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Original Article

Seroprevalence of Anti-*Toxoplasma gondii* Antibodies among Patients with Cancer at Hiwa Cancer Hospital in Sulaimani City, Kurdistan Region, Iraq

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Abstract

Background: *Toxoplasma gondii* is an opportunistic protozoan parasite that causes a life-threatening disease – toxoplasmosis – in immunocompromised individuals, including patients with cancer. This prospective cross-sectional study set out to determine the prevalence of toxoplasmosis in patients with cancer compared with that of healthy individuals.

Methods: A prospective cross-sectional study was conducted in Sulaimani City of Iraq from November 2019 to May 2020. Anti-*T. gondii* IgG and IgM antibodies were measured in the blood samples of 113 patients with cancer (80 with solid organ tumors and 33 with haematological malignancies) entered to Hiwa Cancer Hospital and 82 healthy controls, who were referred to the Directorate of Blood Transfusion for blood donation, using chemiluminescence microparticle immunoassay (CMIA).

Results: The prevalence of anti-*T. gondii* IgG was 39.8% in the patient group and 24.4% in the control group, which amounted to a significant difference (P = 0.024). Only one case of anti-*T. gondii* IgM positivity was observed in the patient group, and no IgM seropositivity was reported in the control group. Moreover, the seroprevalence of anti-*T. gondii* IgG was non-significantly higher (P = 0.102) in the patients with haematological malignancies (51.5%) than in those with solid organ tumors (35%). Occupation was the only risk factor which had a significant association with *T gondii* infection (odds ratio [OR]: 1.3, 95% confidence interval [CI]: 0.6746163 - 2.4282788, P = 0.029).

Conclusion: The prevalence of *T. gondii* infection is higher in patients with cancer than in healthy individuals. Therefore, *T. gondii* screening in patients with cancer is recommended.

Introduction

Toxoplasma gondii is an apicomplexan parasite and an obligatory zoonotic intracellular opportunistic pathogen (1-3). It is ubiquitous, infecting a wide range of warm-blooded animals, including one-third of the world's human population (4). The seroprevalence of toxoplasmosis varies markedly among regions worldwide, ranging between 0.5% and 87.7% (5), depending on the degree of exposure to the oocysts (infective stage) or on climatic conditions, socioeconomic standards, and nutritional behaviors (6-8).

Humans are primarily infected by the disease when they eat undercooked or raw meat that contains tissue cysts from an infected host or any food contaminated with oocysts shed in the feces of infected cats (9). Although uncommon, *T. gondii* can also be transmitted through drinking water (10, 11), mother-to-child transmission during pregnancy (12), blood transfusion, and organ transplants (13-15).

In individuals with a normal immune system, *T. gondii* infection is usually asymptomatic (latent) and harmless and often passes unnoticed; however, in immunosuppressed and immunodeficient individuals (e.g., patients with AIDS, patients receiving hemodialysis, organ transplant recipients, and patients receiving chemotherapy for cancer), it is complicated and fatal, if not treated (16-19). Patients with cancer are often considered immunocompromised because of the nature of the anti-neoplastic treatment they receive (e.g., chemotherapy and/or radiotherapy) or the primary disease. In such cases, the infection is the result of reactivation of the latent phase (chronic infection) and transition into the active phase (acute phase) rather than a primary infection (20, 21).

In immunocompromised patients, IgM antibodies, which are the first antibodies to be detected after 7–15 days of infection, indicate the presence of acute infection; meanwhile, IgG antibodies indicate the presence of chronic infection, and elevated titers may show reactivation of infection (22). Serological techniques are commonly used. They remain the primary routine diagnostic tool for identifying anti-*T. gondii* antibodies in patients with cancer (23). Serological evidence of the presence of anti-*T. gondii* IgG and IgM have been related to numerous cancer cases in various regions worldwide (24-26). For instance, a study conducted in Saudi Arabia showed that the seropositivity rate was 30.6% (anti-*T. gondii* IgM and IgG) among patients with cancer (27). In Jordan, 39.5% and 2.5% of 200 patients with cancer were seropositive for anti-*T. gondii* IgG and IgM compared with 12.2% and 1.1% of 90 controls, respectively (28).

To the best of our knowledge, however, only one study has investigated the seroprevalence and risk factors of *T. gondii* infection in patients with cancer (i.e., leukemia) in the Kurdistan Region (29), while several studies have been carried out on pregnant women and women of childbearing age (30-32). Therefore, this study set out to examine the prevalence of anti-*T. gondii* antibodies in patients with cancer at Hiwa Cancer Hospital (HCH) in Sulaymaniyah City, Kurdistan Region, Iraq, compared with that in healthy individuals.

Materials and Methods

Study Population and Blood Sampling

This prospective cross-sectional study was conducted from November 2019 to May 2020. Serum and plasma samples were collected from 195 individuals aged between 9 and 86 years. The study population consisted of 113 patients with cancer who visited HCH and 82 immunocompetent healthy controls, who were referred to the Directorate of Blood Transfusion for blood donation. The patients with cancer were categorized into two subgroups: patients with solid organ tumors (n =

80) and those with hematological malignancies (n = 33).

The study was approved by the research Ethics Committee of the College of Science, University of Sulaimani (Certificate No. 7/12/4110 dated on 12 Nov. 2019). Before blood sample collection, each participant signed a written consent form.

The aims of the experiment were explained to all participants. Socio-demographic data (age, gender, occupation, residency, and contact with cats) were collected from the patient and control groups using a structured questionnaire. In addition, the type of cancer was recorded for the cancer patients. Venous blood samples were obtained from each participant. After centrifugation, the sera and plasma were separated and preserved in labelled Eppendorf tubes at a temperature of -20°C until further examination.

Anti-Toxoplasma antibody determination:

To identify anti-*T. gondii* IgG and IgM antibodies, chemiluminescence microparticle immunoassay (Abbott ARCHITECT i100-0SR) was performed using ARCHITECT Toxo. IgG and IgM commercial kits (Architect i Systems, Germany), respectively. The procedures were performed following the manufacturer's instructions. Specimens with a concentration of ≥ 3.0 IU/mL were considered reactive (positive) for *T. gondii* IgG antibodies, while those with an index of ≥ 0.60 (≥ 1.00 S/CO) were considered reactive for *T. gondii* IgM antibodies.

Statistical analysis

The collected data of the population groups were entered into Excel sheets and then were analyzed using SPSS for Windows, version 20 (IBM Corp., Armonk, N.Y., USA). The seroprevalence of toxoplasmosis among the patient and control groups was calculated, and its associations with variables were evaluated. The chi-square test was used to compare qualitative variables, and associations that showed *P*-values of < 0.05 were considered significant.

Results

The age of the patient group ranged between 9 and 86 years (M ± SD: 52.6 ± 16.9 years). More specifically, the group consisted of 69 women aged between 18 and 86 years (*M* ± *SD*: 51.8 ± 16.1 years) and 44 men aged between 9 and 82 years (M ± SD: 53.9 ± 18.4 years). Meanwhile, the age of the control group ranged between 10 and 70 years (M ± SD: 36.2 ± 11.2 years). More specifically, the group consisted of 37 women aged between 10 and 69 years (M ± SD: 32.2 ± 11.7 years) and 45 men aged between 22 and 70 years (M ± SD: 39.4 ± 9.8 years).

As shown in Table 1, the difference in the IgG seropositivity between the patient and control groups was significant (*P* < 0.024).

Table 1: Anti-*Toxoplasma gondii* IgG antibodies between the patients with cancer and controls

Anti-Toxoplasma Abs	*Patients n = 113 (%)*		*Controls n = 82 (%)*	*P. value*
IgG	Reactive (positive)	45 (39.8)	20 (24.4)	*0.024
	Non-reactive (negative)	68 (60.2)	62 (75.6)	

The patient group included 80 patients with solid organ tumors and 33 patients with haematological malignancies (Table 2). The prevalence of anti-*T. gondii* IgG antibodies in the patients with haematological malignancies (51.5%; *n* = 17) were non-significantly (*P* < 0.102) higher than that in the patients with solid organ tumors (35%; *n* = 28) (Table 2).

Table 2: Association of the seropositivity for Toxoplasma gondii IgG in all cancer types and its distribution between the patients with haematological malignancies and solid organ tumors

Cancer types (n = 113)	***IgG (+ve)***	***Classification of Tumors***	***No.***	***IgG (+ve)***	
				No.	*P*.value
Hematological malignancies (n = 33)	17 (51.5%)	Leukemia	13	7	0.102
		Lymphoma	8	4	
		Hodgkin lymphoma	5	2	
		Multiple myeloma	5	2	
		Chronic myeloproliferative disea	2	2	
Solid organ tumors (n = 80)	28 (35%)	Breast cancer	20	7	
		Rectum cancer	11	3	
		Colon cancer	10	5	
		Stomach cancer	6	5	
		Ovary cancer	5	0	
		Lung cancer	4	3	
		Prostate cancer	3	1	
		Testis cancer	3	0	
		Other solid organ tumor	3	0	
		Bone cancer	2	0	
		Brain cancer	2	0	
		Gallbladder cancer	2	1	
		Endometrial cancer	2	1	
		Submandibular cancer	1	0	
		Pancreas cancer	1	1	
		Placental cancer	1	0	
		Larynx cancer	1	0	
		Small intestine cancer	1	1	
		Cervical cancer	1	0	
		Thyroid gland cancer	1	0	

As shown in Table 3, the seropositivity rates in the women and men were 55.5% and 44.4% in the patient group and 40% and 60% in the control group, respectively. Thus, there was no significant difference in the seropositivity for anti-*T. gondii* IgG antibodies according to sex (OR: 0.4892367, 95% CI: 0.2452735 - 0.9525424, P = 0.246). The occupation of each group was categorized as follows: non-employee, employee, student, and worker. The seroprevalence of *T. gondii* IgG was significantly higher (OR: 1.3, 95% CI: 0.6746163 - 2.4282788, P = 0.029) in the non-employees (46.1%; n = 30) than in the employees (27.7%; n = 18). In the patient and control groups, 42 (64.6%) and 23 (35.4%) participants resided in urban and rural areas, respectively. Among the patient group, the seropositivity rate in those living in urban areas was higher none significantly (OR: 0.7, 95% CI: 0.3538155 - 1.3990835, P = 0.553) than in those living in rural areas. Furthermore, contact with cats yielded no significant (OR: 1.9, 95% CI: 0.7756925 - 4.6564191, P = 0.1311) difference in the seropositivity rate between the patient and control groups (Table 3). Anti-*T. gondii* IgM was detected in one (1.13%) patient with stomach cancer, while no IgM antibodies were detected among the controls.

Table 3: Association between the different variables and the overall anti-Toxoplasma gondii seropositivity

Variables	*Groups*	IgG Seropositive patients, n = 45 (%)	IgG Seropositive controls, n = 20 (%)	*Total n=65 (%)*	*OR (CI)*	*P. value*	*Patients n =113 vs Controls n = 82*
							P. value
Sex	Male (n = 89)	20 (44.4)	12 (60)	32 (49.2)	1.3 (0.245 - 0.952)	0.246	0.447
	Female (n = 106)	25 (55.6)	8 (40)	33 (50.7)			
Residency	Urban (n = 136)	27 (60)	15 (75)	42 (64.6)	0.7 (0.674-2.428)	0.553	0.321
	Rural (n = 59)	18 (40)	5 (25)	23 (35.4)			
Contact with cat	Yes	35 (77.7)	16 (80)	51 (78.4)	1.9 (0.775-4.656)	0.097	0.1311
	No	10 (22.3)	4 (20)	14 (21.5)			
Occupation	Non-employee	23 (51.1)	7 (35)	30 (46.1)	1.3 (0.674-2.428)	0.029*	0.0029*
	Employee	11 (24.4)	7 (35)	18 (27.7)			
	Student	0 (0.00)	0 (0.00)	0 (0.00)			
	Worker	11 (24.4)	6 (30)	17 (26.1)			

Discussion

The immune system plays a critical role in controlling and clearing parasitic infections (33). Hence, those with weakened immune systems, such as immunocompromised individuals, including patients with cancer, may become infected with life-threatening opportunistic infections such as toxoplasmosis (34).

In the present study, the seroprevalence of anti *T. gondii* IgG antibodies in the patient group (39.8%) was significantly (P = 0.024) higher than that in the control group (24.4%), indicating that patients with cancer are at a higher risk for infection by *T. gondii* than healthy individuals (35). This is attributed to weak immune systems in patients with cancer, which may facilitate the reactivation of previous chronic toxoplasmosis or even increase the potential risk of acquiring new infections (36).

Close to our results, in two previous global (24, 34) systematic review and meta-analysis studies, the IgG seropositivity against *T. gondii* was 30.8% and 42.1%, respectively. Similarly, in another systematic review conducted in Iran (25), the authors reported the prevalence rate of toxoplasmosis in 45.06% of patients with cancer. In consistence with the present study; similar results were found in different geographical populations in the world. For example, in Jordan (21), Turkey (22) and China (37), the seropositivity rates of *Toxoplasma* in patients with cancer were (61.2%), (63%) and (35.56%), respectively, which were significantly higher than that in the healthy individual controls. In contrast to our study, an investigation performed in Iran revealed that the seroprevalence of anti *T. gondii* antibodies in the controls was non-significantly higher than that in patients with cancer (38). These varia-

tions may be associated with the presence of different demographical, social, cultural, and environmental factors (39, 40).

In the current study, the prevalence of toxoplasmosis was found to be non-significantly higher in patients with haematological malignancies (51.5%) than in those with solid organ tumors (35%). This finding is supported by another study that also suggests that haematological malignancy cases have higher seropositivity rates than solid organ tumor cases (36). This may be attributed to the fact that the majority of patients with haematological malignancy have weakened cellular immunity. These patients are regarded as being at high risk for neutropenia because chemotherapeutic drugs kill both rapidly growing cancer cells and healthy white blood cells. Furthermore, the difference in the results may be related to the use of corticosteroids, which are commonly used in most chemotherapy treatments for patients with haematological malignancy (36). However, in contrast to those findings and suggestions, a study conducted in Egypt showed that the seropositivity rate was higher in patients with solid organ tumors than in those with haematological malignancies (41).

Some social and cultural factors affect the prevalence of toxoplasmosis in patients with cancer. For example, concerning sex, the positivity rate in women (50.7%) was non-significantly higher than that among men (49.2%) in both patients and control groups in our study. Similarly, another study (36) showed that women (56.6%) had significantly higher seropositivity rates than men (40.4%). This result may be related to the high indoor activity levels of women along with the consumption of raw or undercooked meat and unwashed vegetables and fruits or farming activities and also more frequent contact with cats or other animals (36). Having said all this, though, in other studies, a higher prevalence of toxoplasmosis has been reported in men than in women (42, 43).

An individual's occupation was found to affect significantly the positivity of *T. gondii* IgG in both the patient and control groups. A significant (P = 0.029) higher positivity rate was observed in the non-employees (46.1%) than in the employees (27.7%) (P = 0.029). In contrast, another study conducted in Iran revealed a non-significant difference in the occupation between patients with cancer and controls (38).

Findings of the current study reported that the seropositive rate in the participants living in urban areas was non-significantly (P = 0.553) higher (64.6%) than that in the rural areas (35.4%). Similar results were obtained in other studies, Ali et al. (36) reported non-significant (0.889) higher seroprevalence (57.6%) for toxoplasmosis in individuals living in urban areas and (42.4%) in those living in rural areas. This may be due to changes in health services and cultural development that have improved rural lifestyles in recent years and brought them closer to urban dwellers (37).

Finally, contact with a cat had no significant (P = 0.097) role in parasitic transmission in either the patient (77.7%) or control (78.4%) group. This result coincides with a previous study performed in Iran, where no significant difference was observed between those who did and those who did not have contact with cats (44). These results may be attributed to the fact that many stray cats are present around the neighborhoods of participants (44). While, in a study conducted in China (45), exposure to cats had a significant effect (0.017) on the incidence of toxoplasma infection in cancer patients compared with the control group. This is because in China, keeping cats indoors is a common habit (46).

Conclusion

The results showed a high prevalence toxoplasmosis among patients with cancer. Therefore, it is recommended for patients with cancer to be screened periodically for toxoplasmosis. Cancer patients with IgG positive re-

sults are at risk of *T gondii* reactivation and patients with negative results are subjected to acute toxoplasmosis. Therefore, oncologists should be aware of this opportunistic parasite in patients with cancer. At the same time, raising awareness among patients with cancer on how to avoid exposure to risk factors can reduce infection with this opportunistic parasite. To better explore the prevalence of toxoplasmosis and its risk factors in patients with cancer and for an effective strategy for the control and prevention of toxoplasmosis, future studies with larger populations are highly recommended.

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Conflicts of Interest

The authors declare that there are no conflicts of interest regarding the publication of this article.

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