



Microbial Science Archives (ISSN: 2583-1666)

Journal homepage: www.microbial.sciencearchives.org



Research Article

<http://dx.doi.org/10.47587/MSA.2022.2103>



Prevalence of *Toxoplasma gondii* among women and ewes in Kirkuk city

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Received: Feb 27 / Revised: Mar 16, 2022 / Accepted: Mar 21, 2022

Abstract

This study was conducted in Kirkuk city during the period from October 2020 to March 2021. The study aims to study the prevalence of *Toxoplasma gondii* infection among pregnant women and ewes. Toxoplasmosis is a common disease between humans and animals and diffused throughout the world by 30-60%, caused by a single-celled parasite called *Toxoplasma gondii*. Toxoplasmosis is a parasite disease that causes a variety of major health concerns in pregnant women. Miscarriages and congenital abnormalities in fetuses are caused by this condition. It may also affect the neurological system and the brain's important functions. In Iraq, the prevalence of the *Toxoplasma* parasite in pregnant women and sheep was measured, and the association between the pathogen and abortion was investigated. According to the findings, the condition is prevalent in 51 percent of pregnant women and 41 percent of ewes. The findings revealed a link between the frequency of abortions in pregnant women and *Toxoplasma gondii* infection, with the greatest infection rate of 38 percent for women who had three abortions, whereas the highest risk of abortion in sheep was 43 percent for one abortion. The study also found that women between the ages of 25 and 40 are the most susceptible to infection, with a 40% infection rate, while the highest risk of infection in sheep was 58 percent for the age group 1-2. In addition, the results illustrated a relationship between sheep infection with *Toxoplasma gondii* and the presence of congenital malformations in aborted fetuses. In other words, the rate of infection among sheep that aborted deformed fetuses was 89.5%. The relation between the level of the hormone's progesterone and oestrogen in infected women was studied. It was noted that the ratio of the two hormones was higher in those who were infected than in healthy women, where the proportion of progesterone was 20 mg/ml, while the proportion of estrogen was pg/ml 92.5.

Keywords *Toxoplasma gondii*, Kirkuk, Ewes, Pregnant women

Introduction

Toxoplasmosis, commonly known as feline disease, is a common contagious disease of humans and animals that is spread all over the world. The main cause of this disease is *Toxoplasma gondii*, a unicellular endoparasite, whose life cycle contains infective oval cysts (Mahfouz et al., 2019). *Toxoplasma gondii* is so named because of its curved shape in its active phase, Tachyzoite, this parasite is an obligatory parasite that lives inside the host's cells and is classified as coccidiosis (Mahfouz et al., 2019).

Toxoplasmosis is a parasitic disease that causes a variety of health problems in both humans and animals. It is particularly dangerous for women, especially pregnant women because transmission of the parasite to the fetus can result in abortion, congenital malformations, or death. It may also affect the nervous system, altering the brain's important activities. In an animal, this could lead to a miscarriage or the animal's death after birth (Lepore, 2019). Humans of the order Primates are intermediate hosts, as well as animals such as sheep of the order Carpini, rats of the order Rodents, pigs of the order Suinae (John and Petri, 2013), domestic and wild cats, and all species belong to the family Felidae. Felidae is the ultimate host of the parasite.

Toxoplasma gondii infection occurs in various ways, including ingestion of contaminated water and food loaded with cat feces, or by eating undercooked meat containing tissue cysts, or the infection may be transmitted from the pregnant mother to the fetus through the placenta. Cats play a major role in the spread of parasite infection, as infected cats continue to secrete large numbers of eggs to the external environment, which give them the ability to cause infection for a very long time when appropriate conditions are provided, as sporangia can infect both humans and animals (Aldabagh et al., 2018).

The goal of the study is to use serological diagnosis to estimate the prevalence of *Toxoplasma gondii* in pregnant women and ewes in Kirkuk city, as well as to evaluate IgG and IgM antibodies in blood samples from toxoplasmosis-infected women and ewes.

Materials and methods

Blood samples were collected from the following medical laboratories: Ibn Al-Nafis, Al-Amal, Al-Shefa', Dalida, Al-Sa'a, Al-Arrab, and Al-Awael Within the period (beginning of October 2020 - end of March 2021). The study included 136 samples of women coming to the above-mentioned places who previously had multiple abortions, as well as the control group, which included 20 samples of noninfected women.

Blood samples of aborted female ewes were collected from the Veterinary Teaching Hospital and some veterinary clinics in Kirkuk, in addition to field visits accompanied by veterinarians to villages and countryside to collect samples within the same period. In addition to the control group, which included 20 samples of healthy female ewes that were not exposed to previous abortions, to take the blood samples from the jugular vein of the ewes that aborted.

The serum was separated using a centrifuge at 3000 rpm for 10 minutes, then the serum was withdrawn with a Pasteur pipette and the serum was placed in small, sterile eppendorf tubes, then the samples were kept in freezing at -20°C, and the necessary tests were carried out.

Serological tests

Blood samples that were taken from pregnant women participating in this study were examined in Al-Razi Laboratory / Kirkuk. Serological examinations were carried out by the MINI VIDAS device, using the ELFA kit manufactured by the French bioMérieux company according to the manufacturer's recommendations.

Statistical analysis

Statistical analysis was carried out using the Statistical Package for Social Sciences (SPSS, version 20).

Results and discussion

Results of the IgG antibody test conducted on 136 women who undergone recurrent miscarriages in Kirkuk city showed that the incidence of toxoplasmosis was 51% (Table 1), as this result was compatible with the results that were recorded in the study conducted in Erbil city, with an infection rate of 54.46% (Hamad and Qadeer, 2013) and Qadisiyah city, where the infection rate in 2016 was 44% (Mikaeel and Al-Saeed, 2019), and Basra city, where infection rate was 43.07% (AL-Sadoon et al., 2018). By comparing the percentage recorded in this study with the rates recorded in different regions of Iraq recently, we find that it is higher than it is in Karbala, where the infection was 21% and Dohuk, the percentage was 28% (Mikaeel and Al-Saeed, 2019), and the percentage of infection was 17.3% in Baquba (Al-Khafaji et al., 2020). When comparing the results of the current study with the results of a previous study that was conducted on women in Kirkuk city, we find that the infection in the previous studies was lower, the previous result that recorded was 33% (Raza et al., 2019).

Returning to the reasons for the high rates of studies that it may happen as a result of the large spread of stray cats in Kirkuk city and eating uncooked meat, unwashed vegetables, and fruits, which are a major source of transmission of infection. In addition to the lack of health and cultural awareness and the lack of sufficient information on how to deal with pets such as cats and raise them inside homes and the risks of allowing them to roam and stay indoors for long periods, which increases the possibility of exposure to surfaces contaminated with their waste that may contain one of the infectious lengths of the parasite (Luci et al., 2009).

Results showed that the infection among 110 sheep with a history of repeated miscarriage was 40.9% (Table 1) among the studied group. It was found Al-Diwaniyah, where the infection rate was 40.56% among sheep (Madhuvrata et al., 2005), and in Dohuk, where the infection was 42.1% (Al Hamada et al., 2019) and in Mosul, 37.64% (Zakaria, 2013)). While this percentage was much higher than the percentage recorded in Kurdistan 8.3% (Partoandazanpoor et al. 2020), Diwaniyah, where the percentage was 16.25% (Al-Shaibani and Alwan Alsheabani 2019), and Duhok, where it was 8.1% (Al. Hamada, Habib et al. 2021). As for Najaf, its results recorded a higher percentage than those found in this study, with the infection rate reaching 62% (AL-Hatami, AL-Kardhi et al., 2018). The findings revealed that parasite infection and the number of abortions differed significantly, with the highest infection rate of 38.7% when the number of abortions was 3, and the lowest infection rate of 1.4 percent when the number of abortions was zero (Table 2).

Table 1. The IgG and IgM positive percentage for infected women and sheep

Samples	IgG+	%	P.value	IgM+	%	P.value	IgG+IgM	%
Women	70	51.5	0.005	45	33	0.0001	21	15
Sheep	45	40.9	0.002	10	9.0	0.004	3	2.7

The study found that there are substantial disparities between the number of abortions a pregnant woman has and the likelihood of infection, with the greatest rate of infection being observed for women who have had three abortions, at 38.7%. Previous studies indicated that the reason for this is the accumulation of IgG antigen in the sera of pregnant women, and it is the only IgG antigen that is transmitted from the pregnant mother to the fetus through the placenta.

The parasite crosses the placenta for fetuses and thus increases the miscarriage rate in pregnant women and from congenital malformations in fetuses due to the incomplete immune system of the fetuses.

These results agreed with the results obtained in a previous study conducted in the governorates of Dhi Qar and Kirkuk, where the percentage of women who had abortions was two to three times higher than those who had one abortion. While other studies showed that there are no significant differences between the incidence of infection and the number of abortions for pregnant women (Al-Adlaan, 2007).

Table 2. The incidence of women infection and its relationship to the number of abortions

Abortion No.	+ Ve	%	- Ve	%
0	1	1.4	18	27.2
1	6	8.6	24	36.3
2	25	35.7	14	21.2
3	27	38.6	9	13.6
3 <	11	15.7	4	1.5
Sum	70	100	66	100
X²: 41.535		P.value: 0.0001		

The findings revealed strong correlations between toxoplasmosis infection and the ages of pregnant women, with the greatest infection rates in age groups 30-34 (40%) and 25-29 (37%) respectively (Table 3).

The findings revealed a link between toxoplasmosis infection and the ages of women who had multiple miscarriages, as the incidence of the disease increases with age, with the greatest rates documented for the age groups of 25 to 29 and 30 to 34. In general, 36.3 percent and 40.2 percent, respectively, were the percentages. These findings are consistent with those of numerous other studies, as research conducted in various countries revealed that the incidence of the disease and recurrent miscarriages rises in the age range of 25 to 40 years

(Saleh, 2011). This percentage, on the other hand, contradicts the findings of previous investigations. In research conducted in Tikrit, the age group of 16-25 years had the highest rate of infection (Ahmed, 2008), whereas, in Baghdad, the age group of 15-24 years had the highest incidence of infection (Madhuvrata et al., 2005; Al-Shikhly, 2010).

Table 3. The incidence of women with toxoplasmosis and its relationship to the age groups

Age group	+ IgG	%	- IgG	%
24 - 20	10	12.9	20	33.8
29 - 25	28	36.3	30	50.8
34 - 30	31	40.2	7	11.8
40 - 35	8	10.3	2	3.3
Sum	77	100	59	100
X²: 19.1		P.value: 0.0001		

The current study's findings revealed that there is a link between the rate of Toxoplasmosis infection in sheep and the age of the ewes, with the rate of infection decreasing as the age of the ewes increases, as the percentage of infection in ewes under two years reached 76.3 percent, while the percentage of infection in ewes over two years was 21.0 percent (Table 4).

Previous research has indicated that this is related to the fact that young ewes have lower disease resistance and are more exposed to the Toxoplasma parasite's oocyst stage, which increases infection rates (Mikail and Al-Barawy, 2014). These findings are consistent with those of earlier studies, which have shown that the risk of Toxoplasmosis infection in sheep reduces as the age of the sheep increases. Furthermore, one study found that sheep infected with Toxoplasmosis develop long-term immunity (Lashari and Tasawar, 2010).

Table 4. The percentage of infected ewes with Toxoplasma according to the age groups.

Age	-Ve	%	+Ve	%
1<-2	27	51.9	29	76.3
≥ 2	25	48.0	8	21
Sum	52	100	38	100
X²: 12.43		P-Value: 0.002		

The findings revealed that infection rates and the frequency of abortion among infected ewes differed significantly, with the highest incidence of infection occurring in ewes that had previously miscarried only once, and the percentage of infection decreasing as the frequency of abortion increased (Table 5). These findings corroborated the findings of numerous prior research, which found that infection rates in ewes who had multiple abortions were lower than in ewes who had only one abortion. In a study conducted by Mikaeel and Al-Saeed (Mikaeel and Al-Saeed, 2020), the rate of infection for ewes that aborted only once was greater than other percentages, and these findings corresponded with those obtained by Mikaeel and Al-Saeed (Mikaeel and Al-Saeed, 2020).

Table 5. The incidence of ewes with toxoplasmosis and its relationship to the number of abortions.

No. of abortions	-Ve	%	+Ve	%
1	3	13.6	43	63.2
2	6	27.4	12	17.6
3≤	13	59	13	19.1
Sum	22	100	68	100
X ² : 52.983		P-Value:0.001		

The current study's findings revealed that there were significant differences between parasite infection and the presence of abnormalities in ewes' aborted fetuses, with the highest infection rate of 89.5 percent in ewes who aborted deformed fetuses and the lowest infection rate of 10.5 percent in sheep who aborted healthy fetuses. The findings revealed that there is a link between the occurrence of anomalies and their severity. In the aborted ewes fetuses infected with Toxoplasmosis infection, the percentage of infected sheep that had previously aborted deformed fetuses was 89.5% compared to 10.5% of those that aborted healthy fetuses. As we mentioned earlier, the reason for this is the parasite's ability to pass from the pregnant mother to the fetus through the placenta (Table 6).

Table 6. The incidence of sheep with toxoplasmosis and its relationship to the presence of malformations in aborted fetuses

Malformations	-Ve	%	+Ve	%
Yes	2	89.5	34	3.8
No	50	10.5	4	96.1
Sum	52	100	38	100
X ² : 64.436		P-Value:0.002		

Conclusions

The results of this study show a high prevalence of Toxoplasmosis infections in Kirkuk city among pregnant women and ewes compared to the control group, where the percentage of infected women was 51.5%, while the percentage of infected ewes was 40.4%. moreover, the percentage of IgG antibodies that is recorded 51% compared to 33% for IgM antibodies, indicates that the rate of chronic infection in Kirkuk city is the highest. In addition to that, a strong relationship is reported between the ages of pregnant women and infection rate, where the highest rates of infection were for women between 30 and 34 years, with 31%, followed by women between 28%. It also proved that there is a relationship between ewes infection rates and their age, where the highest rate of infection for ewes that are less than one to two years was recorded at 29%. The results show that toxoplasmosis has a significant role in ritual abortion of pregnant women, where the highest incidence of toxoplasmosis was in women who had previously miscarried 3 times, and there is also a relationship between the number of abortions and toxoplasmosis infection. Furthermore, it is proved that there is a significant role for Toxoplasmosis infection in ewes with causing abnormalities in aborted fetuses, as the percentage of infection among ewes that previously aborted distorted fetuses was 34%.

Conflict of Interest

The author hereby declares no conflict of interest.

Consent for publication

The author declares that the work has consent for publication.

Funding support

The author declares that they have no funding support for this study.

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How to cite this article

Ahmad, H.N.M., Abdullah, A.H., Ali, S.A. (2022). Prevalence of *Toxoplasma gondii* among women and ewes in Kirkuk city. *Microbial Science Archives*, Vol. 2 (1), 9-13. <http://dx.doi.org/10.47587/MSA.2022.2103>

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