

Lecture: Introduction to Medical Parasitology. Medical Protozoology

1. Parasitism as a kind of ecological relationship
2. Components of a “parasite – host” system
3. Features of parasitic diseases
4. Introduction in Medical Protozoology
5. Characteristics of pathogenic protozoans

The goals of this lecture are to review the parasitism in its ecological and medical aspects to consider the features of parasitic diseases and diseases caused by pathogenic protozoans.

1. Parasitism as a kind of ecological relationship

Parasitism (Gr., *para*, beside, + *sitos*, food or eating) is a permanent or periodic association between two organisms of different species in which one (a parasite) lives upon or within a body of other (a host), and obtains nourishment from it.

Medical parasitology or **Human parasitology** is restricted to studying those parasites that are living *in* or *on* the body of human, their geographic distribution, the diseases caused by them, clinical picture and the response generated by human against them.

In modern biomedicine, *Medical Parasitology* and *Medical Microbiology* are integrated into *Pathogenic Biology*. Pathogenic biology is comprised in preventive medicine and is also considered as the foundation of clinical infectious or communicable diseases and public health. Since most of parasitic diseases occur mainly in the tropics, the field of Parasitology has tended to overlap with that of *Tropical Medicine*. global distribution of tropical diseases affect more than 1 billion people worldwide

2. Components of a “parasite – host” system

A parasite - a living organism that acquires some of its basic nutritional requirements through its intimate contact with another living organism”.

Parasites may be

- ✓ simple unicellular Protozoa
- ✓ complex multicellular Metazoa

- ✓ *Obligate parasite*: a parasite that cannot lead an independent nonparasitic existence, in contrast to facultative parasite.
- ✓ *Facultative parasite*: an organism of free-living species that may either lead an independent existence or live as a parasite, in contrast to obligate parasite

- ✓ *Endoparasite*: “a parasite that lives within another living organism” – e.g. parasitic worms (helminthes)
- ✓ *Ectoparasite*: “a parasite that lives on the external surface of another living organism” – e.g. lice, ticks

Host: “the organism in, or on, which the parasite lives and causes harm”

- ✓ *Definitive host*: “the organism in which the adult or sexually mature stage of the parasite lives”
- ✓ *Intermediate host*: “the organism in which the larval stage of parasite lives or/and asexual reproduction of parasite occurs”

Vector: “a living carrier (e.g. an arthropod) that transports a pathogenic organism from an infected to a non-infected host”. A typical example is female *Anopheles* mosquito that transmits malaria.

Parasitic diseases are classified into:

- ✓ *Anthroponosis*: parasitic disease, the agents of which are transmitted by vectors exclusively from human to human (e.g. malaria, louse-borne spotted fever)
- ✓ *Zoonosis*: a parasitic disease in which an animal is normally the host but which can also infect human.

Parasites, which constitute roughly 50% of the species on Earth, typically feed on only one or a few host species. In evolution, the parasites as well as their hosts developed special adaptations, defence and counterdefence adaptation.

Parasitic adaptations include:

1. morphological adaptations (e.g., holdfast organs of parasitic worms for attachment to host tissues)
2. biochemical changes and specialized mechanisms for entry. For instance, parasite *Plasmodium* causing malaria invade erythrocytes via a specific invasion pathway often identified with its dependence or independence on sialic acid residues of the host receptor. The invasion process involves multiple receptor-ligand interactions that mediate a complex series of events in a period of approximately 1 min.
3. complex life cycles and transmission opportunities. Some parasites have direct simple life cycles whereas others have indirect complex lifecycles involving a vector or intermediate host. Complex life cycles using two or even three host species, free-living stages, asexual and sexual reproduction are widespread in parasitic helminths. In complex life cycles the parasite's development and progression through the life cycle is adapted to the physiology and behavior of the host. In general the higher the parasite load the greater the severity of disease in the host.
4. mechanisms for immune evasion. Parasites have evolved a variety of strategies to survive or evade the host's defence mechanisms. Plasmodia pass through several discrete developmental stages, each with its own particular antigens. A similar situation is seen in certain helminths such as *Trichinella spiralis*. As a result, each new stage of the lifecycle is seen by the host as a 'new' infective challenge.
5. Impact on host versus impact of host that can be direct or indirect.

Direct effects of the parasite on the host:

- Mechanical injury: The host may be inflicted by a parasite by means of pressure as it grows bigger, e.g the hydatid cyst causes blockage of ducts such as blood vessels producing infraction.
- Deleterious effect of toxic substances: The production of toxic substances may cause rigors and other symptoms, e.g., in malaria caused *Plasmodium falciparum*.
- Deficiency of nutrients, fluids, and metabolites: parasite may generate disease by opposing with the host for nutrients.

Indirect effects of the parasite on the host

- Immunological reaction: tissue injury may be caused by immunological response of the host, e.g. in nephritic syndrome.
- Excessive proliferation of certain tissues due to invasion by some parasites can also cause tissue damage in human, e.g/, fibrosis of liver after deposition of the ova of blood flukes *Schistosoma*.

Host adaptations

In many host/parasite relationships the host defences reduces the parasite load to low levels but fails to eliminate the parasite completely and transmission continues. The defence to parasitic infection involves both non-immune and acquired immune mechanisms:

1. protective outer coverings (e.g., skin);
2. acidic medium of stomach;
3. biochemical changes (e.g., in sickle-cell anaemia);

4. immune system.

Historical aspect of Medical Parasitology

Humans are hosts to nearly 300 species of parasitic worms and over 70 species of protozoa.

The first written records of what are almost certainly parasitic infections come from a period of Egyptian medicine from 3000 to 400 BC. Later, there were many detailed descriptions of various diseases that might or might not be caused by parasites, specifically fevers, in the writings of Greek physicians between 800 to 300 BC, such as the collected works of Hippocrates and from physicians from other civilizations including:

- China from 3000 to 300 BC (e.g., in the oldest medical book over 2200 years ago)
- India from 2500 to 200 BC,
 - the early texts on Ayurveda (Indian traditional medicine) *Charaka Samhita* and *Sushruta Samhita* document malaria and its main symptoms as fever and enlarged spleens
 - The *Bhrihu Samhita* from 1000 BC makes the earliest reference to amebiasis. The symptoms were given as bloody and mucosal diarrhea.
- Rome from 700 BC to 400 AD: *Celsus* (25 BC to AD 50) and *Galen* (Galenus of Pergamon, AD 129 to 200) were familiar with the human roundworms (maw worm and pinworm) and tapeworms.
- the Arab Empire in the latter part of the first millennium.

As time passed, the descriptions of infections became more accurate and Arabic physicians, particularly *Rhazes* (AD 850 to 923) and *Avicenna* (AD 980 to 1037), wrote important medical works that contain a great deal of information about diseases clearly caused by parasites.

In Europe, the Dark and Middle Ages, characterized by religious and superstitious beliefs, held back medical progress until the Renaissance, which released a flurry of activity that eventually led to the great discoveries that characterized the end of the 19th century and the beginning of the 20th. These discoveries included the demolition of the theory of spontaneous generation and the evolution of the germ theory by *Louis Pasteur*, the demonstration by Pasteur that diseases could be caused by bacteria, the introduction by *Robert Koch* of methods of preventing diseases caused by microorganisms, and the incrimination by *Patrick Manson* of vectors in the transmission of parasites. The great personalities of this period made discoveries in a number of fields, and their findings and ideas fed off one another.

3. Features of parasitic diseases

Parasitic diseases are cosmopolitan and may affect all the world population. They kill several million people every year. The migration and tourism make that even tropical diseases can be frequently met outside their geographical distribution area. Except the arthropod-borne infections, the great majority of these diseases are in relation with the faecal contamination of soil, the general level of hygiene and the food practices. The socio-political and climatic upheavals may result in a creeping extension of the geographical limits of many parasites.

Where and how person can get parasites:

- international travel
- contamination of water supply
- immigrants from endemic areas
- popularity of household pets
- increasing popularity of exotic regional foods
- undercooked meat and raw fish
- ingestion of improperly washed fruits, vegetables
- use of antibiotics and immuno-suppressive drugs

Scientific terminology

Life cycle is process of a parasite's growth, development and reproduction, which proceeds in one or more different hosts depending on the species of parasite

Alternation of Generation is the regular alternations of sexual and asexual reproductions in life cycles of some parasites

Source of infection is persons (or animals) who have parasites in their body and usually show clinical symptoms.

Carrier is a person who has parasites in his/her body, not shows symptoms.

Invasive stage (also infective stage) is a stage when a parasite can invade human body and live in it

Route of transmission means how the parasite invades human body.

Common pathways for the transmission of parasites:

- Peroral by food or water contamination (e.g., *roundworm*, *amoebae*)
- Percutaneous, i.e., skin penetration (e.g, *hookworms*, *blood flukes*)
- Transmissible by insect vectors (e.g., mosquito – *malaria*, sand fly – *leishmaniasis*, housefly - *amoebic cysts*)
- Transplacental from pregnant woman to fetus (e.g., *malaria*)
- Sexual intercourse (e.g., *trichomonas*, *amoebae*)
- Air-borne, i.e., inhalation of contaminated dust or air (e.,g, *pinworm*)

Pathogen is a parasite that causes disease

Pathogenicity is disease-producing capacity of a pathogen

Pathogenesis is development of disease

Infection is the establishment of a pathogen in its host after invasion

Virulence is the degree or intensity of pathogenicity

Invasiveness is ability to spread to other tissues

Infectivity is ability to secrete toxins

Pathology is scientific study of disease

Etiology is cause of disease

Septicemia is blood infection

Parasitic diseases are primarily diseases of poverty. The individuals, communities and countries, which are least able to afford the costs of treatment or prevention, are at serious risk. In turn, economic development projects which aim to increase income levels may lead to negative results because of increased transmission of parasitic diseases often results.

Often entire communities may be infected with multiple, different organisms which remain untreated because treatment is neither accessible nor affordable.

Effective prevention and control requires "mass intervention strategies" and intense community education. The strategies include:

1. General improved sanitation: fresh water wells, piped water, pit latrines.
2. Vector control: insecticide impregnated bed nets, spraying of houses with residual insecticides, drainage, landfill
3. Mass screening and drug administration programmes which may need to be repeated at regular intervals

4. Introduction in Medical Protozoology

Medical Protozoology studies the unicellular organisms of Subkingdom Protozoa that are parasites of human beings.

General characteristics of Protozoa

Protozoa are one-celled eukaryotes bounded by only a cell membrane. Protozoa possess typical eukaryotic organelles and in general exhibit the typical features of other eukaryotic cells. Protozoa exhibit a wide variety of morphologies. Many protozoan species exhibit *complex life cycles*

with multiple stages. According to mode of nutrition the species are *heterotrophic*, *autotrophic*, *mixotrophic*. The most common form of reproduction is asexual binary fission. Many protozoa exhibit also sexual reproduction that can involve the production and fusion of gametes in processes similar to higher organisms. The *Ciliophora* undergo a conjugation in which opposite mating types will pair and directly exchange genetic material.

Historically protozoa were divided into four major groups based on their **life cycles** and **locomotion** (motility) (see a table below)

Taxonomic classification of protozoa

<i>Phylum</i>	<i>Class</i>	<i>Examples</i>	<i>Mode of locomotion</i>	<i>Characteristics</i>
<i>Sarcomastigophora</i>	<i>Lobosea</i>	<i>Amoeba proteus</i> , <i>Entamoeba histolytica</i>	Pseudopodia for locomotion and for capturing food	No constant shape. Reproduction is by simple cell division. Cyst production is one of the stages of life cycle.
	<i>Zoomastigophora</i>	<i>Euglena</i> , <i>Lamblia</i> , <i>Trypanosoma</i>	Flagella for locomotion	They are fixed oval, elongate or spherical shape. Reproduction is by longitudinal fission and sexually.
<i>Apicomplexa</i>	<i>Sporozoa</i>	<i>Plasmodium</i> , <i>Cryptosporidium</i> , <i>Cyclospora</i> , <i>Toxoplasma</i>	Have no means of locomotion	Reproduce sexually and asexually
<i>Ciliophora</i>	<i>Rimostomatea</i> (<i>Litostomatea</i>)	<i>Paramecium</i> , <i>Balantidium</i>	Cilia for locomotion	They have two types of nuclei: micro- and macronucleus. Reproduce sexually and asexually

4. Characteristics of pathogenic protozoans

Protozoan parasites of medical importance include a lot of species.

IMPORTANT INTESTINAL PROTOZOA TRANSMITTED BY PERORAL ROUTE THE FAECAL AND CAUSE DIARRHOEA

- ***Entamoeba histolytica*** is transmitted by ingestion of feces containing infectious cysts; may invade the colon and cause bloody diarrhoea – amoebiasis (amoebic dysentery). Also it can lead to complications – amoebic liver abscess. In some cases it is asymptomatic intestinal infection. Estimated 40 million develop intestinal disease or liver abscess annually; 40,000 die from amoebiasis annually.
- ***Giardia lamblia*** (also ***Lamblia intestinalis***) causes lamblia – infection of small intestine – that occurs worldwide but is more prevalent in areas with inadequate sanitary conditions. Prevalence rates range from 2-7% in developed countries and 20-30% in most developing countries. *Lamblia* is transmitted by feces containing infectious cysts. It is less harmful than *E. histolytica*, but chronic damage to intestinal wall can lead to watery diarrhea, steatorrhea and malabsorption syndrome.
- ***Balantidium coli***: a large motile ciliated parasite that lives in colon of pigs, humans and rodents. An invasive stage is cyst. Balantidiasis can progress as asymptomatic intestinal infection or cause colitis, bloody diarrhea (dysentery), and intestinal ulceration. Diagnosis is obtained by microscopic observation of trophozoites in faeces in fresh or formalin concentrated preparation

Cryptosporidium parvum

The first human cases of **cryptosporidiosis** were reported in 1976. *C. parvum* is now one of the most commonly identified intestinal (mainly jejunum and ileum) pathogens throughout the world. Its occurrence is dependent on season, the age and other demographic characteristics of a population. Among children aged 1-5 years with diarrhoea, *C. parvum* may be the most frequently found pathogen.

Cryptosporidium is transmitted *perorally* by ingestion of feces containing infectious oocysts (invasive stage). The oocysts are spherical, 4-6 μm in diameter. The factors of transmission are water or food contaminated by human feces or livestock mammal feces, person-to-person contacts.

Source of infection is infected people and animals, e.g., cattle farmers, veterinarians who come in contact with farm animals, infants and younger children in day-care centers.

Life cycle of *Cryptosporidium* is complex completing within a single host with both sexual and asexual phases, and there are 6 distinct developmental stages. The removal from a cyst (excystation) occurs in small intestine with release of the four sporozoites. The parasite infects the apex of the epithelial cells, residing beneath the cell membrane but outside the cytoplasm. The sporozoites undergo several transformations in an asexual (*merogony*) and a sexual (*gametogony*) reproduction cycle; it is the latter that generates *microgametes* and *macrogametes*. Fertilization occurs initiating sexual replication & development of oocysts.

Pathogenicity. Clinical picture of the disease progresses in different way depending on immunity of a patient.

Normally, *in immunocompetent patients*, it is an acute, yet self-limiting diarrheal illness (1-2 week duration), and symptoms include: frequent, watery diarrhea that can last 3-12 days and, in some cases, up to 3 weeks, nausea, vomiting, abdominal cramps, and low-grade fever

In immunocompromised persons (e.g., HIV-infected) and malnourished children illness is much more severe: debilitating, profuse cholera-like diarrhea (up to 20 liters/day) (life threatening), severe abdominal cramps, malaise, low-grade fever, weight loss, anorexia. If complications infection has also been identified in the biliary tract (causing thickening of the gallbladder wall), pancreas and the respiratory system.

The **laboratory diagnosis** of cryptosporidiosis includes modified acid-fast stain technique to detect the oocysts, staining the trophozoites in intestinal and biopsy specimens, immunological tests, and polymerase chain reaction (PCR).

Cyclospora cayetanensis

Cyclospora is an emerging food- and water-borne pathogen that causes protracted diarrhea in humans. The infection *cyclosporiasis* occurs in many countries, but most common in tropical and subtropical regions. The invasive stages are oocysts that are passed in stool and become infectious (sporulated) during 7-15 days under ideal conditions. Direct person-to-person transmission is unlikely because the oocysts passed are not invasive. The oocysts are resistant to most disinfectants used in food and water processing and can remain viable for prolonged periods in cool, moist environments.

Localization: duodenum and jejunum

Pathogenicity. The incubation period is ~ 7 days (ranges 2-14 days). The main symptoms are watery diarrhea, which can be profuse and protracted; enteritis (intestinal inflammation), with pathological lesions (villus blunting), and malabsorption. Anorexia, nausea, vomiting, substantial weight loss, flatulence, abdominal cramping, myalgia, and prolonged fatigue also can occur. Low-grade fever occurs in ~ 50% of patients. Biliary tract involvement has also been reported. Infection usually is self-limited, but untreated people may have remitting, relapsing symptoms for weeks to months. Asymptomatic infection has been documented most commonly in settings where cyclosporiasis is endemic.

IMPORTANT SYSTEMIC PROTOZOANS DETECTED IN BLOOD OR TISSUES

Leishmania spp

Leishmania is a causative agent of a variety of diseases commonly referred to as *leishmaniasis*. The disease is endemic in 88 countries, widespread in tropics and subtropics. As estimated 1.3 million new cases and 20 000 to 30 000 deaths occur annually.

Leishmania parasite exists in two vital stages: *promastigote* stage that has flagella and *amastigote* stage with no flagella. Human is infected by *transmissive way* through the bites of vector –

sand flies *Phlebotomus*, *Lutzomyia*, and *promastigote stage is invasive*.

Leishmaniasis can result in 3 main variants of disease:

- cutaneous leishmaniasis (CL) caused by parasites of dermatotropic group (*Leishmania tropica*). The CV is also known as Oriental sore, Aleppo, Delhi ulcer, Delhi boil or Baghdad boil. 90% of all CL are in Afghanistan, Iran, Syria, and Saudi Arabia, India (Central & Western India), Brazil, Peru. The disease cause skin and mucous membrane lesions.
- muco-cutaneous leishmaniasis (ML) caused by *L.brasiliensis*. The local names of the ML are espundia, chiclero ulcer. 90% of all MCL are in Bolivia, Brazil and Peru. After an initial skin lesion, that slowly but spontaneously heals, chronic ulcers appear after months or years on the skin, mouth and nose, with destruction of underlying tissue (e.g., nasal cartilage). Tissue destruction with disfigurement can be very severe.
- visceral leishmaniasis (kala-azar) caused by the parasites of viscerotropic group – *L. donovani* and *L.infantum*. 90% of all VL are in Brazil, Bangladesh, Sudan, Nepal, and India. Infection affects liver, spleen and lymph nodes, bone marrow, intestinal mucosa and other organs.

Trypanosoma species are haemoflagellates which cause

- in Africa – African trypanosomiasis, or African sleeping sickness (transmitted by the tsetse fly *Glossina*);
- in South America – American trypanosomiasis, or Chagas disease (transmitted by the Reduviid “kissing” bug *Triatoma Infestanse*).

Two subspecies of *Trypanosoma brucei* – *T.b. gambiense* and *T.b. rhodesiense* cause two clinical variants of **African trypanosomiasis**: chronic form and acute form, respectively. Humans are infected by *transmissive inoculative* route; a factor of transmission is *bite of tse-tse fly*. The chronic variant is antroponotic infection lasting years and affecting countries of Western and Central Africa. The acute zoonotic illness lasts from several weeks to months in countries of Eastern and Southern Africa. The parasites reside in blood and cerebral spinal fluid. Infected person can experience fever, swollen lymph glands, aching muscles and joints, headaches and irritability. In advanced stages, the disease attacks the central nervous system, causing changes in personality, alteration of the biological clock (the sleep/wake cycle), confusion, slurred speech, seizures, and difficulty walking and talking. If not treated, the person will die.

According to WHO, the estimated number of actual cases is currently 30000. About 70 mln people are at different levels of risk of HAT in 36 African countries.

American trypanosomiasis, or Chagas disease, is caused by *Trypanosoma cruzi*, a parasite related to African trypanosome. Chagas disease occurs mainly in Latin America. Humans are infected by *transmissive contaminative route*; a factor of transmission is bite and feces of *Triatoma infestans* (kissing bug), a blood-sucking. Reservoir hosts are opossums, ant eaters, armadillos and other forest animals in northern part of South America. Infection can also be acquired through blood transfusion, congenital transmission (transplacentally) and organ transplantation.

In adults, the parasite causes a chronic illness with progressive myocardial damage leading to cardiac arrhythmias and cardiac dilatation, and gastrointestinal involvement leading to mega-oesophagus and megacolon. *T. cruzi* causes acute illness in children, which is followed by chronic manifestations later in life. The most recognized early marker of acute Chagas disease is Romaña's sign, which includes one-side swelling of eyelid on the face near the bite wound or where the bug feces were deposited or accidentally rubbed into the eye.

Plasmodium: the cause of malaria

There are 5 species that infect human: *Plasmodium vivax*, *P. falciparum*, *P. malariae*, *P. ovale* and *P.knowlesi*. Vectors of malaria are female mosquitoes of Genus *Anopheles* that are definitive hosts. In human body, the parasites reproduce by multiple division *schizogony* in liver, and then infect RBCs.

Malaria is characterized by attacks (paroxysms). Every attack usually has 3 stages: "cold stage" with chills, "hot stage" with fever 40°C - 41°C (104°F-106°F) headache, and vomiting, and "sweating phase" with diaphoresis and resolution of the fever. If not treated, malaria can quickly become life-threatening by disrupting the blood supply to vital organs.

Species	Disease	Asexual reproductive cycle in RBCs, hrs	Duration of attack, hrs
<i>P. vivax</i>	tertian malaria	48	~ 5-8
<i>P. malariae</i>	quartan malaria	72	~ 13
<i>P. falciparum</i>	tropical malaria	48	~ 24-36
<i>P. ovale</i>	tertian malaria, or ovale malaria	48	~ 6
<i>P.knowlesi</i>	knowlesi malaria	24	~ 8-12

Preventive strategy includes (i) chemoprophylaxis, (ii) mosquito control (insecticides, larvicides, remove habitats); (iii) mosquito protection (nets, screens, repellents), (iv) mass treatment

Toxoplasma gondii

Infection caused by *Toxoplasma gondii*, **toxoplasmosis**, is common, but only problematic in AIDS patients or to fetus. Humans are intermediate host, domestic cat and other felines are definitive hosts.

Humans normally become infected perorally either by direct ingestion of oocysts from a cat or by eating raw or undercooked meat. Human and other animals can also be infected by the percutaneous and congenital (transplacental) route. Toxoplasmosis is mostly asymptomatic but clinically it could be *acute acquired form, ocular form, congenital form, cerebral form*. Congenital infections occur in about 1-5 per 1000 pregnancies of which 5-10% results in a spontaneous abortion or stillbirth and acute infection in children can cause severe clinical symptoms: *fever, anemia, pneumonia, hydrocephalus, hepatosplenomegaly, jaundice, blindness and mental retardation*

Direct identification of *Toxoplasma* is difficult so serological test are used. The parasites can also be isolated from blood, CSF, amniotic fluid, tissue biopsies to xenodiagnosis by mouse inoculation.