

Evaluation of *Toxoplasma gondii* seropositivity and the results of IgG avidity test of patients with suspected Toxoplasmosis

Toksoplazmosis şüpheli hastalarda Toksoplasma gondii seropozitifliğinin ve IgG avidite test sonuçlarının değerlendirilmesi

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SUMMARY

Introduction: *Toxoplasma gondii* is an obligate intracellular protozoan belongs to the phylum Apicomplexa. *T. gondii* has two parts in lifecycle. While the sexual part of the lifecycle occurs in domestic and wild cats, asexual part occurs in any mammal.

Eating undercooked meat containing tissue cysts or drinking water contaminated with oocytes from feline feces cause toxoplasmosis infection. Clinically, infections can go unnoticed or could cause signs and symptoms vary depending on the immune status of the patient and the clinical setting like ocular disease or congenital toxoplasmosis.

Objective: The aim of this study was to investigate the seroprevalance of *Toxoplasma gondii* IgG and IgM in patients with suspected toxoplasmosis.

Method: In this study, seropositivity of antibodies against *T.gondii* in patients with suspected toxoplasmosis was retrospectively evaluated in Medical Microbiology Laboratory of Selcuk University Medical Faculty between January 2013 and December 2016. Anti-*T. gondii* immunoglobulin M (IgM), immunoglobulin G (IgG) antibodies and IgG avidity tests were studied in 7051 serum samples by using the VIDAS (BioMérieux, France) kits and the Enzyme Linked Fluorescence Assay technique.

Results: In all 7051 patients, seropositivity rates of anti-*T. gondii* IgM and IgG antibodies were 2.44 and 29.53 %, respectively. High avidity rate was 78.36 % in 171 patients. Seropositivity rates of toxoplasma IgM antibodies in female and male patients were 2.80 and 1.14%, while IgG seropositivity rates were 34.17 and 22.91 %, respectively.

Conclusions: The importance of searching the seropositivity of toxoplasmosis, which is still considered as an important health problem in especially reproductive women, must be emphasized. In addition, measures should be taken to raise awareness of hygiene in our region in terms of water, foods and sanitation.

Keywords: Avidity, Ig G, Ig M, *Toxoplasma gondii*, seroprevalance

ÖZET

Giriş: *Toxoplasma gondii*, Apicomplexa filumuna ait zorunlu bir hücre içi protozoondur. *T. gondii*, yaşam döngüsü iki evreden oluşur. Yaşam döngüsünün seksüel evresi evcil ve vahşi kedilerde ortaya çıkarken, aseksüel evresi ise herhangi

bir memelide ortaya çıkabilir. İnsanlarda *T. gondii*'ye bağlı enfeksiyonlar, ookist içeren kedi dışkısı ile kontamine yiyeceklerin pişmemiş veya az pişmiş yenmesi ile meydana gelmektedir.

T. gondii enfeksiyonları genellikle asemptomatiktir. Hastanın bağışıklık durumuna ve oküler hastalık veya konjenital toksoplazmozis gibi klinik durumuna bağlı olarak değişik belirti ve semptomlara da neden olabilir.

Amaç: Bu çalışmanın amacı, toksoplazmozis şüpheli hastalarda *Toxoplasma gondii* IgM ve IgG seroprevalansının araştırılmasıdır.

Yöntem: Bu çalışmada, toksoplazmozisten şüphelenilen hastalarda Ocak 2013 ile Aralık 2016 tarihleri arasında Selçuk Üniversitesi Tıp Fakültesi Tıbbi Mikrobiyoloji Laboratuvarı'nda toksoplazma antikorlarının seropozitifliği retrospektif olarak değerlendirildi. Anti-*T.gondii* immünoglobülin M (IgM), immünoglobülin G (IgG) antikorları ve IgG avidite testleri 7051 serum örneğinde VIDAS (BioMérieux, Fransa) kitleri ve Enzim Bağlı Floresans Testi tekniği kullanılarak çalışıldı.

Bulgular: 7051 hastada seropozitiflik oranları anti-*T. gondii* IgM ve IgG antikorları için sırasıyla % 2.44 ve % 29.53'tü. Yüksek avidite oranı 171 hastanın % 78.36'sında saptandı. Kadın ve erkek hastalarda toksoplazma IgM antikorlarının seropozitiflik oranları sırasıyla %2.80 ve 1.14 iken IgG seropozitiflik oranları ise sırasıyla % 34.17 ve % 22.91 idi.

Sonuç: Özellikle üreme çağındaki kadınlarda önemli bir sağlık problemi olarak görülen toksoplazmozisin seropozitifliğinin araştırılmasının önemi vurgulanmalıdır. Ayrıca bölgemizde su, gıda ve temizlik açısından hijyen farkındalığının artırılması ile ilgili önlemler alınmalıdır.

Anahtar sözcükler: Avidite, Ig G, Ig M, *Toxoplasma gondii*, seroprevalans

INTRODUCTION

Toxoplasma gondii is an obligate intracellular protozoan parasite that belongs to the phylum Apicomplexa, subclass coccidia. *T. gondii* has two parts in lifecycle. While the sexual part of the lifecycle occurs in domestic and wild cats, asexual part occurs in any mammal. Human beings can be infected with *T. gondii* by ingestion or handling of undercooked or raw meat containing tissue cysts, drinking water contaminated with oocysts from feline feces, transplantation of infected organs or ingestion of oocysts through close contact with infected cat or feline feces¹.

Most individuals are infected inadvertently, thus the specific route of transmission cannot usually be established. Variations in seroprevalence of *T. gondii* seem to correlate with eating and hygiene habits of a population. This finding lends support to the contention that the oral route is the major source of infection².

It's known that approximately 1/3 of the world population is infected by *T. gondii*. Clinically, infection can go unnoticed or could cause signs and symptoms that vary depending on the immune status of the patient and the clinical setting like ocular disease or congenital toxoplasmosis^{3,4}. As an effective vaccine has not yet been developed, continuous and detailed epidemiological surveillance is required to estimate the risk of infection, especially in pregnant women, and the likelihood of reactivation in immunocompromised individuals⁵.

MATERIAL AND METHODS

In this study, seropositivity of antibodies against toxoplasma in patients with suspected toxoplasmosis was retrospectively evaluated in Medical Microbiology Laboratory of Selçuk University Medical Faculty between January 2013 and December 2016. Blood samples of patients were centrifuged at 4.500 rpm for 15 minutes. Then serum fraction of blood samples was extracted. Anti-*T. gondii* immunoglobulin M (IgM), immunoglobulin G (IgG) antibodies and IgG avidity were studied in 7051 serum samples by using the VIDAS (BioMérieux, France) kits and the Enzyme Linked Fluorescence Assay technique. For IgG, >8 UI/ml test value and for IgM, >0.65 index test value were accepted positive. >0.3 avidity index was interpreted as high.

RESULTS

In all 7051 patients, seropositivity rates of anti-*T. gondii* IgM and IgG antibodies were 2.44 and 29.53%, respectively. At Table 1, there are all rates of anti-*T. gondii* IgM and IgG antibodies. Seropositivity rates of toxoplasma IgM antibodies in female and male patients were 2.80 and 1.14%, respectively. IgG seropositivity rates in female and male patients were 34.17 and 22.91%, respectively.

Seropositivity rate of anti-*T. gondii* IgM antibodies in women at fertility ages was shown at Table 2. The rate of anti-*T. gondii* IgM positivity was 2.92% in 3249 women of fertile age.

As seen at Table 3, high avidity rate was 78.36% in 134 of 171 patients. 40 patients had anti-*T. gondii* IgM antibodies.

Table 1. Prevalence of IgG and IgM *Toxoplasma gondii*-specific antibodies by gender

Gender	IgM-positive No (%)	IgM-negative No (%)	IgG-positive No (%)	IgG-negative No (%)	Total No (%)
Female	108 (2.80)	3754 (97.20)	392 (34.17)	755 (65.83)	5152 (73.06)
Male	12 (1.14)	1039 (98.86)	184 (22.91)	619 (77.09)	1899 (26.94)
Total	120 (2.44)	4793 (97.56)	576 (29.53)	1374 (70.47)	7051 (100)

Table 2. IgM rates of women of childbearing age (15-49 ages)

	Ig M	
	No	%
Positive	95	2.92
Negative	3149	96.92
Equivocal	5	0.15
Total	3249	100

Table 3. Distribution of IgG and IgM compared to IgG avidity patterns in all patients

IgG Avidity	IgG-positive IgM-positive No (%)	IgG-positive IgM-negative No (%)
High Avidity (>0.3) 134/171 (78.36%)	40 (29.85)	94 (70.15)
Low Avidity (0.2<) 27/171 (15.39%)	13 (48.14)	14 (51.86)
Borderline (0.2-0.3) 10/171 (6.15%)	7 (70)	3 (30)
Total 171/171 (100%)	60 (35.08)	111 (64.92)

DISCUSSION

The seropositivity of toxoplasmosis varies in many countries and even in different regions within a country, depending on the differences in socioeconomic situation, development levels, climate, and geography, which has been reported to range between 12-90%. The IgG seropositivity was reported to be 37%, 29%, and 24% in 1984, 1997, and 2004, respectively in Greece, which is in the same climate zone as Turkey⁶. Researchers

have related this decreasing trend to socioeconomic development.

IgG and IgM seropositivity has been reported as 23.3–37.1% and 0.1–1.9% in patients pre-diagnosed with toxoplasmosis in Turkey^{7,8,9}. This difference may be attributed to the size of the working groups, socio-demographic characteristics, and habits, as well as the climatic characteristics and test sensitivity. In this study, seropositivity of IgG was higher than of IgM. IgG and IgM seropositivity were found to be 29.53

and 2.44 %, respectively. In our region, these seropositivity rates show similarity with seropositivity rates of the other studies in Turkey. This contribute that *T. gondii* IgG antibodies are reflection of past or previous infection, while IgM reflects recent or acute *T. gondii* infection.

Many studies have investigated the relationship between *T. gondii* seropositivity and gender both in Turkey and in the world. In a study conducted in Brazil, IgG seropositivity was found to be 63.4% in women and 79% in men¹⁰. In a study from United States, the seropositivity was found to be 23.3% and 21.8% in women and men, respectively¹¹. In our study, seropositivity rates of toxoplasma IgM and IgG antibodies were 2.80 and 34.17% in women and 1.14 and 22.91% in men, respectively. This may be attributed to the fact that as opposed to men, women contact contaminated raw food and are at a higher risk of exposure to oocysts disseminated by cats during household chores and gardening work.

The IgG avidity tests that have been in practical use in recent years allow reliable differentiation between acute primary infection, reactivation, and/ or re-infection in a single serum sample. This differentiation has clinical significance, especially in pregnant women and immunocompromised patients. IgG antibodies against the antigen on first exposure during the primary infection show low avidity in the first weeks and then acquire gradually higher avidity with increased maturity.

Alver et al.¹² found the high avidity in 136 (81.9%), borderline in 17 (10.2%), and low in 13 (7.9%) patients in their study. In our study we found the high avidity in 134 (78.36%), borderline in 10 (6.15%), and low in 27 (15.39%) of 171 samples. In our study, 60 patients (35.08%) had IgM positivity, of whom 13 (21.66%) had low avidity, suggesting a recent previous infection, while 40 (66.66%) had high avidity, suggesting a past or previous infection. Avidity of seven patients who had IgM positivity was borderline (11.66%). Also 111 patients (64.92%) had IgM negativity. 94 (70.15%) of them had high avidity.

In pregnant women, multiparity, older age, and a history of poor obstetric outcome have been previously described as risk factors for *T. gondii* infections¹³⁻¹⁴. In a study conducted in Turkey, seroprevalence of *T. gondii* IgM were determined among women of childbearing age (15-49 ages). Positive rate of the *T. gondii* IgM antibodies was found as 4.6 %. In our study, Ig M rate was found lower as 2.92%¹⁵. Enter et al.¹⁶ found *Toxoplasma gondii* Ig M rate as 1.5% seropositivity among pregnant women.

CONCLUSION

Toxoplasma gondii is able to infect all warm-blooded animals and chronically infects approximately one-third of the world's human population. *T. gondii* can cause serious clinical consequences and even death in immunocompromised individuals or patients undergoing immunosuppressive treatments or during pregnancy¹⁷. Regular surveillance studies on toxoplasmosis seroprevalence must continue in our country in order to prediction of congenital toxoplasmosis risk, determination of screening strategies and important for informing the seronegative pregnancies on the prevention methods.

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