

Seroprevalence of *Toxoplasma gondii* antibodies using Sabin-Feldman dye test among equines in Isparta province, Turkey

Original
Article

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ABSTRACT

Background: Toxoplasmosis is a protozoan disease widely spread among different animal species with an incidence of up to 90%. The presence of this agent among equine species has been reported to be about 46.3%. *T. gondii* infection in equids could favor the spreading of the infection as equine carcasses are used for animal diets in zoos, as well as dead equine carcasses left outside villages are eaten by stray dogs and cats. This study was conducted to determine the seroprevalence of *T. gondii* among equine species (horses, donkeys, and mules) raised in Isparta province of Turkey by using Sabin Feldman dye test (SFDT) which is a reference test widely used for detecting *T. gondii* antibodies in different animal species.

Objective: The aim of this study was to investigate the seroprevalence of *T. gondii* among equids in Isparta province, Turkey.

Material and Methods: Blood samples were collected from 192 animals including horses (n=72), donkeys (n=88) and mules (n= 32) from January to December of 2016. Separated sera were tested for anti-*T. gondii* antibodies using SFDT.

Results: Out of 192 samples, 44 (22.9%) reacted positive. Sero-positivity was 27.8%, 18.2% and 25.0% in horses, donkeys, and mules, respectively with no statistical difference. According to age of animals, the seroprevalence was 14.3%, 32.8%, and 20.8% in age groups 0-5, 6-10 and over 11 years, respectively with a statistically significant difference. Sero-positivity in male animals was 19.44% and in females 25.0%, with no statistical difference.

Conclusion: The prevalence of exposure to *T. gondii* was determined as 22.9% in equids in Isparta province.

Keywords: donkeys, horses, Isparta, mules, serology, toxoplasmosis, Turkey.

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INTRODUCTION

Toxoplasmosis is a parasitological disease of zoonotic character which is wide spread in the World as well as in Turkey^[1]. End host of *T. gondii* are members of felidae family while it causes disease in intermediate hosts such as humans, birds and other mammalian species. It grows in intermediate hosts in two phases. In the first phase it rapidly proliferates through endodyogeny and develops tachyzoites. In the second phase, it divides slowly as bradyzoites in tissue cysts^[2]. The cysts settle in tissues such as skeletal or cardiac muscles, brain, lungs, liver and kidneys and cause latent toxoplasmosis that may become reactivated^[2]. The infection occurs widely by ingestion of contaminated foods and raw or soft-boiled meats. Infection through congenital transmission from mother to offspring and organ transplantation has also been observed^[1]. Sabin Feldman dye test is a quite sensitive test for detection of IgG antibodies against *T. gondii*. It has been reported that a titer of 1:16 or 4 IU should be accepted as positive for toxoplasmosis^[3].

Equids (donkeys, mules, horses) are especially important domestic animals in Turkey used for transportation and racing. Industry of racehorses in Turkey is estimated to be worth 1 billion dollars yearly^[3]. The total number of equids in Isparta province was reported as 2880 including 598 horses, 230 mules, 2052 donkeys^[4]. *T. gondii* infections in horses is generally of subclinical course. Several case studies reported elevated titers in horses with chorioretinitis and in one horse with optic nerve atrophy^[5]. However several studies showed no association of antibody titers with ocular lesions^[6,7] or with Equine Recurrent Uveitis (ERU) in horses^[8]. Toxoplasmosis in equids poses no direct risk for human health because equine meat is not consumed in Turkey. However *T. gondii* infection in equids could favor the spreading of the infection as equine carcasses are used for animal diets in zoos, as well as dead equine carcasses left outside villages are eaten by stray dogs and cats^[9]. Therefore, investigating the prevalence of *T. gondii* infection in equine species could contribute to monitoring this infection in the population.

Sabin Feldman dye test is a reference test which is performed in reference laboratories where tachyzoites obtained from cell cultures or experimental mice are used^[3]. The aim of this study was to determine the seroprevalence of *T. gondii* infection in equids from Isparta province, Turkey.

MATERIAL AND METHODS

This descriptive analytical study was conducted at the Parasitology Laboratory of Turkish Public Health Institution. where SFDT is performed as a routine for diagnosis of toxoplasmosis, during the period from January 2016 to December 2016.

Study area and animals: Isparta province is located in the western and inner parts of the Mediterranean Region of Turkey and is also named as Region of Lakes (38° 17'58.1028" N-31° 10' 32.8944" E). This location lies in the transition zone between Mediterranean climate and the continental climate of Central Anatolia. Blood samples were collected from 192 equids (72 horses, 88 donkeys and 32 mules) into sterile tubes from the punctured jugular vein.

Serology: Sera were obtained by centrifuging the blood samples at 25°C at 4000 x g for 10 min, and stored at -20°C until analysis using SFDT^[10]. Dilutions of 1/16 and above were evaluated as positive.

Statistical analysis: Statistical analysis was performed with SPSS version 20. Differences with respect to seroprevalence among the equine species, age groups or sexes were assessed using Chi-square test. Statistical significance was considered when *P* values were less than 0.05.

Ethical Statement: This study was carried out according to the article 10 of Directives on Duties, Working Procedures and Principles of the Provincial Organization of the Ministry of Food, Agriculture and Livestock.

RESULTS

Out of all 192 samples, 44 (22.9%) reacted positive including 20 (27.8%) of the 72 horses, 16 (18.2%) of the 88 donkeys and 8 (25%) of the 32 mules. Irrespective of equine species, seropositive titers were determined at 1/16 dilution in 30 (15.6%) samples, 1/64 dilution in 8 (4.2%), and 1/256 dilution in 6 (3.1%) samples. Seropositivity among males and females was 14 (19.4%) and 30 (25%), respectively. There was no significant difference between the sex groups (*P*>0.05). Regarding age of animals, the recorded prevalence was 14.3% in 0-5 age group, 32.8% in 6-10 age group and 20.8% in age group over 11 years. Statistically significant difference was observed among age groups (*P*>0.05) (Table 1).

Table 1. The serological results of the equines tested for *T. gondii* seropositivity.

Variable	Sample size (No.)	Seropositive equines		Statistical analysis (<i>P</i> values)
		No.	%	
Species	Horses	72	20	0.340 (NS)
	Donkeys	88	16	
	Mules	32	8	
Gender	Male	72	14	0.240 (NS)
	Female	120	30	
Age (years)	0-5	56	8	0.048 (S)*
	6-10	64	21	
	≥ 11	72	15	

No.: Number; NS: Not significant; S: Significant.

DISCUSSION

Several studies aiming for the detection of prevalence of *T. gondii* in different animal species by various serologic tests including latex agglutination test^[11], indirect hemagglutination test^[12], modified agglutination test^[12], indirect fluorescent antibody test^[13] and SFDT^[14] have been reported from different countries throughout the World.

The prevalence of *T. gondii* infection in horses was reported to range from 0% to 80% in different countries^[15,16,17]. The prevalence of seropositive samples for *T. gondii* in horses found in the present study (27.8%) was higher than that reported from

Spain (10.8%)^[18], Mexico (6.1%)^[19], United States of America (6.9%)^[20], South Korea (2.6%)^[21], and Sweden (1%)^[22]. However, the prevalence observed in our study was lower than that reported in Saudi Arabia (31.6%)^[23], Southwestern China (30.5%)^[24] and Sudan (30.04%)^[25].

Seropositivity for *T. gondii* infection in donkeys was reported as 30% in Northwestern Algeria^[24], 58.7% in Pakistan^[26], 47% in Iran^[27], 65.6% in Egypt^[28] and 5%-8% in Italy^[29]. While the seroprevalence in mules was found to be 23.8% in Brazil^[30] and 15% in Spain^[18].

Numerous studies were also carried out to record the seroprevalence of *T. gondii* in equines in Turkey.

The studies and the determined rates are given in table (2).

The recorded data showed that the highest rate was found in Erzurum province with 62%^[39] and the lowest rate was in Van province with 1.7%^[33]. Seroprevalence observed in the present study for horses (27.8%) was higher than those reported from Malatya, Van, Kayseri, Kars, Şanlıurfa and Niğde provinces while it was similar to those reported from Ankara and Hakkari provinces (Table 2). On the other hand, seroprevalence found in the present study for donkeys was lower than those reported from Kayseri and Erzurum provinces. The seroprevalence in females and males were 25% and 19.4%, respectively. No statistically significant difference was found between *T. gondii* infections and

gender ($P=0.240$). Similar findings were also reported by other studies^[18,40,41].

The seroprevalence of *T. gondii* infection showed a statistically significant difference among age groups and the maximum seroprevalence was observed in the age-group of 6-10 years. This finding was in concordance with Rouatbi *et al.*^[42], Güçlü *et al.*^[37] and Machacova *et al.*^[29]. The results indicated an increase in the risk of toxoplasmosis in correspondence with the increase in age^[29,37,42].

In this study, it was concluded that seroprevalence of *T. gondii* was determined as 22.9% in equines in Isparta province, Turkey. The *T. gondii* infection was found in 27.8% of horses, 18.2% of donkeys and 25% of mules.

Table 2. Seroprevalence of *T. gondii* antibodies in equines in Turkey.

Cities	Seroprevalence			
	Method	Species	%	Reference
Kars	SFDT	Horse	20.6	[9]
Ankara	SFDT	Horse	2.0	[31]
Malatya	SFDT	Horse	6.4	[32]
Van	IHA	Horse	1.7	[33]
Kayseri	SFDT	Horse	10.4	[34]
Kayseri	SFDT	Mule	10.0	[34]
Kayseri	SFDT	Donkey	42.42	[34]
Şanlıurfa	SFDT	Horse	7.5	[35]
Hakkari	IHA	Horse	13.5	[36]
Hakkari	SFDT	Horse	28.3	[36]
Ankara	SFDT	Horse	28.0	[37]
Niğde	SFDT	Horse	7.0	[38]
Erzurum	SFDT	Donkey	62	[39]

SFDT: Sabin Feldman dye test, IHA: Indirect hemagglutination test

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