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Toxoplasmosis in man and animals

Abdel-Rahman M.A.M.

*Parasitology Department, Animal Health Research Institute, Dokki, Giza.

Abstract

Toxoplasmosis is one of the most common parasitic zoonoses world-wide, infecting most species of warm blooded animals, including humans. It is caused by a single-celled protozoan parasite, *Toxoplasma gondii*, an obligate intracellular parasite. It was first observed in the spleen, liver, and blood of gondis, a species of rodents in North Africa. Cats and other felines are the only final hosts, while other animals and humans act as intermediate hosts. The disease is of major medical and veterinary importance; it may cause congenital disease and abortion in humans and domestic animals. Infection in domestic animals is a threat to public health from food-borne outbreaks and causes a great economic loss, as it may lead to abortion, stillbirth and neonatal loss. It has developed several potential routes of transmission within and between different intermediate host species. The major routes of transmission are different in human populations with differences in culture and eating habits.

Introduction

Toxoplasmosis is a common parasitic zoonotic disease world-wide, caused by a single-celled protozoan parasite, *Toxoplasma gondii*, an obligate intracellular parasite. The parasite is a facultatively heteroxenous, polyxenous protozoon that has developed several potential routes of transmission within and between different host species. It can probably infect all warm-blooded animals (mammals and birds) and humans **(Tenter et al., 2000).** The first comprehensive description of *T. gondii* merozoites was given by **Nicolle and Manceaux in 1908** in the spleen, liver, and blood of gondis, a species of rodents in North Africa**.** Evidence for the coccidian nature of *T. gondii* came first from EM studies carried out in the 1960s. These studies revealed ultrastructural similarities between extraintestinal merozoites of *T. gondii* and intestinal merozoites of *Eimeria* species, and thus indicated a coccidian-like life cycle for *T. gondii* **(Tenter and Johnson, 1997).** Disease in humans caused by *T. gondii* was first recognised in the late 1930s. Later, congenital transmission was found to occur in numerous other species, particularly in sheep and rodents**.** *T. gondii* is prevalent in most areas of the world and is of veterinary and medical importance, because it may cause abortion or congenital disease in its intermediate hosts. *T. gondii* has been studied most intensively among the coccidia as a causative agent of a zoonosis due to its great importance **(Dubey, 2009)**.

In 1970, knowledge of the coccidian life cycle of *T. gondii* was completed by the discovery of sexual stages in the small intestine of cats **(Dubey and Beattie, 1988).** It was finally revealed that *T. gondii* is a tissue cyst-forming coccidium with a heteroxenous life cycle in which an asexual phase of development in various tissues of herbivorous or omnivorous intermediate hosts is linked to a sexual phase of development in the intestine of carnivorous definitive hosts.

T. gondii has been generally considered as the only valid species of the genus Toxoplasma **(Tenter and Johnson, 1997).** More recently, molecular epidemiological studies have provided evidence that there are at least two clonal lineages within *T. gondii*, one comprising strains that are virulent in mice and another comprising strains that are avirulent in mice **(Johnson, 1999).**

It has developed several potential routes of transmission within and between different intermediate host species. *T. gondii* may be transmitted vertically by tachyzoites that are passed to the foetus via the placenta. Horizontal transmission of *T. gondii* may involve different routes, i.e. ingesting infectious oocysts from the environment or ingesting tissue cysts or tachyzoites which are contained in meat or primary offal (viscera) of many different animals, or unpasteurized milk **(Tenter et al., 2000).** Transmission may also occur via tachyzoites contained in blood products, tissue transplants. However, it is not known which of these routes is more important epidemiologically. The major routes of transmission are different in human populations with differences in culture and eating habits **(Hill and Dubey, 2002).**

Nearly one-third of humanity has been exposed to this parasite and more than 60 million people in the United States may be infected with the Toxoplasma parasite. In most adults it does not cause serious illness, but it can cause blindness and mental retardation in congenitally infected children and devastating disease in immune-compromised individuals **(Hill and Dubey, 2002).**

This review focuses on probable routes of transmission of *T. gondii* from animals to humans, symptoms, immunological aspects, treatment and preventive measures to control toxoplasmosis.

Prevalence of *T. gondii* infections in humans

The prevalence of *T.gondii* infection varies according to age, environmental factors, diagnostic method, and cultural habits of the consumers **(Dubey and Beattie, 1988).**

T. gondii infections are highly prevalent in adult human populations throughout the world. Sero-prevalences are low in areas with a cold climate. Different serological methods used to for diagnosis of *T.gondii* infection vary in sensitivity, specificity, and predictive values. As a consequence, no two tests produce the same results in all cases, even when carried out in the same laboratory **(Jackson and Hutchison, 1989).**

Cultural and eating habits: - The sources of infection vary greatly with differences in culture and eating habits in human populations and different prevalences of infection in meat-producing animals in these regions. The high prevalence of *T. gondii* infection in Central and South America is probably due to high levels of contamination of the environment by oocysts.

The consumption of undercooked beef in France **(Baril et al., 1999)**, undercooked lamb in Norway **(Kapperud et al., 1996)**, undercooked pork in Poland **(Paul, 1998),** was a stronger risk factor in these countries.

Prevalence of *T. gondii* infections in animals

Sheep, goats, and pigs are more susceptible to disease following *T. gondii* infection than other livestock species and can harbor tissue cysts for life, whereas horses and cattle are less susceptible to infection and cysts containing bradyzoites are rarely detected in their tissues. Several reports of *T. gondii* oocysts from the environment contaminating oceans and being a source of fatal infection in marine mammals as these animals has very little resistance to the parasite **(Dubey and Thulliez, 1993)**.

Epidemiology:-

There are three infectious stages in the life cycle of *T. gondii,* i.e. tachyzoites, bradyzoites contained in tissue cysts, and sporozoites contained in sporulated oocysts. All three stages are infectious for both intermediate and definitive hosts **(Tenter et al., 2000).**

Oocysts:-

1-Shedding:-

Domestic cats are the major source of contamination. Following a primary infection with *T. gondii* cats will shed millions of oocysts in their faeces into the environment **(Dubey and Frenkel, 1972).** Congenitally infected kittens can excrete oocysts **(Hill and Dubey, 2002).** Around 3-10 days after infection, cats start to shed oocysts for two-three weeks. Oocysts are shed for only a short period (1–2 weeks) in the life of the cat. Almost all cats that have been infected primarily with tissue cysts shed

oocysts after a prepatent period of 3–10 days, with patency lasting for up to 20 days. About one third of cats that have been infected primarily with oocysts shed other oocysts after a prolonged prepatent period of 18–49 days for up to 10 days (**Evans, 1992**).

Cats may also become infected by ingesting large numbers (≥1000) of tachyzoites which may result in shedding of oocysts after 15–19 days for up to 7 days (**Literák et al., 1998**). Following this primary shedding of oocysts, cats do develop some immunity and therefore are unlikely to shed further oocysts upon re-infection or reshed oocysts from the original infection.- However this immunity may not last for the lifetime of the cat and research has shown that cats may shed further oocysts when re-challenged around six years after their primary infection.- Experimentally, reshedding of oocysts may be induced by superinfection with other coccidia, e.g. *Isospora* species, after immuno-suppression and due to application of high doses of corticosteroids **(Frenkel, 2000).**

2-Detection:-

Concentration methods (e.g. flotation in high-density sucrose solution) are often used because the number of *T. gondii* oocysts in cat feces may be too few to be detected by direct smear. Examination of faeces is not an appropriate procedure as most cats will shed the majority of oocysts during only 1–2 days, while the whole period of patency may last for up to 20 days (**Boothroyd and Sibley, 1993**).

A serologically negative result suggests that the cat has not yet been exposed to *T. gondii* and is still susceptible to infection in the future. The majority of cats with detectable levels of IgG antibodies to *T. gondii* are likely to be immune and, thus, will not shed oocysts in the near future (**Tibayrenc, 1993**).

3-Sporulation:-

Oocysts shed by cats are unsporulated and, thus, are not immediately infectious. Under environmental conditions with sufficient aeration, humidity, and warm temperature oocysts sporulate and become infectious within 1–5 days **(Evans, 1992).**

4-Resistance:-

Sporulated oocysts survive for long periods under most ordinary environmental conditions. They can survive in for months and even years **(Dubey and Beattie, 1988).** They survive short periods of cold and dehydration, and remain infectious in moist soil or sand for up to 18 months. Under laboratory conditions, sporulated oocysts survived storage at 4°C for up to 54 months and freezing at −10°C for 106 days **(Johnson, 1997).**

However, they were killed within 1–2 min by heating to 55–60°C. Sporulated oocysts also are highly impermeable and, therefore, are also very resistant to disinfectants **(Johnson, 1999).**

5-Distribution:-

Flies, cockroaches, dung beetles and earthworms can mechanically spread these oocysts and even carry them onto food. Oocysts are distributed in the environment through wind, rain, and surface water, or harvested feeds. Hay, straw, and grain which had been contaminated with cat faeces have been identified as sources of infection**.** Infections in aquatic mammals indicate contamination and survival of oocysts in sea water **(Boothroyd, 1993)**.

6-Infection:-

Humans may become infected via contact with contaminated soil. Eating unwashed raw vegetables and fruits was associated with an increased risk of primary infection during pregnancy **(Howe and Sibley, 1995).** Geophagia was strongly associated with an outbreak of acute toxoplasmosis in preschool-aged children of an extended family who played in the same sandy yard of their grandmother's house (**Frenkel and Ambroise-Thomas, 1997**). An outbreak of toxoplasmosis in 35 of 98 military trainees was linked to ingestion of oocyst-contaminated water during training in a jungle environment in Panama (**Sulzer et al., 1986**).

Tissue cyst:-

A recent European multicenter study indicated that up to 60% of *T. gondii* infections may be attributed to the consumption of undercooked or cured meat products from animals infected with the parasite **(Cook et al. 2000).**

T. gondii infection is common in many animals used for food, including sheep, pigs and rabbits, cattle, horses and water buffaloes.

Tissue cysts may develop as early as 6–7 days after infection of intermediate hosts by both oocyte and other tissue cysts. They probably persist for the life of the host.

The number of *T. gondii* tissue cysts in meat from food animals is very low. Therefore, digestion of meat samples in trypsin or pepsin is used to concentrate *T. gondii* in meat.

However, the numbers of tissue cysts that may develop inside a certain host and the locations parasitized vary with the intermediate host species (**Dubey et al., 1998**).

Tissue cysts of *T. gondii* contained in meat of livestock are an important source of infection for humans. In meat-producing animals, tissue cysts of *T. gondii* are most frequently observed in tissues of infected pigs, sheep, and goats, and less frequently in infected poultry, rabbits, dogs, and horses. Tissue cysts are found only rarely in beef or buffalo meat, although antibodies in up to 92% of cattle and up to 20% of buffaloes are evidence of past exposure to the parasite (**Tenter et al., 2000**).

Infection in humans often results from ingestion of tissue cysts contained in undercooked meat. Tissue cysts in the transplanted tissue or in the latently infected transplant patient are probably the source of the infection. Bradyzoites of *T. gondii* are more resistant to digestive enzymes, (pepsin and trypsin) than tachyzoites. Therefore, ingestion of viable tissue cysts by a non-immune host will usually result in an infection with *T. gondii* (**Dubey et al., 1998**).

Although tissue cysts are less resistant to environmental conditions than oocysts, they are relatively resistant to changes in temperature and remain infectious in refrigerated (1–4°C) carcasses or minced meat for up to 3 weeks, i.e. probably as long as the meat remains suitable for human consumption. Tissue cysts also survive freezing at temperatures between −1 and −8°C for longer than a week. Most tissue cysts are killed at temperatures of −12°C or lower, but occasionally some tissue cysts may survive deep-freezing.

It has also been suggested that some strains of *T. gondii* may be resistant to freezing. By contrast, tissue cysts in meat are killed by heating to 67°C, while salting alone is probably not sufficient to prevent transmission to humans via tissue cysts.

Tissues cysts of *T. gondii* in venison (deer) and other meat of wild animals, including hares, kangaroos, and bears are another potential source of infection for humans (**Kotula et al., 1991**).

Tachyzoites

Tachyzoites play the major role in vertical transmission of *T. gondii.*

Resistance

Tachyzoites are very sensitive to environmental conditions and are usually killed rapidly outside the host. Tachyzoites are sensitive to proteolytic enzymes and usually are destroyed by gastric digestion**.** Therefore, infants who have a lower concentration of proteolytic enzymes in the digestive tube are more susceptible to toxoplasmosis than adults **(Johnson, 1997)**.

Transmission

Transplantation of heart, kidney, liver, and bone marrow may be complicated by *T. gondii* infections. Tachyzoites or tissue cysts may be involved. Tachyzoites of *T. gondii* have also been transmitted via blood products, and by accidental injection in the laboratory **(Dubey and Beattie, 1988).**

Tachyzoites of *T. gondii* have been found in the milk of several intermediate hosts, including sheep, goats, and cows but acute toxoplasmosis in humans has been associated only with consumption of unpasteurized goat's milk.

Tachyzoites may enter the host by penetration of mucosal tissue and thereby gain access to the host's circulation or lymphatic system before reaching the stomach (**Sacks et al., 1982**). In addition to blood and milk, tachyzoites have been detected in other body fluids, including saliva, sputum, urine, tears, and semen, but there is currently no evidence of horizontal transmission of *T. gondii* to humans via any of these routes. An early study reported that *T. gondii* tachyzoites may be isolated from raw chicken eggs **(Jacobs and Melton, 1966).**

Life cycle of *Toxoplasma gondii* (Dubey, 2009).

The life cycle of *T. gondii* is facultatively heteroxenous, intermediate hosts are probably all warm-blooded animals including most livestock, and humans. Definitive hosts are members of the family Felidae, for example domestic cats.

During different periods of its life cycle, individual parasites convert into various cellular stages, with each stage characterized by a distinct cellular morphology, biochemistry, and behavior. These stages include the tachyzoites, bradyzoites (found in tissue cysts), and sporozoites (found in oocysts).

When the intermediate host consumes tissue cyst (containing bradyzoites) or an oocyst (containing sporozoites). the parasites first invade cells in and surrounding the intestinal epithelium, and inside these cells, the parasites differentiate into tachyzoites, the motile and quickly multiplying cellular stage of *T. gondii*.

T. gondii undergoes two phases of asexual development. In the first phase, during acute stage, tachyzoites multiply rapidly in many different types of host cells. Inside host cells, the tachyzoites replicate inside specialized vacuoles (called the parasitophorous vacuoles) created during parasitic entry into the cell. Tachyzoites multiply inside this vacuole until the host cell dies and ruptures, releasing and spreading the tachyzoites via the blood stream to all organs and tissues of the body, including the brain.

During the chronic stages of infection, pressure from the host's immune system causes tachyzoites of the last generation stage-convert to bradyzoites to form tissue cysts. Tissue cysts in tissues such as brain and muscle tissue form approximately 7–10 days after initial infection.

Although bradyzoite-containing tissue cysts can form in virtually any organ, tissue cysts predominantly form and persist in the brain, the eyes, and striated muscle (including the heart). However, specific tissue tropisms can vary between species; in pigs, the majority of tissue cysts are found in muscle tissue, whereas in mice, the majority of cysts are found in the brain. Cysts usually range in size between five and 50 μm in diameter, (with 50 μm being about two-thirds the width of the average human hair).

Within the tissue cyst, bradyzoites (or cystozoites) multiply slowly by endodyogeny, they are the terminal life-cycle stage in the intermediate host and are immediately infectious. In some intermediate host species, they may persist for the life of the host. Tissue cysts break down periodically, with bradyzoites transforming to tachyzoites that reinvade host cells and again transform to bradyzoites within new tissue cysts.

When a feline definitive host consumes a tissue cyst (containing bradyzoites), bradyzoites convert into merozoites inside intestinal epithelial cells. The merozoites initiate another asexual phase of proliferation which consists of initial multiplication by endodyogeny followed by repeated endopolygeny in epithelial cells of the small intestine. The terminal stages of this asexual multiplication initiate the sexual phase of the life cycle.

Gamogony and oocyst formation also take place in the epithelium of the small intestine.

Unsporulated oocysts are released into the intestinal lumen and passed into the environment with the faeces and sporogony occurs outside the host.

Route of infection

There are three main route of infection for *T. gondii:-*

(A) Oral ingestion of infective stage of the parasite.

People can accidentally swallow the oocyst from the environment by accidental ingestion of oocysts after cleaning a cat's litter box when cat has shed Toxoplasma in its feces, ingestion of oocysts after touching or ingesting anything has come into contact with a cat's feces or ingestion of oocysts in contaminated soil (e.g., not washing hands

after gardening or eating unwashed fruits or vegetables from a garden). Also, infection occurs when drinking water contaminated with the Toxoplasma parasite **(Boothroyd and Sibley, 1993).**

Tissue cysts of *T. gondii* contained in meat or primary offal (viscera) of intermediate hosts are an important source of infection for humans **(Dubey et al., 1998)** and drinking milk containing tachyzoites from infected animal **(Dubey and Beattie, 1988).**

(B) Vertically by transplacental transmission of tachyzoites. A woman who is newly infected with *Toxoplasma* during pregnancy can pass the infection to her unborn child (congenital infection). The woman may not have symptoms, but there can be severe consequences for the unborn child, such as diseases of the nervous system and eyes **(Tenter et al., 2000).**

(C) Organ transplant recipients can become infected by receiving an organ from a *Toxoplasma*-positive donor. Rarely, people can also become infected by receiving infected blood via transfusion. Laboratory workers who handle infected blood can also acquire infection through accidental inoculation **(Siegel et al., 1971).**

Thus, *T. gondii* may be transmitted from definitive to intermediate hosts, from intermediate to definitive hosts, as well as between definitive and between intermediate hosts. Its life cycle may continue indefinitely by transmission of tissue cysts between intermediate hosts (even in the absence of definitive hosts) and also by transmission of oocysts between definitive hosts, even in the absence of intermediate hosts (**Tenter et al., 2000**).

Peoples at risk:-

These individuals are at risk of developing acute lethal infection if left untreated, the unborn child if the mother becomes infected while pregnant, individuals who are immunosuppressed due to tissue transplants, AIDS, certain types of cancer, and those treated with certain forms of cancer therapy. Also, consumption of raw or undercooked meat was consistently identified as a risk for acquiring toxoplasmosis. The very young and very old may also be more susceptible (**Tenter et al., 2000**).

Symptoms of toxoplasmosis in human.

Most people who become infected with *Toxoplasma gondii* are not aware of it because their immune system usually keeps the parasite from causing illness. Some people who have toxoplasmosis may feel as if they have the "flu" with swollen lymph

glands or muscle aches and pains that last for weeks to months and then go away **(Flegr et al., 2014).** Other symptoms include fever, confusion, headache, nausea, and poor coordination **(Hill and Dubey, 2002).** However, the parasite remains in their body in an inactive state. It can become reactivated if the person becomes immunosuppressed due to various causes. Severe toxoplasmosis occurred leading to damage of the brain, eyes, or other organs. Encephalitis is the most important manifestation of toxoplasmosis in immunosuppressed patients as it causes the most severe damage to the patient**.** Toxoplasma infection can reactivate in immune-compromised pregnant women who were infected with Toxoplasma before their pregnancy, and this can lead to congenital infection **(Dubey and Beattie, 1988)**.

Eye disease includes pain, sensitivity to light and tearing of the eyes. Moreover, there are reports that *T. gondii* may be associated with psychiatric disorders and may affect human behavior, personality, and other phenotypic traits. A number of studies have suggested that subtle behavioral or personality changes may occur in infected humans **(Flegr, 2013)**, and infection with the parasite has recently been associated with a number of neurological disorders, particularly schizophrenia **(Webster et al., 2013).** In some studies, it was found that suicide attempters have significantly higher IgG antibody levels to *T. gondii* as compared with patients without a suicide attempt **(Arling et al., 2009).** Correlation has also been observed between seroprevalence of *T. gondii* in humans and increased risk of traffic accidents. Infected subjects have a 2.65 times higher risk of getting into a traffic accident **(Kocazeybek et al., 2009).** *T. gondii* has been shown to alter the behavior of infected rodents in ways that increase the rodents' chances of being preyed upon by felids. Support for this "manipulation hypothesis" stems from studies showing *T. gondii*-infected rats have a decreased aversion to cat urine **(Berdoy et al., 2000).** Preliminary evidence suggests that *T. gondii* infection can induce some of the same alterations in the human brain as those observed in mice **(Gale et al., 2015).**

Symptoms of toxoplasmosis in animals.

Clinical symptoms in sheep include foetal death, production of a mummified foetus,still born lamb or birth of a live but weak lamb **(Buxton and Rodger, 2008)**. If infection of the foetus occurs early in gestation, while the foetal immune system is still relatively immature foetal death is the usual outcome.

A Toxoplasma infection occurring at mid-gestation typically results in a stillborn or weak lamb accompanied by a small mummified foetus, whereas infection later in

gestation may result in the lamb being born live, but infected and immune **(Innes et al., 2009)**.

Congenital toxoplasmosis

Congenital toxoplasmosis may cause abortion, neonatal death or fetal abnormalities **(Remington et al., 1995).** The risk of intrauterine infection of the foetus, manifestation of symptoms and the severity of the disease depends on:- the immunological competence of the mother, the time of maternal infection during pregnancy, the number and virulence of the parasites transmitted to the foetus and the age of the foetus at the time of transmission.

The immune-status of the mother:-

-In immune-competent hosts, infection with *T. gondii* usually results in life-long immunity against toxoplasmosis. In the primary *T. gondii* before pregnancy, protective immunity will usually prevent vertical transmission to the foetus on subsequent exposures. In the immunocompromised women, seropositive individuals have transmitted *T. gondii* congenitally.

During pregnancy, *T. gondii* may also be transmitted to the foetus in immunocompetent women **(Chatterton, 1992)**.

The time of maternal infection during pregnancy:-

- The risk of intrauterine infection of the foetus increases during pregnancy (14% after primary maternal infection in the first trimester to about 59% after primary maternal infection in the last trimester).
- The effects on the foetus are more severe if transmission occurs at an early stage of pregnancy than those acquired in the second and third trimester.
- About 10% of prenatal infections result in abortion or neonatal death.
- About 10–23% of prenatally infected newborns show clinical signs of toxoplasmosis at birth.
- About 10% of newborns show--------- (hydrocephalus).
    - About 12–16% of these newborns die from the disease.
    - The surviving infants suffer from progressive mental retardation.
- If transmission occurs at a late stage of pregnancy, effects on foetus are less severe.

-The prenatally infected infants appear healthy at birth; they may develop clinical symptoms involving eyes, the CNS and ear later in life **(Remington et al., 1990).**

Immunological aspect:-

The host immune system is activated at a very early stage after infection, and following infection most animals develop adaptive humoral and cell-mediated immune responses. As *T. gondii* is an obligate intracellular parasite, cellular immunity has been considered the major response to eliminate the parasite within the host, yet humoral immunity also plays an important role in shaping the immune responses **(Denkers and Gazzinelli, 1998).** Cell-mediated immune responses are involved in protective immunity and recovery from a primary infection, while specific antibody may be more important in defending the host following a secondary challenge **(Innes and Vermeulen , 2006).**

As a consequence of the host immune response, the parasites are retained in tissue cysts and transform to bradyzoites, which multiply slowly within the cyst. In sheep and other livestock species, the tissue cysts can remain for the lifetime of the host, and this is also thought to be the case with humans **(Innes, 1997).** Tachyzoites may persist longer in the spinal cord and brain than in visceral tissues because immunity is less effective in neural organs, and this persistence varies depending on the strain of *T. gondii* and the host species **(Dubey et al., 2012).**

Toxoplasmosis in an immunocompetent host induces lifelong protective immunity to reinfection **(Verma and Khanna, 2013).** Chronically infected women can exhibit a degree of immunity normally sufficient to protect their fetus from congenital infection even if re-exposed during pregnancy **(Alexander et al., 1996).**

The development of protective immunity to *T. gondii* following natural infection in many host species has led researchers to look at vaccination as a strategy to control disease, parasite multiplication and establishment in animal hosts **(Innes et al., 2009).** However, immunity to *T. gondii* is complex, and involves many facets of the immune system and our understanding of the immune mechanism as well as pathogenesis and host cell invasion mechanisms remain incomplete. While these are lacking, vaccine development is considerably more problematic **(Liu et al., 2012).**

Diagnosis:-

Clinical signs of toxoplasmosis are non-specific and are not characteristic for a definite diagnosis. Diagnosis of toxoplasmosis in humans is made by biological, serological, histological, or molecular methods as described by **Hill and Dubey (2002).**

Serological procedures include:-

- Sabin–Feldman dye test,
-Indirect fluorescent antibody assay (IFA),
-Direct agglutination test,

-Latex agglutination test (LAT).

- Enzyme immunoassays (ELISA, immunoblots)

Diagnosis of toxoplasmosis is usually made by detection of Toxoplasma-specific IgG, IgM, or IgA antibodies. The IgM antibodies appear sooner after infection than the IgG antibodies and disappear faster than IgG antibodies after recovery **(Remington et al., 1995).**

It is best to collect two samples from the same individual, the second collected 2–4 weeks after the first. A higher antibody titer in the second sample indicates an acute infection **(Hill and Dubey, 2002).**

Diagnosis can be made by direct observation of the parasite in stained tissue sections of biopsy material and cerebrospinal fluid (CSF). These techniques are used less frequently because of the difficulty of obtaining these specimens.

Toxoplasma gondii can be isolated from patients by inoculation of laboratory animals for microscopical examination of the parasite and for PCR **(Grover et al., 1990).**

A rapid diagnosis may be made by microscopic examination of impression smears of lesions. After drying for 10–30 min, the smears are fixed in methyl alcohol and stained with Giemsa stain. In sections, the tachyzoites usually appear round to oval. Tissue cysts are usually spherical, lack septa, and the cyst wall can be stained with a silver stain **(Hill and Dubey, 2002).**

Toxoplasma in milk

Previous studies indicated that consumption of raw milk from infected animal, especially goats, represent a vehicle for transmission of toxoplasmosis as unpasteurized milk is considered an important food source in rural areas**.** It has been shown that milk promoted higher infectivity, revealing probably better preservation or efficiency of *T.gondii* as milk nutrients maintains tachyzoites viable for longer time and protects them from gastric juice **(Riemann et al., 1975 and Sacks et al., 1982)**.

Toxoplasma antibodies are detectable in serum two weeks post infection, whereas milk antibodies were observed between 7-10 days post infection **(Thierry et al., 1990).** Toxoplasma antibodies were detected in the milk of naturally infected lactating women **(Azab et al., 1992),** in working donkeys **(Haridy et al., 2010)** and experimentally in mice **(Thierry et al., 1990)** and in naturally infected lactating goats **(Abdel-Rahman et al., 2012).**

Studies indicated that demonstrable antibodies in milk were higher than that detected in serum samples. It has been shown that milk antibodies reflect local antigenic stimuli to the infection. The systemic antibody response may be boosted by repeated infections, and high-titer serum antibodies may persist for years even in the absence of continuous infection. Whereas the systemic response has a longer-lasting immunological memory, the local immunological memory is short lasting, although reinfection may boost memory of a somewhat longer duration. Therefore, although serum antibodies indicate past or present invasive disease, milk antibodies are more likely to suggest present or recent infection **(Grundy et al., 1983).**

The presence of positive IgM in goat serum samples suggests recently acquired or active infection, demonstrating that these animals can constitute an important source of transmission to man, since they are able to present *T. gondii* tachyzoites in milk **(Abdel-Rahman et al., 2012).**

Treatment of Toxoplasmosis.

In healthy person who is not pregnant, this form of the disease is usually self-limited and treatment usually is not needed. Most healthy people recover from toxoplasmosis without treatment. If symptoms occur, they typically go away within a few weeks to months. For pregnant women or persons who have weakened immune systems, medications are available to treat toxoplasmosis.

Pregnant women, newborns, and infants can be treated, although the parasite is not eliminated completely. The parasites can remain within tissue cells in a less active phase; their location makes it difficult for the medication to completely eliminate them.

Persons with compromised immune systems need to be treated until they have improvement in their condition.

Treatment may be indicated for 2 to 4 weeks. Pyrimethamine, plus sulfadiazine, plus folinic acid (leucovorin). Clindamycin can be used instead.

The fixed combination of trimethoprim with sulfamethoxazole has been used as an alternative. Corticosteroids are sometimes prescribed in addition to antiparasitic agents **(Daffos et al., 1988).**

Spiramycin is recommended for the first and early second trimesters or pyrimethamine/sulfadiazine and leucovorin for late second and third trimesters **(Desmonts and Couvreur, 1979).**

Sulphadiazine and pyrimethamine (Daraprim) are two drugs widely used for treatment of toxoplasmosis **(Dabil et al., 2001).**

While these drugs have a beneficial action when given in the acute stage of the disease process when there is active multiplication of the parasite, they will usually not eradicate infection. Certain other drugs, diaminodiphenylsulphone, atovaquone, spiramycin and clindamycin, are also used to treat toxoplasmosis in difficult cases **(Hill and Dubey, 2002).**

Prevention & Control

Preventive measures can significantly reduce the risk of acquiring an infection with *T. gondii*, includes three ways of measures.

1-Reduce risk from food and water.

T. gondii organisms in meat can be killed by exposure to extreme heat or cold, so avoid eating of undercooked meat, cook food to safe temperatures. Tissue cysts in meat are killed by heating the meat throughout to 67C° or by cooling to -13 C° **(Dubey et al., 1990)**. Meat of any animal should be cooked to 67C° before consumption, and tasting meat while cooking or while seasoning should be avoided. Also, meat must be frozen for several days at sub-zero (0° F) temperatures before cooking to greatly reduce chance of infection **(Kotula et al., 1991).**

Fruits and vegetables must be peeled or washed thoroughly before eating, avoid drinking untreated water and unpasteurized goat's milk **(Foulon et al., 1994)**.

2-Reduce risk from the environment.

To prevent infection of human beings by *T.gondii*, the hands of people e handling meat or gardening or come in contact with soil or sand should be washed thoroughly with soap and water. All surfaces and utensils come in contact with soil or sand or raw meat should be washed with hot soapy water also. Washing is effective because the stages of *T. gondii* in meat are killed by contact with soap and water **(Dubey and Beattie, 1988).**

When gardening and during any contact with soil or sand gloves must be used, and cats fed only canned or dried commercial food or well-cooked food **(Hill and Dubey, 2002).**

In these programs, pregnant women or immune-compromised persons must be informed about the epidemiology of *T. gondii* infections as well as preventive measures and should avoid contact with cats, soil and raw meat **(Foulon et al., 1994)**. All children of mothers who seroconverted during pregnancy should be evaluated for congenital toxoplasmosis and treated if infection is still evident **(Couvreur et al., 1993)**.

3-Vaccination: - There is an approved live vaccine for sheep available called Toxovax (MSD Animal Health) that provides lifetime protection. Whereas, there is no currently licensed human vaccine exists against *T. gondii* and research on human vaccines is ongoing **(Verma and Khanna, 2013).**

References

Abdel-Rahman, M.A.M., Soheir, M. El-Manyawe, Khateib. A.M. and Sahar, E. Saba (2012): Occurrence of Toxoplasma antibodies in caprine milk and serum in Egypt**.** Assut Vet. Med. J.85 (133) 145-152.

Alexander, J., Jebbari, H., Bluethmann, H., Satoskar, A. and Roberts, C.W. (1996): Immunological control of *Toxoplasma gondii* and appropriate vaccine design. Curr. Top. Microbiol. Immunol. 219:183–95.

Arling, T.A., Yolken, R.H., Lapidus, M., et al. (2009): "*Toxoplasma gondii* antibody titers and history of suicide attempts in patients with recurrent mood disorders.". The Journal of Nervous and Mental Disease. 197 (12): 905–8.

Azab, M.E., Kamel, A.M., Makled, K.M., Khattab, H., el-Zayyat, E.A., Abo-Amer, E.A., and Samy, G. (1992): Naturally occurring toxoplasma antibodies in serum and milk of lactating women. J. Egypt. Soc. Parasitol. Aug; 22(2):561-568.

Baril, L., Ancelle, T., Goulet, V., Thulliez, P., Tirard-Fleury, V. and Carme, B. (1999): Risk factors for Toxoplasma infection in pregnancy: a case-control study in France. Scand J Infect Dis.;31:305–309.

Berdoy, M., Webster, J. and Macdonald, D. (2000): Proceedings of the Royal Society B: Biological Sciences. 267 (1452): 1591–1594.

Boothroyd, J.C. (1993): Population biology of Toxoplasma: clonality, virulence, and speciation (or not) Infect. Agents Dis. 2:100–2.

Boothroyd, J.C. and Sibley, L.D. (1993): Population biology of *Toxoplasma gondii*. Res. Immunol. 144:14–6.

Buxton, D. and Rodger, S.M, (2008): Toxoplasmosis and neosporosis. In Diseases of sheep. 4th ed (Aitken ID ed) Wiley-Blackwell, Hoboken, p. 112-118.

Chatterton, J.M.W. (1992): Pregnancy. In: Ho-Yen DO, Joss AWL, editors. Human toxoplasmosis. Oxford: Oxford University Press; 144–83.

Cook, A.J., Gilbert, R.E., Buffolano, W., Zufferey, J. Petersen, E., Jenum, P.A., Foulon, W., Semprini, A.E. and Dunn, D.T. (2000): Sources of Toxoplasma infection in pregnant women: European multi centre case-control study. European Research Network on Congenital Toxoplasmosis.

Couvreur, J., Thulliez, P., Daffos, F., et al. (1993): In utero treatment of toxoplasmic fetopathy with the combination pyrimethamine-sulfadiazine. Fetal Diagn. Ther; 8:45–50.

Dabil, H., Boley, M.L., Schmitz, T.M. et al. **(**2001**):** Validation of a diagnostic multiplex polymerase chain reaction assay for infectious posterior uveitis. Arch Ophthalmol.119: 1315–22.

Daffos, F., Forestier, F. and Capella-Pavlovsky, M. (1988): Prenatal management of 746 pregnancies at risk for congenital toxoplasmosis. N. Engl. J. Med.; 318:271–5.

Denkers, E.Y. and Gazzinelli, R.T. (1998): Regulation and function of T-cell-mediated immunity during *Toxoplasma gondii* infection. Clin. Microbiol. Rev.11:569–88.

Desmonts G. and Couvreur J. **(**1979**):** Congenital toxoplasmosis: a prospective study of the offspring of 542 women who acquired toxoplasmosis during pregnancy: pathophysiology of congenital disease. In: Thalhammer O, Baumgarten K, Pollak A, editors. Perinatal medicine: sixth European Congress; Stuttgart: Georg Thieme Verlag;. 51–60.

Dubey, J.P. (2009):"History of the discovery of the life cycle of *Toxoplasma gondii*". International Journal for Parasitology. 39 (8): 877–82.

Dubey, J.P. and Frenkel, J.K. (1972):Cyst-induced toxoplasmosis in cats. J. Protozool;19:155–77.

Dubey. J.P. and Beattie, C.P. (1988): Toxoplasmosis of Animals and Man. Boca Raton, FL: CRC Press,

Dubey, J.P. and Thulliez, P. (1993): Persistence of tissue cysts in edible tissues of cattle fed *Toxoplasma gondii* oocysts. Am. J. Vet .Res. 54(2):270–273.

Dubey. J.P., Kotula, A.W., Sharar, A.K. et al. (1990): Effect of high temperature on infectivity of *Toxoplasma gondii* tissue cysts in pork. J. Parasitol. 76: 201–204.

Dubey. J.P., Lindsay, D.S. and Speer, C.A. (1998): Structures of *Toxoplasma gondii* tachyzoites, bradyzoites, and sporozoites and biology and development of tissue cysts. Clin. Microbiol. Rev.11:267–99.

Dubey. J.P., Lago, E.G., Gennari, S.M., Su, C., Jones, J.L. (2012): Toxoplasmosis in humans and animals in Brazil: high prevalence, high burden of disease, and epidemiology. Parasitology. 139(11):1375–1424.

Evans, R. (1992): Life cycle and animal infection. In: Ho-Yen DO, Joss AWL, editors. Human toxoplasmosis. Oxford: Oxford University Press;. pp. 26–55.

Flegr, J. (2013): "Influence of latent Toxoplasma infection on human personality, physiology and morphology: Pros and cons of the Toxoplasma-human model in studying the manipulation hypothesis". The Journal of Experimental Biology. 216 (Pt 1): 127–33.

Flegr, J., Prandota, J., Sovičková, M. and Zafar, H. Israili (2014): Toxoplasmosis – A Global Threat. Correlation of Latent Toxoplasmosis with Specific Disease Burden in a Set of 88 Countries. PLoS One. 2014; 9(3): e90203.

Foulon, W., Naessens, A. and Derde, M.P. (1994): Evaluation of the possibilities for preventing congenital toxoplasmosis. Am. J. Perinatol; 11: 57–62.

Frenkel, J.K. (2000): Biology of *Toxoplasma gondii*. In: Ambroise-Thomas P, Peterse E, editors. Congenital toxoplasmosis: scientific background, clinical management and control. Paris: Springer-Verlag;. pp. 9–25.

Frenkel, J.K. and Ambroise-Thomas P. (1997): Genomic drift of *Toxoplasma gondii*. Parasitol Res. 83:1–5.

Gale, S.D., Erickson, L.D., Brown, B.L. and Hedges, D.W. (2015): "Interaction between Helicobacter pylori and latent toxoplasmosis and demographic variables on cognitive function in young to middle-aged adults. PLoS ONE. 10 (1): e0116874.

Grover, C, M., Thulliez, P., Remington, J.S. and Boothroyd, J.C. (1990): Rapid prenatal diagnosis of congenital Toxoplasma infection by using polymerase chain reaction and amniotic fluid. J. Clin. Microbiol; 28: 2297–301.

Grundy, M.S., Cartwright-Taylor, L., Lundin, L., Thors, C. and Huldt, G. (1983): Antibodies Against *Entamoeba histolytica* in Human Milk and Serum in Kenya. J. Clinical Microbiology, p. 753-758.

Haridy, F.M., Saleh, N.M., Khalil, H.H. and Morsy, T.A. (2010): Anti-*Toxoplasma gondii* antibodies in working donkeys and donkey's milk in greater Cairo, Egypt. J. Egypt. Soc. Parasitol. Aug; 40(2):459-464.

Hill, D. and Dubey, J. P. (2002): *Toxoplasma gondii*: transmission, diagnosis and prevention. Clin Microbiol Infect; 8: 634–640.

Howe, D.K. and Sibley, L.D. (1995): *Toxoplasma gondii* comprises three clonal lineages: correlation of parasite genotype with human disease. J. Infect. Dis. 172:1561–6.

Innes, E.A. (1997): Toxoplasmosis: comparative species susceptibility and host immune response. Comp. Immunol. Microbiol. Infect. Dis. 20(2):131–138.

Innes, E.A. and Vermeulen, A.N. (2006): Vaccination as a control strategy against the coccidial parasites Eimeria, Toxoplasma and Neospora.Parasitol 133: 145-168.

Innes, E.A., Bartley, P.M., Maley, S. Katzer, F. and Buxton, D. (2009): Veterinary vaccines against *Toxoplasma gondii*. Mem. Inst. Oswaldo Cruz, Rio de Janeiro Mar, 104(2): 246-251.

Jackson, M.H. and Hutchison, W.M. (1989): The prevalence and source of Toxoplasma infection in the environment. Adv. Parasitol.28:55–105.

Jacobs, L. and Melton, M.L. (1966): Toxoplasmosis in chickens. J. Parasitol. 52:1158–62.

Johnson, A.M. (1997): Speculation on possible life cycles for the clonal lineages in the genus Toxoplasma. Parasitol. Today.13:393–7.

Johnson, A.M. (1999): Is there more than one species in the genus Toxoplasma? Tokai J. Exp. Clin. Med. 23:383–9.

Kocazeybek, B., Oner, Y., Turksoy, et al. (2009): "Higher prevalence of toxoplasmosis in victims of traffic accidents suggest increased risk of traffic accident in Toxoplasma-infected inhabitants of Istanbul and its suburbs". Forensic Science International. 187 (1–3): 103–108.

Kapperud, G., Jenum, P.A., Stray-Pedersen, B., Melby, K.K., Eskild, A. and Eng, J. (1996): Risk factors for *Toxoplasma gondii* infection in pregnancy: results of a prospective case-control study in Norway. Am. J. Epidemiol; 144:405–412.

Kotula, A.W., Dubey, J.P., Sharar, A.K. et al. (1991): Effect of freezing on infectivity of *Toxoplasma gondii* tissue cysts in pork. J. Food Protection. 54: 687–90.

Literák, I., Rychlík, I., Svobodová, V. and Pospíšil, Z. (1998): Restriction fragment length polymorphism and virulence of Czech *Toxoplasma gondii* strains. Int J Parasitol. 28:1367–74.

Liu, Q., Singla, L.D. and Zhou, H. (2012): Vaccines against *Toxoplasma gondii*: Status, challenges and future directions. Hum. Vaccin. Immunother. 8(9): 1305–1308.

Nicolle, C. and Manceaux, L. (1908):Sur une infection à corps de Leishman (ou organismes voisins) du gondi. C R Hebd Séances Acad Sci.; 147:763–6.

Paul, M. (1998): Potential risk factors for *Toxoplasma gondii* infection in cases with recently acquired toxoplasmosis. Przegl Epidemiol.; 52:447–54.

Remington, J.S., Desmonts, G., Klein, J.O. and editors. (1990): Toxoplasmosis. In: Infectious diseases of the fetus and newborn infant. 3. Philadelphia: WB Saunders; 1990, 89 –195.

Remington, J.S. McLeod, R. and Desmonts, G. (1995): Toxoplasmosis. In: Remington JS, Klein JO, eds. Infectious Disease of the Fetus and Newborn Infant. Philadelphia: W.B. Saunders Company, 140–267.

Riemann, H. P., Meyer, M. E., Theis, J. H., Kelso, G. and and Behymer, D. E. (1975): Toxoplasmosis in an infant fed unpasteurized goat milk. Journal of Pediatrics. 87 (4) 573-576.

Sacks, J.J., Roberto, R.R. and Brooks, N.F. (1982): Toxoplasmosis infection associated with raw goat's milk. *J.A.M.A.*248:1728–1732.

Siegel, S., Lunde, M., Gelderman, A., Halterman, R., Brown, J.,Levine, A. and Graw, R. (1971): "Transmission of Toxoplasmosis by Leukocyte Transfusion". Blood. 37 (4): 388–394.

Sulzer, A.J., Franco, E.L., Takafuji, E., Benenson, M., Walls, K.W., Greenup, R.L. (1986): An oocyst-transmitted outbreak of toxoplasmosis: patterns of immunoglobulin G and M over one year. Am. J. Trop. Med. Hyg. 35:290–6.

Tenter, A.M. and Johnson, A.M. (1997): Phylogeny of the tissue cyst-forming coccidia. Adv Parasitol.; 39:69–139.

Tenter, A.M., Anja R. Heckeroth and Louis M. Weissb (2000): *Toxoplasma gondii*: from animals to humans. Int J Parasitol. 30(12-13): 1217–1258.

Thierry, C., Isabelle bourguin, Marie-Noelle Mevelec, Jean-Francois Dubremetz, and Daniel Bout (1990): Antibody Responses to *Toxoplasma gondii* in Sera, Intestinal Secretions, and Milk from Orally Infected Mice and Characterization of Target Antigens. Infection and Immunity. 1240-1246

Tibayrenc, M. (1993): Entamoeba, Giardia and Toxoplasma: clones or cryptic species? Parasitol Today. 9:102–5.

Verma, R. and Khanna, P. (2013): Development of *Toxoplasma gondii* vaccine, A global challenge. Human Vaccines & Immunotherapeutics 9:2, 291–293.

Webster, J.P., Kaushik, M., Bristow, G.C. and McConkey, G.A. (2013): The Journal of Experimental Biology. 216 (1): 99–112.