

# Evaluation of Anti-*Toxoplasma gondii* Antibodies in Hemodialysis Patients with Chronic Kidney Disease in Sari, Iran

Seyyed Hosein Seyyedpour,<sup>1</sup> Parvaneh Afshar,<sup>2,\*</sup> Ayoub Barzegarnejad,<sup>3</sup> Shamsi Kalhori,<sup>4</sup> and Reza Agah<sup>5</sup>

<sup>1</sup>Deputy of Health, Mazandaran University of Medical Sciences, Sari, IR Iran

<sup>2</sup>Referral Laboratory, Mazandaran University of Medical Sciences, Sari, IR Iran

<sup>3</sup>Department of Urology, Medical School, Mazandaran University of Medical Sciences, Sari, IR Iran

<sup>4</sup>Fatemeh Zahra Hospital, Mazandaran University of Medical Sciences, Sari, IR Iran

<sup>5</sup>Deputy of Treatment, Mazandaran University of Medical Sciences, Sari, IR Iran

\*Corresponding author: Parvaneh Afshar, Referral Laboratory, Mazandaran University of Medical Sciences, Sari, IR Iran. Tel: +98-9111531715, Fax: +98-1133390353, E-mail: Afshar164@yahoo.com.afshar@mazums.ac.ir

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## Abstract

**Background:** *Toxoplasma gondii* has worldwide distribution and is one of the most prevalent infectious agents in humans.

**Objectives:** The aim of this study was to determine the prevalence of anti-*Toxoplasma gondii* antibodies in hemodialysis patients with chronic kidney disease (CKD) in the hemodialysis unit of Fatemeh Zahra hospital at the Mazandaran University of Medical Sciences in Sari, Iran.

**Methods:** Seventy-three patients with CKD and 145 healthy volunteers were assessed for anti-*Toxoplasma gondii* (IgG, IgM, and IgA) antibodies using a conventional ELISA technique.

**Results:** The anti-*Toxoplasma gondii* IgG antibody was detected in 80.8% of the cases in the patient group, while 31.5% of the cases in the CKD patient group and 31.8% in the control group were found to be positive for the anti-*Toxoplasma gondii* IgA antibody. All the patients in the CKD group were negative for the anti-*Toxoplasma gondii* IgM antibody, although 2.76% of the healthy volunteers were found to be positive. The present study suggests that there was no significant difference between the hemodialysis patient group and the healthy volunteers.

**Conclusions:** The epidemiological data collected in this study could serve as a reference for future studies and may be useful in developing preventive and educational strategies, and consequently reducing healthcare expenditure.

**Keywords:** Toxoplasmosis, Hemodialysis, ELISA, IgM, IgG, IgA

## 1. Background

*Toxoplasma gondii* is disseminated worldwide and is one of the most prevalent infectious agents in humans. *Toxoplasma gondii* is an opportunistic parasite in which acute infection is usually asymptomatic in immunocompetent patients, and spontaneous recovery is the norm (1, 2). Generally, toxoplasmosis is dependent on many factors, such as sanitation levels, temperature, humidity, and contact with soil and domestic animals, with a seroprevalence that varies from 0% to 95% (3, 4). The prevalence of *Toxoplasma gondii* is about 22.5%, 0.27% - 12.9%, 23.1%, 80%, and 50% - 84% in the USA (5), South Korea (6), Turkey (7), and Brazil (8, 9), respectively. According to Assmar et al. (10), the seroprevalence of *Toxoplasma gondii* in Iran is 51.8%. Other studies have indicated that 55.7% of the population in the provinces of Mazandaran, Gilan, and

Khozestan (11), 17.7% in Fars and Azarbayjan provinces (11, 12), and 68.4% in the southern region of Tehran (11, 13) are seropositive. Acute toxoplasmosis is asymptomatic in 80% of healthy hosts and only 20% of adults and children exhibit symptomatic infection (14). However the existence of *Toxoplasma* tissue cysts in hosts may contribute to the maintenance of immunity against reinfection, and their presence may also cause symptomatic toxoplasmosis under certain conditions, especially in infants with congenital toxoplasmosis and immunocompromised patients (15). Studies have shown that renal failure patients, such as those on hemodialysis, are immunodeficient, and such patients are therefore at high risk for numerous infections (16-18).

## 2. Objectives

No studies have been conducted regarding toxoplasmosis in hemodialysis patients in the northeast of Iran. In the present study, we therefore aimed to determine the prevalence of anti-*Toxoplasma gondii* IgG, IgM, and IgA antibodies in hemodialysis patients with chronic kidney disease (CKD) in Sari, Iran.

## 3. Methods

From July to September 2013, this cross-sectional study was carried out on 73 patients with CKD undergoing regular hemodialysis in the hemodialysis unit of Fatemeh Zahra hospital at the Mazandaran University of Medical Sciences in Sari, Iran. A total of 145 healthy volunteers with normal creatinine and BUN levels and without any underlying renal disease (control group) who matched the CKD patients according to age, gender, and geographical area were selected. The study was approved by the local ethics committee, and informed consent was obtained from all the participants. Medical records as well as data such as age, sex, and the length of time on hemodialysis treatment were obtained from all the hemodialysis patients. To determine the seroprevalence of *Toxoplasma* infections, 10 mL of peripheral blood was taken under aseptic conditions from all the patients in the hemodialysis and control groups, and each sample was left for 30 - 60 minutes at room temperature for spontaneous clotting before being centrifuged at 1500 rpm for 10 minutes. The serum samples were stored in acid-washed polyethylene tubes and kept frozen at  $-20^{\circ}\text{C}$  until tested in Referral Laboratory of Mazandaran University of Medical Sciences. Anti-*Toxoplasma gondii* IgG, IgM, and IgA were determined using a conventional enzyme-linked immunosorbent assay (ELISA) technique. ELISA kits were purchased from the commercial manufacturer Pishtazteb, Iran, for the IgG and IgM tests, and EI 2410-9601 A (Euroimmun, Germany) was procured for the IgA test. According to the manufacturers' recommendations, IgG absorbance levels  $< 9$  were considered negative,  $9 - 11$  was considered borderline, and  $> 11$  was positive; IgM absorbance levels  $< 0.9$  were considered negative,  $0.9 - 1.1$  was assumed to be borderline, and  $> 1.1$  was positive; and an IgA ratio  $< 0.8$  was considered negative, a ratio  $\geq 0.8$  but  $< 1.1$  was assumed to be borderline, while  $\geq 11$  was positive. In the event of a borderline test result, an additional blood sample was obtained 2 weeks later and retested in parallel with the first suspect sample. Using the results of both samples allowed for the proper evaluation of titer changes. The statistical analysis was performed using SPSS software version 18. The data were analyzed using

a chi-squared test, and  $P < 0.05$  were considered statistically significant.

## 4. Results

Of the 73 CKD patients, there were 40 males and 33 females aged between 28 and 88 years (mean:  $62.32 \pm 12.06$  years). The length of time on hemodialysis treatment was 3 - 13 weeks (mean:  $12.05 \pm 1.97$  weeks). In the control group comprising 145 healthy volunteers, there were 63 males and 82 females aged between 27 and 84 years (mean:  $55.52 \pm 11.43$  years).

In the present study, 59 of the 73 (80.8%) cases in the patient group and 132 of the 145 (91%) healthy volunteers (control group) were found to be positive for IgG antibodies, and 23 (31.5%) cases in the patient group and 46 (31.8%) healthy volunteers (control group) were found to be positive for IgA antibodies. The percentage of participants who were anti-*Toxoplasma gondii* IgG-antibody positive in the control group was found to be significantly greater than in the CKD patient group, although the difference was not statistically significant for the IgA antibody ( $P < 0.05$ ). Meanwhile, all the hemodialysis patients were negative for the anti-IgM antibody, as were four (2.76%) of the control group subjects, and no statistically significant difference was observed between the groups. The seroprevalence distribution of the three antibody groups and a comparison of the anti-*Toxoplasma* antibodies present among the hemodialysis patients and healthy volunteers are shown in [Tables 1 and 2](#), respectively. The relationship between duration of hemodialysis treatment and anti-*Toxoplasma gondii* antibody seropositivity was also verified, and we found a significant difference between these two parameters ( $\chi^2 = 5.24$ ,  $P = 0.155$ ).

## 5. Discussion

Toxoplasmosis, which is an indexed global public health concern (8), is an opportunistic infection that causes non-symptomatic infections in immunocompetent individuals (7). Almost equal percentage prevalence, namely 80.6% and 91%, of anti-*Toxoplasma gondii* IgG antibody was revealed in the hemodialysis patients and healthy volunteers in this study, respectively. The results of the patient group share a similar frequency with figures reported in Turkey (76.5%) (17). However, the data in our study were higher than those reported by Sahinl et al. (16) (59.1%) and Maraghi et al. (18), Ebrahim Zadeh et al. (19) and Rezavand et al. (20) of 59.1%, 40.67%, 56.7% and 60%, respectively in Iran, and Aufy et al. (21) and Alvarado-Esquivel et al. (22) of 56.06%, 56.7% and 10%, respectively in Turkey,

**Table 1.** Seroprevalence of Positive *Toxoplasma* Antibodies in the Hemodialysis patients and Healthy Volunteers

Group	Hemodialysis Patients (n = 73)	Healthy Volunteers (n = 145)	$\chi^2$	P Value
IgG, No. (%) (M/F)	59 (80.8) (34/25)	132 (91) (55/77)	27.901	0
IgM, No. (%) (M/F)	-	4 (2.76) (0/4)	-	-
IgA, No. (%) (M/F)	23 (31.5) (11/12)	46 (31.8) (22/24)	7.667	0.006

Abbreviations: M, male; F, female; Ig, immunoglobulin.

**Table 2.** A Comparison of the Anti-*Toxoplasma* Antibodies in the Hemodialysis Patients and Healthy Volunteers

Group	IgG+				IgG-			
	IgM+ IgA+	IgM- IgA+	IgM+ IgA-	IgM- IgA-	IgM+ IgA+	IgM- IgA+	IgM+ IgA-	IgM- IgA-
Hemodialysis patients (n = 73)	-	21 (28.8)	-	38 (52.1)	-	2 (2.7)	-	12 (16.4)
Healthy volunteers (n = 145)	-	46 (31.7)	1 (0.7)	85 (58.6)	-	-	3 (2.1)	10 (6.9)

Egypt and Mexico; whereas in our results the percentage of Anti-*Toxoplasma gondii* IgG antibody in the volunteer group was more than other studies (17-22). Furthermore, in our study, there were no positive cases of anti-*Toxoplasma gondii* IgM antibody in the patient group and only 2.76% of the healthy volunteer showed the IgM antibody. This result is the same as that of Ocak et al. (17) who studied hemodialysis patients and healthy volunteers in Turkey. On the other hand, Afuy et al. (21), Sahinl et al. (16), Maraghi et al. (18), Ebrahim Zadeh et al. (19), Rezavand et al. (20) and Nissap-torn et al. (23) studies determined 16.7%, 6.8%, 8.6%, 13.5%, 3.3% and 5.7% respectively in the patient groups, which is not compatible with our results.

The anti-*Toxoplasma gondii* IgA antibody was prevalent in 61.5% of the hemodialysis patients and 31.8% of the healthy volunteers in our study. These data were higher than those reported by Sahinl et al. (16) across both groups. The outcome in our study was different from those reported in certain studies where such differences may have been related to local environmental conditions.

Additionally, our study indicated that there was no significant association between anti-*Toxoplasma gondii* due to IgG antibody frequency and the duration of hemodialysis treatment in either sex, whereas such a correlation was shown in the studies of Sahinl et al. (16) in Iran and Ocak et al. (17) in Turkey.

### 5.1. Conclusion

We conducted a case-controlled study to determine the prevalence of anti-*Toxoplasma gondii* IgG, IgM, and IgA antibodies in hemodialysis patients with CKD in Sari, Mazandaran Province, Iran. Nowadays hemodialysis patients should be tested for *Toxoplasma gondii* regularly. Clinicians

should become more aware of these patients, and parasitological surveys should be carried out steadily to eliminate the risk of severe toxoplasmosis. Additionally, it is recommended that this study be extended to include a large sample of CKD patients on hemodialysis from other cities in Mazandaran Province, Iran. The epidemiological data collected in this study could serve as a reference for future studies and may be useful in developing preventive and educational strategies, and consequently reducing health-care expenditure.

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### Footnote

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