

# Seroprevalence, Histologic Grades and Stages in Breast Cancer Patients with Toxoplasmosis in Thi-Qar Province/ Southern Iraq

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**Abstract:** Toxoplasmosis is one of the most common diseases in the world. It is found all over the world and affects a considerable proportion of people. It is known for being asymptomatic and affects all warm-blooded animals, including humans. *T. gondii* parasite is responsible for the disease. Patients with weakened immune systems, such as those with AIDS or cancer, are most at risk for infection. The study's main goal is to detect *T. gondii* infection in BC patients using the serological method (ELISA). This study involves measuring the *T.gondi* (IgG) levels of 150 patients with breast cancer whom entered oncology center of Al-Haboubi Teaching Hospital in Nasiriya City (southern Iraq ), and 54 healthy controls. The results demonstrated that 47.3% of breast cancer patients were infected with *T. gondii* compared with the control group (33.3%). The age group with the highest infection rate was (41–50) years old among BC patients infected with *T.gondii* (38%), while the age range between 71–80 years old had the lowest infection rate (4%). Additionally, there was a higher rate of toxoplasmosis among patients with breast cancer whom were lived in urban (57.75%) compared with those living in rural. The results showed a high rate of breast cancer patients with toxoplasmosis (59.15%) in grade 2 compared with other grades. Stage III of breast cancer showed a high rate of infection with toxoplasmosis (66.19%) compared with other stages.

**Keywords:** Toxoplasmosis, breast cancer, serology, histological grade.

**Introduction:** *T.gondii* is a ubiquitous intracellular parasite that is commonly found in a wide variety of warm-blooded species, including humans. Its common hosts are found in different parts of the world (1). The main human infection route is predominantly arises from the ingestion of water or foodstuffs that have been contaminated with oocysts expelled by felids, or by the consumption of inadequately cooked or raw meat that contains tissue cysts of *T. gondii* (1). Congenital transmission, which occurs when women have the primary infection during pregnancy, may lead to dangerous adverse consequences like stillbirth, miscarriage, or severe damage to the fetus (3).

In healthy people, the infection is typically self-limited and doesn't cause any symptoms (asymptomatic) (4). People with weakened immune systems, such as cancer patients, organ

transplant recipients, and people living with HIV/AIDS, are at high risk of developing toxoplasmosis, which may cause fatal results (5, 6).

Moreover, the reactivation of latent infection can lead to severe illness in individuals with weakened immune systems (immunocompromised patients), which induces encephalitis or disseminated infection (7, 8). In light of this, occurrences of latent toxoplasmosis reactivation have been documented in patients with various kinds of cancer (9, 10). Breast cancer is one of the most common diseases among women worldwide, with an anticipated 2.3 million new cases in 2020 (2).

### **Material and method:**

**1. Collection of blood sample :** Blood samples were taken from 150 breast cancer patients who visited oncology center of Al-Haboubi Teaching Hospital in Nasiriya City (southern of Iraq ), between July to September 2023. An additional 54 patients were included in a control group. The samples were collected and allowed to remain at room temperature for 30 minutes in sterile gel tubes, then centrifuged for five minutes at 3000 rpm to isolate the serum. The samples were placed in an eppendorf tube and kept at -20 °C until required (11).

**2. Detection of *T.gondii* (IgG):** The *Anti-T.gondii* (IgG) antibodies were detected using ELISA technique, which was performed in accordance with the manufacturer's instructions using a commercial ELISA kit (CAMP, Romania). Cut-off values were used for establishing the diagnostic threshold for IgG antibodies: values less than one indicated a negative result (<1), while values more than one indicated a positive result ( $\geq 1$ ).

**3. Histological grades and stages:** Histological grades and stages were studied by following up on patients data.

**Statistical Analysis:** Statistical analyses were conducted using the Statistical Package for the Social Sciences (SPSS, version 27, IBM). Categorical variables were compared using the Chi-square test at  $P \leq 0.05$ .

### **Results :**

The results showed that 47.3% of the breast cancer patients were infected with *T.gondii* (IgG),

and 52.7 without *T.gondii* (IgG), based on the serological analysis with the ELISA test , reporting a significant differences (  $\chi^2 = 4.08$  ,  $P \leq 0.05$  ) As listed in Tables (1).

**Table (1): *T.gondii* IgG antibodies among patients with breast cancer.**

Case	Toxo Igg	No. (%)	Mean ± Sd
Patient	Positive	71(47.3%)	2.783 ± 0.02
	Negative	79 (52.7%)	0.655 ± 0.01
Control	Positive	18 (33.3%)	3.013 ± 0.05
	Negative	36(66.7%)	0.424 ± 0.02
<b>Total: Patients (150)</b>		<b>Total: Control (54)</b>	

$\chi^2 = 4.08$                       DF = 1                      P-value = 0.043

S: significant difference  $P \leq 0.05$

The results showed that patients with breast cancer who were aged between (41 and 50) had the greatest risk of having a toxoplasmosis infection (38%), followed by patients who were aged between (51 and 60) , while patients who were aged between (21-30) had the lowest infection rates (3%). The findings revealed a significant variation (  $\chi^2 = 0000$  ,  $P \leq 0.05$  ) in age between patients with breast cancer and *T. gondii* infections as listed in table (2) .

**Table (4-6): Age of breast cancer with *T.gondii* (IgG)**

Age(Years)	No. Patients With <i>T.Gondii</i> (Lgg) (%)
21-30	4 ( 6% )
31-40	9 (13%)
41-50	27 (38%)
51-60	20 (28%)
61-70	8 (11%)
71-80	3 (4%)
<b>Total</b>	<b>71</b>

$\chi^2 = 54.20$       df = 5      P-value = 0.000

S: significant difference  $P \leq 0.05$

Geographic distribution for breast cancer patient infected with *T.gondii* (IgG) were not revealed a significant differences (  $\chi^2 = 2.56$  ,  $P > 0.05$  ) when the study attended the relationship between the infection with *T.gondii* (IgG) and the residency of breast cancer patient , as explained in the

following table :

**Table (3): Residency of breast cancer patients infected with *T.gondii* (IgG) .**

Residency	No. Patients (%)
<b>Urban</b>	<b>41 ( 57.75)</b>
<b>Rural</b>	<b>30 (42.25)</b>
<b>Total</b>	<b>71</b>

$\chi^2 = 2.56$                       df = 1                      P-value = 0.110

**N.S:** Non-Significant differences P>0.05

There were a significant difference (  $\chi^2 = 40.58$  ,  $P \leq 0.05$  ) based on the histologic grading of breast cancer patients with *T.gondii* (IgG) where The result showed a high rate of breast cancer patients with toxoplasmosis (59.15%) in grade 2 compared with grade 1 and grade 3, which have (7.04%) and (33.8%), respectively, as shown in table (4).

**Table (4) :Histologic grading of breast cancer patients with *T.gondii* (IgG) .**

Grades	No. Patients With <i>T.Gondi</i> (Lgg) (%)
<b>Grade 1: Low Grade Or Well-Differentiated</b>	<b>5 (7.04%)</b>
<b>Grade 2: Moderate Grade Or Moderately Differentiated</b>	<b>42 (59.15 %)</b>
<b>Grade 3: High Grade Or Poorly Differentiated</b>	<b>24 (33.8 %)</b>
<b>Total</b>	<b>71</b>

$\chi^2 = 40.58$               df = 2                      P-value = 0.000

**S:** significant difference  $P \leq 0.05$

The result of the current study shows a high rate of breast cancer patients infected with toxoplasmosis (66.19%) in Stage III compared with other stages (8.45% , 14.08% and 11.26% respectively ) , indicating a significant difference (  $\chi^2 = 90.64$  ,  $P \leq 0.05$  ) based on the staging of breast cancer patients with *T.gondii* (IgG) as listed in tables (5) .

**Table (5): Stages of Breast Cancer Patients with *T.gondii* (IgG).**

Stages	No. Patients with <i>T.Gondii</i> (Lgg) (%)
<b>Stage I</b>	<b>6 (8.45 %)</b>
<b>Stage II</b>	<b>10 (14.08 %)</b>
<b>Stage III</b>	<b>47 (66.19%)</b>
<b>Stage IV</b>	<b>8 (11.26 %)</b>
<b>Total</b>	<b>71</b>

$\chi^2 = 90.64$                       DF = 3                      P-value = 0.000

**S:** Significant Difference  $P \leq 0.05$

**Discussion:** *T.gondii* is an obligatory intracellular protozoan parasite that may infect warm-blooded animals, including humans, and appears to have a broad range of hosts (12). All non-feline vertebrate species act as intermediate hosts, whereas the cat family acts as both intermediate and definitive hosts (13). Breast cancer (BC) is a kind of cancer that develops when breast cells grow out of control and proliferate to create a lump or tumor (14).

Cancer patients are susceptible to infection with a variety of infections, including *T. gondii* (15). A meta-analysis's conclusion shows that the risk of toxoplasmosis is three times higher in cancer patients than in healthy people (4). Toxoplasmosis is an opportunistic infection that can occur in patients with immunocompromised circumstances. Therefore, the immune-suppressive effects of radiotherapy and hormone therapy may raise the chance of contracting toxoplasmosis. The immunomodulatory effects of radiotherapy and hormone therapy are well documented (16).

The present study showed that the rate of *T.gondii* (IgG) was 47.3% among breast cancer patients compared with the control group (33.3%) due to a persistent latent infection that was previously acquired, it is related to elevated IgG levels (17) .

The present study was comparable to a previous study in the Thi-Qar Province of Iraq to identify immunoglobulins (IgG) specific for *T. gondii*, and the results showed that the rate of *T. gondii* (IgG) was 36% among breast cancer patients (18). The present study was supported by another study that was accomplished at Beni-Suef Hospital in Egypt, and it was found that the rate of *T. gondii* infection was 47.1% among breast cancer patients (19). Recently, a study was conducted in Iran, and the findings revealed the rate of *T. gondii* (IgG) was 45.8% among breast cancer patients (20).

The current study was incompatible with other study in Nasiriyah city, Iraq that used ELISA to detect IgG levels in serum to determine the prevalence of *T. gondii* infection in breast cancer patients through the examination of 150 blood specimens of patients with cancer. The result revealed IgG concentrations were (87.5%), according to ELISA test findings (21).

The prevalence of toxoplasmosis differs because of many factors, including regional variance, the variation in climate, habits, and cultural practices in different regions of Iraq, differences in genetic susceptibility, and how toxoplasmosis is acquired (22).

The current study's findings indicate that the age range of 41 to 50 years old is the most

commonly affected by *T. gondii* infection. This is because cancer patients have weakened immune systems, which is the main reason why toxoplasma infections are becoming more common among cancer patients (23).

The results of another study that was carried out in Basrah Province, Iraq, to determine the prevalence of *T. gondii* infection was revealed a high incidence of *T. gondii* infection among patients in the 41–50 age group (24). Furthermore, the results of a previous study that examined 90 cancer patient samples from Baghdad, Iraq, in order to determine the prevalence of toxoplasmosis infection among cancer patients and its findings demonstrated that among breast cancer patients, the age range of 31 to 40 years old had the highest *T. gondii* infection, which is inconsistent with the present study (25).

The current study found that among breast cancer patients with toxoplasmosis, the residency rate was higher in urban areas (57.75%) than in rural areas (42.25%). It's possible that cultural barriers and socioeconomic factors keep women from obtaining treatment when they need it, which raises the risk of advanced breast cancer. Also Increased infection rates may result from the parasite *T. gondii* being more common in the environment (26).

This finding agrees with the findings of a Saudi Arabian study that found that cancer patients who resided in urban regions had a greater prevalence of toxoplasmosis infection (73.8%) than those who lived in rural areas (27). Another study was done on cancer patients in Egypt to estimate the prevalence of *T. gondii* infection. The result revealed a high rate of infection with *T. gondii* inside cities (53%), compared with those who lived outside of cities (46.9%). This is similar to the present study (19).

The present results showed that were a high incidence of *T. gondii* in grade II among breast cancer patients (59.15%) compared with grade I and grade III (7.04%) (33.8%), respectively. Due to the tumor burden and systemic effects of cancer, patients with advanced breast cancer (such as those in stage III) may have a weakened immune response. Breast cancer patients may be more susceptible to opportunistic diseases like toxoplasmosis (18).

This result was in agreement with a previous study in Thi-Qar, Iraq (2020), which showed a high incidence of *T.gondii* in grade II breast cancer (42.4%) compared with other grades (28). Recently, a study was conducted in Baghdad, Iraq, to estimate the prevalence of *T. gondii* infection among breast cancer .Findings revealed the IgG concentrations for *T. gondi* in grade III breast cancer were highest compared with other groups (25).

The current study's findings demonstrate a substantial variation in the prevalence of toxoplasmosis among women with breast cancer in pathologic stages, with a higher percentage (66.19%) in stage III when compared to other stages. It's possible that cultural barriers and socioeconomic factors keep women from obtaining treatment when they need it, which raises the risk of advanced illness. Increased infection rates may result from the parasite *T.gondii* being more common in the environment (26).

A previous study in Iran supports this current study, as a study was conducted in Iran on breast cancer patients infected with toxoplasmosis, and a high percentage of patients with breast cancer and toxoplasmosis were in stage III (72.97%) compared to the other stages (20).

The results of this study are dissimilar to those of another study in Guangzhou, China, which recorded a higher infection rate of toxoplasmosis among breast cancer patients in stage II (50.6%) compared to other stages (29).

**Conclusion:** The results demonstrate a high seroprevalence of toxoplasmosis among breast cancer patients. Specifically, *T.gondii* (IgG) is more frequently detected in individuals with moderately differentiated tumors (Grade II). Additionally, *T.gondii* (IgG) positivity is more common in the advanced stages of breast cancer, particularly Stage III. These findings suggest a potential link between *T.gondii* infection and breast cancer progression, underscoring the need for additional research to explore the role of this infection in disease advancement.

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