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Association of Host Interferon-γ Gene Polymorphism with *Toxoplasma gondii* Infection in Pregnant Women of Bangladesh

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Abstract

Human toxoplasmosis is caused by the intracellular protozoan parasite *Toxoplasma gondii.* Although *T. gondii* infection is generally asymptomatic for most of the immunocompetent adults, severe complications may occur particularly in pregnant women and immunocompromised individual. Host cell immunity plays a critical role in parasite differentiation and persistence in the host. Therefore, genetic polymorphism in the host immune genes, for instance interferon-γ gene could be linked with possibility of *T. gondii* infection. The objective of the study was to verify the link between the single nucleotide polymorphisms (SNPs) in the IFN-γ gene of pregnant women and *T. gondii* infection through correlating with anthropometric and sociodemographic parameters. In this study, ninety-two (N = 92) pregnant women (16 - 40 years) and healthy controls (N = 95) with similar age ranges were included. Among them, 25% (n = 23) pregnant women were seropositive for *T. gondii* IgG antibodies by Rapid Test Assay. Allelic and genotypic frequencies of IFN-γ +874T/A (rs2430561) SNPs were evaluated by using ARMS-PCR. The distribution of the A and T alleles in the specific position of the IFN-γ gene in the *T. gondii*-infected pregnant women and the control groups did not differ significantly, according to the data. However, we found a higher frequency (13.04%) of A/A genotype in *T. gondii* infected pregnant women as compared to non-infected individuals (8.70%), demonstrating that *T. gondii* infection susceptibility may be increased by homozygosity for the A allele. Further studies are to be needed to find out the link between host gene polymorphism and *T. gondii* infection in Bangladesh.

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Keywords

T. gondii, Pregnant Women, Seroprevalence, IFN-γ Gene, Polymorphism

1. Introduction

Toxoplasma gondii, a food-borne protozoan parasite causes toxoplasmosis in humans. It is known as one of the most prevalent parasites throughout the world due to its wide host range, pervasive environmental contamination, and various infection methods [1]. *T. gondii* has a distinct parasite life cycle that involves both asexual and sexual reproduction. There are normally three stages of infection development: the fast replicating acutely infected stage called "tachyzoite", the slow replicating, metabolically dormant & tissue cyst forming "bradyzoite" and environmental form "Oocysts" [2]. Transmission of *T. gondii* occurs predominantly by oocyst contaminated food, water and vegetables, ingestion of tissue cysts containing undercooked meats and through congenital transmission [3]. After primary infection in pregnant women, the parasite develops congenital toxoplasmosis through crossing placenta into developing fetus [4]. Transmission of the parasite may also be affected by the gestational stages of pregnancy for instance infection in first trimester affects more severely than 3rd trimester because of anatomical and immunological factors [5]. Generally, the placenta barrier of a woman is thick and gradually reduces at the end of pregnancy. These ultimately allow more tachyzote to infiltrate trophoblasts easily at the end of gestation. Toxoplasmosis is typically asymptomatic in immune-competent individuals; however, it may develop influenza-like symptoms and other non-specific clinical indications [6]. The systemic nature of this congenital infection may result in premature delivery, abnormal brain development of the fetus, intrauterine growth restriction, hepatosplenomegaly, thrombocytopenia, or involvement of the eyes and brain and eventually fetal death [7]. Chorioretinitis, meningoencephalitis, hydrocephaly, microcephaly, or calcifications of previously necrosed sites are examples of ocular or encephalic illness manifestations in the developing fetus.

Host immune responses against *T. gondii* are similar to other intracellular pathogens. First, *T. gondii* promotes innate immunity and then develops acquired immunity afterwards. Once acquired immunity is activated, host immune cells release plenty of the pro-inflammatory cytokine, for instance interferon gamma (IFN-γ), which ultimately triggers a group of IFN-γ inducible proteins including GTPases. IFN-γ inducible GTPases accumulates in parasitophorous vacuole membranes and are important for inhibition of *T. gondii* replication and clearance [8]. Polymorphisms of host cytokine genes are critical to regulate the expression of these compounds and may play a crucial role in gene expression that may lead to increase rate of parasite infection, developing resistance or susceptibility to illnesses such as toxoplasmosis. SNPs in the promoter region of

cytokine genes are associated with the improvement of toxoplasmosis; for instance, gene encoding TNF-α polymorphism (rs1799964T > C) in the locus -1031 at promoter region, representing a transition from the thiamine (T) to cytosine (C), leads to down-regulate its transcriptional activity of TNF-α. This polymorphism has an association, for instance, toxoplasma cretinochoroiditis (TR) pathogenesis [9]. Functional polymorphisms in the promoter regions IL-17A (rs2275913: G > A) gene, representing a transition from the guanine (G) to Adenine (A), up-regulate its expression [10]. Another study conducted by Albuquerque et al showed that SNPs in IFN-γ gene at position +874 T/A play an important role in developing symptoms of toxoplasmosis [11]. However, there is limited research on Toxoplasma in Bangladesh. Recently we have shown that more than 25% pregnant women in Bangladesh are chronically infected with *T. gondii* but there is no data available whether polymorphism in host cytokine gene has any role in the susceptibility of *T. gondii* infection and severity. Therefore, for the first time we investigated the role of host genetic factors *i.e.*, SNPs in IFN-γ gene at position +874T/A in susceptibility of *T. gondii* infection and severity in pregnant women.

2. Materials & Methods

2.1. Study Design and Sample Collection

A total of one hundred eighty-seven (N = 187) adult women were enrolled in this study. Among them ninety-two (N = 92) were fetus bearing pregnant women and ninety (N = 95) were age matched health women as control. Pregnant women were visited as patients at outdoor department of gynecology of "Bangladesh Institute of Research and Rehabilitation in Diabetes, Endocrine and Metabolic Disorders (BIRDEM)" and Dhaka metropolitan area whereas healthy women were recruited from Dhaka metropolitan area. Inclusion criteria were aged between 16 - 45 years; Patients with various forms of chronic diseases and children were not included in the study. A written consent was obtained from the study participants for their enrollment and collection of blood for this research. The study was conducted after taking approval from the Ethical Review Board of the department of Biochemistry and Molecular Biology, University of Dhaka (BMBDU/ERC/EC/20).

2.2. Separation of Serum from Whole Blood

Serum was isolated from whole blood through centrifugation. Briefly, after collecting the whole blood, it was left at room temperature for 30 - 60 minutes without any disturbance. Then centrifugation was done at 2000 xg for 15 minutes, the resulting supernatant is designated as serum which was collected into microcentrifuge tube and stored at −20˚C for further experimental analysis.

2.3. Genomic DNA Extraction

Genomic DNA was extracted from 3 - 5 ml of whole blood of pregnant women

and healthy control subjects using standard phenol-chloroform method. Briefly, 500 µl of EDTA containing whole blood was mixed with 800 µl of RBC lysis buffer and was centrifuged for 2 min at 7000 rpm. The supernatant was discarded and repeated this above step. After discarding supernatant, 400 µl of nuclear lysis buffer was added and mixed with the cell pellet carefully. Afterwards, 100 µl of saturated NaCl & 600 µl Phenol:chloroform:isoamyl alcohol PCIA) were added and mixed gently. The tube was centrifuged for 2 min at 7000 rpm. 400 µl of supernatant was collected and mixed with 800 µl ice cold absolute ethanol. After centrifugation at 12,000 rpm for 1 minute, the supernatant was discarded and mixed with 50 µl 70% ethanol for washing. Finally, the pellet containing genomic DNA was collected after centrifugation at 12,000 rpm for 1 minute and dissolved at 50 µl TE buffer. NanoDropTM 1000 Spectrophotometer (Thermo Fisher Scientific Inc., USA) was used to measure the quality and quantity of extracted DNA. It measured absorbance of DNA concentration at 260 nm and protein at 280 nm. Absorbance ratio at 260/280 nm ~1.8 confirmed the purity of DNA. Then, DNA samples were also stored at −20˚C for genetic analysis.

2.4. Rapid Anti-Toxo Antibody Test

The serum samples from pregnant women and healthy controls were assessed for diagnosing *Toxoplasma gondii* infection using IgM/IgG Rapid Test Cassette (Cortez Diagnostics, USA). Briefly, one drop of serum sample was drawn about 1 cm above the upper end of the nozzle and transferred to the wells in the leveled cassettes. Then 2 drops of provided buffer solution were added and waited for 15 minutes for colored line to appear. Results had to be interpreted through visualizing the colored line. The concentration of the anti-Toxo antibody present in the sample is directly proportional to the observed color. The presence of one colored band at the C region (control line) and one- or two-colored band at the IgG or/and IgM region (test line) indicates positive.

2.5. IFN-γ Gene Polymorphism

In this study, an SNP at the +874 position was used to type the IFN-γ T/A polymorphism. Using a single-stranded oligonucleotide made to cover a 24bp area for each allele, the T and A polymorphism sequences were found. With a total volume of 25 µl, an amplification refractory mutation system by polymerase chain reaction (ARMS-PCR) was carried out according to the study of Abu EK [12]. This contains 12.5 µl of Go Taq® Green Master Mix (Promega, USA), 0.5 µl of generic primer (100 pmol/µL), 0.5 µl of specific A or T primer (100 pmol/µL) 0.5 µl of internal control 1 and 2 (10 pmol/µL) and 20 ng/µl of genomic DNA with various amount of ddH20. ARMS-PCR was performed in a thermo-cycler with the cycles: 95˚C for 5 min (Initial Pre-heating), 20 cycles of 95˚C for 30 s (denaturation), 56˚C for 40 s (annealing), and 72˚C for 45 s (extension), followed by 25 cycles of 95˚C for 25 s (denaturation), 50.5˚C for 45 s (annealing), and 72˚C for 45 s (extension), 72˚C for 5 min (Final extension) and 4˚C (hold temperature). ARMS-PCR amplified products were visualized in 2% agarose gel

electrophoresis followed by staining with fluorescent dye "Midori green" using an ultraviolet transilluminator.

2.6. Statistical Analysis

Data were analyzed using Graph Pad Prism version 9.5.0. The odds ratio (OR) and confidence interval (CI) was determined using Fisher's exact test to calculate for evaluating the risk related to genotypes and alleles. P-value < 0.05 was considered statistically significant. Direct counting method was used to calculate the frequencies for SNPs of genotype and alleles.

3. Results

3.1. Sociodemographic Features of Pregnant Women Subjects

In this study, we investigated the seroprevalence of *T. gondii* infection in pregnant women and healthy subjects to understand the role of parasite on congenital toxoplasmosis in Bangladeshi population. A*nti-T. gondii* IgG antibodies was detected in plasma of case and control group to determine the seroprevalence. The sociodemographic characteristics of the pregnant women (N = 92) and the prevalence of *T. gondii* infection are given in Table 1.

Table 1. Seroprevalence of IgG antibodies against *T, gondii and* sociodemographic features of pregnant women subjects (N = 92).

Parameter	Cases (Pregnant Women)		Anti-*T. gondii* IgG Positive	
	Number	Percentage (%)	Number	Percentage (%)
Gender				
Female	92	100	23	25
Age Sub-groups:				
Group-A: 16 - 25 Years	23	25	6	26
Group-B: 26 - 35 Years	59	64	16	27
Group-C: >36 Years	10	11	1	10
Stage of Pregnancy:				
First Trimester	21	23	5	24
Second Trimester	26	28	2	8
Third Trimester	45	49	16	36
Number of Pregnancy:				
First	24	26	5	21
Second	33	36	7	21
Third	25	27	10	40
Fourth	10	11	1	10
Cat Contact:				
Yes	24	26	16	17
No	68	74	7	8
Occupation:				
Housewife Service	81	88	21	26
Cases (Pregnant Women)	11	12	2	18

3.2. *T. gondii*-Specific IgG Seroprevalence in Pregnant Women and Healthy Controls

T. gondii infection was determined by detecting *T. gondii* specific IgG and IgM antibodies in plasma. Here, twenty three of the 92 plasma samples from pregnant women were IgG antibody positive. Three of them also had IgM antibody positivity. Therefore, 25% of the pregnant women were seropositive for *T. gondii* antibodies. Only seven (7.37%) of the 95 samples in the control group had IgG antibodies to *T. gondii* (Figure 1). This data confirms that the higher rate of seropositivity of chronic *T. gondii* infection in pregnant women than in healthy control. Our data also showed that anti-IgM antibodies against *T. gondii* was found only 3% pregnant women suggesting acute infection (data not shown). However, a higher rate of *anti-T. gondii* IgG in pregnant women, which is significantly higher than that of non-pregnant control participants ($P < 0.05$), revealed a chronic infection.

3.3. Evaluation of PCR Amplified Products of Targeted IFN-γ Gene

A polymerase chain reaction assay was performed to amplify the targeted region of IFN-γ. By comparing the target gene band to a 100 bp DNA ladder (Promega, USA), the appropriate size of the product was determined. Amplified DNA was visualized under UV transilluminator, and gel image was captured (Data not shown).

3.4. Genotype and Allele Distribution of IFN-γ Gene Polymorphisms in *T. gondii* Infected and Non-Infected Pregnant Women and Healthy Control Subjects

Genotypic and allelic distribution of IFN-γ gene polymorphisms in and *T. gondii* infected and non-infected pregnant women and healthy controls are given in Table 2. For the SNP rs2430561 for the IFN-γ gene, 90.22% of the control subjects had the A/T genotype and 9.78% had the A/A genotype. For the pregnant women, 97.84% had the A/T genotype and 3.1% had the A/A genotype (Figure 2). In contrast, neither the case nor the control group had the T/T genotype of SNP rs2430561 for the IFN-gene, when considering A/A as the wild type of genotype.

Table 2. Genotype and allele distribution of IFN-γ gene polymorphism in *T. gondii* infected and non-infected pregnant women and healthy control Subjects.

Gene	Genotype and Allele	Case No (%), N = 92	Control No (%), N = 95	OR (95% CI)	Fisher's P-value
IFN-γ	A/A	9 (9.78)	3 (3.16)	1.0 (Ref.)	
	A/T	83 (90.22)	92 (97.84)	3.32 (0.86 - 11.66)	0.078 (NS)
	T/T	0 (0)	0 (0)		>0.99 (NS)
	A	101 (54.89)	98 (51.58)	1.14 (0.76 - 1.72)	0.535 (NS)
	T	83 (45.11)	92 (48.42)		

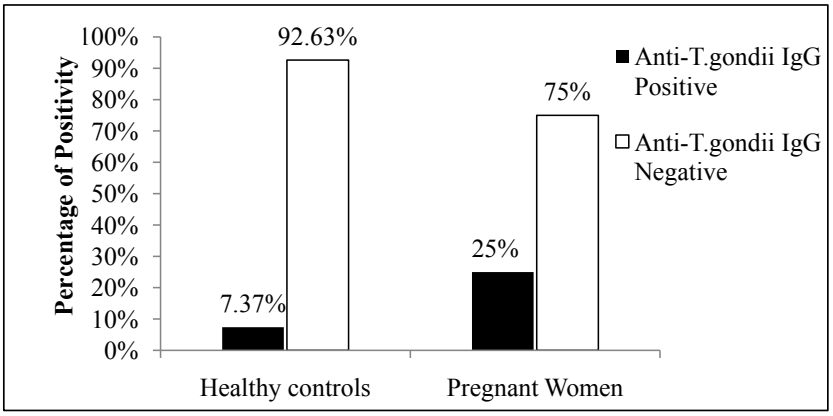

Figure 1. Seroprevalence of *T. gondii* infection in plasma of pregnant women and healthy sunject. The rate of seropositivity of *T. gondii* infection was investigated in pregnant women (N = 92) and healthy control subjects (N = 95). Percentages of anti-*T. gondii* IgM and IgG positive samples were calculated for both pregnant women and control subjects. Using a non-parametric T-test (often referred to as Fisher's exact test), significant differences between pregnant women and non-pregnant control individuals were calculated.

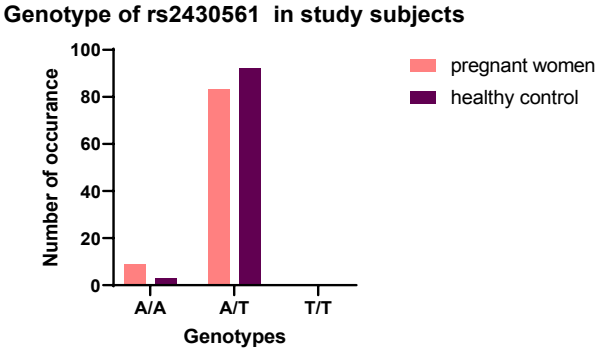

Figure 2. Genotypic distribution of rs2430561 in pregnant women and healthy control subjects. The AA genotypes are significantly lower in case and control subjects then the AT genotype. And the T/T genotypes are absent both in the case and control subjects.

So, it was found that there was no significant association between *T. gondii* infection in pregnant women and allelic variation of SNP rs2430561 (OR = 3.32 at 95% Confidence Interval 0.86 - 11.66, and P Value = 0.078 at Fisher's Exact Test), considering A/A as wild type genotype.

3.5. Genotype and Allele Distribution of IFN-γ Gene Polymorphisms in *T. gondii* Infected and Non-Infected Pregnant Women

Table 3 shows information on the genotypic and allelic distribution of IFN-gene polymorphisms in pregnant women with and without *T. gondii* infection. Infected individuals had 86.96% A/T, 13.04% A/A genotypes and non-infected pregnant women had 91.30% A/T, 8.70% A/A genotypes for the SNP rs2430561

for IFN-γ gene, When A/A is considered as the wild type of genotype. However, T/T genotype of SNP rs2430561 for IFN-gene was lacking in both case and control participants (Figure 3). When considering A/A as the wild type of genotype, it was discovered that there was no significant correlation between *T. gondii* infection in pregnant women and allelic variation of SNP rs2430561 (OR = 1.58 at 95% Confidence Interval 0.40 - 5.84, and P Value = 0.68 at Fisher's Exact Test).

3.6. Genotype and Allele Distribution of IFN-γ Gene Polymorphisms in *T. gondii* Infected and Non-Infected Pregnant Women Based on Different Ages

Table 4 presents the genotypic and allelic distribution of IFN-γ gene polymorphisms in pregnant women with and without *T. gondii* infection based on their age. About the IFN-γ gene, 87.5% of infected people between the ages of 26 and 35 had the A/T genotype, and 12.5% had the A/A genotype.

Table 3. Genotype and allele distribution of IFN-γ gene polymorphism in *T. gondii* infected pregnant women and non-infected pregnant women.

Gene	Genotype and Allele	Infected No (%), N = 23	Non infected No (%), N = 69	OR (95% CI)	Fisher's P-value
IFN-γ	A/A	3 (13.04)	6 (8.70)	1.0 (Ref.)	
	A/T	20 (86.96)	63 (91.30)	1.58 (0.40 - 5.84)	0.68 (NS)
	T/T	0 (0)	0 (0)		>0.99 (NS)
	A	26 (56.52)	75 (54.35)	1.09 (0.57 - 2.14)	0.86 (NS)
	T	20 (43.48)	63 (45.65)		

Table 4. Genotype and allele distribution of IFN-γ gene polymorphism in *T. gondii* infected pregnant women and non-infected pregnant women based on different ages.

Gene	Age	Genotype and Allele	Infected No (%), N = 23	Non infected No (%), N = 69	OR (95% CI)	Fisher's P-value
IFN-γ	16 - 25 Years	A/A	0 (0)	2 (11.76)	1.0 (Ref.)	
		A/T	6 (100)	15 (88.28)	0 (0 - 6.2)	>0.99 (NS)
		A	6 (50)	19 (55.88)	0.79 (0.24 - 2.69)	0.75 (NS)
		T	6 (50)	15 (44.12)		
	26 - 35 Years	A/A	2 (12.5)	4 (9.30)	1.0 (Ref.)	
		A/T	14 (87.5)	39 (90.40)	1.39 (0.24 - 6.56)	0.66 (NS)
		A	18 (56.25)	47 (54.65)	1.07 (0.49 - 2.42)	>0.99 (NS)
		T	14 (43.75)	39 (45.35)		
	>35 Years	A/A	1 (100)	0 (0)	1.0 (Ref.)	
		A/T	0 (0)	9 (100)		0.10 (NS)
		A	2 (100)	9 (50)	1.0 (Ref.)	
		T	0 (0)	9 (50)		0.48 (NS)

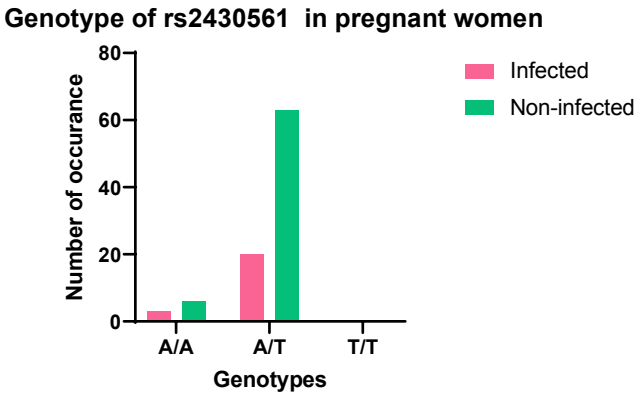

Figure 3. Genotypic Distribution of rs2430561 in *T. gondii* infected and non-infected pregnant women. The AA genotypes are significantly lower in *T. gondii* infected and non-infected pregnant women then the AT genotype. And the T/T genotypes are absent both infected and non-infected pregnant women.

And among the people who were not infected, 90% had the A/T genotype and 12.5% had the A/A genotype. When A/A was considered the wild type of genotype for SNP rs2430561 for the IFN-γ gene, neither infected nor non-infected people had the T/T genotype. It was found that there was no significant association between toxoplasmosis susceptibility and allelic variation of SNP rs2430561 for ages 26 - 35 (OR = 1.39; 95% CI = 0.24 - 6.56; P Value = 0.66 at Fisher's Exact Test), with A/A as the wild type of genotype.

3.7. Genotype and Allele Distribution of IFN-γ Gene Polymorphisms in *T. gondii* Infected and Non-Infected Pregnant Women Based on Stage of Pregnancy

Genotypic and allelic distribution of IFN-γ gene polymorphisms in and *T. gondii* infected and non-infected pregnant women based on different Trimester are given in **Table 5**. In non-infected individuals, the A/T genotype frequency was 87.5% in the first trimester and 91.67% in the second. Again, the occurrence of the A/A genotype in the First and Second Trimesters was 12.5% and 8.33%, respectively. A/A genotype was absent in both the First and Second Trimester of infected individuals, but A/T genotype was 100% in both. Considering A/A as the wild-type genotype, the T/T genotype of SNP rs2430561 for the IFN-gene was absent in both infected and uninfected individuals in all three trimesters. Therefore, no significant correlation was found between toxoplasmosis susceptibility and allelic variation of SNP rs2430561 of the First and Second Trimesters

Among the infected subjects of Third Trimester, A/A genotype was 18.75% and genotype for A/T was 81.25%. Again, among the non-infected subjects of Third Trimester, A/A genotype was 6.9% and genotype for A/T was 93.1%. Therefore, it was found that there was no significant association between toxoplasmosis susceptibility and allelic variation of SNP rs2430561 of Third Trimester (OR = 3.12; 95% CI = (0.56 - 18.7); P Value = 0.33 at Fisher's Exact Test), considering A/A as wild type genotype.

Table 5. Genotype and Allele Distribution of IFN-γ Gene Polymorphisms in *T. gondii* infected and non-infected pregnant women based on Stage of Pregnancy.

Gene	Stage of Pregnancy	Genotype and Allele	Infected N = 23	Non infected N = 69	OR (95% CI)	Fisher's P-value
IFN-γ	First Trimester	A/A	0 (0)	2 (12.5)	1.0 (Ref.)	
		A/T	5 (100)	14 (87.5)	0.0 (0.0 - 7.136)	>0.99 (NS)
		A	5 (50)	18 (56.25)	0.78 (0.19 - 3.21)	>0.99 (NS)
		T	5 (50)	14 (43.75)		
	Second Trimester	A/A	0 (0)	2 (8.33)	1.0 (Ref.)	
		A/T	2 (100)	22 (91.67)	0.0 (0.0 - 31.32)	>0.99 (NS)
		A	2 (50)	26 (54.17)	0.85 (0.13 - 5.76)	>0.99 (NS)
		T	2 (50)	22 (45.83)		
	Third Trimester	A/A	3 (18.75)	2 (6.9)	1.0 (Ref.)	
		A/T	13 (81.25)	27 (93.1)	3.12 (0.56 - 18.7)	0.33 (NS)
		A	19 (59.38)	31 (53.45)	1.27 (0.53 - 2.91)	0.66 (NS)
		T	13 (40.62)	27 (46.55)		

3.8. Genotype and Allele Distribution of IFN-γ Gene Polymorphisms in *T. gondii* Infected and Non-Infected Pregnant Women Based on Cat Contact

Table 6 shows the genotypic and allelic distribution of the IFN-γ gene polymorphism in *T. gondii* infected and uninfected pregnant women based on cat interaction. Regarding the IFN-γ gene, 13.33% of infected individuals in contact with cats had the A/A genotype, whereas 87.67% had the A/T genotype. And among the non-infected participants, the A/A genotype was 11.1% and the A/T genotype was 88.89%. In contrast, the T/T genotype of SNP rs2430561 for the IFN-γ gene was absent in both infected and uninfected individuals when the wild-type genotype A/A was considered. No significant connection was detected between toxoplasmosis susceptibility and allelic variation of SNP rs2430561 in individuals in contact with cats (OR = 1.23; 95% CI = (0.13 - 19.84); P Value > 0.99 at Fisher's exact test), considering A/A as the wild-type genotype.

4. Discussion

The IFN-γ +874T/A polymorphism and IFN-γ production levels are correlated, according to several studies. According to previous research, it has been shown that the T/T genotype is linked to high levels of IFN-γ production, whilst the A/T genotype is linked to medium levels and the A/A genotype is linked to low levels [13] [14] [15]. Many studies showed that polymorphism at position +874T/A in the IFN-γ gene is linked to the development of several disorders. For instance, the A/A genotype is linked to hepatitis B in China [16] tuberculosis in Spain [13], helicobacter pylori gastritis in Italy [17], type 2 diabetes mellitus in Greece [18], and Wegener' [19]. It has been demonstrated that the A/T and T/T genotypes are associated with a variety of diseases, including hepatitis C in Taiwan [14], breast cancer in Iran [20], and Hashimoto's disease in Japan [21].

Table 6. Genotype and Allele Distribution of IFN-*γ* Gene Polymorphisms in *T. gondii* infected and non-infected pregnant women based on cat contact.

Gene	Cat Contact	Genotype and Allele	Infected N = 23	Non infected N = 69	OR (95% CI)	Fisher's P-value
IFN-*γ*	Yes	A/A	2 (13.33)	1 (11.11)	1.0 (Ref.)	
		A/T	13 (86.67)	8 (88.89)	1.23 (0.13 - 19.84)	>0.99 (NS)
		A	17 (56.67)	10 (55.56)	1.05 (0.34 - 3.14)	>0.99 (NS)
		T	13 (43.33)	8 (44.44)		
	No	A/A	1 (12.5)	5 (8.33)	1.0 (Ref.)	
		A/T	7 (87.5))	55 (91.67)	1.57 (0.12 - 10.68)	0.54 (NS)
		A	9 (56.25)	65 (54.17)	1.09 (0.39 - 2.87)	>0.99 (NS)
		T	7 (43.75)	55 (45.83)		

To our knowledge, toxoplasmosis has been linked to the +874T/A polymorphism in the gene producing IFN-*γ*. In our study it was found that 90.22% of the population had the A/T genotype and 9.78% had the A/A genotype. Similar to this study, the A/T genotypes were found as the predominant group in Brazilian populations. According to Albuquerque *et al.* study [11], the genetic profile consisted of 27% A/A genotype, 63.7% A/T genotype, and 8.9% T/T genotype. Our findings concurred with the study of LaguilaVisentainer *et al.* [22] in Brazil. The A/A genotype was found in 30.3%, the A/T genotype was found in 55%, and the T/T genotype was found in 14.7% in this population. Similarly, Matos *et al.* [23] observed a genotypic distribution of 38.4%, 45%, and 16.6%, and Visentainer *et al.* [24] identified a profile of 31.8%, 54% and 14.2% of each genotype. In contrast to the current data, it was reported in India [25] that the A/A genotype was the most prevalent (46.2%), followed by the A/T genotype (35%) and the T/T genotype (18.2%). Amim *et al.* [26] reported a similar genotype distribution profile (45% had the A/A, 32% had the A/T, and 23% had the T/T genotype) in their population. In a population of Rio de Janeiro, Brazil, Neves *et al.* [27] identified a distinct profile with 40% of the A/A genotype, 42.5% of the A/T genotype, and 17.5% of the T/T genotype. Interestingly, in Egypt, Hussein *et al.* [28] revealed that the T/T genotype (53%) was the most prevalent, followed by the A/T genotype (33%) and the A/A genotype (14%).

5. Conclusion

This case-control study demonstrates that pregnant Bangladeshi women are more likely to have *T. gondii* infection than the healthy control group suggesting that pregnant women may acquire more *T. gondii* infection. We did not find any statistically significant correlation between IFN-*γ* gene polymorphism with susceptibility to *T. gondii* infection. However, further studies are needed to perform with large sample size to find out the association of *T. gondii* infection with cytokine gene polymorphisms.

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Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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