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Original Article

Prevalence of Toxoplasmosis among Antenatal Patients Attending the Outpatient Department of a Tertiary Care Health Facility

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Abstract *- Acute infection with Toxoplasma gondii during early pregnancy presents a substantial risk of transplacental transmission, which may lead to severe fetal complications. Detecting specific IgG and IgM antibodies through serological assays is critical for differentiating recent infections from prior exposures. This cross-sectional study involved 50 pregnant women across all three trimesters who attended an antenatal care (ANC) clinic in India. Serum samples were assessed for T. gondii-specific IgG and IgM antibodies using two automated platforms: the VITEK Immunodiagnostic Assay System (VIDAS, employing the ELFA method) and the Architect i2000 system (utilizing the CMIA method). Of the participants, 18% (9 out of 50) were seropositive for T. gondii-specific IgG antibodies (P = 0.038). Seropositivity was most prevalent in the third trimester (66.66%), significantly higher than in the first and second trimesters (11.11%) (P < 0.05). A higher rate of IgG positivity was also observed among non-vegetarian participants (77.77%) compared to vegetarians (22.22%) (P = 0.272), and among primigravida women (55.55%) compared to multigravida women (22.22%) (P = 0.735). In conclusion, both the Architect i2000 (CMIA) and VIDAS TOXOM (ELFA) systems demonstrated high sensitivity and specificity, confirming their utility in the serodiagnosis of toxoplasmosis among pregnant women, particularly those with adverse obstetric histories. The study also emphasizes the need to improve expectant mothers' awareness of transmission risks. Targeted educational programs and preventive counselling could play a crucial role in mitigating the incidence of congenital toxoplasmosis. Furthermore, public health initiatives should aim to control T. gondii infections in livestock, particularly sheep and goats, which serve as key zoonotic sources.*

Keywords *- Toxoplasma gondii, Trimesters of pregnancy, Serological immunoassay test, Technical automated system, CLIA, VIDAS.*

1. Introduction

Nicolle and Manceaux, in the mononuclear cells of the spleen and liver of a North African rodent species, Ctenodactylus gundi, first identified Toxoplasma gondii in 1908. The name "Toxoplasma" originates from Greek, with "toxon" meaning "arc" and "plasma" meaning "form." (Parija, 2009; Sudan, Jaiswal, and Shanker, 2013). The infection caused by this obligate intracellular protozoan parasite is known as toxoplasmosis—a globally prevalent zoonotic disease affecting both humans and warm-blooded animals, including birds and mammals. *T. gondii*, the sole species in the genus *Toxoplasma*, belongs to the phylum *Apicomplexa*. Its capacity to infect a wide range of hosts is due to its ability to invade various cell types (Sakikawa et al., 2012; Vinhal et al., 1994). The parasite's life cycle consists of three infectious stages: the rapidly dividing tachyzoite, the slowly replicating bradyzoite within tissue cysts, and the environmentally resilient sporozoite, found in oocysts. The parasite reproduces asexually in intermediate hosts (mammals and birds) and sexually in its definitive hosts—members of the Felidae family, particularly domestic cats. Cats play a crucial role in the life cycle of *T. gondii*, as they are the only known hosts capable of shedding oocysts containing sporozoites into the environment (Halonen and Weiss, 2013; Black and Boothroyd, 2000). Transmission to humans and animals typically occurs through: (Robert-Gangneux and Dardé, 2012; Torrey, Bartko, and Yolken, 2012)

- Ingestion of undercooked or raw meat harboring tissue cysts
- Consumption of contaminated food, unwashed vegetables, or water containing oocysts from cat faeces
- Vertical transmission from an infected mother to her fetus via the placenta

T. gondii infection is common globally, with human seroprevalence rates ranging from 16% to 80%, though the

majority of cases remain asymptomatic. Human infection typically occurs via ingestion of tissue cysts or oocysts, followed by intestinal epithelial invasion and intracellular multiplication. Clinically, toxoplasmosis is of particular concern in immunocompromised individuals and in cases of congenital transmission. The severity of congenital infection is influenced by the gestational age at the time of maternal infection, potentially leading to spontaneous abortion, visual impairment, or neurodevelopmental disorders in the newborn. Diagnosis involves serological testing: (Tortora and Funke, 1989) IgM antibodies, indicative of recent infection, are detected using the Enzyme-Linked Fluorescent Assay (ELFA) method, performed on the MiniVIDAS automated platform using BIOMÉRIEUX kits. IgG antibodies signal past or chronic infection and are identified through the Chemiluminescent Microparticle Immunoassay (CMIA) method using the Architect i2000 automated analyzer. This study aims to assess the incidence of toxoplasmosis among pregnant women attending antenatal care at a tertiary health facility, with the specific goal of identifying *T. gondii* infection during pregnancy to help prevent congenital transmission. The study further intends to evaluate seroprevalence using IgG and IgM markers among antenatal attendees. In addition, this prospective study was conducted at a large tertiary care hospital from July to October 2013. A total of 50 blood samples were collected from pregnant women attending the Antenatal Care (ANC) clinic after obtaining informed consent and recording relevant clinical histories, including age, diet, gestational trimester, and history of abortion. Under aseptic conditions, 3–5 ml of venous blood was drawn using sterile disposable syringes and collected in clot activator tubes. Samples were centrifuged at 3,300–3,400 RPM, and the separated serum was stored at 4°C until serological testing was performed (Sanati et al., 2012; Rahbari et al., 2012; Dubey and Jones, 2008; Dubey, 2008)

2. Research Theoretical Framework

Toxoplasma gondii, a protozoan parasite with a global distribution, was first discovered in 1908 by Nicolle and Manceaux in *Ctenodactylus gundi*, a rodent native to North Africa. That same year, Splendore identified the parasite in rabbit tissue in Brazil (Dubey, 2016). The name *Toxoplasma gondii* is derived from Greek—"toxon" meaning arc or bow and "plasma" referring to form or life—chosen based on the organism's curved morphology, formally proposed in 1909. The first viable isolation of *T. gondii* from an animal was achieved by Sabin and Olitsky in 1937, and the first human isolate was reported by Wolf and colleagues in 1939. Immunity to *T. gondii* involves both innate and adaptive responses. In the 1940s, researchers discovered that humoral antibodies could neutralize extracellular but not intracellular forms of the parasite (Sabin & Feldman, 1948; Sabin & Olitsky, 1937).

2.1. Taxonomy and Classification

T. gondii is a coccidian parasite belonging to the phylum Apicomplexa, class Sporozoasida, subclass Coccidiasina, order Eimeriorina, family Toxoplasmatidae, and genus *Toxoplasma*. It is the only recognized species in its genus (Waree, 2010). The parasite demonstrates a complex life cycle, which includes both asexual and sexual phases, depending on the host species.

2.2. Morphology and Life Cycle

The parasite exhibits three major forms throughout its life cycle: tachyzoites, bradyzoites (in tissue cysts), and sporozoites (within oocysts).

- Tachyzoites are crescent-shaped, rapidly multiplying forms that dominate the acute phase of infection. They actively invade host cells and replicate through endodyogeny within a parasitophorous vacuole. Ultrastructural analysis reveals various organelles such as the pellicle, conoid, rhoptries, micronemes, mitochondria, and an apicoplast, all of which aid in host cell penetration and survival.
- Bradyzoites are slow-growing forms found within tissue cysts during chronic infection. Cysts can be small (as little as 5 μm) or large, containing hundreds of bradyzoites. These are typically located in neural and muscular tissues, including the brain and heart.
- Oocysts, produced by sexual reproduction, are excreted in the feces of felines, the definitive hosts. They are initially non-infective but become infective after sporulation in soil over several days, eventually developing into sporozoite-containing oocysts.

2.3. Definitive and Intermediate Hosts

The life cycle of *T. gondii* involves two hosts:

- Definitive hosts**:** Domestic cats and other felids support both asexual (schizogony) and sexual (gametogony) reproduction in the enteric cycle. They acquire the parasite by ingesting tissue cysts or oocysts and subsequently shed oocysts in their feces.
- Intermediate hosts**:** Humans, birds, and various mammals acquire infection via ingestion of contaminated food or undercooked meat. Only asexual stages (tachyzoites and bradyzoites) occur in these hosts.

2.4. Life Cycle Phases

1. Enteric Cycle (in felids)**:** After ingesting tissue cysts or oocysts, cats undergo cycles of schizogony and gametogony in the intestinal mucosa, resulting in the formation and excretion of oocysts. These oocysts become infective after sporulation in the environment.
2. Exoenteric Cycle (in intermediate hosts)**:** Upon ingestion of sporulated oocysts or tissue cysts, the parasite's invasive forms (sporozoites or bradyzoites) disseminate through the bloodstream, invading tissues where they transform into tachyzoites. These then multiply and later

develop into bradyzoites, forming tissue cysts that sustain chronic infection.

2.5. Modes of Transmission

Transmission to humans and animals can occur through:

- Horizontal transmission, via ingestion of undercooked meat containing tissue cysts or contaminated food and water carrying oocysts.
- Vertical transmission, where tachyzoites cross the placental barrier during pregnancy, potentially leading to congenital toxoplasmosis.

2.6. Pathogenicity and Congenital Infection

T. gondii infections in humans are often asymptomatic but can have severe consequences in immunocompromised individuals and fetuses. Congenital toxoplasmosis results from maternal infection during pregnancy, with severity depending on the gestational age at the time of infection. Outcomes range from miscarriage and stillbirth to long-term neurological and visual impairments in the newborn.

2.7. Placental Role in Toxoplasma Transmission

The placenta serves a dual function during pregnancy: as a physiological barrier protecting the fetus and as a site for parasitic replication. Its effectiveness is strongest during early gestation, with transmission rates below 10% in the first trimester. However, placental permeability increases throughout pregnancy, resulting in transmission rates of approximately 30% in the second trimester and 60–70% in the third trimester. Notably, the severity of fetal complications is inversely related to the timing of transmission.

2.8. Toxoplasmosis in Immunocompromised Individuals

In immunocompromised patients, toxoplasmosis can be life-threatening. Disease reactivation due to cyst rupture poses a greater risk than primary infection. In individuals with HIV, susceptibility increases when CD4+ T-cell counts fall below 100 cells/µL. The most common manifestation is Toxoplasma encephalitis (TE), which presents with neurological symptoms such as headache, fatigue, memory loss, coordination issues, seizures, and sometimes dementia—often accompanied by fever (Robert-Gangneux & Dardé, 2012).

2.9. Global Prevalence of Toxoplasmosis

T. gondii infects an estimated 25–30% of the global population. Prevalence varies significantly:

- Low (10–30%): North America, Southeast Asia, Northern Europe, and the Sahel region.
- Moderate (30–50%): Central and Southern Europe.
- High: Latin America and tropical Africa.

Climatic and environmental conditions (warm, humid climates), along with human behavior (dietary habits, hygiene), largely influence regional prevalence (Robert-Gangneux & Dardé, 2012).

2.10. Immune Response to Toxoplasma gondii

- Humoral Immunity: IgM antibodies appear within two weeks; IgG peaks within 6–8 weeks. IgA and IgE may also be present early. IgG positivity indicates past exposure.
- Cell-Mediated Immunity: The intracellular nature of the parasite necessitates a strong T-cell response. IFN-γ activates macrophages, which inhibit parasite growth via reactive oxygen species and tryptophan degradation. IL-12 enhances NK and T-cell activation, boosting IFN-γ and TNF-α production (Jones et al., 2001).

2.11. Diagnosis of Toxoplasmosis

- Microscopy: Useful in immunosuppressed individuals; Giemsa-stained smears can show the parasite.
- Serological Tests:
    - Sabin-Feldman Dye Test: Gold standard but involves live tachyzoites.
    - LAT, IFAT, ELISA, VIDAS, and CMIA: Various commercial kits and automated platforms provide rapid, sensitive results based on IgG/IgM detection.

2.12. Treatment of Toxoplasmosis

The standard regimen includes pyrimethamine + sulfonamides, effective against tachyzoites but not bradyzoites. Spiramycin is used prophylactically in pregnancy. Folinic acid is co-administered to reduce bone marrow suppression. Clindamycin, vancomycin, and corticosteroids may be used in specific cases, including ocular or congenital toxoplasmosis. Treatment in immunocompromised individuals is urgent and tailored based on clinical severity (Waree, 2010).

2.13. Risk Factors for Toxoplasmosis

Key risk factors include:

- Consumption of undercooked lamb/goat meat.
- Exposure to contaminated soil or cat feces.
- Lack of awareness about transmission—especially in pregnant women.

Educational campaigns significantly enhance awareness and reduce congenital transmission (Chintapalli & Padmaja, 2013).

3. Research Data Collection

This study analysed all collected serum samples for *Toxoplasma gondii*-specific IgG and IgM antibodies using two automated immunoassay platforms to ensure diagnostic accuracy and serological differentiation.

3.1. VITEK Immunodiagnostic Assay System (VIDAS TOXO IgM)

Detection of anti-*Toxoplasma* IgM antibodies was performed using the Enzyme-Linked Fluorescence Assay (ELFA) via the VIDAS platform, employing the BioMérieux SA reagent kit. This method integrates an immuno-capture step with subsequent fluorescence-based detection. Each assay utilizes a Solid Phase Receptacle (SPR) as the reaction surface and pipetting unit. All reagents are preloaded and ready-to-use within sealed strips. The automated system performs all procedural steps, including multiple cycles of reagent exchange through the SPR. Initially, serum samples are diluted, and polyclonal anti-IgM antibodies coating the SPR interior capture IgM antibodies. Anti-Toxoplasma IgM is detected using inactivated RH strain antigens, which are subsequently targeted by an alkaline phosphatase-conjugated murine monoclonal antibody (anti-P30). During the final detection phase, the substrate 4-methylumbelliferyl phosphate is enzymatically converted into the fluorescent product 4-methylumbelliferone, whose intensity—measured at 450 nm—correlates directly with the antibody concentration in the sample. Interpretation criteria:

- Negative: Index < 0.55
- Equivocal: Index 0.55 – 0.64
- Positive: Index ≥ 0.65

A positive result signifies recent or acute *T. gondii* infection due to the presence of specific IgM antibodies, while a negative result suggests no current infection.

3.2. Architect i2000 System – Chemiluminescent Microparticle Immunoassay (CMIA) for IgG Detection

Quantifying T. gondii-specific IgG antibodies using the Architect i2000 automated analyzer via CMIA (Chemiluminescent Microparticle Immunoassay) technology, utilizing the Abbott Architect kit. The Architect Toxo IgG assay is a two-step immunoassay based on recombinant antigens (P30/SAG1 and P35/GRA8). In the first step, the serum sample undergoes an automated 1:10 dilution and is mixed with recombinant antigen-coated paramagnetic microparticles. IgG antibodies specific to *T. gondii* bind to these coated microparticles. Following a wash step, a murine acridinium-labelled anti-human IgG conjugate is introduced. In the second phase, additional wash cycles are added by chemiluminescent trigger solutions, initiating a light-emitting reaction measured in Relative Light Units (RLUs). The luminescence intensity is directly proportional to the amount of IgG present in the sample. Interpretation criteria:

- Non-reactive: < 1.60 IU/mL
- Equivocal (Gray zone): 1.60 – 2.99 IU/mL
- Reactive: ≥ 3.00 IU/mL

A positive IgG result confirms prior exposure and indicates chronic toxoplasmosis, while a negative result suggests no evidence of past infection.

4. Research Data Analysis

4.1. Results and Observations

- A total of 50 antenatal women were included in this study.
- Of this, 9 (18%) women were found seropositive for *Toxoplasma gondii*-specific IgG antibodies.
- One case (2%) was found equivocal(intermediate) for specific IgM antibodies, and three cases were in the gray zone for IgG.
- In this study, the Grayzone and equivocal are ignored for both IgG and IgM because no significant results were obtained.
- In this study, none of the test serum samples were found to have anti-Toxoplasma gondii IgM antibodies.

Table 1. Seroprevalence of *Toxoplasma* Immunoglobulin (IgG) in different age groups, n=50

Age group	IgG			Total
	Non-reactive	Gray zone	Reactive (%)	
15 – 19	3	0	(11.11%) 1	4
20 – 24	27	0	(44.44%) 4	31
25 – 29	7	2	(33.33%) 3	12
30 – 34	1	1	(11.11%) 1	3
Total	38	3	9	50

The P-value is 0.038 (Using Fisher's exact test), which is significant.

The majority of seropositive cases in the study group were in the age group 20-24 years (44.44%). Among the seropositive cases, IgG seropositivity was higher in the 20-24 age group than in the 30-34 age group (11.11%).

Fig. 1 IgG results with respect to Age group

Table 2. Seroprevalence of Toxoplasma Immunoglobulin (IgM) in different age groups. Casesn=50

Age group	IgM		Total
	Negative	Equivocal	
15 – 19	4	0	4
20 – 24	30	1	31
25 – 29	12	0	12
30 – 34	3	0	3
Total	49	1	50

The P-value is 0.999 (Using Fisher's exact test), which is not significant.

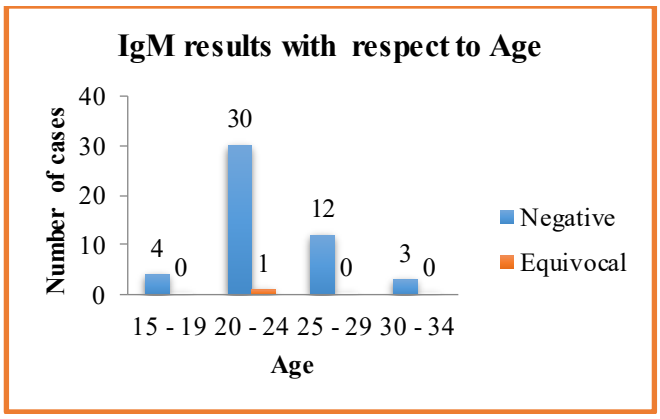

Fig. 2 IgM result with respect to age

Table 3. Stage of pregnancy Toxoplasma gondii IgG Seroprevalence in pregnant women of ANC clinic n=50

Trimester	IgG group			Total
	Non-reactive	Gray zone	Reactive (%)	
First	4	0	(11.11%) 1	5
Second	10	0	(22.22%) 2	12
Third	24	3	(66.66%) 6	33
Total	38	3	9	50

P-value is 0.952. Fisher's exact test was not significant at P< 0.05; IgG seropositivity was high in the third trimester of gestation (66.66%) as compared to the first and second trimesters (11.11%). (Table 3)

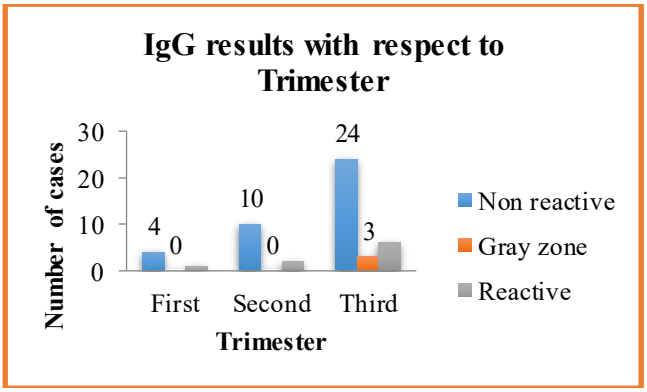

Fig. 3 IgG results with respect to trimester

Table 4. Stages of pregnancy T.gondii IgM Seroprevalence in pregnant women of ANC clinic n=50

Trimester	IgM group		Total
	Negative	Equivocal	
First	5	0	5
Second	12	0	12
Third	32	1	33
Total	**49**	**1**	**50**

The P-value is 0.999 (Using Fisher's exact test), which is not significant.

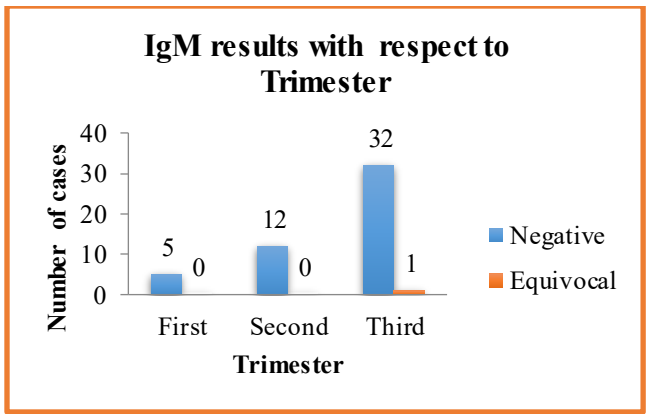

Fig. 4 IgM results with respect to trimester

Table 5. Association of Diet and Toxoplasma Seroprevalence IgG cases n=50

Food	IgG group			Total
	Non-reactive	Gray zone	Reactive (%)	
Vegetarian	20	2	(22.22%) 2	24
Non-Vegetarian	18	1	(77.77%) 7	26
Total	**38**	**3**	**9**	**50**

The P-value is 0.272 (Using Fisher's exact test). Not significant IgG Seropositivity was high among the non-vegetarian group (77.77%), compared to the vegetarian group (22.22%). (Table 5)

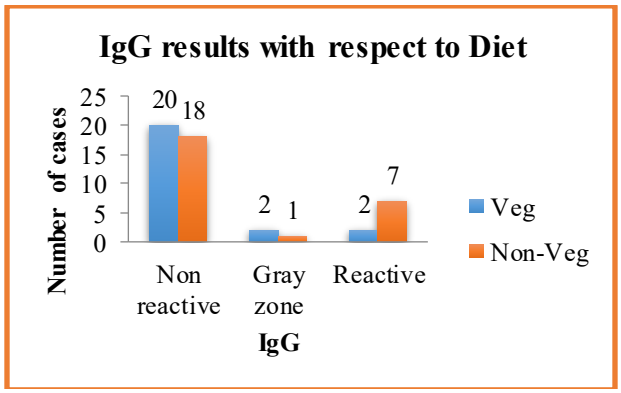

Fig. 5 IgG result with respect to diet

Table 6. Association of Diet and Toxoplasma Seroprevalence IgM cases n=50

Diet	IgM group		Total
	Negative	Equivocal	
Vegetarian	24	0	24
Non-Vegetarian	25	1	26
Total	49	1	50

The P-value is 0.999 (Using Fisher's exact test), which is not significant.

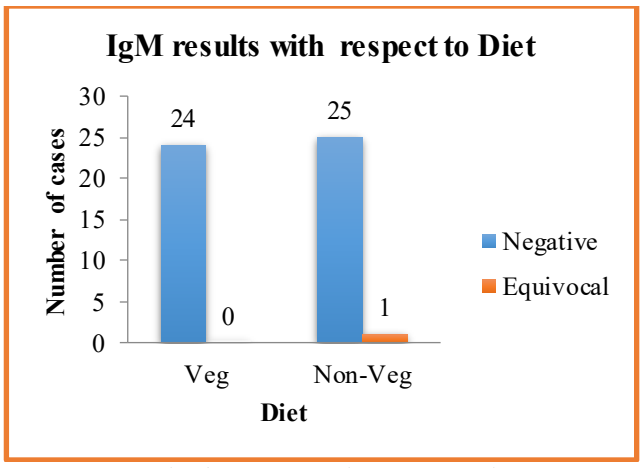

Fig. 6 IgM results with respect to diet

Table 7. Analysis of results (IgG) in relation to parity, n=50

Gravida	IgG group			Total
	Non-reactive	Gray zone	Reactive (%)	
I	22	2	(55.55%) 5	29
II	13	1	(22.22%) 2	16
III	3	0	(22.22%) 2	5
Total	38	3	9	50

P-value 0.735 (Fisher's exact test) not significant. IgG seropositivity was higher in primigravida (55.55%) than in Gravida II and III (22.22%).

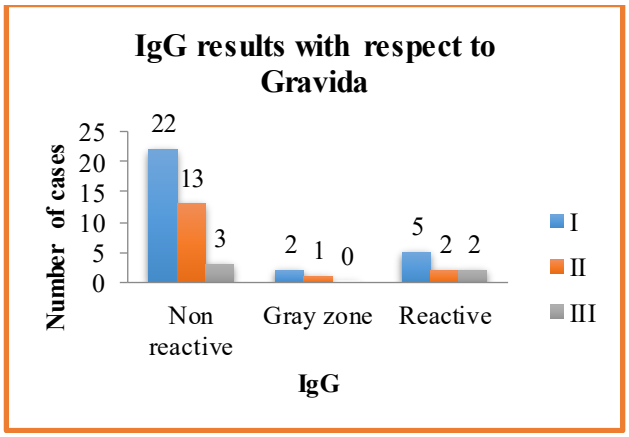

Fig. 7 IgG with respect to Gravida

Table 8. Analysis of results (IgM) in relation to parity, n=50

Gravida	IgM		Total
	Non-reactive	Gray zone	
I	28	1	29
II	16	0	16
III	5	0	5
Total	49	1	50

P-value 0.680 (Fisher's exact test) not significant

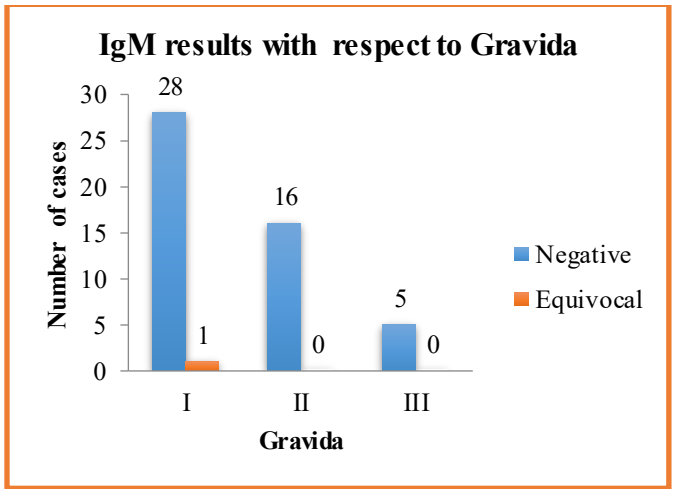

Fig. 8 IgM Result with respect to Gravida

Table 9. Clinical spectrum of Seropositive IgG in Bad Obstetric History (BOH) cases n=50

Abortion	IgG			Total
	Non-reactive	Gray zone	Reactive	
Yes	3	0	0	3
No	35	3	9	47
Total	**38**	**3**	**9**	**50**

P-value is 0.999 (Fisher's exact test used). Not significant. In this study, there is no seropositivity for IgG antibodies. *Toxoplasma gondii* infection was not found to be related to abortion and bad obstetric history.

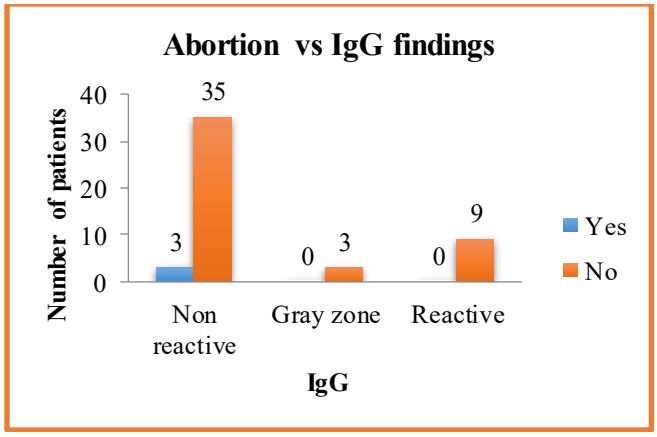

Fig. 9 Abortion vs IgG finding

Table 10. Clinical spectrum of Seropositive IgM in Bad Obstetric History (BOH) cases n=50

Abortion	IgM		Total
	Negative	Equivocal	
Yes	3	0	3
No	46	1	47
Total	**49**	**1**	**50**

In this study, there is no seropositivity for IgM antibodies. *Toxoplasma gondii* infection is not found to be related to abortion and bad obstetric history.

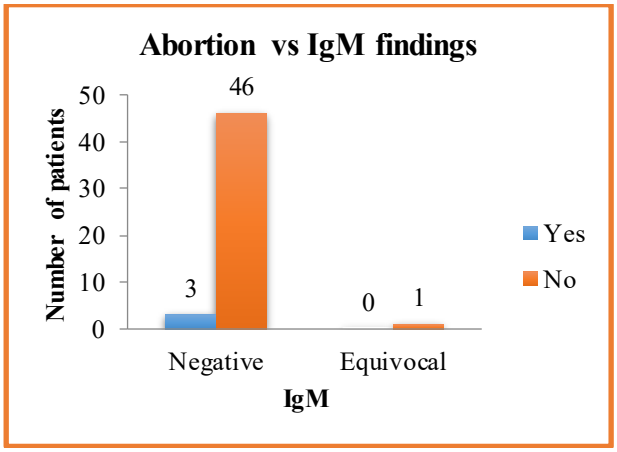

Fig. 10 Abortion vs IgM finding

4.2. Statistical Data Analysis

Data analysed using SPSS 17.0 (Statistical Package for Social Science). We have used Fisher's exact test to determine the association between IgG and IgM results with various parameters. P-value < 0.05 is considered significant.

P-value> 0.05, No significant, No difference between groups
P-value < 0.05 Significant, different between groups.

Emerging evidence supports the association between *Toxoplasma gondii* infection and fetal loss in pregnant women. The parasite exhibits a strong affinity for nucleated cells, particularly within muscle tissues, the intestinal epithelium, and the placenta. Transplacental transmission can occur if a woman acquires the infection during pregnancy, potentially resulting in spontaneous abortion, stillbirth, or congenital malformations. Diagnosis is typically made via serological testing, where the presence of *Toxoplasma*-specific IgM antibodies signifies a recent infection, whereas IgG antibodies are indicative of a chronic infection. Early identification of primary toxoplasmosis, especially during the first trimester, is critical for initiating timely therapeutic interventions aimed at preventing vertical transmission.

Accordingly, determining a woman's serological status either before conception or during early pregnancy is essential. In this study, the combined use of the VITEK Immunodiagnostic Assay System (VIDAS TOXO IgM) and the Architect i2000 CMIA-based system proved effective in distinguishing between acute and chronic infections during the first trimester. The study revealed a seroprevalence rate of 18% among pregnant women, which is lower than previously reported rates from other Indian regions, ranging between 5% and 46.7%. This lower prevalence may reflect better education, improved sanitation, and reduced agricultural exposure among women in this region compared to those in North India, where rates have reached as high as 77%. Among the seropositive participants, the highest infection rate occurred in the 20–24-year age group, with 44.44% testing positive for *Toxoplasma*-specific IgG. This rate was significantly higher than the 11.11% observed in the 30–34 age group (Fisher's Exact Test, p = 0.038). These findings are consistent with those of Chintapalli and Padmaja (2013), who reported a similar age-related trend in IgG seropositivity. None of the women screened during pregnancy tested positive for anti-*Toxoplasma gondii* IgM, whereas 44.44% (4/9) were positive for IgG only, indicating latent infection.

A comparable finding was reported by researchers at King Khalid University Hospital in Riyadh (2011), where no IgM seropositivity was found among women under 25, though 38% showed IgG positivity. One case (2%) yielded an intermediate or "gray zone" result for IgG and IgM. Although routine repeat screening during pregnancy is costly, it may help prevent congenital infections in approximately 10 per 10,000 births. Eighty percent (40/50) of participants were seronegative for both IgG and IgM, suggesting that poor obstetric outcomes in these cases might be attributable to other infections such as cytomegalovirus, herpes simplex virus, or rubella. Although the highest IgG seropositivity (66.66%) was observed during the third trimester, the association between gestational age and toxoplasmosis was not statistically significant (p = 0.952). IgG seropositivity was notably higher among non-vegetarian women (77.77%) compared to vegetarians (22.22%), although the difference was not statistically significant (p = 0.272). Dietary habits likely influence transmission, as meats such as lamb and goat are more frequently infected with viable tissue cysts compared to beef or chicken. This pattern has been observed in studies from Norway and France, where undercooked lamb and beef were associated with increased risk. Gravida status was not significantly associated with IgG seropositivity (p = 0.735); however, a higher prevalence (55.55%) was observed among primigravidae.

This supports the hypothesis that *T. gondii* may persist across pregnancies, possibly contributing to recurrent miscarriages or stillbirths due to reactivation of encysted parasites in the uterine endometrium during placentation. Chronic infection, therefore, remains a potential cause of fetal loss. Seroprevalence in Indian pregnant populations varies widely, from as low as 5% to as high as 80%. Despite its clinical significance, routine testing is uncommon, and interpretation of results often lacks standardization. In countries where educational interventions have been integrated into prenatal care, reductions in infection rates up to 50% have been reported. Enhanced public awareness and healthcare education are critical. Preventive measures such as proper food hygiene, handwashing, and access to clean water can substantially reduce infection risks. Early screening and timely treatment during pregnancy can prevent fetal transmission. Carter et al. demonstrated the effectiveness of a short audiovisual and handout-based educational program in

improving pregnant women's behavior toward risk factors such as food preparation and cat handling. Breugelmans et al. reported a 60% reduction in *T. gondii* seroconversion through sustained education over two decades.In the present study, participants were educated on the risks and significance of toxoplasmosis screening. Based on our findings, it is recommended that pregnant women undergo routine screening and receive education regarding the transmission and prevention of toxoplasmosis.

4.3. Summary of Findings

- Seropositivity for *T. gondii* IgG antibodies was 18% (9/50).
- The highest IgG positivity (44.44%) was seen in the 20–24 age group.
- 66.66% of IgG-positive cases were in the third trimester.
- Non-vegetarian women showed higher IgG seropositivity (77.77%).
- Primigravidae exhibited a higher rate of IgG positivity (55.55%).

5. Research Conclusion

This study concludes that both the Chemiluminescent Microparticle Immunoassay (CMIA) system—demonstrating a sensitivity and specificity of 99.7%—and the Enzyme-Linked Fluorescent Assay (ELFA) system—with a specificity of 99.25% and sensitivity of 96%—are reliable and effective serological tools for the detection of *Toxoplasma gondii* infection in pregnant women, particularly those with a history of adverse obstetric outcomes. The observed low incidence of toxoplasmosis among the participants may be attributed to avoiding undercooked or raw meat and adherence to satisfactory hygiene practices. It is recommended that all antenatal patients undergo routine serological screening for *Toxoplasma gondii*–specific IgG and IgM antibodies, as well as other TORCH infections (Toxoplasma, Rubella virus, *Chlamydia*, and Herpes simplex virus). Early diagnosis through such screening can significantly reduce the risk of fetal loss and other complications during pregnancy. Furthermore, the study highlights a lack of awareness among many pregnant women regarding the risk factors for toxoplasmosis transmission.

Educational interventions and counselling focused on preventive measures can substantially reduce the incidence of congenital toxoplasmosis. Public health strategies should include reducing *T. gondii* infection rates in livestock (particularly sheep and goats), representing a significant zoonotic reservoir. Preventive measures should emphasize cooking meat to internal temperatures above 66°C or subjecting it to repeated freezing at temperatures below −20°C, thoroughly washing or peeling fruits and vegetables, and disinfecting kitchen surfaces and utensils that come into contact with raw food products. Finally, the development of an inactivated (killed) parasite-based vaccine should be prioritized as a potential long-term control strategy against toxoplasmosis.

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