

Presence of Anti-*Toxocara* Antibodies in Sheep from the State of Mexico

Camilo Romero¹, Rafael Heredia², Saira Aguilar¹, Nadyeli Nava², Jocelyn Pineda¹

Universidad Autónoma Del Estado de México, ¹Centro Universitario UAEM Amecameca, Amecameca,
²Doctorado en Ciencias Agropecuarias y Recursos Naturales, Toluca, México

Received September 8, 2015

Accepted August 31, 2016

Abstract

Toxocariasis is a parasitic zoonosis caused by the nematode *Toxocara canis*, and less frequently *Toxocara cati*, whose final hosts are the dog and cat, respectively. It is acquired by the ingestion of embryonated parasite eggs; the ingestion of meat from animals carrying cystic larvae plays a central role in this disease. The study was conducted in Ayapango, Mexico. Ninety-two sheep were used, of which 72 were females and 20 males. The total prevalence of anti-*Toxocara* antibodies was 15.21% (14/92), ranging from 17.24% in the one to six months age group to 14.28% in the group older sheep six months, with a higher percentage in females (19.44%) compared to males (5.0%), with a significant difference between positive males and females older than six months of age (Chi-square test = 4.22, $P < 0.05$). The prevalence of anti-*Toxocara* antibodies in sheep suggests that a high number of animals are infected with *Toxocara* spp. The consumption of meat from paratenic hosts, including sheep, is considered a means of transmission of toxocariasis to humans.

Seroprevalence, larva migrans, zoonoses, ELISA

Toxocariasis is one of the most commonly reported helminth zoonotic infections throughout the world. Its main aetiological agent is the *Toxocara canis* nematode, and less frequently *Toxocara cati*, whose final hosts are the dog and cat, respectively (Rubinsky 2004; Radman et al. 2006; Marino et al. 2011). It is acquired through the ingestion of embryonated eggs exposed to infected soil, geophagy, poorly washed hands, onychophagy, as well as the ingestion of vegetables with animal faeces and larvae encysted in the tissue of paratenic hosts (various animals) (Del Valle et al. 2002; Strube et al. 2013; Ferrero et al. 2014). These animals host hypobiotic larvae in the liver and other tissues, which can be transmitted to humans through the ingestion of raw or undercooked meat (De La Fé et al. 2006; Lescano et al. 2012). However, there is little information available on the prevalence of *T. canis* infection in humans through paratenic hosts such as horses, sheep, birds, etc. (Alvarez et al. 2011). The objective of the present study was to determine the presence of *Toxocara* antibodies in sheep of Ayapango, Mexico State, Mexico.

Materials and Methods

This study was conducted from September to November 2013 in the municipality of Ayapango, Mexico (19°10' N, 98°45' W and 2,450 metres above sea level). Ninety-two sheep were used, within the age range from one to 24 months, of which 72 were females and 20 were males. Jugular venipuncture blood samples were taken to measure the level of anti-*Toxocara* antibodies. The selection of all of the animals included in this study followed random allocation criteria.

The anti-*Toxocara* IgG antibody levels were evaluated using a commercial kit SCIMEDX *Toxocara* Microwell Serum ELISA indirect format, using antigen excretion/secretion of *Toxocara* larvae 2, with a dilution of 1:64 test samples, and using protein "A" conjugated to peroxidase. Reading of optical densities was conducted with a spectrophotometer (Victor Wallack 120) (Heredia et al. 2014), using a 0.3 cut-off point diopter for the positives. All the data obtained were registered to compare the prevalence between sexes and ages, using a test of Chi-square and ODDS ratio to measure the relative risk for the same variables, using a 95% confidence interval and significance of $P < 0.05$.

Address for correspondence:

Camilo Romero Núñez, Ph.D
Centro Universitario UAEM Amecameca
Universidad Autónoma Del Estado de México
Carrtera Amecameca-Ayapango km. 2.5
56900, Amecameca de Juárez, Estado de México

Phone: +15 979 782 158
E-mail: cromeron@uaemex.mx
<http://actavet.vfu.cz/>

Ethical statement

The research protocol was approved by the Ethics Committee of the Autonomous University of the State of Mexico.

Results

The total prevalence of anti-*Toxocara* antibodies was 15.21% (14/92), ranging from 17.24% in the age group of one to six months to 14.28% in the group of sheep older than six months, with a higher percentage in females (19.44%) compared to males (5.0%). The frequency of antibodies was directly proportional to the age of the animals, with no significant differences between sexes in animals aged from one to six months. A difference was found in males (Chi-square = 4.58, $P < 0.03$) older than six months, yet none of the variables (age and sex) were shown to be a risk factor for the presence of antibodies against *Toxocara* (Table 1). A significant difference was found (Chi-square = 4.22, $P < 0.03$) comparing the differences between positives from different sexes in sheep over six months of age (Table 2).

Table 1. Seroprevalence of *Toxocara canis* association and risk factor between ages and sexes of sheep from Ayapango, Mexico, from September to November 2013.

	Positive n = 14	Negative n = 78	Chi-square	P-value	OR	P-value	CI (95%)
0 to 6 months							
Males	1 (7.1%)	4 (5.1%)	0.08	0.7	1.42	0.76	0.14-13.6
Females	4 (28.5%)	20 (25.6%)	0.03	0.86	1.16	0.81	0.32-4.11
Over 6 months							
Males	0 (0.0%)	15 (19.2%)	4.58	0.03	0.48	0.63	0.02-9.5
Females	9 (64.2%)	39 (50.0%)	0.28	0.59	1.8	0.32	0.55-5.85

OR - ODDS ratio, CI - confidence interval, $P \leq 0.05$

Table 2. Comparison between ages and sexes positive for *Toxocara canis* antibody in sheep from Ayapango, Mexico of September to November 2013.

Positive	Males (20)	Females (72)	Chi-square	P-value
Total	1	13	1.63	0.20
Under 6 months of age	1	4	0.008	1
Over 6 months of age	0	9	4.22	0.03

$P \leq 0.05$

Discussion

Literature data on the prevalence of *Toxocara* spp. in paratenic guests are limited; however, some studies of sheep report higher prevalence rates than those found in this study, such as the study by Lloyd et al. (2006) carried out in the country of Wales. Four hundred serum samples were evaluated with ELISA test reporting a *T. canis* antibody positivity of 13% in six-month-old sheep, which is still less than the 17.24% found in the present report. Ten-month-old sheep were reported to have a 16% positivity rate in the sample; and 31% positivity rate was found in 15-month-old sheep. Both values exceed

those found in our study of sheep older than six months (14.28%). In both studies an increase of positive samples in older animals was noted, which could be related to greater animal contact with infective forms of *T. canis*.

Kantzoura et al. (2013) found in a study conducted in Thessaly, Greece 42.9% seroprevalence of *Toxocara canis* in sheep (361/155), triple than that found by us. The authors attribute the prevalence to temperature and rainfall which facilitate incubation and subsequent dissemination of *Toxocara* eggs. Another mentioned risk drivers are the conditions such as the sheep's feed; seroprevalence increases when animals graze and decreases when they are stabled.

A study carried out in Brazil on anti-*T. canis* antibodies in sheep reported that the overall prevalence of antibodies was 50.1% (183/365), which exceeds the number reported in this study, where it was found in 15.21% of the tested sheep in Ayapango, Mexico. The same study reported 6.01% of positive samples in the group aged one to six months and 61.2% in the group aged between 10 and 15 months, which is contrary to the results presented in this study where the sheep under six months of age had higher positive samples (17.24%) compared with samples from sheep over six months of age (14.28%) (Alvarez et al. 2011).

Another study in the south of Brazil, with 1,642 sheep serum samples, reported that the frequency of anti-*Toxocara* antibodies was 29.0% (477/1642), which is still higher than what was found in this investigation. These results indicate that infection by *T. canis* is widely distributed among sheep, representing a potential risk for human health (Muñoz et al. 2010; Overgaauw et al. 2013; Rassier et al. 2013).

The prevalence of anti-*Toxocara* antibodies in sheep suggests that a high number of animals are infected with *Toxocara* spp.; these animals can host hypobiotic larvae in the liver and other tissues that could be transmitted to humans by ingestion of raw or undercooked meat. In addition, transmission of larvae from meat to wild and domestic animals could perpetuate the parasites' life cycle in their definitive hosts (Radman et al. 2006; Alvarez et al. 2011).

Consumption of meat from paratenic hosts, including sheep, is considered a transmission route of toxocariasis to humans (Salem and Schantz 1992). Plenty of studies have considered the consumption of raw or undercooked meat as a risk factor because of the *Toxocara* larvae (Alvarez et al. 2011).

The overall prevalence of anti-*Toxocara* antibodies (15.2%) in sheep in Ayapango, Mexico, is lesser than that reported in studies from other parts of the world. However, these results highlight a little-studied problem, which could participate in the perpetuation of toxocariasis in their final and paratenic hosts, including humans. Thus, monitoring the animals' health status and providing guarantees regarding appropriate production and animal conditions is important.

Conflict of Interests

The authors declare they have no conflict of interest.

References

- Alvarez V, Fabris PA, Leme BE, Rubinsky-Elephant G, Guiffreda R 2011: Anti-*Toxocara* spp. antibodies in sheep from southeastern Brazil. *Vet Parasitol* **179**: 283-286
- Del Valle GM, Radman NE, Burgos L, Fonrouge R, Archelli SM 2002: *Toxocara canis*: Migración larval y eosinofílica en el hospedador paraténico. *Parasitol latinoam* **57**: 46-49
- De La Fé P, Duménigo RB, Brito AE, Aguiar SJ 2006: *Toxocara canis* y síndrome larva migrans visceral. *Rev Electron Vet* **7**: 1-42
- Ferrero M, Sánchez RJ, Pizzi RD, Pizzi HL 2014: Determinación de anticuerpos IgG anti-*Toxocara canis* en estudiantes de medicina. *Rev Salud Publica* **18**: 36-43
- Heredia R, Romero C, Mendoza GE, Ponce M, Portal A, González L, Bautista LG 2014: Occurrence of *Toxocara canis* in students of veterinary and graphic design in a Mexican University. *Acta Sci Vet* **42**: 1-6

- Kantzoura V, Diakou A, Kouamc MK, Feidas H 2013: Seroprevalence and risk factors associated with zoonotic parasitic infections in small ruminants in the Greek temperate environment. *Parasitol Int* **62**: 554-560
- Lescano SA, Nakhle MC, Ribeiro MC, Chieffi PP 2012: IgG antibody responses in mice coinfecting with *Toxocara canis* and other helminths or protozoan parasites. *Rev Ins Med Trop São Paulo* **54**: 145-152
- Lloyd S 2006: Seroprevalence of *Toxocara canis* in sheep in Wales. *Vet Parasitol* **137**: 269-272
- Marino GL, Bojanich MV, López MD, Alonso JM 2011: Prueba de avidéz de los anticuerpos de IgG en la infección por *Toxocara canis*. *Acta Bioquim Clin L* **145**: 323-327
- Muñoz MA, Alba HF 2010: Secretory-excretory antigens of *Toxocara canis* recognized by puppies of the Mexico City metropolitan area. *Vet Mex* **41**: 59-64
- Overgaaauw PA, Van Knapen F 2013: Veterinary and public health aspects of *Toxocara* spp. *Vet Parasitol* **193**: 398-403
- Radman NE, Archelli MS, Burgos L, Fonrouge RD, Del Valle M 2006: *Toxocara canis* en caninos, prevalencia en la ciudad de La Plata. *Acta Bioquim Clin L* **40**: 41-48
- Rassier GL, Pappen FG, Borsuk S, Scaini CJ, Gallina T, Pinto JS, Rodrigues S, Berne ME 2013: *Toxocara* spp. seroprevalence in sheep from Southern Brazil. *Parasitol Res* **112**: 181-187
- Rubinsky-Elephant G 2004: Human toxocariasis: Humoral response (IgG, IgA and IgE) anti-*Toxocara canis* and clinical-laboratorial correlation in patients following chemotherapy. *Rev Ins Med Trop São Paulo* **46**: 76-81
- Salem G, Schantz P. 1992: *Toxocara* visceral larva *Migrans* after ingestion of raw lamb liver. *Clin Infec Dis* **15**: 743-744
- Strube C, Heuer L, Janecek E 2013: *Toxocara* spp. infections in paratenic hosts. *Vet Parasitol* **193**: 375-389