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Serological detection of *Toxoplasma gondii* in some intermediate hosts (sheep and goats) in Thi-Qar province, Southern Iraq

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Abstract:

Iraq considers sheep and goats a significant source of meat in daily meals and during some holly celebrations. These animals are suitable reservoir hosts of *Toxoplasma gondii* and play a critical role in transmitting the parasite to humans. Thus, this study investigated the prevalence of *Toxoplasma gondii* infection in sheep and goats in Thi-Qar province in southern Iraq. In the study conducted from October 2020 to February 2021, two hundred eighty goat and sheep blood samples were collected from Nasiriyah and Shatrah slaughter house. Antibodies type IgG against *Toxoplasma gondii* were investigated in serum using the Enzyme-Linked Immunosorbent Assay (ELISA). Furthermore, 134 (47.85%) of the 280 animals showed signs of toxoplasmosis. The infection rate of *Toxoplasma* in goats and sheep was 50.69% and 44.85%, respectively. Prevalence of the antibodies for *toxoplasma gondii* type IgG was found to be variant in animals regarding animals' ages and sex. The current study showed that animals with periods of more than two years and females significantly have a higher infection rate compared to males younger than two years.

Keywords: Toxoplasmosis, goat and sheep, IgG, Thi-Qar province.

Introduction:

An obligatory intracellular protozoan parasite called *Toxoplasma gondii* causes serious illness in humans, small ruminants, other warm-blooded mammals, and birds [1]. Between 15 and 85% of the human population has an asymptomatic infection from the parasite, which is widespread in people and animals [2]. All animal species act as intermediate hosts, except feline species, which serve as definitive and intermediate hosts [3]. This parasite is a single-celled member of the animal phylum protozoa that may parasitize the cells of another host creature reliant on the host's food supply [4].

Toxoplasma gondii has three infectious stages that can infect humans and animals. The first stage is the sporozoite, found in the oocyst. The second stage is the tachyzoites, characterized by the ability to divide rapidly during acute infection. The last stage is the bradyzoite, characterized by slow dividing during chronic illness[5]. Infection of *Toxoplasma gondii* can be transmitted by eating undercooked meat and

drinking contaminated water, food, and soil. Furthermore, vertically from the mother to the baby during pregnancy. Only a small number of people have clinical ailments [6, 7].

Since the *Toxoplasma gondii* cyst stage can be found in some animals' skeletal and cardiac muscles, these animals are considered intermediate hosts like sheep and goats [8]. *Toxoplasma gondii* is also a significant contributor to reproductive failure in goats, sheep, and pigs [6, 8].

Many risk factors participate in the transmission of the parasite causing infection, including consuming raw or undercooked meat [9-11]. Many studies have shown the prevalence of *Toxoplasma gondii* in humans in Thi-Qar province. However, there was no study investigating the infection rate of this parasite among intermediate hosts like goats and sheep, which are two important sources of meat in the region. As a result, the study aim was to detect antibodies against *Toxoplasma gondii* in sheep and goats in the province of Thi-Qar.

Material and methods:

Sample collection:

The current study aimed to assist the seroprevalence of *Toxoplasma gondii* in some intermediate hosts, which involved goats and sheep in Thi- Qar province, southern Iraq. The samples were collected from October 2020 to February 2021 from Al-Nasiriyah, and Al-Shatrah slaughter houses. Two hundred eighty blood samples have been collected from goats and sheep, and all required information for each sample has been collected. Five milliliters of blood were collected directly from the heart chambers after slaughtering. Blood samples were placed in a sterilized plain tube and left to stand for 30 minutes at room temperature; then, all samples were centrifuged at (3000 rpm) for 10 minutes to collect serum, which was dispensed in Eppendorf – tubes by using a micropipette. Samples were stored at -20°C until used for Enzyme-linked immunosorbent assay (ELISA) to detect anti-*Toxoplasma gondii* IgG antibodies.

Serological investigation

According to the manufacturer's instructions, the enzyme-linked immunosorbent assay (ELISA) kit from IDvet (310, rue Louis Pasteur- Grabels, France) was used to detect anti-*Toxoplasma gondii* IgG antibodies in serum samples.

Statistics analysis:

SPSS software V 26 has been used for statistical analysis. Chi-square was used as a statistical test to assess the significant difference between study groups. The statistical correlation is considered significant when the P value is less than 0.05 and insignificant when the P value is more than 0.05.

Results:

1-Prevalence of *Toxoplasma gondii* in Sheep and Goats in Thi –Qar province.

This study included 280 animals (goats and sheep). Samples were investigated for anti-*Toxoplasma gondii* IgG antibodies using specific tests ELISA IgG. Data show 134 (47.85%) of the 280 animals have *toxoplasma gondii* infection. On the other hand, 61 (44.85%) of the 136 goats and 73 (50.69%) of the 144 sheep examined were positive for anti-*toxoplasma gondii* IgG antibodies. Although sheep had a more significant infection rate than goats, the difference was insignificant (table -1).

Table (1): Prevalence of *Toxoplasma gondii* in Sheep and Goats in Thi –Qar province.

Animals	Positive		Negative		Total	
	No.	%	No.	%	No.	%
Goats	61	44.85	75	55.15	136	48.57
Sheep	73	50.69	71	49.31	144	51.43
Total	134	47.85	146	52.15	280	100
$CalX^2 = 0.956$ $TabX^2 = 3.84$ $DF= 1$ $p. value 0.328$						

2-Age-specific distribution of goats and sheep infected with *Toxoplasma gondii*

The seroprevalence of anti-*toxoplasma gondii* antibodies among types of animals showed significant differences in different age groups of goats. Goats older than two years showed the highest prevalence, 31(56.36%) out of 55. On the other hand, in sheep, the high prevalence was showed in age over two years, but the differences were insignificant at different ages. In contrast, statistical analysis showed significant differences between goats and sheep for different age groups (table -2).

Table (2): Age-specific distribution of goats and sheep infected with *Toxoplasma gondii*.

Animals	Gender	Positive		Negative		Total	
		No.	%	No.	%	No.	%
Goats	< 1 year	8	22.22	28	77.78	36	12.85
	1-2 year	22	48.88	23	51.52	45	10.07
	> 2 years	31	56.36	24	43.64	55	19.64
Goats Statistic $CalX^2 = 10.696$ $TabX^2 = 5.99$ $DF= 2$ $p. value 0.005$							
Sheep	< 1 year	10	34.48	19	65.52	29	10.35
	1-2 year	25	50.0	25	50.0	50	17.85
	> 2 years	38	58.46	27	41.54	65	23.21
Total		134	47.85	146	52.15	280	100
Sheep Statistic $CalX^2 = 4.628$ $TabX^2 = 5.99$ $DF= 2$ $p. value 0.099$							
Both statistic $CalX^2 = 16.194$ $TabX^2 = 11.07$ $DF= 5$ $p. value 0.006$							

3-Distribution of *Toxoplasma gondii*-infected goats and sheep by sex.

Out of 70 goats tested for toxoplasma antibodies, 25 (35.71%) were positive. Furthermore, out of 66 goats, data showed 36 females (45.54%) positive for IgG, and the infection rate was higher in females than in males, with significant differences.

On the other hand, out of 74 sheep males, 42 (56.75%) were positive for IgG, while 70 sheep females showed 31 (44.28%) positive for IgG. The prevalence of Toxoplasmosis was higher in female sheep than in male sheep. Moreover, the differences were not statistically significant, and there was a significant difference between the males and females of sheep and goats (table -3).

Table (3): Distribution of *Toxoplasma gondii*-infected goats and sheep by sex.

Animals	Sex	Positive		Negative		Total	
		No.	%	No.	%	No.	%
Goats	Male	25	35.71	45	64.29	70	25.0
	Female	36	54.54	30	45.46	66	23.57
Goats Statistic		CalX ² = 4.870	TabX ² = 3.84	DF= 1	p. value 0.027		
Sheep	Male	42	56.75	32	43.25	74	26.43
	Female	31	44.28	39	55.72	70	25.0
Total		134	47.85	146	52.15	280	100
Sheep Statistic		CalX ² = 2.238	TabX ² = 3.84	DF= 1	p. value 0.135		
Both statistic		CalX ² = 8.026	TabX ² = 7.81	DF= 3	p. value 0.045		

Discussion:

Sheep and goats are significant sources of meat production in Iraq. These animals are also suitable reservoir hosts of *Toxoplasma gondii* [12]. In this study, the overall *Toxoplasma gondii* infection rate in Thi -Qar province in sheep and goats was 47.85%. The current data came similar to the results reported by [13] in Peshawar, Pakistan, where they recorded a total infection rate of 47.44%, higher than that reported by [14] in central Ethiopia (20%) and [15] in Kashan, central Iran.

The relative cat densities, the access of goats and sheep to tainted feed and water, the age and sex of the animals examined, and the agro-climatic variation. Moreover, the diagnostic techniques employed and other factors could all contribute to the differences in prevalence between the present study and the earlier studies [16, 17].

The current analysis discovered that sheep had a seroprevalence of *Toxoplasma gondii* (51.43%) greater than goats (48.57%). These data are similar to the previously reported by [18] in Ghana, [14] in Ethiopia, and [13] in Peshawar, Pakistan. This difference might result from the different feeding patterns, as goats prefer browsing while sheep graze closer to the ground; this feature increases the risk of infection from the pasture [14].

The flowing results showed that animals of both sheep and goats older than two years were more likely than younger ones to be seropositive for *Toxoplasma gondii*. This finding is similar to what was discovered by [14], who reported a significantly high prevalence in adult sheep than in younger sheep in central Ethiopia [19], who reported that older animals older than three years have an infectious rate (70.84%) with statistical significant more than the younger animal that recorded infectious rate (22.36%) in Tunisia. Furthermore, a study conducted in Peshawar, Pakistan, according to [13], showed the prevalence was highest in goats aged three years (56.90%), followed by aged 1-2 years (45.59%), and lastly aged one year (28.18%). The high significantly prevalence in adult animals than young animals is due to the reason that might be high probability of exposure to the source of infection as age increases. It suggests that most sheep acquire the infection post-natal [6, 20-22].

Furthermore, that might probably be due to the increased exposure to infective oocysts in the environment [18, 23]. The current study showed increased infection rates in female animals compared to males. Data findings came consistent with [19] observations in southern Tunisia and [13] in Peshawar, Pakistan. These results may be related to the female's greater vulnerability to protozoan parasites [24].

Conclusion:

According to current findings, sheep and goats in Thi-Qar- Iraq, serve as significant *Toxoplasma gondii* reservoirs. Since sheep and goats are important meat producers in Thi-Qar, Iraq, good cooking of meat should be encouraged to avoid contracting the parasite.

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