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# PRESENCE OF ANTI-Toxocara canis ANTIBODIES IN SHEEP

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PRESENTA:

SAIRA BERENICE AGUILAR LÓPEZ

ASESOR:

Dr. CAMILO ROMERO NÚÑEZ

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## PRESENÇA DE ANTICORPOS ANTI-Toxocara canis EM OVINOS PRESENCE OF ANTI-Toxocara canis ANTIBODIES IN SHEEP

#### Aguilar López Saira, Romero Núñez Camilo

Centro Universitario UAEM Amecameca. Universidad Autónoma Del Estado de México.

CORRESPONDECE: C. Romero: [cromeron@uaemex.mx – Fax + 52 (001) 597-978-2158]. Centro Universitario UAEM Amecameca, Universidad Autónoma Del Estado de México, 56900 Amecameca, México.

**Background:** 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.

However there is little information available on the prevalence of *T. canis* infection in humans trough paratenic hosts such as horses, sheep, birds, etc. The aim of the present study was to determine the presence of *T. canis* antibodies in sheep of Ayapango, Mexico State, Mexico.

Materials, methods and results: This study was conducted in the municipality of Ayapango, Mexico. Ninety-two sheep where used, from one to 24 months of age, of which 72 were females and 20 males and the sheep race used was Rambouillet. The selection of all of the animals included in this study followed random allocation criteria. The anti-Toxocara antibodies were measured using an ELISA test (SCIMEDX Toxocara Microwell Serum ELISA, commercial kit). Optical densities reading was conducted with a spectrophotometer (Victor Wallac K120), using a 0.3 cut-off point diopter for the positives. The total prevalence of anti-Toxocara canis 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%) than in 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).

**Discussions:** The prevalence of *Toxocara* spp. in paratenic guests is limited, however there are studies of sheep where prevalence rates reported are higher than those found in this study, such as report carried out in the country of Wales, four hundred serum samples were evaluated with the ELISA test reporting a T. canis antibody positivity of 13% in sheep of six months of age, which is still less than the 17.24% found in the present report. The 10-month-old sheep were reported to have a 16% positivity rate in the sample and 31% at 15 months of age. Both values are higher than those found in this investigation concerning sheep older than six months (14.28%). A study carried out in Brazil on anti-T. canis antibodies in sheep, reported that the overall prevalence of the antibodies was 50.1% (183/365), which is greater than the number reported in this study, where it was found in 15.21% of tested sheep in Ayapango, Mexico. 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 the sheep, and it represents a potential risk for human health. The prevalence of anti-*Toxocara* antibodies in sheep suggests that a high number of animals are infected with *Toxocara* spp.; these animals can harbor hipobiotic larvae in the liver and other tissues that could be transmitted to humans by ingestion of raw or undercooked meat. The consumption of meat from paratenic hosts, including sheep, is considered a means of transmission of toxocariasis to humans. The positivity rate of 15.2% from this study on sheep is lower than that reported in other countries, as Wales reported 47% in 2006 and Brazil reported 50.1% in 2011.

**Key words:** Toxocara, antibodies, sheep, zoonosis, Mexico.

#### INTRODUCTION

Toxocariasis is one of the most commonly reported helminth zoonotic infections throughout the world. Its main etiologic agent is the *Toxocara canis* nematode, and less frequently *Toxocara cati*, whose final hosts are the dog and cat respectively [6, 9, and 11]. 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 feces and larvae encysted in the tissue of paratenic hosts (various animals) [2,3,14]. These animals host hipobiotic larvae in the liver and other tissues, which can be transmitted to humans trough the ingestion of raw or undercooked meat [1, 4]. However there is little information available on the prevalence of *T. canis* infection in humans trough paratenic hosts such as horses, sheep, birds, etc. [13]. The objective of the present study was to determine the presence of *T. canis* antibodies in sheep of Ayapango, Mexico State, Mexico.

#### MATERIAL AND METHODS

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

The anti -Toxocara canis antibody levels were evaluated using antigen excretion / secretion of T. canis larvae 2 with the commercial kit SCIMEDX Toxocara Microwell Serum ELISA; optical densities reading was conducted with a spectrophotometer (Victor Wallac K120), using a 0.3 cut-off point diopter for the positives. The research protocol was approved by the ethics committee of the Autonomous University of the State of Mexico. All the data obtained were registered to compare the prevalence between genders and ages, using a test of Chi square and ODD ratio to measure the relative risk for the same variables, using a 95% confidence interval and significance of 5%.

#### **RESULTS**

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

**Table 1.** Association and factor of risk between seroprevalence of Toxocara canis in age and gender of sheep from Ayapango, Mexico from September to November 2013.

	Positive	Negative	Chi	p-Value	OR	p-Value	IC (95%)
	N=14	N=78	Square				
0 to 6 mor	nths						
Male	1 (7.1%)	4 (5.1%)	0.08	0.7	1.42	0.76	0.14-
							13.6
Females	4	20	0.03	0.86	1.16	0.81	0.32-
	(28.5%)	(25.6%)					4.11
Over 6 mo	onths						
Males	0 (0.0%)	15	4.58	0.03	0.48	0.63	0.02-9.5
		(19.2%)					
Females	9	39	0.28	0.59	1.8	0.32	0.55-
	(64.2%)	(50.0%)					5.85
n<0.05							

p≤0.05

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

Positives	Males (20)	Females (72)	Chi Square	p-Value
Totals	1	13	1.63	0.20
Under to six months	1	4	0.008	1
Over to six months	0	9	4.22	0.03

p≤0.05

#### **DISCUSSION**

The literature data on the prevalence of *Toxocara* spp. in paratenic guests is limited; however there are studies of sheep where prevalence rates reported are higher than those found in this study, such as Lloyd's report [5] carried out in the country of Wales. Four hundred serum samples were evaluated with the ELISA test reporting a *T. canis* antibody positivity of 13% in sheep of six months of age, which are still less than the 17.24% found in the present report. The 10-month-old sheep were reported to have a 16% positivity rate in the sample and 31% at 15 months of age. Both values are higher than those found in this investigation concerning 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*.

In a study carried out in Brazil on anti-*T. canis* antibodies in sheep, reported that the overall prevalence of the antibodies was 50.1% (183/365), which is greater than the number reported in this study, where it was found in 15.21% of tested sheep in Ayapango, Mexico. The same study reported a 6.01% of positive samples in the animal group from one to six months and 61.2% in the animal group between

10 and 15 months, which is contrary to the results presented in this study where the animals under six months of age had higher positive samples (17.24%) compared with samples from animals over six months of age (14.28%) [13].

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 the sheep, and it represents a potential risk for human health [7, 8, and 10].

The prevalence of anti-*Toxocara* antibodies in sheep suggests that a high number of animals are infected with *Toxocara* spp.; these animals can host hipobiotic 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 [9, 13].

Consumption of meat from paratenic hosts, including sheep, is considered a transmission route of Toxocariasis for humans [12]. Plenty of studies have considered the consumption of raw or undercooked meat as a risk factor because of the Toxocariasis larvae [13].

#### CONCLUSION

The overall prevalence of anti-*T. canis* antibodies (15.2%) in sheep in Ayapango, Mexico, is less than that reported in published accounts 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 animal status and providing guarantees on appropriate production and animal conditions is important.

#### SOURCES AND MANUFACTURERS

**Declaration of interest:** The authors report no conflicts of interest.

<sup>&</sup>lt;sup>1</sup>Toxocara Microwell Serum ELISA, commercial kit – SCIMEDX, Denville, NJ, USA.

<sup>&</sup>lt;sup>2</sup>Victor Wallac K120 – PerkinHelmer, Waltham, MA, USA.

#### REFERENCES

- 1 De la Fé P., Duménigo R.B., Brito A.E. & Aguiar S.J. 2006. Toxocara canis y síndrome larva migrans visceral. Revista Electrónica de Veterinaria.7 (4): 1-42.
- 2 Del Valle G.M., Radman N.E., Burgos L., Fonrouge R. & Archelli S.M. 2002. Toxocara canis: Migración larval y eosinofílica en el hospedador paraténico. Parasitología latinoamericana. 57 (1.2): 46-49.
- 3 Ferrero M., Sánchez R.J., Pizzi R.D. & Pizzi H.L. 2014. Determinación de anticuerpos IgG anti-*Toxocara canis* en estudiantes de medicina. *Revista de Salud Pública*. (18) 1: 36-43.
- 4 Lescano S.A., Nakhle M.C., Ribeiro M.C. & Chieffi. P.P. 2012. IgG antibody responses in mice coinfected with *Toxocara canis* and other helminths or protozoan parasites. *Instituto de Medicina Tropical de Sao Paulo*. 54(3): 145-152.
- **5 Lloyd S. 2006.** Seroprevalence of *Toxocara canis* in sheep in Wales. *Veterinary Parasitology.* 137 (3-4): 269-272.
- 6 Marino G.L., Bojanich M.V., López M.D. & Alonso J.M. 2011. Prueba de avidez de los anticuerpos de IgG en la infección por *Toxocara canis*. *Acta Bioquímica Clínica Latinoamericana*. 45(2): 323-327.
- **7 Muñoz M.A. & Alba H.F. 2010.** Secretory-excretory antigens of *Toxocara canis* recognized by puppies of the Mexico City metropolitan area. *Veterinaria México*. 41(1): 59-64.
- **8 Overgaauw P.A. & Van Knapen F. 2013.** Veterinary and public health aspects of *Toxocara* spp. *Veterinary Parasitology*. 193(4): 398-403.
- 9 Radma N.E., Archelli M.S., Burgos L. Fonrouge R.D. & Del Valle M. 2006. *Toxocara canis* en caninos, prevalencia en la ciudad de La Plata. *Acta Bioquímica Clínica Latinoamérica*. 40(1): 41-48.

- 10 Rassier G.L., Pappen F.G., Borsuk S., Scaini C.J., Gallina T., Pinto J.S., Rodrigues S. & Berne M.E. 2013. Toxocara spp. seroprevalence in sheep from southern Brazil. Parasitology Research. 112(9): 181-187.
- 11 Rubinsky-Elephant G. 2004. Human toxocariasis: Humoral response (IgG, IgA and IgE) anti-*Toxocara canis* and clinical-laboratorial correlation in patients following chemotherapy. *Instituto de Medicina Tropical de Sao Paulo*. 46(2): 76-81.
- **12 Salem G. & Schantz P. 1992.** *Toxocara* visceral larva migrans after ingestion of raw lamb liver. *Clinical Infectious Diseases*. 15(4): 743-744.
- 13 Santarém V.A., Chesine P.A., Lamers B.E., Rubinsky-Elephant G. & Guiffrida R. 2011. Anti-*Toxocara* spp. antibodies in sheep from southeastern Brazil. *Veterinary Parasitology*. 179(1-3): 283-286.
- **14 Strube C., Heuer L. & Janecek E. 2013.** *Toxocara* spp. infections in paratenic hosts. *Veterinary parasitology*. 193: 375-389.