- Phylum: **Metamonada**
  - Class: Diplomonadea
    - Order: Diplomonadida
      - Family: Hexamitidae
      - Genus: *Giardia*
      - Genus: *Hexamita,*
      - Genus: *Octomits,*
      - Genus: *Spironucleus*

Hexamitidae

- This family includes protozoan species that have 2 nucleuses and 8 whips.
- The function of the double nucleus has not yet been clarified.
- It has been found that each of the nuclei in *Giardia* species contains a complete genome.

Genus: Giardia

- The disease caused by the *Giardia* species is called as giardiosis. They can cause disease in human and animals.
- There are vegetative and cyst forms in life cycle of *Giardia* species.

Giardia: morphology

- There are trophozoite and cyst forms.
- Infection is caused by cysts.

Species

- ***Giardia duodenalis (=G.intestinalis, G.lamblia),*** human and various animals
- ***Giardia muris,*** rodents.
- ***Giardia agilis***, amphibians
- ***Giardia psittaci,*** parrots
- ***Giardia ardeae,*** birds

Giardia

- **Trophozoites**
  - **Localization: Duodenum, jejunum and ileum.**
  - **Host: human, other primates, cat, dog, calf, beaver, rabbit.**
  - **They live attached to the mucosa but do not enter the cell.**
    - **Attach to the mucosa with the adhesive discs.**
    - **Provides flow of food through flagellums.**
  - **They reproduce very quickly with simple longitudinal division.**
    - **14 billion trophozoites can be observed in acute diarrheal stool.**
    - **300 million cysts in chronic cases.**
- **Cysts**
  - Trophozoites usually encysted by forming a cyst wall in the end of the jejunum and occasionally in large bowel.
  - **The agents begin to encysted during migration towards the colon.**
  - **Cysts are infective and take as fecal-oral.**

Giardia

- **Cysts**
  - **The cyst is thick-walled oval, 8-10 µm and has 4 nuclei.**
  - **Cyst is an infective stage. It is transmitted by fecal-oral route.**
  - 2 trophozoites are released from each cyst with effect of the low acidic pH in stomach and bile in the duodenum after cysts are taken by susceptible hosts.
  - Prepatent period is usually short (4-16 days in dog and cat; 4-7 days in calf; 10-21 days in lamb)
  - In contrast, patent period is usually long. Infection can last for weeks, even months.

Giardiasis

- The disease is very widespread in the world.
- It is a development criteria for countries.
  - 20% in developing countries
  - 1-6% in developed countries
- Giardia spp. is the most common protozoa in human feces.
  - ~200 million cases per year
- Giardiasis
  - Generally asymptomatic
  - Acute or chronic diarrhea

Pathogenesis and clinical manifestations

- Incubation period is ~10 days.
- 10 cysts may be enough for infection (100 cysts-guarantee)

Clinical manifestations

- Abdominal swelling
- Gas pains
- Diarrhea
- Nausea
- Weight loss
- Stomachache

Pathogenesis:

- Epithelial destruction
  - Villus atrophy
  - Crypt cell hypertrophy
  - Cell infiltration
  - Adsorption and digestion disorders in the intestine.
  - Deterioration of bacterial flora in the intestine.
- Malabsorption (lipid and A,D,E,K vitamins)

Pathogenesis and clinical manifestations

- Giardiasis is usually asymptomatic in domestic animals.
- It is characterized by chronic (rarely acute) catarrhal and intermittent diarrhea especially in young animals.
- The incubation period is lees than 10 days.
- It is seen that the feces are mucus and oily, and some times mixed with blood in dogs and cats.
- Along with these, there is usually vomiting.
- Calves and lambs have weight loss and diarrhea that last for many days with low feed consumption.
- Until today, several giardiasis cases have been reported in horses and there have been no symptoms other than chronic diarrhea and loos of appetite.

Diagnosis, Treatment and Prevention

- Both cysts and trophozoites can be detected by flotation methods ($ZnCl_2$ or $ZnSO_4$: density 1.3).
- Stool examinations should be repeated with intervals of 1-days because of the intermittent disposal of the cysts.
- Fenbendazole is used for treatment in calves and lamps (5 mg/kg p.o. per day-3 days), and cats and dogs (50 mg/kg p.o. per day- 3-5 days).
- The re-infections can be occur in contaminated environmental after treatment.
- General hygiene regulations must be implemented in the struggle with this disease.
- The cysts die rapidly at over 60°C or dry conditions.

Phylum: Parabasala

- Order: Trichomonadida
  - Family: Monocercomonadidae
    - Genus: *Cercomonas, Dientamoeba, Histomonas, Monocercomonas, Parahistomonas*
  - Family: Trichomonadidae
    - Genus: *Pentatrichomonas, Tetratrichomonas, Trichomitus, Trichomonas, Tritrichomonas*

Histomonas meleagridis

- has amoeboid shape and one nucleus.
- has one whip and it emerges from the basal granule near the nucleus.

Histomonas meleagridis

- The disease caused by this protozoon is called histomonosis or Black head (infectious enterohepatitis) in turkeys.
- The protozoa are located in liver and cecum
- It also found in chickens, partridges, quails, peacocks, pheasants and African chickens along with turkeys.
- Deaths can be seen in turkey poults up to 100%. Therefore, it causes huge economic losses.
- This disease is widespread in the world and the presence of this pathogens has been reported from Turkey.

Histomonas meleagridis

- The morphology of this parasite, which has both amoeboid and flagellar activity and is polymorphic, may vary depending on the location, environmental factors, life stages and infection stages.
- Cyst form of this parasite is absent.
- There are 4 stages of this parasite.
  - **Invasive stage**
  - **Vegetative stage**
  - **Resistant stage**
  - **Flagellar stage**

Histomonas meleagridis

- **Invasive stage:** It is seen on the edges of newly formed lesions in the cecum and liver.
  - In this stage, the parasite has not a whip and is extracellular.
  - 8–17 μm in size
  - Nucleus and blefaroblast exist
  - It moves like an amoeba.
  - Protoplasm is granular in the middle, transparent in the edges.

Histomonas meleagridis

- **Vegetative stage:** It occurs in the middle of the lesions in the intestine (in chronic wounds).
  - They are found in tissues as cluster and cause tissue destruction.
  - 12–21 μm.
  - has cytoplasmic bodies in their cytoplasm.

Histomonas meleagridis

- **Resistant stage:** the parasite is surrounded by a compact, thick membrane.
  - The cytoplasm is filled with small granules.
  - 4–11 µm.
  - Cyst does not occur.

Histomonas meleagridis

- **Flagellar stage**: The parasite has a whip. It is found extracellularly in the cecum.
  - It has a granular endoplasm and a transparent ectoplasm.
  - Bacteria or erythrocytes may be present in the parasite cytoplasm.
  - 9–27 μm.

Histomonas meleagridis

- No cyst form
- The parasites multiply with binary fission in the cecum mucosa of birds.
- It leads to lesions in the cecum.
- In the meantime, some parasites reach the liver through the blood, and sometimes the organs such as the kidney and the lung.
- They cause ulcers which have pinhead size.

Histomonas meleagridis

- The parasites in the cecum enter egg of *Heterakis gallina*rum, a nematote and are thrown out with stool together with the eggs.
- The disease occurrs by taking of *H. gallinarum* eggs infected with *H. meleagridis*.

Histomonas meleagridis

- This protozoon also multiplies in eggs of *H. gallinarum*.
- However, not all *H. gallinarum* eggs are infected, about 1% of them are infected.
- The egg develops in the outer environment and infective larva forms inside.
- When the larva leaves the egg, the parasite are released in the cecum lumen.

Histomonas meleagridis

- Since the trophozoites of *H. meleag*radis do not transform to cyst form, they are not resistant to the outdoor environment. These trophozoites need to be taken by other turkeys for infection shortly after exiting from host.
- Earthworms play transporter role by eating the eggs of *H. gallinarum*.
- Current studies reveal that histomonosis can spread easily with directly touch (cloacal drinking).

Histomonas meleagridis

- The mortalities are between 80-100% in 3–12 weeks old turkey poults.
- The prepatent period is 8 days in acute infections.
- Debility, fluffy feathers, flaccid wings are seen in infective turkeys.
- There is a yellow-green diarrhea.
- Cyanosis can be formed in head and crista. Therefore, this disease is called as “Black head”.

Histomonas meleagridis

- Lesions occur in cecum after being infection.
- At the beginning, ulcers occur in small, pin-shaped-sized and they gradually enlarge. One or both cecums can be infected. The wall of cecum become thick.
- Lumen of cecum is filled by a serous and hemorrhagic exudate.
- The cecum enlarges like a hard sausage
- Sometimes these ulcers can be punctured and peritonitis occurs.

Histomonas meleagridis

- Ten days after the infection, necrotic areas of grayish-yellow or green color and degenerations of the tissue occur in liver.
- The edges of these necrotic fields are puffed, and the center is hollow. They are about 1 cm. Sometimes they can merge into a bigger view.
- Apart from the liver, lesions can also be found especially in other organs such as the lung, pancreas, heart, spleen and kidney.
- Death is occurred by impaired liver function and/or peritonitis.

Histomonas meleagridis

- **Diagnosis:** Tissue scrape samples are taken from the liver and cecum and prepared a smear by crushing. The smears are stained with Giemsa. One or 4 whipped forms of parasites are searched.
- Histopathologic smears can be prepared from the liver and cecum. The disease can also be diagnosed by the appearance of typical necrotic lesions in the liver during the necropsy.
- **Treatment:** Nifursol, nitarsone, carbasone, other nitrofurans, dimetridazole, ipronidazole, ronidazole, thiazol and nitrimidazole.

Trichomonas

- Phylum: **Parabasala**
  - Order: Trichomonadida
    - Family: Trichomonadidae
      - Genus: Tritrichomonas
        Tritrichomonas foetus
      - Genus: Trichomonas
        Trichomonas vaginalis
        Trichomonas gallinae
      - Genus: Tetratrichomonas
      - Genus: Pentatrichomonas

Trichomonadida

- Pear-like, mononuclear
- 3-5 anterior flagellums
- posterior flagellum with undulated membrane
- Parabasale body
- Costa and Axostyle
- No mitochondria
- No cyst form
- are located in digestive and genital systems.

➢ **Important species**

- ✓ ***Tritrichomonas foetus*: cattle, genital s.**
- ✓ ***Trichomonas vaginalis*: human, genital s.**
- ✓ ***Trichomonas gallinae*: birds, pharynx**

Tritrichomonas foetus

- Host: cattle (zebu, horse, deer)
- Causes significant economical loss in cattle.
- The protozoa live in vaginal and preputial mucosa.
- Reproduce by simple binary division
- Transmission is occurred by coitus
- Pathogenesis and clinical manifestations
  - Venereal disease: males are asymptomatic carriers
  - Older bulls play an important role in the spread of the disease.
- Vaginitis and uterus infection are seen
- Abort can be seen during the 4th mount of the pregnancy
- Cows can remain infected for years.
- Spontaneous healing may occur when the females are left empty for a period
- Sometimes the infection is prolonged and may result in pyometra and infertility.

Diagnosis and treatment

- The protozoa can be seen in cervical swap or preputial washing.
- There is a special culture material (Diamonds)
- No specific treatment.
  - Female animals can be left empty for a pregnancy period.
  - Carrier bulls can be separated from the herd.
- There are vaccines being developed.
- Prevention
  - Use of young bulls
  - Continuous examination

Trichomonas gallinae

- It is seen mostly in pigeons and doves. It is widely found all over the world. It has been reported from pigeons in Turkey.
- Pear-shaped,
- Small forms are 10 µm x 5 µm, while large forms are 6-19 µm x 2-9.
- There are 4 anterior flagella and 1 posterior flagella characterized with undulated membrane.
- They are located in upper digestive system organs (oral cavity, pharynx and sinuses)
- Infection occurs during feeding of baby by its mother or with infected watery feeder.

Trichomonas gallinae

- Infection is more severe in young birds.
- Adult pigeons can stay to be infected for a year or more and they are source of infection for youngers.
- They transmit the pathogens to their babies during feeding.
- The infection of turkeys and chickens occurs with drinking of infected pigeon's water. However, the survival period of these parasites in the outdoor environment is short and direct contamination is required.
- Wild pigeons and other birds can be an important source of infection for domestic birds.
- Raptors such as hawk, falcon take the parasites by eating infected pigeons and other birds.

Trichomonas gallinae

- Small yellowish lesions are seen in the soft palate and the mouth cavity of pigeons. Later, the number and size of the lesions increase and spread to the esophagus, craw and gizzard.
- Sometimes, the lesions can also be seen in liver.
- In acute phase, deaths may occur before clinical symptoms.
- Wight loses, fluffy feather, anorexia, depression, weakness and weakening are seen in squabs (young birds).
- There is a greenish liquid or cheesecloth material accumulation in the mouth and the crop.
- There is a bad smell in the mouth with drowsiness and the image of sagging crup.
- Dimetridazole and metranidazole can be used in treatment.

Trichomonas vaginalis

- It is a cause of vaginal trichomoniasis in women.
- Pear-shaped and 7-32 μm x 3-12 μm in size.
- Has 4 anterior flagella and one posterior flagella characterized with a undulated membrane.
- The parasite reproduces with binary fusion
- Transmission from person to person happens through sexual intercourse since it has not a cystic form.

Trichomonas vaginalis

- It is a cause of venereal disease in humans.
  - Located in prostate and urethra in man.
  - Located in vaginal mucosa and cause vaginitis in women.
- A common problem in humans. 25% of world population is infected, this situation is 50-70% in prostitutes.
- Pathogenesis and clinical manifestations
- Transmitted by sexual route, but they can also stay alive for a while in clothes and towels (24 hours).
- Man are asymptomatic carriers. Sometimes, urethritis and painful urination can been seen.
- Vaginitis occurs in women
  - inflammation, itching, vaginal discharge

Diagnosis

- Vaginal smear, culture.

Phylum: Amoebozoa

- **Class: Lobosea**
  - **Order: Entomobida**
    - **Family: Entamoebidae**
      - **Genus: *Entamoeba***
    - **Family: Hartmannellidae**
    - **Family: Vahlkampfiidae**
    - The cell membrane in species belonging to these families is composed by a thin structure called plasmalemma.
    - Therefore, body shape of these protozoa is not fixed and they move with pseudopods.
    - These organelles are used for food phagocytosis.
    - Ecto and endoplasms in cytoplasm can easily be separated in the species.
    - They generally reproduce by binary division as asexual.

Genus: *Entamoeba*

- Those with 8 nuclei in their cysts (*Entamoeba coli, E. muris, E. wenyoni)*
- Those with 4 nuclei in their cysts (*E. histolitica*, *E. hartmanni, E. equi)*
- Those with one nuclei in their cysts *(E. bovis, E. bubalis, E. suis)*
- Species whose cysts are not detected (*E. gingivalis, E. canibuccalis, E. equibuccalis)*

The most important species of *Entamoeba* genus is *E. histolitica*.

Entamoeba histolitica

- It is more prevalent in tropical and subtropical regions, with widespread worldwide.
- They can infect humans, various primates, cat, dog, swine, cattle and rats.
- They are generally located in intestine of hosts, and sometimes in liver, lung, brain, testis and bladder.

Entamoeba histolitica

- They can be found in different forms in host.
  - precyst,
  - cyst,
  - metacyst
  - trophozoite

Entamoeba histolitica

- **Trophozoites** move actively with pseudopods.
- There is two forms; parasitic pathogen tissue form (magna) and apathogen intestinal form (minuta).
- Pathogen form is 20-35 µm and erythrocytes can be found in their cytoplasm.
- Apathogen form is 12-15 µm and bacteria can be found in their cytoplasm.
- Both forms move rapidly in the stool and can be seen in stained smears.

Entamoeba histolitica

- The small amebae formed by the division of trophozoites become precyst by altering their external structures after they have consumed the nutrients.
- Cysts are formed from precysts and they are 10-20 µm.

Entamoeba histolitica

- The round or oval cysts have 1-4 nuclei.
- As the number of nuclei increases, the cyst shrinks.

Entamoeba histolitica

- Transmission occurs with taking of cysts through the mouth. The cysts open at the and of the small intestine. Amoeba with 4 nucli exist from the cysts. These are called **metacyst**.
- Amoeba with 8 nuclei are formed with division of metacyst's nucleus.
- 8 metacystic trophozoites are formed by dividing of the cytoplasm. They become normal trophozites in the large intestine.
- And then, the trophozoites continue to multiply by binary fusion.

Entamoeba histolitica

- The parasites shrink and take a round shape before they are encysted, and they form a cyst wall.
- The nucleus firstly divides into two, then divides into two again to form a 4-nuclei cyst.
- This cyst (4-nuclei) produces 8 small amoebae.

Entamoeba histolitica

- Some of these amoebae develop into the intestinal form.
- They are transformed into cyst after they divide and multiply.
- Some of them grow more and locate in the intestine mucosa. These large forms destroy the tissue and phagocytose erythrocytes. They multiply by dividing into two in tissue, but they do not form cyst in tissues.
- In order for tissue forms to form cysts, it must first be transformed to intestinal form in the intestinal lumen.

Entamoeba histolitica

- The disease is called as amoebiasis or amoeboic dysentery.
- Pathogenicity occurs when amoebae enter the intestinal wall and tissues.
- Only large forms are pathogenic

Entamoeba histolitica

- There are different symptoms in intestinal and liver amoebiasis.
- The intestinal amoebiasis progresses acute or chronic.
- There is painful, bloody and mucous diarrhea in the patients.
- Vomiting is also seen.
- Stool has bad smell.
- Fatigue and weakness begin, but there is no fever.
- The symptoms disappear after a while.
- Sometimes, instead of dysentery, large intestinal inflammation occurs.

Entamoeba histolitica

- Sometimes, constipation can also be seen beside diarrhea.
- Amoebae are first found as colonies in intestinal mucosa.
- They can then spread to the submucosa and even to the muscle layer.
- Small amoebic ulcers occur in the intestines.
- These ulcers are superficial at the beginning, sometimes they open up to the peritoneum and cause fistula formation.
- The ulcers are generally found in rectum, cecum and sigmoidal region.

Entamoeba histolitica

- Amoeba may go to the liver with mesenteric veins, or to the right heart, brain and other organs via the lymph vessels.
- Amoebae reaching the liver cause apses in this organ and cause hepatitis.
- Fever, jaundice and leukocytosis can be seen with liver growth and pain.
- Peritonitis and liver, lung, brain apses may occur as complications in amoebic dysentery.
- Metronidazole or furazolidone can be used for treatment in humans and animals.
- It should not be forgotten that *E. histolitica* is one of the a few zoonotic protozoa.

Genus: *Malpighamoeba*
Species: *Malpighamoeba mellifica*

- They cause amoebiasis in worker bees.
- The worker bees become infected by taking through the mouth.
- After the cysts are opened in the digestive system of the bee, trophozoites accumulate between the middle intestine and the last intestine.
- They reproduce by binary fusion.
- This parasite that is rapidly transmitted to the colony causes weakness in the worker bees.

The other amoeba

- ***Balamuthia mandrillaris***
  - They can cause granulomatous ameobic encephalitis in humans.
- ***Acanthamoeba***
  - They cause granulomatous ameobic encephalitis in humans.
- ***Naegleria***
  - They cause primer ameobic meningoencephalitis in humans.

Blastocystis hominis

- The place of this species in the taxonomy is controversial.
- There are a lot of opinions that this species is amoeba.
- There are also opinions which regard this species as an anaerobic protozoa.
- It has been reported that contamination can occur with cyst forms.
- It causes various gastrointestinal complaints in humans.