

SEROPREVALENCE OF *TOXOPLASMA GONDII* INFECTION IN DOMESTIC SHEEP IN DURANGO STATE, MEXICO

C. Alvarado-Esquivel, C. García-Machado*, D. Alvarado-Esquivel, J. Vitela-Corrales*, I. Villena†, and J. P. Dubey‡

Faculty of Medicine and Nutrition, Juárez University of Durango State, Avenida Universidad S/N. 34000 Durango, Mexico. e-mail: jitender.dubey@ars.usda.gov

ABSTRACT: The seroprevalence of *Toxoplasma gondii* infection in sheep (*Ovis aries*) in northern Mexico is largely unknown. Antibodies to *T. gondii* were determined in serum samples from 511 sheep from 8 farms in Durango State, Mexico, using the modified agglutination test (MAT). Sheep were raised in 3 geographical regions, i.e., mountainous (n = 68), semi-desert (n = 132), and valley (n = 311). Overall, *T. gondii* antibodies were found in 77 (15.1%) of 511 sheep, with MAT titers of 1:25 in 27, 1:50 in 10, 1:100 in 11, 1:200 in 11, 1:400 in 8, 1:800 in 3, 1:1,600 in 4, and 1:3,200 in 3. The seroprevalence of *T. gondii* infection increased significantly with age, indicating post-natal transmission. In contrast, gender, breed, flock size, and geographic region did not significantly influence the seroprevalence. Seropositive sheep were found in 7 of 8 farms sampled. This is the first report of *T. gondii* infection in sheep in Durango State, Mexico. Results indicate that infected sheep are probably an important source of *T. gondii* infection for humans in Durango State.

Infections with *Toxoplasma gondii* in domestic sheep (*Ovis aries*) have been reported worldwide (Dubey, 2009, 2010). Toxoplasmosis has been recognized as one of the main causes of infective ovine abortions in several countries (Dubey, 2010). Ingestion of undercooked meat from infected sheep could be a source of infection in humans (Cook et al., 2000; Dubey, 2009; Chikweto et al., 2011).

We have been studying the epidemiology of *T. gondii* infections in humans (Alvarado-Esquivel et al., 2009, 2010; Alvarado-Esquivel, Estrada-Martínez et al., 2011) and other animals in Durango State, México (Alvarado-Esquivel et al., 2007; Dubey et al., 2007, 2009; Alvarado-Esquivel, García-Machado et al., 2011; Alvarado-Esquivel, García-Machado, Vitela-Corrales et al., 2011). However, information regarding *T. gondii* infection in sheep in Durango State, Mexico, is lacking. Meat of sheep is widely consumed in Mexico, and many typical Mexican dishes are made from mutton. Epidemiologically, consumption of sheep meat was associated with *T. gondii* infection in patients suffering from liver disease in Durango, Mexico (Alvarado-Esquivel, Torres-Berumen et al., 2011). Therefore, we sought to determine the seroprevalence of *T. gondii* infection in sheep raised in 3 geographical regions in Durango State, Mexico.

MATERIALS AND METHODS

Sheep surveyed

Five hundred and eleven domestic sheep were sampled from 8 farms in 4 municipalities of Durango State (Table I) from November 2010 to August 2011. The number of sheep per farm ranged from 14 to 169 (median = 49.5). One farm was located in the municipality of Pueblo Nuevo in the mountainous region, 3 farms were located in the municipality of Santa Clara in the semi-desert region, and 4 farms (3 in the municipality of Durango and 1 in the municipality of Guadalupe Victoria) were located in the valley region. Sheep from the mountainous region (n = 68) were a

mixed breed; all were females and 12–84 mo of age (median = 24 mo). Sheep from the semi-desert region (n = 132) were pure (Dorper; n = 96) and mixed breed (n = 36) and were 0.5–40 mo of age (median = 12 mo); 98 (74.2%) were females and 34 (25.8%) were males. Sheep from the valley region (n = 311) were pure breed (Pelibuey 30, Dorper 37, Suffolk 28, Rambouillet 16, and Charollais 1) and mixed breed (n = 199) and were 4–60 mo of age (median = 20 mo). These sheep included 290 (93.2%) females and 21 (6.8%) males.

Serological examination

Blood samples were transported to the laboratory on the day of collection or a day after collection. Sera were collected from whole blood by centrifugation and stored at –20 C until tested. Sheep sera were tested for *T. gondii* antibodies using 2-fold serial dilutions from 1:25 to 1:3,200 with the modified agglutination test (MAT; Dubey and Desmonts, 1987) using reagents described by us (Alvarado-Esquivel, García-Machado et al., 2011). A titer of 1:25 was used as the cut-off for seropositivity in MAT.

Statistical analysis

Statistical analysis was performed using Epi Info software version 3.5.1 (Centers for Disease Control and Prevention; <http://wwwn.cdc.gov/epiinfo/>). A chi-square test was used for comparison of the frequencies among groups. $P < 0.05$ was considered statistically significant.

RESULTS

Overall, antibodies to *T. gondii* were found in 77 (15.1%) of 511 sheep, with titers of 1:25 in 27; 1:50 in 10; 1:100 in 11; 1:200 in 11; 1:400 in 8; 1:800 in 3; 1:1,600 in 4; and 1:3,200 in 3. Seropositive sheep were found on 7 of 8 farms. In sheep from the mountainous region, antibodies to *T. gondii* were found in 6 (8.8%) of 68 sheep, with titers of 1:25 in 1; 1:200 in 2; and 1:3,200 in 3. In sheep from the semi-desert, antibodies to *T. gondii* were found in 23 (17.4%) of 132 sheep, with titers of 1:25 in 4; 1:50 in 3; 1:100 in 1; 1:200 in 7; 1:400 in 4; 1:800 in 1; and 1:1,600 in 3. In sheep from the valley region, antibodies to *T. gondii* were found in 48 (15.4%) of 311 sheep, with titers of 1:25 in 22; 1:50 in 7; 1:100 in 10; 1:200 in 2; 1:400 in 4; 1:800 in 2; and 1:1,600 in 1. In general, the seroprevalence of *T. gondii* infection was comparable among geographical regions ($P > 0.05$; Table I). However, sheep from a farm of the semi-desert region (SC-1) had the highest seroprevalence (20.2%); cats (*Felis catus*) were observed on this farm. The seroprevalence of *T. gondii* infection increased significantly ($P = 0.004$) with age (Table II). In contrast, the seroprevalence of *T. gondii* infection was comparable among sheep regardless their sex, breed, and flock size ($P > 0.05$; Table II).

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*Laboratory of Animal Pathology. Regional Livestock Union of Durango. Km 4.5 Carretera Durango-Parral, 34217 Durango, Mexico.

†Laboratory of Parasitology-Mycology, National Reference Centre on Toxoplasmosis, Biological Resources Centre *Toxoplasma*, Hospital Maison Blanche, 45 rue Cognac-Jay, 51092 Reims Cedex, France.

‡Animal Parasitic Diseases Laboratory, Animal and Natural Resources Institute, United States Department of Agriculture, Agricultural Research Service, Building 1001, Beltsville, Maryland 20705-2350.

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TABLE I. Seroprevalence of *Toxoplasma gondii* infection in domestic sheep (*Ovis aries*) in Durango, Mexico.

Region	Municipality	Farms surveyed	No. sheep tested	Seroprevalence of <i>T. gondii</i> infection	
				No.	%
Mountains Semi-desert	Pueblo Nuevo	PN-1	68	6	8.8
		Santa Clara	SC-1	94	19
	SC-2		14	0	0
	SC-3		24	4	16.7
Valley	Durango	All	132	23	17.4
		DGO-1	31	4	12.9
		DGO-2	169	30	17.8
	Guadalupe Victoria	DGO-3	30	3	10
		GV-1	81	11	13.6
		All	311	48	15.4
All			511	77	15.1

* Significantly higher than that in farm in the mountainous region ($P = 0.04$).

DISCUSSION

Here, a 15.1% seroprevalence of *T. gondii* infection in sheep raised in the northern Mexican Durango State is lower than those reported in other Mexican states. However, comparisons of our results with those of previous studies should be made with care because different assays were used in the studies. García-Vázquez et al. (1990) studied 495 sheep from 3 states (Morelos, San Luis Potosi, and Guanajuato) in central Mexico, and they found an overall 30% seroprevalence using indirect immunofluorescent test (IFAT). In a second study by the same research group, IFAT seropositivity was reported at 37.9% in 702 sheep older than 1 yr from Morelos State (Cruz-Vázquez et al., 1992). In a study on a ranch in the eastern part of Mexico, 103 sheep were analyzed by an indirect enzyme-linked immunosorbent assay (ELISA) and an immunoblot for *T. gondii* antibodies, and a seroprevalence ranging from 77 to 84% was observed (Caballero-Ortega, Quiroz-Romero et al., 2008). In a study in western Mexico (Colima State), 29.1% of 351 sheep were positive for anti-*T. gondii*

TABLE II. General characteristics of the 511 sheep (*Ovis aries*) studied and seroprevalence of *Toxoplasma gondii* infection.

Characteristic	No. sheep tested	Seroprevalence of <i>T. gondii</i> infection		P
		No.	%	
Age (mo)				
0.5–1	5	2	40	0.004
3–6	52	4	7.7	
7–12	96	13	13.5	
13–18	30	5	16.7	
20–24	200	29	14.5	
25–36	72	6	8.3	
40–48	39	13	33.3	
54–84	17	5	29.4	
Gender				
Male	55	7	12.7	0.75
Female	456	70	15.4	
Breed				
Pure	208	34	16.3	0.58
Mixed	303	43	14.2	

antibodies by an indirect ELISA (Caballero-Ortega, Palma et al., 2008). Worldwide seroprevalence of *T. gondii* infection in sheep was recently reviewed by Dubey (2010). Since then, Chikweto et al. (2011) reported *T. gondii* antibodies in 44.1% of 204 sheep from Grenada and Carriacou, West Indies, using MAT. Langoni et al. (2011) reported *T. gondii* antibodies in 18.6% of 382 commercial sheep from Sao Paulo, Brazil, using MAT.

Here, seroprevalence increased significantly with age of the sheep, indicating post-natal exposure to *T. gondii* oocysts. The high seropositivity (2 of 5) in new born lambs (Table II) is probably related to transcolostral ingestion of antibodies; passively acquired antibodies generally disappear by 6 mo of age. Therefore, a proportion of 3- to 6-mo-old lambs may have had transcolostral antibodies (Table II). Cats were observed on the farm with the highest seroprevalence of *T. gondii* infection, but farm cats were not tested for *T. gondii* infection. However, in a separate study, antibodies to *T. gondii* were found in 21% of 105 (Alvarado-Esquivel et al., 2007) and 9.3% of 150 (Dubey, Velmurugan et al., 2009) domestic cats from Durango, Mexico. These seropositive cats would have shed oocysts in the environment.

Results of the present study indicate that *T. gondii* infection is common in sheep in Durango regardless their gender, breed, flock size, and the geographical region. Results are of concern because sheep meat is widely eaten in Mexico in general and in Durango State in particular. In 2009, Mexico had 7,306,600 sheep, and 111,551 of these animals were in Durango State (<http://www.inegi.org.mx/sistemas/TabuladosBasicos/Default.aspx?c=17177&s=est>). In addition, Mexico produced 53,737 tons of sheep meat in 2009 (<http://www.inegi.org.mx/sistemas/mexicocifras/MexicoCifras.aspx?e=0&m=0&src=0&ent=0&sec=M&ind=1009000054&enn=Estados%20Unidos%20Mexicanos&ani=2009>). Most, if not all, sheep meat produced in Mexico is consumed in Mexico. Consumption of sheep meat was recently associated with *T. gondii* infection in patients suffering from liver disease in Durango, Mexico (Alvarado-Esquivel, Torres-Berumen et al., 2011). Therefore, further research on the role of sheep meat in human infections and pathogenesis of *T. gondii* is needed.

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