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Preliminary study on association between toxoplasmosis and breast cancer in Iran

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PEER REVIEW

Peer reviewer

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Comments

This cross-sectional study actually investigated and compared the seroprevalence and serointensity of anti-*T. gondii* antibodies (IgG and IgM) in three study arms; and showed higher rates of seropositivity and serointensity in breast cancer patients than in healthy controls, and reported no association between human *T. gondii* infection and breast cancer. Such a study would provide insight into the management of breast cancer patients.

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ABSTRACT

Objective: To investigate the possible association between *Toxoplasma gondii* (*T. gondii*) infection and breast cancer by examining the seropositivity and serointensity rate of anti-*T. gondii* antibodies in breast cancer patients and healthy volunteers.

Methods: This study was carried out on 66 women with breast cancer which consists of 29 newly diagnosed patients (Group 1) and 37 cases undergoing treatment and regular checkups (Group 2). Also, 60 healthy women (Group 3) with no history of cancer confirmed by clinical examination and imaging participated in this study. The participants were tested for *T. gondii* immunoglobulin G (IgG) and immunoglobulin M (IgM) antibodies by enzyme-linked immunoassays.

Results: The mean age of Groups 1, 2 and 3 were 43.3±6.8, 41.8±5.5 and 42.3±4.9, respectively (P=0.72). Overall, 104 (82.5%) and 8 (6.3%) out of 126 women were positive for anti-*T. gondii* IgG and IgM antibodies, respectively. Higher seropositivity rate of anti-*T. gondii* antibodies (IgG) was seen in breast cancer patients (86.4%) compared with control group (78.3%) (P=0.24). IgG antibodies were detected in 89.2% of cancer patients under treatment, 82.7% of newly diagnosed patients (P=0.18). IgM antibodies were found in 3 (10.3%), 2 (5.4%) and 3 (5%) in Groups 1, 2 and 3. No significant difference was found between the mean titers of *T. gondii* IgG antibody among these groups (P=0.87).

Conclusions: This study did not find any significant association between toxoplasmosis and breast cancer besides higher rates of seropositivity and serointensity in patients compared with healthy volunteers.

KEYWORDS

Breast cancer, Malignancy, Toxoplasmosis, *Toxoplasma gondii*

1. Introduction

Toxoplasmosis is the most common parasitic disease in the world. The most common form of the disease in humans is asymptomatic but it may be fatal in immunocompromised patients and congenitally infected fetuses and newborns[1]. Recent data showed that latent toxoplasmosis is not only hazardous for human, but also may play various roles in the etiology of several

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diseases such as mental disorders[1-3]. Moreover, reactivation of the parasite or disseminated acute infection may occur in patients with acquired immunodeficiency virus, in organ transplant recipients, and in patients with malignancy such as leukemia, lymphoma, and solid tumors[4-10]. Generally, these patients have been treated with corticosteroids and cytotoxic agents which reduced immune system response and lead to reactivation of the dormant parasite[11]. On the other hand, *Toxoplasma gondii* (*T. gondii*) has been shown to be able to activate cellular immunity in the animals[12]. Recently, several studies have demonstrated that parasitic or bacterial infections possess antitumor activities against certain types of cancers[13,14]. For instance, a study showed that injection of *T. gondii* and *Toxocara canis* antigens is associated with a significant reduction in the tumor size in comparison with the control group[13]. While the results obtained from the other studies strongly indicated that *T. gondii* potentially increases the risk of brain cancer in humans or in adult patients with brain cancer aged 55 years or older. Also, the regional mortality rates have positively correlated with the local seroprevalence of *T. gondii,* which suggested that *T. gondii* should be investigated further as a possible oncogenic pathogen to humans[15,16].

Breast cancer is the most common cancer in women throughout the world. In 2008, 22.9% of all new cancers were related to breast cancer. In spite of good prognosis of this malignancy, it was also the most common cause of death related to cancer. Based on the annual report of Health Deputy of Ministry of Health and Medical Education in Iran, breast cancer in Mazandaran Province is the most common cancer of women[17]. On the other hand, *T. gondii* infection has been reported very high in this region and most women get infection before 25 years old[18,19]. The role of *T. gondii* to cause severe symptoms in immunocompromised cases has been studied widely, but toxoplasmosis in immunocompromised patients with cancer has not been considered well. The current study aimed to investigate the association between *T. gondii* infection and breast cancer by testing the seropositivity rate of anti-*T. gondii* antibodies [immunoglobulin G (IgG) and immunoglobulin M (IgM)] in breast cancer patients and healthy volunteers.

2. Methods and materials

2.1. Population study

This cross sectional study was performed on women with breast cancer that are between 20 to 50 years old, referring to Ayatolah Rohani Hospital and Shaheed Rajaee Hospital from January 21st, 2012 to January 21st, 2013. The participants of this study were divided into three groups. Group 1 consists of 29 patients with breast cancer recently diagnosed. Group 2 consists of 37 patients with breast cancer undergoing radiotherapy or chemotherapy and under regular checkups. Group 3 consists of 60 healthy women with no history of cancer confirmed by clinical examination and imaging. In addition to demographic data, the outcome of disease, stage of cancer, period of chemotherapy were recorded.

2.2. Sample collection

A total of 2 mL blood samples were taken from each participant. The collected blood samples were centrifuged at 2000 r/min for 5 min to gain serum. The serum was separated and then stored at -20 °C until required. After sample collection, the frozen sera were thawed at room temperature and tested for existence of anti-*T. gondii* antibodies, IgG and IgM, using a commercial ELISA kits (Genesis *Toxoplasma* IgG, and Genesis *Toxoplasma* IgM, UK). Based on the manufacturer's instruction, the optical density of IgG and IgM antibody titers were read at 450 nm using automatic ELISA reader. The sera IgG titer <8 IU/mL was considered negative for anti-*T. gondii* IgG antibodies and >8 IU/mL positive. Additionally, the sera that were IgG positive were further divided into two groups: low positive sera (IgG titer between 8-50 IU/mL) and high positive sera (IgG titer >50 IU/mL).

2.3. Statistical analysis and ethic consideration

The data were processed and analyzed by SPSS version 18.0. One-way ANOVA, Fisher's exact test and *Chi*-square tests were used to analyze the data at 95% confidence level and a *P* value less than 0.05 was considered statistically significant. This study was approved by the Ethic Committee of the Research Council of Babol University of Medical Sciences, Babol, Iran. The purpose of the study was explained in details to all the participants, and all the contributors consent to share.

3. Results

In total, 126 women participated in this study, including 29, 37 and 60 cases in Groups 1, 2 and 3, respectively. The mean age and standard deviation of women in Groups 1, 2 and 3 were 43.3±6.8, 41.8±5.5 and 42.3±4.9, respectively. There were no statistically significant differences between the mean age of patients and healthy cases (P=0.72, F=0.328, df=123). All participants lived in urban areas. Overall, 104 (82.5%) and 8 (6.3%) women were positive for anti-*T. gondii* IgG and IgM antibodies, respectively. Anti-*T. gondii* IgG and IgM antibodies detected in cancer patients and controls were presented in Table 1. The results showed that higher seropositivity rate of anti-*T. gondii* antibodies (IgG) was 86.4% in breast cancer patients compared with the controls (78.3%). The difference of IgM seropositivity rate among studied groups was not statistically significant (P=0.24).

Furthermore, prevalence rate of anti-*T. gondii* antibodies (IgG) in patients under treatment and regular checkups (89.2%) was higher than newly diagnosed patients (82.7%), but this difference was not statistically significant (P=0.18).

Five out of 66 (7.6%) breast cancer patients and three out of 60 (5.0%) healthy women were positive for anti-*T. gondii* IgM antibodies (Table 1). Among patients group, three out of 29 (10.3%) newly diagnosed patients and two out of 37 (5.4%) under treatment cases were positive for IgM. These cases were positive for IgG antibodies, simultaneously. No statistically significant difference of IgM seropositivity rate was found among the studied groups (P=0.60). Moreover, any clinical manifestations of toxoplasmosis were not observed in all studied groups.

Table 1
The distribution of seroprevalence of *T. gondii* IgG and IgM antibodies in the cancer patients and control groups.

Antibodies	Patients group (n=66)	Control group (n=60)	P value
IgG(+)	57 (86.4%)	47 (78.3%)	0.24
IgG(-)	9 (13.6%)	13 (21.7%)	
IgM(+)	5 (7.6%)	3 (5.0%)	0.60
IgM(-)	61 (92.4%)	57 (95.0%)	

P value is obtained by *Chi*-square and Fisher's exact test.

Mean titers of anti-*T. gondii* IgG antibody in Groups 1, 2 and 3 were 27.6 IU/mL, 28.2 IU/mL and 25.4 IU/mL, respectively. No statistically significant difference of mean titers was seen among

the studied populations (F=0.139, df=125, P=0.87). Furthermore, serointensity rate of anti-*T. gondii* IgG antibody (>50 IU/mL) was higher in cancer patients 15.8% (9 out of 57) compared with healthy women 6.4% (3 out of 47) (P=0.11) (Table 2). Relationship between serointensity rate of anti-*T. gondii* IgG antibody and age were also shown in Table 2.

Table 2
Relationship between serointensity rates of anti-*T. gondii* antibodies (IgG) and breast cancer and age.

Variables		Number	Serointensity rate		*P* value
			High *n* (%)	Low *n* (%)	
Groups	Patients	57	9 (15.8)	48 (84.2)	0.11
	Control	47	3 (6.4)	44 (93.6)	
Age groups	>43	49	3 (6.1)	46 (93.9)	0.09
	<43	55	9 (16.4)	49 (83.6)	

P value is obtained by *Chi*-square and Fisher's exact test.

Furthermore, cancer metastasized to liver and bone in three patients. All these three patients were positive for anti- *T. gondii* IgG antibody. The IgG titers ranged from 12 to 16.6 IU/mL indicating that the infection had occurred long time ago.

4. Discussion

Toxoplasma infection in association with different malignancy has been described by several studies[7,10,20,21]. An early study showed that 93.7% women with genital tract and breast cancer were positive for anti-*T. gondii* antibodies and 20.46% of them had antibody titers at 1:400 or higher[22]. A recent study reported higher seroprevalence rate of toxoplasmosis in breast cancer patients following leukemia and lymphoma[7]. The current work is the first preliminary study in Iran that investigated the seroprevalence rate of *T. gondii* in breast cancer patients in order to find out any possible association between this infection and breast cancer. The findings demonstrated that the positivity rate for *T. gondii* IgG antibodies was higher in patients (86.4%) compared with the healthy women (78.3%). It also showed that seroprevalence rate of toxoplasmosis was higher in patients who were under treatment and regular checkups than newly diagnosed patients. But statistically significant differences were not observed. These findings suggest that not only *Toxoplasma* infection but also treatment did not have significant relationship between breast cancer and seropositivity rates of toxoplasmosis. With excepting the kind of cancer, these findings are in agreement with other studies which reported that seroprevalence rate of toxoplasmosis in cancer patients was higher than that in healthy individuals[11,23]. However, these results are in contrast with other reports which found significant relationship between *Toxoplasma* infection and cancer. This significant association was particularly seen between *T. gondii* infection and brain cancer[10,15,16,24,25].

On the other hand, the positivity rates of anti-*T. gondii* IgG in breast cancer patients (86.4%) and healthy volunteers (78.3%) were much higher than that reported by other researchers[7,10,11]. These findings could be due to many reasons such as the geographical variation, customs and habits, difference in genetic susceptibility and the possible risk factors contributing to the acquisition of *Toxoplasma* infection[23]. Indeed, several seroepidemiological studies have reported that there is higher prevalence rate of *Toxoplasma* infection in Northern Iran where the current study was performed[19,26]. In addition, age is an important associated factor in the epidemiology of *Toxoplasma* infection which made limitation in the current work. Many studies demonstrated that the seroprevalence rate of toxoplasmosis increases with age and the peak level was seen in cases older than 50 years[26,27]. Also, the incidence of breast cancer increases rapidly with age during the reproductive years and then increases at a slower rate after about age of 50 years[28]. However, this study showed that breast cancer is more prevalent (69.7%) in women who are more than 40 years of age. According to *T. gondii* IgM antibodies results, there was not statistically significant difference between the breast cancer patients and healthy women, which was consistent with the other reported data[10,23].

In addition, the high serointensity rate of anti-*T. gondii* IgG antibody was not statistically higher in cancer patients (15.8%) compared with healthy women (6.4%) (P<0.11). Although, *Toxoplasma* has been considered as an opportunistic infection, we did not find significant association between *Toxoplasma* infection and breast cancer. Age is a critical factor for both breast cancer and toxoplasmosis which are more prevalent in women aged over 40 years. Indeed, *Toxoplasma* infection occurred in people at lower age in our region where the study preformed[26,27]. Therefore, we did not find any significant association between toxoplasmosis and breast cancer as the mean age of the studied population was 42.3 years. Another possible reason is small sample size.

It was also not statistically higher in cases older than 43 years (16.4%) compared with individuals younger than 43 years (6.1%) (P<0.09). This finding is in line with other study demonstrating that the seroprevalence rate of toxoplasmosis increase with age[26].

In conclusion, the current study did not find any significant association between toxoplasmosis and breast cancer besides the higher rates of seropositivity and serointensity in patients compared with healthy volunteers. Furthermore, no clinical manifestations of toxoplasmosis were seen in the studied groups. This work also suggested that further studies should be performed to establish any association between *T. gondii* and breast cancer.

Conflict of interest statement

We declare that we have no conflict of interest.

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Comments

Background

Toxoplasmosis, a global parasitic disease, is usually asymptomatic in healthy subjects, but is fatal in immunocompromised patients, including HIV patients, cancer patients, organ transplantation recipients, *etc*. This study examined the possible correlation between *T. gondii* infection and risk of breast cancer.

Research frontiers

This study detected seroprevalence and serointensity of anti-*T. gondii* antibodies IgG and IgM in recently diagnosed breast cancer patients, breast cancer patients undergoing radiotherapy or chemotherapy, and healthy controls. The results showed higher rates of seropositivity and serointensity in breast cancer patients than in healthy controls, and reported no association between human *T. gondii* infection and breast cancer.

Related reports

The seroprevalence and serointensity of anti-*T. gondii* antibodies IgG and IgM have been extensively investigated in cancer patients.

Applications

Based on the results from this study, it is concluded that the seroprevalence and serointensity of anti-*T. gondii* antibodies IgG are higher in breast cancer patients than in healthy individuals.

Peer review

This cross-sectional study actually investigated and compared the seroprevalence and serointensity of anti-*T. gondii* antibodies (IgG and IgM) in three study groups, and showed higher rates of seropositivity and serointensity in breast cancer patients than in healthy controls, and reported no association between human *T. gondii* infection and breast cancer. Such a study would provide insight into the management of breast cancer patients.

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