Evaluation of immunological response in women infected with *Toxoplasma gondii*

Alaa Adnan Aljanabi

College of Nursing, University of Kerbala , Kerbala , Iraq

E-mail: alaa.adnan@uokerbala.edu.iq

Received: 04/04/2026
Accepted: 13/05/2026
Published: 30/06/2026
DOI:
https://doi.org/10.65682/kjnhs.v2.i2.136-145

Abstract

Abstract :

Background: *Toxoplasma gondii* parasite is a source of concern for health authorities, especially women of reproductive age, and cytokines play a key role in the immune response against the parasite and have an effective role in eliminating it and limiting its multiplication.

Objective: Measurement of cytokine levels (IL-17A and IFN-γ) in the serum of women infected with *Toxoplasma gondii* during their reproductive years . Comparison their levels in infected women with those in a control group . Assessment of the relationship between these cytokine levels and certain epidemiological factors in infected women .

Methods: Serum samples (90) were collected from women blood and divided into patient group (60)and control group (30) . Epidemiological factors (age, place of residence, educational level, economic status, pregnancies and whether or not they contact with cats) were recorded by structured questionnaire . Serum concentrations of the cytokines IL-17A and IFN-γ were calculated using (ELISA) technique .

Results: There was significant increase in IL-17A and IFN-γ levels in the patient group compared to the control group ($P < 0.05$). These results indicate the presence of activated immune response represented by Th1 and Th17 cells in the patient group.

Conclusion: *Toxoplasma gondii* is closely linked to an immune response and elevated levels of IL-17A and IFN-γ. Epidemiological factors were associated with higher rates of parasite infection and stimulation of the immune response. These results reinforce the importance of studying the relationship between epidemiological factors and immune response and underscores the need to identify preventive methods .

Keywords: *Toxoplasma gondii*; IL-17A; IFN-γ; cytokines; epidemiological factors; rural residence; immune response.

1. Introduction

Toxoplasma gondii is a widespread parasite, affecting approximately one-third of the world's population. It is an obligate intracellular parasite. Infection occurs through the consumption of undercooked meat containing parasite cysts and or through contaminated water and food with parasite oocysts contaminate by cats feces . Individuals with a healthy immune system may not experience any symptoms, In women, it can cause serious complications, especially in pregnant women, leading to miscarriage or other complications(Montoya & Liesenfeld, 2004 ; Flegr, *et al*., 2014) . The most common methods for diagnosing toxoplasmosis are immunological methods, as the detection of immunoglobulins definitively and conclusively determines the state of infection. If the infection is new, immunoglobulin M (IgM) appears, while if the infection is old, immunoglobulins G (IgG) appear, Detecting these two types of immunoglobulins in women of childbearing age is very important to avoid certain diseases and their risks to the fetus (Robert-Gangneux & Dardé, 2012 ; Rostami, *et al*., 2020) . In addition to humoral immunity, cellular immunity is crucial in toxoplasmosis infection. One of the key cytokines against the disease is IFN-γ, which activates phagocytic cells and stimulates internal processes that inhibit parasite replication. The lower the level of this cytokine, the greater the parasite's multiplication, leading to more severe symptoms in the patient (Hunter & Sibley, 2012; Yarovinsky, 2014) .

IL-17A is primarily secreted by TH17 cells, a potent pro-inflammatory agent that plays a significant role in the body's immune response. Elevated levels of this cytokine disrupt the inflammatory response, as observed in women with recurrent miscarriages. It attracts immune cells and exacerbates inflammation, potentially leading to pregnancy loss. Elevated TH17 levels in women with recurrent miscarriages indicate a strong TH17 response, which may negatively impact pregnancy in cases of parasitic infection (Liu *et al*., 2021) . Understanding the body's immune response, both humoral and cellular, helps in understanding the immune response triggered by *Toxoplasma gondii* . immunoglobulin IgG & IgM play a role in determining the stage of infection, while cytokines IFN-γ & IL-17A indicate the functional immune status of the infected body, The balance between the body's protective and inflammatory immune responses is key to the severity of the disease (Robert-Gangneux & Dardé, 2012; Yarovinsky, 2014) .

IFN-γ acts as a macrophage activator and promoter of intracellular parasite elimination. It also regulates the innate and adaptive immune response, and thus it is a key cytokine in the body’s resistance to Toxoplasma gondii. Many recent studies have shown the important pathways of this cytokine in resisting parasites and their reproduction within the body, and thus reducing disease complications (Lüder, 2024; Chen *et al*., 2024) . While IL-17A attracts neutrophils and amplifies the local immune response, it is a pro-inflammatory cytokine. Recent studies show that it contributes to protective immunity against Toxoplasma gondii infection. Excessive production of this cytokine causes inflammatory tissue damage, and individuals infected with Toxoplasma gondii have been found to have high concentrations of it, which helps to establish it as an immunological marker for parasite infection (Silva et al., 2024). Demographic factors may influence an individual's susceptibility to infection and the extent of the cytokine response to this infection. Age, place of residence, educational level, and direct contact with cats can also affect the host's response to the parasite and influence differences in IFN-γ and IL-17A synthesis in infected women (Robert-Gangneux & Dardé, 2012) .

Therefore, assessing the levels of these cytokines in the presence of these factors may help

improve the understanding of the immune responses associated with *Toxoplasma gondii* infection (Robert-Gangneux & Dardé, 2012; Petersen et al., 2010).

2. Material & Methods

2.1. Study design

This study was designed as a case-control study to determine the values of the immune response in women infected with *Toxoplasma gondii* . The sample size was determined to be 90 women infected with the parasite Toxoplasma gondii and 30 healthy women as a control group, based on the patient turnout at the hospital during the study period.

2.2. Study population

The study included women who visited the Obstetrics and Gynecology Hospital for the period from (10-2-2026) to (1-6-2026). 90 blood samples were collected from patients and healthy women as control (60 patients & 30 healthy) , and their data on age and disease history were taken using a questionnaire.

2.3. Sample collection

Approximately 5 ml of venous blood was taken from the female patients. The serum was then separated from the blood using a centrifuge at 3000 rpm for 10 minutes. The samples were then transferred to Eppendorf tubes and stored at -20°C until analysis.

2.4. Serological analysis

The presence of the *Toxoplasma gondii* in serum samples was detected using enzyme-linked immunosorbent assay (ELISA) kit (Elabscience, USA) according to the manufacturer's instructions, by the presence or absence of IgG and/or IgM immunoglobulins. The women were divided into two groups: a patient group with positive IgG and/or IgM levels, and a control group with negative IgG and/or IgM levels (Montaya &Liesenfeld,2004 ; Sensini, 2006) .

2.5. Cytokine measurement

Serum IFN-γ and IL-17A levels were measured using commercial ELISA kits. The steps were performed according to the manufacturer's instructions, and the concentrations were calculated from the titration curves and measured in pg/ml.

2.6. Ethical approval

Ethical approval for this study was obtained from the Scientific and Ethical Committee of the College of Nursing - University of Kerbala, Iraq, through written consent from the committee members , Ethical approval bears the symbol (UOK.CON.26.0128) .

2.7. Inclusion Criteria

Samples were taken from women aged 18-45 years who had tested positive for the disease via serological diagnosis (IgG and/or IgM) and who consented to provide a sample for this study.

2.8. Exclusion Criteria

The study excluded women with autoimmune diseases, malignancies, chronic infections, acute non-parasitic infections such as *Toxoplasma gondii* , or women taking immunosuppressive therapy.

2.9. Statistical analysis

The data were analysed using IBM SPSS statistics for windows, version 26.0 and the results were expressed as mean ± standard deviation. Then, the study groups were compared using appropriate statistical tests, and a p-value of less than 0.05 was adopted as a statistically significant , IBM Corp. (2019)

3. Results

The epidemiological factors obtained from the study groups of patients and control showed several important results, as shown in table no. (1). There was no significant difference between the two groups in terms of age, as the average age of infection for the patient group was 29.3 years and for the control group 29.2 years, with a p-value of 0.907. Thus, it is clear that age has no effect on infection.

However, the results showed that the mean number of pregnancies in the patient group (3.17) was higher than in the control group (2.23), with a significant difference (p = 0.002). This suggests a possible association between reproductive history and the incidence of the disease. Regarding environmental and behavioural factors, the results showed that patient contact with cats had a clear impact on increasing the disease incidence. The infection rate with contact cat in patient group was 63.30%, compared to 23.30% in the control group (p = 0.0059).

As for housing, living in rural areas had a clear effect on infection at a rate of 66.70% compared to 33.30% in the control group, with a value of p = 0.0059.

Table 1. Distribution of epidemiological factors, IgG & IgM in women infected with *Toxoplasma gondii* comparison to control.

Factor	Patient Group	Control Group	P-Value
Age (Mean)	29.3 years	29.2 years	0.907
Pregnancies (Mean)	3.17	2.23	0.002
Cat Contact (%)	63.30%	23.30%	0.0007
Rural Residence (%)	66.70%	33.30%	0.0059
Education (%)	*61.7%*	36.7%	0.0497
Socioeconomic (%)	50.0%	16.7%	0.0154
IgG Positive (%)	75.00%	0%	<0.0001
IgM Positive (%)	78.3%	0%	0.0038

Regarding education data, the results showed that the level of education influenced the increase in infection rates, with infection rates reaching 61.7% among those with low education, compared to 16.7% in the control group. Furthermore, individual economic income also impacted

infection rates, with infection rates reaching 50% among low-income patients, compared to 16.7% in the control group.

As for the values of immunoglobulin IgG and IgM , the results showed significant differences between the patient group and the control group. IgG appeared in 75.00% of the patients, while it was 0% in the control group, with a value of p = 0.0001. Also, IgM appeared in 78.3% of the patients and was not present at all in the control group , with a value of p = 0.0038. This indicates the existence of significant differences in the measures of immune response between the two groups of patients and control .

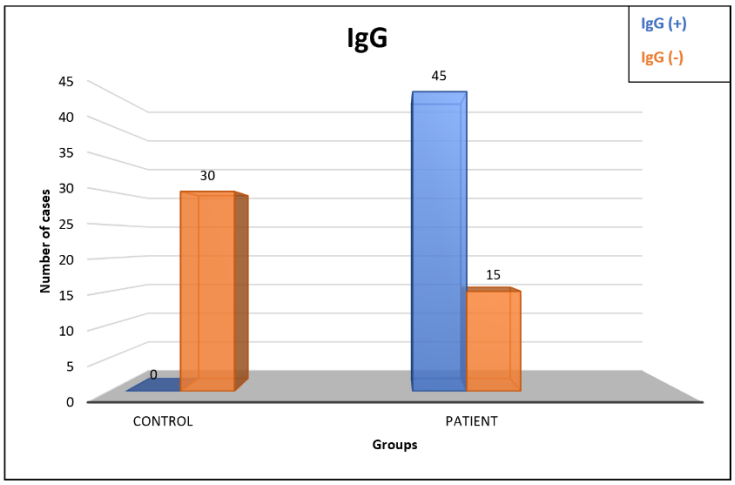

Figure 1. Distribution of IgG seropositivity in infected women group comparison to control group.

Figure 1. shows that the control group did not show the presence of immunoglobulin IgG (30 individuals), meaning they had not been previously infected with the parasite. In contrast, the patient group was much more positive, with a high percentage of 45 individuals testing positive for immunoglobulin IgG compared to only 15 negative individuals. This indicates that the patient group had experienced a previous infection more frequently than the control group, and that this infection was chronic, not acute, as IgG appears after an acute infection and remains in the body for a long period .

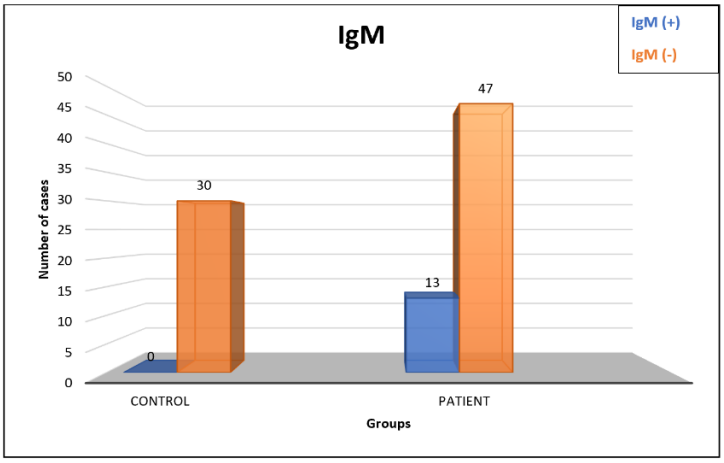

Figure 2. Distribution of IgM seropositivity in infected women group comparison to control group.

Figure 2. shows that all members of the control group (30 individuals) were IgM-negative, while in the patient group, 47 individuals were IgM-positive and 13 were IgM-negative. As illustrated in Figure 2, the results revealed that the IgM positivity rate reached 78.3%. This high percentage indicates the presence of a recent infection within the group of infected women, as the body produces these immunoglobulins during the early stages of parasitic infection. Conversely, the negative IgM results observed in the control group indicate the absence of infection among the women in that group.

Table 2. Comparison of IL-17A & IFN-γ concentration between infected women and healthy controls.

Group	**No.**	**Mean ± SD**	
		IL-17A	**IFN-γ**
Patient	60	160 ± 36.23	134.25 ± 25.09
control	30	77.22 ± 26.01	50.93 ± 10.61
LSD value		15.56*	10.17

*significant at P < 0.05 according to LSD test

The results, as shown in Table (2), indicate that the mean of IL-17A level increased in the patient group compared to the control group, reaching 160.13 ± 36.23 compared to 77.22 ± 26.01, with a highly significant difference (p < 0.001. , LSD 15.56) . The results also showed that IFN-γ levels increased significantly in the patient group compared to the control group, with a mean IFN-γ of 134.25 ± 25.09 in the patient group compared to 50.93 ± 10.61 in the control group, a highly significant difference (p < 0.001, LSD 10.17).

The results showed that both cytokines were significantly elevated in patients compared to controls, indicating an immune response due to the infection.

4. Discussion

The results showed no significant age difference between the study groups, which is consistent with Pappas *et al.* (2009), who indicated no association between age at birth and infection. Furthermore, many epidemiological studies have shown that cumulative exposure to diseases increases with age, but because this study was limited to age at birth, no clear significant difference was observed (Jones *et al*., 2001) .

The study's results were consistent with those of Tenter *et al*. (2000) who demonstrated that the number of pregnancies increases cumulative exposure to the disease. This was also the conclusion of the study, as the number of pregnancies in the patient group had a clear effect on increasing exposure to the disease compared to the control group . Montoya & Liesenfeld (2004) further corroborated this, linking reproductive history to an increased likelihood of developing the disease, particularly in areas where it is endemic.

The incidence of the disease is higher in rural areas compared to urban areas because individuals are more exposed to soil contamination due to animal husbandry (Robert-Gangneux & Dardé, 2012). Soil contamination with oocysts, as well as water contamination and the ability of the

oocysts to survive for extended periods, are key factors in disease transmission .

Higher levels of education contribute to increased preventative behaviours such as thoroughly cooking meat and exercising caution when approaching animals (Pappas et al., 2009). Jones *et al*. (2001) found that lower levels of education among patients are logical indicators, as they directly affect health awareness, hygiene practices, and the ability to determine food safety, all of which are beneficial as preventative measures against disease & this aligns with the results of the current study.

The study showed that infection rates increase with declining economic status, and this was confirmed by Tenter *et al*. (2000) who indicated that poor sanitation, population growth, and weak quality control of food are all contributing factors to increased infection. Economic decline also contributes to reduced access to preventive and health care services for members of the community (Montoya & Liesenfeld, 2004) .

Contact with cats plays a major role in the transmission of the infection, as cats are the definitive host of the parasite. The oocysts that cats excrete with feces are well resistant to unfavourable environmental conditions, making them a major cause of disease transmission (Robert-Gangneux & Dardé, 2012). Pappas et al. (2009) considered direct or indirect contact with cats to be a risk factor for the transmission of the infection .

When the level of immunoglobulin (IgG) rises in a patient, this indicates that he has been exposed to a previous or latent infection, and these immunoglobulins continue to be present in the body for life, giving an indication of a chronic infection (Montoya & Liesenfeld, 2004 ; Lopes, *et al*., 2007). If the level of immunoglobulin (IgM) rises, it indicates the presence of a recent or acute infection. If IgM continues to appear, it requires a precise clinical interpretation (Robert-Gangneux & Dardé, 2012).

The level of IL-17A was significantly elevated in the patient group, indicating an immune response via Th17 helper T cells due to *Toxoplasma gondii* infection. IL-17A is considered essential for stimulating inflammation, attracting neutrophils, and enhancing inflammatory responses and defense pathways employed by the body against intracellular pathogens (Kelly *et al*., 2005 ; Evangelista, *et al*., 2021; Silva, *et al*.,2024). Studies have shown that IL-17A plays a crucial role in regulating and balancing protective immunity and inflammation, and that its increase is associated with heightened immune response and tissue inflammation caused by parasite infection (Stumhofer, *et al*., 2006). In addition, this may explain why antigenic stimulation persists in the infected group. It is noted that immunity continues to be activated in the parasitic infection, as cytokines help the body control the parasitic infection (Dupont *et al*., 2012).

IFN-γ has a major protective role when infected with the parasite, and therefore its levels rise in infected individuals. It is the most important in resisting the disease as it helps in stimulating phagocytic cells and contributes to eliminating the parasite inside cells and inhibits the replication of tachyzoites (Suzuki *et al*., 1988). It stimulates type 1 Th1 helper T cells, which help in eliminating intracellular infections of protozoa. Studies have shown that a deficiency of this cytokine in the body increases the severity of the parasite infection (Hunter & Sibley, 2012), and if its concentration rises in the infected body, it will cause a strong and protective immune response. This is consistent with the numerous studies that have shown that this cytokine is a key factor in the body's resistance (Gazzinelli *et al*., 1993 ; Sturge & Yarovinsky, 2014) .

Elevated levels of IL-17A and IFN-γ indicate an immune response to infection, as demonstrated by the current study, which also showed the influence of environmental, social, and economic factors. In this study, age was not significantly associated with parasite infection, possibly because the age range used was narrow (18-45%), which reduced the possibility of detecting age-related differences. (Pappas *et al*., 2009). However, place of residence influences the likelihood of infection, as rural areas have increased contact with animals, particularly cats. Soil contamination with parasite cysts enhances antigenic stimulation , which activates the Th1 and Th17 immune response (Robert-Gangneux & Dardé, 2012 ; Hill & Dubey, 2002) .

Overall, the results showed that IL-17A and IFN-γ concentrations increase upon exposure to infection, in addition to epidemiological factors that may promote infection.

5. Conclusion

In conclusion, the current study showed that women infected with *Toxoplasma gondii* have changes in the immune response, as the levels of IFN-γ and IL-17A, caused by the activation of Th1 and Th17 immune cells due to the infection with the parasite, have increased. When infected with intracellular parasites, these cytokines play a major role in the immune response. At the same time, If they are produced in quantities exceeding the need , may be associated with inflammatory responses during infection . These results show the importance of these cytokines in the immune response, It may be used as an indicator of the severity of infection and immune activity .

6. Acknowledgements

I extend my sincere thanks to everyone who helped me and contributed to the completion of this research, including the staff at the hospital and medical laboratories, who played an important role in collecting samples and analysing results. Thanks, are also due to everyone who provided advice, guidance, or moral support and played a role in completing the study.

7. Recommendations

Based on the results of the current study, a number of recommendations can be made, including: future studies should examine larger groups of individuals and additional immunological markers related to the immune response to parasitic infection, and assess inflammatory and regulatory cytokines. Studies on the dynamics of these cytokines during infection and pregnancy can also be conducted. All of these studies could help in early diagnosis and understanding the immunological mechanism of the disease.

8. References

Dupont, C. D., Christian, D. A., & Hunter, C. A. (2012). Immune response and immunopathology during toxoplasmosis. *Seminars in Immunopathology*, *34*(6), 793–813. https://doi.org/10.1007/s00281-012-0339-3

Evangelista, F. F., Nishi, L., Colli, C. M., de Laet Sant'Ana, P., Higa, L. T., Muniz, L. H. G., & Falavigna-Guilherme, A. L. (2021). Increased levels of IL-17A in serum and amniotic fluid of pregnant women with acute toxoplasmosis. *Acta Tropica*, *222*, 106019. https://doi.org/10.1016/j.actatropica.2021.106019

Flegr, J., Prandota, J., Sovičková, M., & Israili, Z. H. (2014). Toxoplasmosis – A Global Threat. Correlation of Latent Toxoplasmosis with Specific Disease Burden in a Set of 88 Countries. *PLoS ONE*, *9*(3), e90203. https://doi.org/10.1371/journal.pone.0090203

Gazzinelli, R. T., Hieny, S., Wynn, T. A., Wolf, S., & Sher, A. (1993). Interleukin 12 is required for the T-lymphocyte-independent induction of interferon gamma by an intracellular parasite and induces resistance in T-cell-deficient hosts. *Proceedings of the National Academy of Sciences*, *90*(13), 6115–6119. https://doi.org/10.1073/pnas.90.13.6115

Hill, D., & Dubey, J. P. (2002). Toxoplasma gondii: transmission, diagnosis and prevention. *Clinical Microbiology and Infection*, *8*(10), 634–640. https://doi.org/10.1046/j.1469-0691.2002.00485.x

Hunter, C. A., & Sibley, L. D. (2012). Modulation of innate immunity by Toxoplasma gondii virulence effectors. *Nature Reviews Microbiology*, *10*(11), 766–778.

Jones, J. L., Lopez, A., Wilson, M., Schulkin, J., & Gibbs, R. (2001). Congenital toxoplasmosis: a review. *Obstetrical & Gynecological Survey*, *56*(5), 296–305.

Kelly, M. N., Kolls, J. K., Happel, K., Schwartzman, J. D., Schwarzenberger, P., Combe, C., Moretto, M., & Khan, I. A. (2005). Interleukin-17/Interleukin-17 Receptor-Mediated Signaling Is Important for Generation of an Optimal Polymorphonuclear Response against Toxoplasma gondii Infection. *Infection and Immunity*, *73*(1), 617–621. https://doi.org/10.1128/IAI.73.1.617-621.2005

Liu, Y., Chen, H., Feng, L., & Zhang, J. (2021). Interactions between gut microbiota and metabolites modulate cytokine network imbalances in women with unexplained miscarriage. *Npj Biofilms and Microbiomes*, *7*(1), 24. https://doi.org/10.1038/s41522-021-00199-3

Lopes, F. M. R., Mitsuka-Breganó, R., Gonçalves, D. D., Freire, R. L., Karigyo, C. J. T., Wedy, G. F., Matsuo, T., Reiche, E. M. V., Morimoto, H. K., & Capobiango, J. D. (2009). Factors associated with seropositivity for anti-Toxoplasma gondiiantibodies in pregnant women of Londrina, Paraná, Brazil. *Memórias Do Instituto Oswaldo Cruz*, *104*, 378–382.

Montoya, J., & Liesenfeld, O. (2004). Toxoplasmosis. *The Lancet*, *363*(9425), 1965–1976. https://doi.org/10.1016/S0140-6736(04)16412-X

Pappas, G., Roussos, N., & Falagas, M. E. (2009). Toxoplasmosis snapshots: Global status of Toxoplasma gondii seroprevalence and implications for pregnancy and congenital toxoplasmosis. *International Journal for Parasitology*, *39*(12), 1385–1394. https://doi.org/10.1016/j.ijpara.2009.04.003

Robert-Gangneux, F., & Dardé, M.-L. (2012). Epidemiology of and Diagnostic Strategies for Toxoplasmosis. *Clinical Microbiology Reviews*, *25*(2), 264–296. https://doi.org/10.1128/CMR.05013-11

Rostami, A., Riahi, S. M., Gamble, H. R., Fakhri, Y., Shiadeh, M. N., Danesh, M., Behniafar, H., Paktinat, S., Foroutan, M., & Mokdad, A. H. (2020). Global prevalence of latent toxoplasmosis in pregnant women: a systematic review and meta-analysis. *Clinical Microbiology and Infection*, *26*(6), 673–683.

Silva, Z. M., Toledo, D. N. M., Pio, S., Machado, B. A. A., dos Santos, P. V., Hó, F. G., Medina, Y. N., de Miranda Cordeiro, P. H., Perucci, L. O., & de Castro Pinto, K. M. (2024). Neuroserpin, IL-33 and IL-17A as potential markers of mild symptoms of depressive syndrome in Toxoplasma gondii-infected pregnant women. *Frontiers in Immunology*, *15*, 1394456.

Stumhofer, J. S., Laurence, A., Wilson, E. H., Huang, E., Tato, C. M., Johnson, L. M., Villarino, A. V, Huang, Q., Yoshimura, A., Sehy, D., Saris, C. J. M., O'Shea, J. J., Hennighausen, L., Ernst, M., & Hunter, C. A. (2006). Interleukin 27 negatively regulates the development of interleukin 17–producing T helper cells during chronic inflammation of the central nervous system. *Nature Immunology*, *7*(9), 937–945. https://doi.org/10.1038/ni1376

Sturge, C. R., & Yarovinsky, F. (2014). Complex Immune Cell Interplay in the Gamma Interferon Response during Toxoplasma gondii Infection. *Infection and Immunity*, *82*(8), 3090–3097. https://doi.org/10.1128/IAI.01722-14

Suzuki, Y., Orellana, M. A., Schreiber, R. D., & Remington, J. S. (1988). Interferon-γ: the Major Mediator of Resistance Against Toxoplasma gondii. *Science*, *240*(4851), 516–518. https://doi.org/10.1126/science.3128869

Tenter, A. M., Heckeroth, A. R., & Weiss, L. M. (2000). Toxoplasma gondii: from animals to humans. *International Journal for Parasitology*, *30*(12–13), 1217–1258. https://doi.org/10.1016/S0020-7519(00)00124-7

Yarovinsky, F. (2014). Innate immunity to Toxoplasma gondii infection. *Nature Reviews Immunology*, *14*(2), 109–121. https://doi.org/10.1038/nri3598