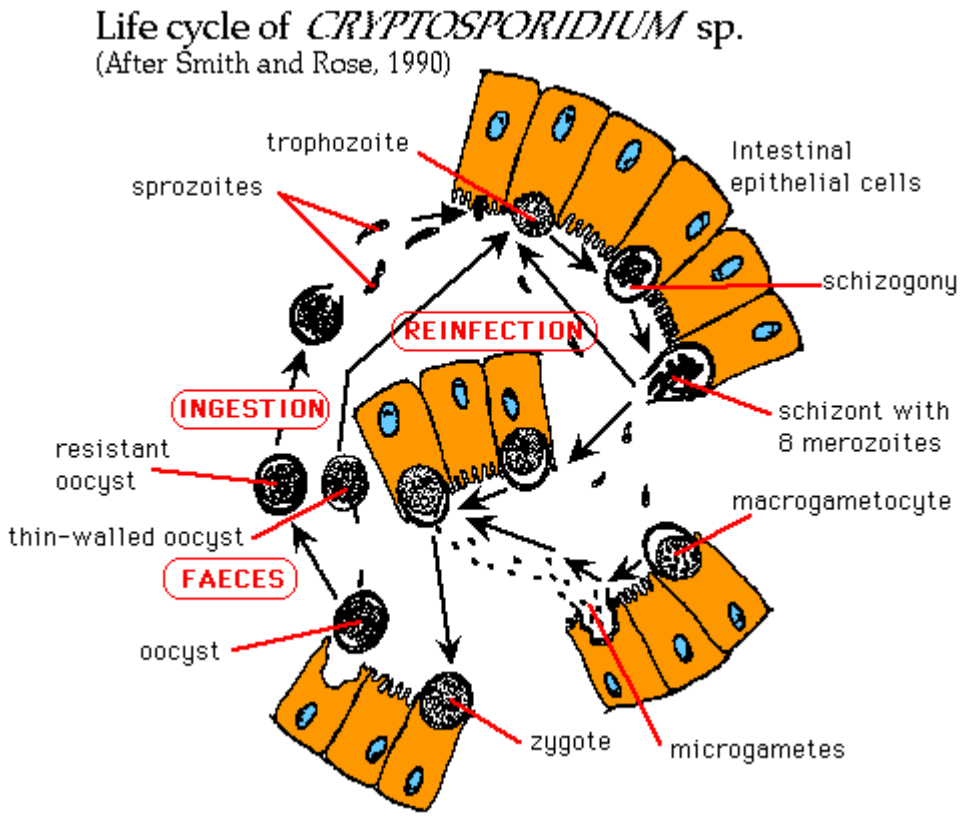
General description

- ✓ The parasite is minute, color less and transparent protozoal parasite.
- ✓ Currently differentiated in to 13 valid spp based on their genetic profile and the spp of the host from which they were originally isolated. But only two spp *C. parvum* and *C. andersoni* are involved in causing disease.
- ✓ *C. parvum* measures 5x4.5µm and infects small intestine of mammals including humans.
- ✓ *C. adersoni* infects the gastric glands of laboratory rodents and several mammalian spp and measures about 7.5 x 5.6 µm. previously it was named as *C. muris*.
- ✓ *Cryptosporidium* spp are not host specific.
- ✓ Develops just under the surface membrane of the host cell or within its brush border rather than in the cell proper.

Life cycle

- ❖ Infective, thick oocysts containing 4 sporozoites are discharged in the feces and serve to disseminate the infection.
- ❖ The oocysts remain viable for months unless exposed to extremes of temperature (below 0 C^o and above 65 C^o) and desiccation.
- ❖ Unlike Eimeria and isospora spp, which are intracellular, cryptosporidium spp are intramenbraneous and resides with in the brush border of the intestinal epithelial cells.
- ❖ When ingested by suitable host, the thick walled oocysts exists to release the four sporozoites that invade the microvillus boarder of the gastric glands or lower half of the small intestine,

- ❖ In the parasitophorous vacuoles of the microvillus border the cryptosporidium undergoes schizogony, gametogony and sporogony.
- ❖ About 20% of the oocysts produced have thin walls and break in the GIT thus releasing their sporozoites which reinvade host cell (auto infection).
- ❖ The other 80% are thick –walled oocysts that pass out with the feces.



Treatment

- ✓ Antibiotics for human being are better than anticoccidial drugs and spiromycin is better drug of choice.
- ✓ Broad spectrum antibiotics also used to avoid bacterial complication
- ✓ Fluid and electrolyte administration for diarrheic patients

Diagnosis

- ✓ Oocysts may be demonstrated by using Zeal nelson staining of fecal smear in which the sporozoites appear as bright red granules.
- ✓ Serological tests like IFA is usually used.