Congenital toxoplasmosis: Clinical features, outcomes, treatment, and prevention

Sarman Singh
Division of Clinical Microbiology and Molecular Medicine, All India Institute of Medical Sciences, New Delhi, India

ABSTRACT
Toxoplasmosis is caused by a coccidian parasite, Toxoplasma gondii. The parasite is highly prevalent both in humans and in warm-blooded animals. Cat family animals are definitive host, and these animals excrete the infective oocysts in their feces. Humans, though not definitive host, get infection by consuming water or food contaminated with cat feces. Rarely, infection can also take place through transfusing the infected blood, through transplantation of infected organs, or transplacentally from infected mother to fetus. Transplacental infection can cause congenital infection with varied degree of clinical manifestations, which depend on the age of fetus when infection took place. Diagnosis of congenital toxoplasmosis is difficult to establish until it is suspected and laboratory investigations are carried out. In more than 75% of cases, acute infection is missed due to very mild or unnoticeable clinical symptoms and signs. In India, a prevalence rate of 22.4% (8.8–37.3%) has been reported with an overall IgM positivity of 1.43%. It is estimated that approximately between 56,737 and 176,882 children per year are born in India with a possible risk of congenital toxoplasmosis. The diagnosis of congenital toxoplasmosis can be made by serological methods which are most commonly used. The other methods are parasite isolation by culture and molecular methods. Toxoplasmosis is treatable and transplacental transmission can be prevented by spiramycin, which concentrates in the placenta. However, if infection has done any damage to the fetus or the parasite has passed the placenta, spiramycin cannot reverse the damage. Prevention remains the best remedy.

KEY WORDS
Effect of climate, food-borne toxoplasmosis, incidence, India, live births, oocysts, prevalence, sexual mode of transmission

INTRODUCTION
Toxoplasmosis is an important constituent of Toxoplasma, Others (syphilis, Parvovirus B19, Varicella zoster, Hepatitis B virus), Rubella, Cytomegalovirus, Herpes viruses (TORCH) group of infections that if acquired during pregnancy, it can cause congenital infections and may lead to permanent disability or defects in the fetus and even fetal loss.\(^1,2\) While three other pathogens are viruses, Toxoplasma is a protozoan parasite. Despite

Access this article online

Quick Response Code:

Website:
www.tropicalparasitology.org

DOI:
10.4103/2229-8070.190813

For reprints contact: reprints@medknow.com

This is an open access article distributed under the terms of the Creative Commons Attribution-NonCommercial-ShareAlike 3.0 License, which allows others to remix, tweak, and build upon the work non-commercially, as long as the author is credited and the new creations are licensed under the identical terms.

How to cite this article: Singh S. Congenital toxoplasmosis: Clinical features, outcomes, treatment, and prevention. Trop Parasitol 2016;6:113-22.

DOA: 07-08-2016, DOP: 19-09-2016
its significant impact on pregnancy outcomes, the toxoplasmosis remains neglected, especially in the Indian subcontinent. Toxoplasma is also an important opportunistic pathogen in immunocompromised hosts and also a major concern for animal health. However, the topic is vast and it is beyond the scope of this review to give a comprehensive and exhaustive account of toxoplasmosis; hence, I will try to focus only on the management of acute Toxoplasma infection occurring during pregnancy and possibilities of congenital toxoplasmosis.

CAUSATIVE AGENT AND LIFE CYCLE

The infection is caused by a coccidian parasite Toxoplasma gondii, most common of all the protozoan infections and having widest host range. The parasite was first described in 1908 by Nicolle and Manceaux in the blood, spleen, and liver of North African rodent - gundi (Clenodactylus gundi) which was captured in Tunisia. However, in 1900, a similar structure was reported by Leveran in the spleen and bone marrow of Java sparrow (Padda oryzivora). However, this structure was considered as reproductive form of Haemamoeba danilewskyi.[3] Because the parasite looked like leishmania promastigotes and because it was discovered in rodent C. gundi, Nicolle and Manceaux named it as Leishmania gondii. However, after 1 year in 1909, they reconsidered and renamed it as T. gondii (toxon = arc, plasma = form).[4]

In fact, Dubey et al.[3] in 1970 first time established the life cycle of this parasite. They showed that the parasite multiplies in the intestinal mucosa of a cat. Later on, it was found that all cat family (Felidae) animals act as definitive host. The young kittens are highly infectious. However, the oocysts are shed in the kitten feces only in the first 2–3 weeks of infection. However, even during acute infection, the cat does not exhibit any symptom. Other animals (in fact, almost all warm-blooded animals) including humans get accidentally infected.[3,4]

MODES OF TRANSMISSION

When the life cycles were not fully known, the major question was how the humans get infected from the cat. However, at present, it is well established that there are three primary routes of transmission of T. gondii by (1) ingesting the food and water contaminated with cat feces, containing oocysts of the parasite, (2) ingesting uncooked or undercooked meat containing tissue cysts of the parasite, and (3) congenitally from mother-to-fetus through placenta.[1,2,6-12] Transplacental infection occurs when an uninfected mother acquires infection during pregnancy. There are other rare modes of transmission such as through organ transplantation, blood transfusion, and laboratory acquired.[13,14] In India, this infection is more common in socioeconomically backward and tribal populations, particularly in those who work barefoot and bare hands in the fields and consume contaminated water or vegetables.[6,15] It has also been found to be more prevalent in populations where iodine deficiency is more pronounced.[12]

INCIDENCE AND PREVALENCE OF TOXOPLASMOsis IN India

In India, serologic studies reveal that a large section of the population has had the disease at one time or the other. A wide variation in the prevalence and incidence rates of this infection has been reported.[6,12,15-21] Our group earlier studied its seroprevalence in various cohorts and found prevalence rates ranging from 26% in Delhi to as high as 77% in Sub-Himalayan parts of India, when indirect hemagglutination test (IHAT) was used.[15] A prevalence rate of 41.75% was noted in primigravida woman cohort from a nearby rural area of Delhi, when direct agglutination test (DAT) was used.[16] Using combination of several serological methods and internationally approved standards, another study carried out in 2004 found a seroprevalence rate of 45% and incidence rate of <3% in a North Indian cohort of a woman with pregnancy of <4 months duration.[21] Overall, approximately 1% of the general human population becomes infected congenitally. In a recent study, we found that estimated number of children born with risk of congenital toxoplasmosis per year in India is 387,904.[3]

TOXOPLASMA AS A FOOD-BORNE PATHOGEN

The expenditures on human illness due to food-borne pathogens are estimated to be US$ 6.5–34.9 billion annually in the United States of America (USA) alone. The presence of food-borne pathogens in a country’s food supply not only affects the health of the local population but also represents hygienic conditions in that country. This also possesses risk of spread of infection to the visitors to that country and to consumers of countries where these food products are exported. In the USA, food-borne toxoplasmosis is considered to be the third leading food-borne cause of death.

Vegetables, salads, and water as source of Toxoplasma infection

The oocyst-mediated infection plays major role in developing countries. The oocysts that sporulate in the soil are infectious to intermediate hosts including grazing animals and humans. These oocysts once passed out from cat intestine through feces, these may remain in the soil or the contaminated soil is admixed with water, can adhere to the land vegetables and fruits which are consumed raw by unaware children and adults without washing. Commonly used ground vegetables and fruits that can get contaminated in this way are carrots, redish, sweet potatoes, red beet, cauliflower, spinach, etc. Even other
vegetables and fruits can be contaminated by humans through the use of soil-contaminated utensils, knives, and if the cut fruits are left open and allowed to access by flies and other insects. Food handlers and vegetable vendors have also been reported to serve as source of Toxoplasma infection as they clean the soiled vegetables with bare hands often with waste water.\textsuperscript{[6,11,12]}

Contaminated water, if consumed as such, carries high risk of Toxoplasma infection or can lead to contamination of the food and thus facilitate the transmission of \textit{T. gondii} in various ways. Transmission through water is most difficult to distinguish from meat-borne infections. In developing countries where safe drinking water is not available to more than half of the population, water itself or vegetables irrigated and washed with such water and food served using contaminated water can lead to oocysts ingestion. Several epidemiological studies have supported this hypothesis. We reported that in Sub-Himalayan areas of India where natural spring water is drunk by the local inhabitants, the seroprevalence of toxoplasmosis is highest in India. In this area, the natural water sources are common and are shared by both humans and animals including feline animals. The cats and other members of the cat family are definitive host of this parasite after drinking the water, taking bath, and even defecating in the same water body. The water flows from higher altitude to lower residential areas, and the inhabitants without knowing that this water is contaminated consume it. In this area, seroprevalence of toxoplasmosis was found to be 77% in women as well as in sheep of the area.\textsuperscript{[13]} In another study, we did not find any difference in the prevalence between nonvegetarian and vegetarian Indians, suggesting that contaminated vegetable and water are major sources of infection in the developing world.\textsuperscript{[14]}Several well-documented outbreaks of water-borne toxoplasmosis have been reported from both developing as well as developed world.\textsuperscript{[7,10]}

\textbf{Animal meat as source of Toxoplasma infection}

Toxoplasma is known to be encysted in tissues of both humans and animals. Each cyst contains many viable trophozoites of the parasite, and these are highly infectious and capable of invading the intestinal wall of the carnivores/omnivores. It is interesting that as early as 1923 (only 15 years after its discovery), frequent eating of rabbit meat was considered as a contributory factor for congenital toxoplasmosis. Serological studies also confirm that several animals are infected with \textit{T. gondii} and they transmit this infection when their meat is consumed. Outbreaks of toxoplasmosis associated with raw meat consumption have been reported. On animal toxoplasmosis, maximum work has been done by Dr. Dubey, a doyen in the field. More details of his findings can be found in his recent books and book chapters.\textsuperscript{[3,4]}

\textbf{Sheep meat}

Sheep are commonly infected with \textit{T. gondii}. Sheep from five flocks in Yorkshire, UK, were identified as having Toxoplasma infection. The prevalence of antibody in the hill flock was lower than the grassland or Worrall flocks. Similarly, 5% of Navajo sheep were positive for antibody as compared to 56% of Kentucky sheep. The Navajo sheep are reared in dry, mountainous areas whereas Kentucky sheep are reared on rich grassland; therefore, it was postulated that environmental factors might have played major role in these varied prevalence rates. Similar findings were reported from India. High Toxoplasma seroprevalence of 77% was reported in sheep and goats of Kumaon region of India,\textsuperscript{[22]} whereas from the Rajasthan area 28% of sheep and 13.3% of goats were reported seropositive.\textsuperscript{[23]} The Kumaon region is a Sub-Himalayan terrain with rich vegetation, humid environment, and high population of wandering felines, but the Rajasthan province is dry, desert, and extremely hot area. The seroprevalence trends of human toxoplasmosis in the respective areas have also shown concordance. These trends are indicative of oocyst infection; the presence of cats and survival of oocysts in those environmental conditions. Toxoplasma has been isolated from tissues (skeletal muscle and brain tissue) of naturally infected sheep; parasite was more commonly seen in the musculature than the brain. However, even though mutton (word used for both sheep and goat meats) is most commonly consumed meat in most of the Asian countries including India, the chances of toxoplasmosis through meat as such are rare because in the Indian subcontinent meat is cooked well and semi-cooked and raw meat is rarely consumed. However, in low socioeconomic populations, precooking processing and cleaning of meat and improper cleaning of hands and meat utensils can be potential sources of toxoplasmosis. For a comprehensive data on toxoplasmosis in sheep, readers are encouraged to go through a recent review by Dubey.\textsuperscript{[24]}

\textbf{Goat meat}

Goats are commonly infected with \textit{T. gondii}. Various experimental and histopathological studies have revealed that most commonly infected tissues of goat are skeletal muscle, diaphragm, intestine, heart, kidneys, and liver, in the same order. A few outbreaks of food-borne human toxoplasmosis have been reported after consuming raw goat meat. Although in Indian continent goat meat (called mutton in this continent) is the most common meat consumed, the chances of toxoplasmosis through this route are rare as the meat is cooked adequately. Raw meat in these countries is used extremely rarely during religious functions where some tribes consume sheep and goat’s blood raw. In several other countries, the natives hunt and consume raw meat of wild goats. Studies from the USA had shown a
prevalence of anti-Toxoplasma IgG in goats up to 22%, but the seroprevalence data from India indicate that 32–77% of Indian goats are seropositive for \( T. gondii \).[3,4,22] Further, in countries such as India where automated slaughtering is not available, prevalence rate of 70% has been reported in the butchers of goats and sheep.[23] This may be explained by the fact that butchers usually handle their food with unclean and often contaminated hands and it is likely that some \( T. gondii \) tissue cysts are also ingested with food.

**Pig meat (pork)**
The seroprevalence of toxoplasmosis in pigs is reported to vary from <1% to as high as 100%.[3] It has also been reported that prevalence varies from herd to herd as well as with the age of the animals. The seroprevalence of toxoplasmosis in breeding swines was reported as high as 96.8% as compared to 20% in growing/finishing pigs. The access of cats to the pig farms has unequivocally been associated with infection of Toxoplasma in pigs. Toxoplasma has been isolated from various tissues (skeletal muscle, heart, and brain) of naturally infected pigs. Outbreaks of human toxoplasmosis after consuming uncooked pork have been reported from Asian countries also.[1,4]

From India and other Southeast Asian countries, very few studies are published. The major reason is that these countries eat pork minimally due to their cultural and religious beliefs. A few studies done in India using IHAT in the 1970s and early 1980s showed that seroprevalence of Toxoplasmosis in Indian pigs ranges from 14% to 31%.[3] No studies were done later on.[25]

**Cattle meat (beef)**
Some doubt has been cast over the accuracy of prevalence rates of toxoplasmosis in cattle as most of these studies were based on serological testing. The dye test (DT) is too unreliable for the diagnosis of toxoplasmosis in cattle as false positive results are caused by bovine serum protein. Although the prevalence of tissue cysts in cattle is low, beef is one of the most common meats eaten in Britain.[3,4,11] In India where beef is rarely eaten by Hindus, but in Pakistan and other Islamic regions where beef is the main meat, seroprevalence is reported to vary from 8% to 25%. However, no work has been done to find the prevalence rate of toxoplasmosis in snow cattle, for example, yaks which are used not only as baggage careers and source of wool but also as source of meat for Himalayan tribes.[26–28]

**Poultry**
The work on prevalence of Toxoplasmosis in poultry is almost confined to only one laboratory in the USA. Dr. Dubey et al. have done extensive work on this neglected area. These studies indicated that prevalence rates varied from as low as 6.2% in Mexico, 16.9% in Ohio state of the USA, 17.9% in India, 40–65.15% in Brazil, 40.4–47.2% in Egypt, and as high as 65.5% in Argentina. These workers also found that 70% isolates from chicken were genotype I and 27% isolates from chicken were genotype III. Most of these studies were done on free-ranging chicken, and the authors concluded that prevalence of \( T. gondii \) in free-ranging chickens is a good indicator of prevalence of \( T. gondii \) oocysts in the environment of the area because these chickens feed from ground.[3,6,11]

Seroprevalence studies in other meat birds have also been done but scarcely, showing seroprevalence of 50% in ducks (\( Anas \) sp.) and 59.5% in turkeys of Egypt. Similarly, 18% gees (\( Anseranas semiarguldactyla \)) of Texas, USA, were reported seropositive.[3,6,11] We, in India, screened 66 wire-caged quails from a North Indian veterinary institution and found 59% of these were found seropositive for \( T. gondii \) by modified DAT. The quail meat is used as special feast in India.

**Other meats and meat products**
Only a little information is available about Toxoplasma infection in other animals such as horses, camels, kangaroos, water buffaloes, and elephants whose meat is often consumed in some countries.[6,4]

There are anecdotal reports of toxoplasmosis associated with consumption of raw milk and hen eggs. However, parasite has not yet been isolated from the eggs commonly consumed by human. If at all, the infection can occur through contaminated and broken egg shell if eggs are laid on the soil-containing Toxoplasma oocysts. Although Toxoplasma has been demonstrated in the milk of experimentally infected cat, sheep, goat, and mice, whether these reports can be considered indicative of milk-borne transmission in human is yet to be established. The survival of Toxoplasma has been reported for up to 10 days in milk and unprocessed home-made cheese. For more details, readers are encouraged to read a recent review by Singh and Munawwar.[13]

**Clinical manifestations of congenital infection**
The severity and likelihood of infection depend on the trimester of pregnancy the mother becomes infected with \( T. gondii \). Toxoplasmosis is more severe in infants whose mothers become infected during the first trimester than those during the third trimester.[1,2,15,21] Transmission of \( T. gondii \) from preconception seropositive mothers to their babies is rare, but occasional reports of seropositive mothers transmitting Toxoplasma to their child are on record. Although most congenitally infected children are asymptomatic at birth, they may develop some symptoms later in life. Loss of vision is the most common (up to 95%) sequel in congenitally infected children. Hydrocephalus, retinochoroiditis,
Singh: Congenital toxoplasmosis

Infection of the mother before pregnancy rarely, if not ever, results in birth of a congenitally infected child. However, in countries such as France and Austria, all seropositive women are also tested every trimester for rising IgG titers. Those women who seroconvert during pregnancy are followed clinically and their fetuses are examined for T. gondii infection by ultrasound and amniocentesis and for the presence of T. gondii in amniotic fluid and fetal blood. For more details, refer to previous review.[2]

Important findings from these studies are as follows:
- Infection of the mother before pregnancy rarely, if not ever, results in birth of a congenitally infected child.
- Half of the women who acquire T. gondii infection during pregnancy do not transmit the parasite to their fetus.
- T. gondii is transmitted more frequently during the latter part of gestation, but the disease is more severe if infection is acquired during the first and second trimesters.
- Detection of T. gondii in amniotic fluid is possible with polymerase chain reaction (PCR)
- Except in rare instances, T. gondii does not cause abortion or sterility in women.

Is Toxoplasma infection sexually transmitted?
Although it has not been proved following the Cox postulate, there are enough epidemiological and case studies which show that females’ partners of males with positive evidence of toxoplasmosis have high chances of becoming Toxoplasma positive. In a recent study, we found a significantly high prevalence of toxoplasmosis in married women as compared to unmarried women.[4]

There is also evidence of finding Toxoplasma DNA in semen samples.

Multiple factors lead to Toxoplasma infection
The incidence and prevalence of toxoplasmosis are determined by a number of factors. These factors as mentioned above include environmental factors, parasite strain, and the host.[6,11] The environment plays most crucial role in the dissemination and transmission of the parasite. Within the environmental conditions are factors such as heat and humidity, number of feral cats, opportunity of the cat feces to mix with the soil, and access of intermediate hosts to this contaminated soil. Further, the recent studies also suggest that virulence of the parasite may vary with genotype of the parasite involved. Nonetheless, host factors are also equally important. These host factors are age and immune status of the host. The dietary habits and cultural and religious background of the host can also play major roles. These factors may influence the epidemiology of Toxoplasmosis in humans.

Chorioretinitis, intracerebral calcification, mental retardation, loss of hearing, and very rarely death may also occur.[1,2,4]

Clinical manifestations of postnatal-acquired infection
Although most postnatal-acquired infections are asymptomatic, manifestations of toxoplasmosis include mild lymphadenopathy (particularly of the cervical region), headaches, muscle aches, and sore throat. Because these symptoms are nonspecific, postnatal toxoplasmosis is rarely diagnosed. Toxoplasmosis in AIDS patients and other immunocompromised patients can be life-threatening. Heart and other organ transplantation recipients are at risk for developing toxoplasmosis because of lowering of host resistance by immunosuppressive medication. Similarly, cancer patients are also at risk of developing clinical toxoplasmosis. In most of these immunocompromised patients, toxoplasmosis results from reactivation of a latent infection, especially due to the rupture of tissue cysts which lead to the renewed multiplication of the parasite’s tissue forms. Although any organ may be involved, encephalitis is the predominant presentation of toxoplasmosis in AIDS patients.[4]

Toxoplasma as a cause of repeated bad obstetric outcome
There is considerable confusion and uncertainty concerning T. gondii as a cause of multiple abortions, sterility, and other reproductive failures in India. The term bad obstetric history (BOH) or bad obstetric outcome has been coined to associate Toxoplasmosis by Indian microbiologists and obstetricians, based on serology findings. However, my team has been against associating Toxoplasma with these conditions. There are several flaws in linking habitual abortion with Toxoplasma infection. In most of these studies which associated prevalence of anti-Toxoplasma antibodies with multiple abortions, the number of patients was low, studies were uncontrolled, and serologic data before pregnancy were not obtained. In many cases, poor-quality test kits were used which yielded high false positivity.[6,21] We realize that there are technical difficulties in conducting a well-controlled prospective study in India that is necessary to establish a causal relationship between toxoplasmosis and abortion. Even isolation of T. gondii from the endometrium several weeks after abortion does not prove congenital toxoplasmosis because T. gondii has been found in the uterus of nonpregnant women. Furthermore, even when the placenta is infected, the fetus may escape infection.[4]

Much that is known about diagnosis and management of T. gondii during pregnancy has come from the studies in Austria and France where it is compulsory by law to test all pregnant women for T. gondii. Women are tested for T. gondii antibodies on their first visit to their gynecologist. Seropositive women are not tested further during pregnancy in countries where no national screening programs exit; however, in countries such as France and Austria, all seropositive women are also tested every trimester for rising IgG titers. Those women who seroconvert during pregnancy are followed clinically and their fetuses are examined for T. gondii infection by ultrasound and amniocentesis and for the presence of T. gondii in amniotic fluid and fetal blood. For more details, refer to previous review.[2]
New challenges to the safety of the food supply require new strategies for evaluating and managing food safety risks. Changes in pathogens, food preparation, distribution, and consumption, and population immunity have the potential to adversely affect human health. Risk assessments may provide prediction of the impact on the provision of safe food and health of consumers. Risk assessment models facilitate the evaluation of active or passive changes in how foods are produced, processed, distributed, and consumed.

ASSOCIATION OF GEO-CLIMATIC AND SOCIODEMOGRAPHIC CONDITIONS

In a recent study, we selected four regions of India [Figure 1, map] with maximum diversity in cultural and climatic conditions to see if these environmental factors are associated with variation in the incidence and prevalence of toxoplasmosis.[6]

The overall seroprevalence of toxoplasmosis in Indian women of reproductive age amounted to 22.4%. The prevalence rates varied significantly among four regions, being highest in South India (37.3%) and least in West India (8.8%). The difference was highly significant. Most women (97.1%) belonged to low-income or lower middle-income group, and most women from South India and East India resided in mud-plastered houses and consumed tube well/hand pump water without using any disinfectant. The difference was highly significant ($P < 0.001$) in the living conditions between South India and North India and South India and West India. Most women were involved in housekeeping. About one-third (29.1%) women gave a history of contact with animals, but pet animals, mainly cats, were significantly more common (53.5%) in South Indian households. Consumption of raw salad was very common (92.89%) across the whole country. Seroprevalence increased with age. Most of the seropositive women were multigravida (74.2%), suggesting that most of the obstetricians request TORCH tests only after multiple abortions or nonliving issues.

Incidence rate was very low (1.43%) which found to be positive for anti-$T. gondii$ IgM antibodies. We observed that the incidence of toxoplasmosis was not homogeneously among the four regions. Incidence was highest (2.9%) in South India, 0.6% in Western India, 0.4% in North India, and 0.8% in Eastern India. The study showed that the socioeconomic background of North Indian women was better than others as most of them were from urban background (Delhi and national capital region). While in rest three regions, the population was predominantly rural with lower socioeconomic background. It is unfortunate that in India, most women seek medical advice after undesirable pregnancy outcome and results from such studies have created a myth that Toxoplasma is highly associated with multiple pregnancy losses or BOH, the term which I do not subscribe to. The fact is that higher prevalence rate is associated with advanced age rather than it has any relation with multiple pregnancy outcomes.

The climatic conditions in South India favor sustenance and proliferation of Toxoplasma oocysts. A highly significant number of households owned pet cats in this region. Moreover, as a social culture, South Indian population does not wear shoes and most often are barefooted or only put sleepers. On the contrary, West India (Gujarat) is a dry, arid climatic zone where temperature in May–June touches 46°C. Socioculturally also, population in West India must wear shoes due to high temperature and sandy soil. These climatic conditions are detrimental for $T. gondii$ to maintain its life cycle. Earlier also, we reported that seroprevalence was highly significantly lower in dry arid areas of Rajasthan which is adjacent to Gujarat as compared to prevalence in animals of the Sub-Himalayan region, which has humid tropical climatic conditions.[22] The role of environmental conditions on prevalence to toxoplasmosis is well established.[23]

We also found that true incidence rate of toxoplasmosis in India is indeed very low (1.43%), but exaggerated figures are published in various other studies from India. These alarmingly high incidence rates are observed in
Although the incidence rate we observed may seem very low, cumulative figures are scary. We estimated that child births (living or still) with risk of congenital toxoplasmosis would be approximately 387,904 per annum. We also concluded that carrying out only IgM test without IgG testing is not an advisable approach of investigating toxoplasmosis, and all patient must be tested first for IgG and if found positive only in that case, laboratory should perform IgM and/or avidity test.\[^6\]

**DIAGNOSIS**

Diagnosis of acute toxoplasmosis is often missed because acute infection does not present with any pathognomonic symptom or sign. It can present with very mild flu-like features with or without lymphadenopathy. Very rarely, a crop of central rash may be noticed on careful examination. Because of these reasons, most of the patients ignore the acute infection. Therefore, strong clinical suspicion is the key for laboratory investigations, which play crucial role. The laboratory diagnosis can be made by doing serology, culture isolation (in animal models or cell lines), or molecular methods such as PCR.

**SERODIAGNOSIS OF ACUTE TOXOPLASMOSIS IN A PREGNANT WOMAN**

The detection of Toxoplasma-specific antibodies is the primary diagnostic method to determine infection with Toxoplasma. Toxoplasma antibody detection tests are performed by numerous serologic tests, and most of these are commercially available to detect IgG and IgM *T. gondii* specific antibodies. The Sabin–Feldman DT, indirect fluorescent antibody test (IFAT), IHAT, latex agglutination test (LAT), DAT, and enzyme-linked immunoabsorbent assay (ELISA) are some of the tests used to detect *T. gondii* antibodies. Although the DT is the most specific, it is rarely used now because it uses live virulent *T. gondii*. The IFAT is nearly as sensitive as the DT, but it requires a fluorescent microscope. The IFAT, LAT, DAT, and ELISA are used more commonly.\[^2,6,13,21,29-31\] An algorithm for the immunodiagnosis of toxoplasmosis for individuals >1 year of age has been published with the help of a flowchart in 2003.\[^2\] The immunofluorescence assay and enzyme immunoassay (EIA) tests for IgG and IgM antibodies are the tests most commonly used today. Persons should be initially tested for the presence of Toxoplasma-specific IgG antibodies to determine their immune status. The presence of IgG antibodies only means exposure because asymptomatic humans can develop very high (>100,000) *T. gondii* antibody titers, and titers may remain elevated for several years or even whole life if repeated exposures are encountered. Although an 8-fold rise in antibody titers, taken 2 weeks apart, is indicative of a recent infection, this is seldom achieved in practice because by the time patients are seen in the clinic, antibody titer has usually peaked. Compared to IgG antibodies, IgM antibodies are short-lived and they appear before IgG antibodies.\[^21,29-31\]

If more precise knowledge of the time of infection is necessary, then an IgG-positive person should have an IgM test performed by a procedure with minimal nonspecific reactions, such as IgM-capture EIA. A negative IgM test essentially excludes recent infection, but a positive IgM test is difficult to interpret because Toxoplasma-specific IgM antibodies may be detected by EIA for as long as 18 months after acute acquired infection. A major problem with Toxoplasma-specific IgM testing is lack of specificity. There are two possibilities: (i) persons with a positive IgM but negative IgG and (ii) individuals with positive IgG and IgM results. In the first situation, a positive IgM result with a negative IgG result in the same specimen should be viewed with great suspicion; the patient’s blood should be redrawn 1–2 weeks after the first and tested together with the first specimen. If the first specimen was drawn very early after infection, the patient should have highly positive IgG and IgM antibodies in the second sample. If the IgG is negative and the IgM is positive in both specimens, the IgM result should be considered as a false positive and the patient should be considered as not infected. In the second situation, a second specimen should be drawn and both specimens submitted together to a reference laboratory which employs a different IgM testing system for confirmation.

**IGG AVIDITY TEST**

Avidity of IgG (binding capability of the antibody with the antigen) is very useful test.\[^30,34\] Avidity of IgG can be tested by either ELISA or commercially available Vitek Immuno-Diagnostic Assay System (VIDAS, BioMerieux SA, France). The latter is more standardized and has its own system controls and standards. In a pregnant woman whose sample is taken in the second trimester rather than ideally in the first trimester, and she is found IgG positive but IgM negative, it is more advisable to perform IgG avidity test. High avidity IgG tests indicate that she acquired the infection more than 4 months ago.\[^21,32,34\] However, the low avidity is not a confirmatory test for recent infection. Before initiation of patient management for acute toxoplasmosis, all IgM positives should be verified by a reference laboratory with experience in toxoplasmosis such as Toxoplasma reference laboratory, Department of Laboratory Medicine, All India Institute of Medical Sciences, New Delhi. In either case, the clinical microbiologist must counsel the patient patiently, a practice lacking in
the Indian subcontinent, as most clinical microbiologists do not interact with patients directly.

SERODIAGNOSIS OF CONGENITAL TOXOPLASMOSIS IN A NEWBORN

Diagnosis of congenital toxoplasmosis in a newborn child presents many difficulties because of the transfer of material IgG antibodies to the fetus, low sensitivity of serologic tests, and lack of availability and cost of T. gondii-specific IgA detection kits. Newborn babies and infants with suspected congenital toxoplasmosis should be tested by both IgM- and IgA-capture EIA. Detection of Toxoplasma-specific IgA antibodies is more sensitive than IgM detection in congenitally infected babies. Considering the high cost of a screening program for congenital toxoplasmosis during pregnancy, a program to screen cord blood for IgM antibodies to T. gondii, coupled with screening for other congenital infections, might be more cost-effective than screening for toxoplasmosis alone.

TREATMENT

Toxoplasmosis can be treated with rovamycin (spiramycin), a drug which is marketed in the form of tablets each containing 3 MIU (1 g) of spiramycin salt. Spiramycin is a macrolide antibiotic, which binds irreversibly to the 50s ribosomal subunit inhibiting microbial protein synthesis. It is one of the very few antimicrobials capable of intracellular penetration, including the macrophages, thus eliminating intracellular pathogens in active as well as carrier stages of the disease. The drug is given orally. Gastric acid appears to have minimal effect, but coadministration with food significantly reduces bioavailability and delays the time to peak concentration. In contrast to the high levels found in most tissues and body fluids, it does not penetrate into the cerebrospinal fluid. Placental transfer is poor and only 9-16% found in the maternal blood concentration appears in the amniotic fluid. The drug is eliminated from the body slowly, the majority being inactivated in the tissues. Since it is a macrolide, it is contraindicated in persons known to have hypersensitivity to this group of medicine. Otherwise, the drug is well tolerated and serious adverse reactions are extremely rare. There are no specific contraindications to its usage in lactation.

In pregnant women, 6 MIU to 9 MIU (2-3 tablets) daily in 2 or 3 divided doses for 3 weeks are given. This 3-week course should be repeated after a 2-week interval till parturition. In pregnant women with confirmed Toxoplasma infection, oral spiramycin 1 g 3 times daily has been administered to prevent transmission to the fetus. In these women, the drug should be started as soon as possible after proven or suspected maternal diagnosis and continued throughout pregnancy. Monitoring for the presence of fetal infection is indicated. The newborns and infants with confirmed congenital toxoplasmosis and adults males with acquired toxoplasmosis should be treated with pyrimethamine plus sulfadiazine. Other drugs showing variable cure rate are azithromycin, atovaquone, and dapsone.[35] Spiramycin has no role in postnatal toxoplasmosis. In AIDS patients, clarithromycin gives best results. However, due to low efficacy of all the currently available drugs[36] and associated side effects with sulfa + pyrimethamine drug combination, search for new compounds has been expedited.[36]

PREVENTION

Since there is no effective vaccine against human toxoplasmosis so far, the best way to prevent Toxoplasmosis in pregnant women is to practice the following measures:

- Cook meat thoroughly before consumption
- Wash hands and utensils thoroughly after handling uncooked meat
- Wash fruits and vegetables before eating
- Pregnant and immunocompromised individuals must consume ground vegetables (e.g., reddish, carrot, sweet potato etc.) only after blanching these. It is misconception that cabbage consumption carries a risk. In fact cabbage is one of the safest ground vegetable
- Wear gloves while gardening where applicable
- Deep frozen meat is preferred over fresh meat as freezing at <−20°C is highly detrimental for T. gondii tissue cysts
- Cats become infected either from other cats or from eating the flesh of infected birds or mammals. Therefore, pet cats should not be fed raw meat and should be prevented from hunting or scavenging. Dispose of cat feces and litter daily before the Toxoplasma organism have a chance to become infective. Feces can be flushed down the toilet or deeply buried; litter can be sealed in a plastic bag in a manner that will not disperse litter dust (and possibly the T. gondii organism) into the air. Disinfect litter pans daily with scalding water. Unless they are known to have immunity to toxoplasmosis, pregnant women should avoid cleaning litter pans and avoid contact with cats that have an unknown feeding history
- Outdoors, one should wear gloves when gardening. Prevent cats from gaining access to sandboxes used by children; change sand if it is contaminated.

CONCLUSIONS

T. gondii is a coccidian parasite and most widely prevalent in warm-blooded animals including humans.
If the infection is acquired during pregnancy, it can lead to congenital toxoplasmosis, which can cause mild to severe congenital abnormalities and even fetal loss. However, the infection acquired once has no role in multiple pregnancy losses. Spiramycin (rovamycin) is the drug of choice which prevents the transplacental transmission of the parasite. However, if the parasite has already crossed the placenta, spiramycin cannot reverse the damage caused to the fetus. For treating the postnatal toxoplasmosis (e.g., nonpregnant prospective mothers or males), sulfadiazine plus pyrimethamine combination is the best option. As for any other infection, prevention is better than treating it. Best methods of preventing the infection are proper personal hygiene, consumption of properly cooked meat and properly washed vegetables and fruits, and safe drinking water.

Financial support and sponsorship
Nil.

Conflicts of interest
There are no conflicts of interest.

REFERENCES
1. Remington JS, Klein JO. Infectious Diseases of the Foetus & Newborn Infants. 4th ed. Philadelphia: W.B Saunders Company; 1995. p. 140-266.
2. Singh S. Mother-to-child transmission and diagnosis of Toxoplasma gondii infection during pregnancy. Indian J Med Microbiol 2003;21:69-76.
3. Dubey JP, Beattie CP. Toxoplasmosis of Animals and Man. 2nd ed. Florida, Boca Raton: CRC Press; 2010. p. 336.
4. Dubey JP. Toxoplasmosis. In: Cox FE, Kreier JP, Wakelin D, editors. Topley Wilson's Microbiology and Microbial Infections: Parasitology. 9th ed. New York: Oxford University Press; 2010. p. 303-18.
5. Dubey JP, Miller NL, Frenkel JK. Characterization of the new feline form of Toxoplasma gondii. J Parasitol 1970;56:447-56.
6. Singh S, Munawwar A, Rao S, Mehta S, Hazarika NK. Serologic prevalence of Toxoplasma gondii in Indian women of child bearing age and effects of social and environmental factors. PLoS Negl Trop Dis 2014;8:e2737.
7. Dubey JP. Toxoplasmosis – A waterborne zoonosis. Vet Parasitol 2004;126:57-72.
8. Jones JL, Dubey JP. Foodborne toxoplasmosis. Clin Infect Dis 2012;55:845-51.
9. Palanisamy M, Madhavan B, Balasundaram MB, Andavar R, Venkatapathy N. Outbreak of ocular toxoplasmosis in Coimbatore, India. Indian J Ophthalmol 2006;54:129-31.
10. Bahia-Oliveira LM, Jones JL, Azevedo-Silva J, Alves CC, Oréfice F, Addiss DG. Highly endemic, waterborne toxoplasmosis in north Rio de Janeiro state, Brazil. Emerg Infect Dis 2003;9:55-62.
11. Singh S, Munawwar A. Human toxoplasmosis: A food borne parasitic disease. Proc Natl Acad Sci India 2009;79:207-20.
12. Singh S, Singh N, Pandav R, Pandav CS, Karmarkar MG. Toxoplasma gondii infection & its association with iodine deficiency in a residential school in a tribal area of Maharashtra. Indian J Med Res 1994;99:27-31.
13. Singh S, Lodha R, Passi GR, Bhan MK. Cholestatic jaundice due to congenital Toxoplasma gondii infection. Indian J Pediatr 1998;65:154-7.
14. Derouin F, Pelloux H; ESCMID Study Group on Clinical Parasitology. Prevention of toxoplasmosis in transplant patients. Clin Microbiol Infect 2008;14:1089-101.
15. Singh S, Nautiyal BL. Seroprevalence of toxoplasmosis in Kumaon region of India. Indian J Med Res 1991;93:247-9.
16. Akotjam BS, Kant S, Singh S, Kapoor SK. Seroprevalence of toxoplasma infection among primigravid women attending antenatal clinic at a secondary level hospital in North India. J Indian Med Assoc 2002;100:591-2, 604.
17. Hingorani V, Prakash O, Chowdhry P, Kamalad TS. Toxoplasmosis: Abortions and stillbirths. Indian J Med Res 1970;58:967-74.
18. Mittal V, Bhatia R, Singh VK, Sehgal S. Prevalence of toxoplasmosis in Indian women of child bearing age. Indian J Pathol Microbiol 1995;38:143-5.
19. Pal MN, Aggarwal DS. Toxoplasmosis and abortion. J Obstet Gynaecol India 1979;29:59-61.
20. Sharma P, Gupta I, Ganguly NK, Mahajan RC, Malla N. Increasing toxoplasma seropositivity in women with bad obstetric history and in newborns. Natl Med J India 1997;10:65-6.
21. Singh S, Pandit AJ. Incidence and prevalence of toxoplasmosis in Indian pregnant women: A prospective study. Am J Reprod Immunol 2004;52:276-83.
22. Dubey JP, Somvanshi R, Jitendran KP, Rao JR. High seroprevalence of Toxoplasma gondii in goats from Kumaon region of India. J Vet Parasitol 1987;7:17-21.
23. Singh S, Singh N. Seroepidemiology of toxoplasmosis in sheep and goats of Rajasthan state and their butchers. In: Somvanshi R, Lokeshwar RR, editors. Current Advances in Veterinary Sciences and Animals Production in India. Lucknow: International Book Distributing Co.; 1994. p. 204-12.
24. Dubey JP. Toxoplasmosis in sheep – The last 20 years. Vet Parasitol 2009;163:1-14.
25. Dubey JP. Toxoplasmosis in India. In: Sen AB, Katiyar JC, Guru P, editors. Perspectives in Parasitology. Delhi: CBS Publishers & Distributers; 1988. p. 131-52.
26. Mirdha BR, Samantaray JC, Pandey A. Seropositivity of Toxoplasma gondii in domestic animals. Indian J Public Health 1999;43:91-2.
27. Rajkhowa S, Sarma DK, Rajkhowa C. Seroprevalence of Toxoplasma gondii antibodies in captive mithuns (Bos frontalis) from India. Vet Parasitol 2006;135:369-74.
28. Sharma S, Sandhu KS, Bal MS, Kumar H, Verma S, Dubey JP. Serological survey of antibodies to Toxoplasma gondii in sheep, cattle, and buffaloes in Punjab, India. J Parasitol 2008;94:1174-5.
29. Sensini A. Toxoplasma gondii infection in pregnancy: Opportunities and pitfalls of serological diagnosis. Clin Microbiol Infect 2006;12:504-12.
30. Hedman K, Lappalainen M, Seppäiä I, Mäkelä O. Recent toxoplasmosis in sheep, cattle, and buffaloes in northern Finland. Scand J Infect Dis 2009;41:436-41.
VIDAS system (bioMérieux). Diagn Microbiol Infect Dis 1998;32:69-73.

32. Lappalainen M, Koskela P, Koskineni M, Ammälä P, Hillemaa V, Teramo K, et al. Toxoplasmosis acquired during pregnancy: Improved serodiagnosis based on avidity of IgG. J Infect Dis 1993;167:691-7.

33. Liesenfeld O, Montoya JG, Kinney S, Press C, Remington JS. Effect of testing for IgG avidity in the diagnosis of Toxoplasma gondii infection in pregnant women: Experience in a US reference laboratory. J Infect Dis 2001;183:1248-53.

34. Singh S. Avidity testing to pinpoint timing of maternal infections during pregnancy. In: Deka D, editor. Congenital Intrauterine Infections. New Delhi: J P Brothers Medical Publishers Pvt. Ltd.; 2011. p. 131-9.

35. Wei HX, Woi SS, Lindsay DS, Peng HJ. A Systematic review and meta-analysis of the efficacy of anti-Toxoplasma gondii medicines in humans. PLoS One 2015;10:e0138204.

36. Antczak M, Dzitko K, Dlugonska H. Human toxoplasmosis-Searching for novel chemotherapeutics. Biomed Pharmacother 2016;82:677-84.