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9	ORIGINAL ARTICLE / ARTÌCULO ORIGINAL
10	ZOONOTIC FOCI OF TOXOCARA SP.: COMPARISON OF TWO TOURIST REGIONS IN
11	THE STATE OF SUCRE, VENEZUELA
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13	FOCOS ZOONÓTICOS DE TOXOCARA SP.: COMPARACIÓN DE DOS REGIONES
14	TURÍSTICAS EN EL ESTADO SUCRE, VENEZUELA
15	
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ABSTRACT

Toxocariasis is a human infection caused by the larvae of the nematodes *Toxocara canis* (Werner, 1782) or *Toxocara cati* (Schrank, 1788), which are parasitic in animals, and represents a serious public health problem. The objective of this study was to investigate the information and prevalence of parasitic toxocariasis in the state of Sucre, Venezuela, particularly in the municipalities of Montes and Sucre, in communities considered vulnerable due to social and economic deficiencies within the epidemiological/sanitary monitoring. In these municipalities, there are many stray dogs living near homes and maintaining contact with other domestic dogs. The seroprevalence of *T. canis* was evaluated through the ELISA method. A seroprevalence of IgG anti-*T. canis* was obtained in the parish of San Juan of 90.12% and in the parish of Cumanacoa of 82.14%. In the parish of San Juan, when evaluating the prevalence of parasites by species, *T. canis* was present in 18.37% of the feces of the canines evaluated. On the other hand, in the parish of Cumanacoa, *T. canis* was present in 9.90% of the canine samples analyzed; similarly, the presence of *Toxocara* sp. in soils in both parishes. In summary, the determination of two zoonotic foci of *Toxocara* sp. in the state of Sucre, Venezuela, since all the factors necessary for the transmission of toxocariasis are present.

Keywords: Dogs – Epidemiology – Larva migrans – Parasitic disease – *Toxocara canis* –

62 Zoonosis

RESUMEN

La toxocariasis es una infección humana causada por las larvas de los nematodos *Toxocara canis* (Werner, 1782) o *Toxocara cati* (Schrank, 1788), que son parásitos en los animales, y representa un grave problema de salud pública. El objetivo de este trabajo fue investigar la información y prevalencia de toxocariasis parasitaria en el estado Sucre, Venezuela, particularmente en los municipios de Montes y Sucre, en comunidades consideradas

vulnerables por deficiencias sociales y económicas en el monitoreo epidemiológico/sanitario. En estos municipios hay muchos perros callejeros que viven cerca de las casas y mantienen contacto con otros perros domésticos. La seroprevalencia de *T. canis* se evaluó mediante el método ELISA. Se obtuvo una seroprevalencia de IgG anti-*T. canis* en la parroquia San Juan de 90,12% y en la parroquia Cumanacoa de 82,14%. En la parroquia San Juan, al evaluar la prevalencia de parásitos por especie, *T. canis* estuvo presente en el 18,37% de las heces de los caninos evaluados. Por otra parte, en la parroquia Cumanacoa, *T. canis* estuvo presente en el 9,90% de las muestras caninas analizadas; Asimismo, la presencia de *Toxocara* sp. en suelos de ambas parroquias. En resumen, la determinación de dos focos zoonóticos de *Toxocara* sp. en el estado Sucre, Venezuela, ya que están presentes todos los factores necesarios para la transmisión de la toxocariasis.

Palabras clave: Epidemiología – Enfermedad parasitaria – Larva migrans – Perros – *Toxocara canis* – Zoonosis

INTRODUCTION

Toxocariasis is a human infection caused by the larvae of the nematodes *Toxocara canis* (Werner, 1782) or *Toxocara cati* (Schrank, 1788), which are parasitic in animals. The clinical manifestations of the disease are usually asymptomatic, which contributes to the disease being neglected. And yet, toxocariasis has been shown to be especially prevalent among children from socioeconomically disadvantaged populations, both in the tropics and subtropics and in industrialized nations (Macpherson, 2013; Rostami *et al.*, 2019; Ma *et al.*, 2020).

In humans, the infection is acquired accidentally, as there is no normal development of the parasite in humans, only the larval stage survives as it is a paratenic host in this parasitosis (Taylor *et al.*, 2017). Infection occurs by ingestion of infective eggs found in poorly washed vegetables and fruit originating from a contaminated environment, geophagy, ingestion of

undercooked paratenic hosts (chicken, beef or lamb), putting contaminated objects in the mouth, as well as not washing hands after playing with or petting infected dogs (Holland *et al.*, 1995; Vázquez *et al.*, 1997; Ulloque-Badaracco *et al.*, 2023). Children are the most infected due to their poor hygienic habits and are often in contact with dogs and puppies, as well as exposed to the pets' living environment (Cabral-Monica *et al.*, 2022).

Toxocariasis is a widely distributed zoonosis, in which the human need to keep pets and companion animals such as dogs and cats around ensures the persistence of the parasite and infection in humans over time. with rates ranging from 1% in Spain to 86% in Santa Lucia; from 3.6% to 24.7% in Brazil; 47.5% in Colombia; from 34.9% to 66.6% in Venezuela and from 37.9% (Roldán *et al.*, 2010a). In Argentina, a study carried out in children showed a 67.7% seroprevalence of anti-*Toxocara* IgG antibodies (López *et al.*, 2005), while in Paraguay a 78% seroprevalence for toxokaryosis was reported (Rivarola *et al.*, 2009). In Peru, a few reports have informed of frequencies from 7.8% to 44.92% in rural populations from different places (Roldán *et al.*, 2010a).

Toxocara canis infection in dogs has worldwide distribution rates ranging from 0 to 99.4% (Barriga, 1988; Heymann & American Public Health Association, 2004; Borges & Ferreira, 2020). In Venezuela, studies on *T. canis* infection were carried out in urban areas such as Caracas, reporting prevalences of 66.6%, in rural and suburban communities of the Capital District and the State of Miranda 20% and 70% (Incani, 1996; Delgado & Rodríguez-Morales, 2009). In the Brazilian Amazon, the prevalence in urban children < 5 years of age has been reported to be 21.5%, increasing to 26.7% in the rural general population, and to 26.8% or 52% in the riverine population (Oliart-Guzmán *et al.*, 2014). Furthermore, studies on canine toxocariasis carried out in Maracaibo reported a prevalence of 11.4% (Ramírez-Barrios *et al.*, 2004) and 63,16% in soil samples from leisure parks (Cazorla-Perfetti *et al.*, 2007). In eastern Venezuela, particularly in the state of Anzoátegui, (Fernández *et al.*, 2009) carried out a study

on children aged 6 to 8 years, obtaining a prevalence of 16%, and (Gómez, 2004) carried out a study in the Nueva Toledo neighborhood of El Peñón, state of Sucre, on children aged 2 to 15 years, with prevalences of 77.14%.

Thus, the present study evaluated the seroprevalence of toxocariasis in the municipalities of Montes and Sucre, which are regional tourist destinations in Venezuela. These regions were selected to be considered vulnerable due to social and economic deprivation, the large number of stray dogs found in the area, and the fact that most family groups have dogs as pets.

MATERIAL AND METHODS

Study area

The present study was conducted in the municipalities of Sucre and Montes, in the state of Sucre, Venezuela (Fig. 1). The municipality of Sucre is located in a mountainous area with a hot, semi-arid climate, with average temperatures of 27°C and 440 mm of precipitation with a rainy season from July to November (Pérez, 2006). The second site was Cumanacoa in the municipality of Montes. It is characterized by a hot sub-humid climate with temperatures ranging from 21°C to 27°C and annual precipitation between 900 and 1500 mm.



Figure 1. Map of the state of Sucre, Venezuela with the respective municipalities evaluated: Sucre (10,24198° N, 64,27980° O), Cumanacoa (10,24720° N, 63,92019° O), Google Maps.

Population sample

To calculate the representative sample size, a formula proposed by Martínez (2003) for finite populations, with a known sampling frame, was used. The prevalence for toxocariosis of 74.14% was considered (Gómez, 2004). In the parish of San Juan, 172 blood and faeces samples were taken from people who voluntarily attended the survey. The same occurred in the parish of Cumanacoa, where 100 blood and stool samples were taken, respectively. A clinical-epidemiological survey was administered to each individual, regardless of gender and age, in each parish, with prior informed consent (OPAS, 1993).

Human faecal samples

All the people who atended and participated in the collection were asked to provide a stool sample for the evaluation of the different helminths including *Ascaris lumbricoides* (Linnaeus, 1758). As an ascaridium, *A. lumbricoides* cross-reacts with *T. canis* when using immunological techniques using sera to evaluate for anti-*T. canis* antibodies. Serum samples of individuals presenting with *Ascaris lumbricoides* eggs in their stool was excluded from the study.

Human blood samples

Blood samples were obtained from the participants by venous puncture and transferred to the parasitology laboratory at the Instituto de Investigaciones en Biomedicina y Ciencias Aplicadas "Dr. Susan Tai", Universidad de Oriente (IIBCA-UDO), Venezuela, where they were processed for serum collection. The collected sera were frozen at -20°C until analysis.

Determination of anti-T. canis IgG antibodies by ELISA method

A commercial Toxocara-IgG ELISA kit (RIDASCREEN Toxocara-IgG), which uses

excretion-secretion antigens, was used for serological diagnosis. No absorption of sera with *Ascaris suum* (Goeze, 1782) antigen was performed. However, all individuals with *A. suum* eggs in their faeces were excluded from analysis. For the evaluation and interpretation of the results, the sample was considered as positive for anti-*T. canis* antibodies when its index was higher than 1.10, as a negative sample, that with an index lower than 0.90 and as an indeterminate sample that with an index between 0.90 – 1.10 (Fernández *et al.*, 2009).

Dog Fecal Samples

In both parishes, with the consent of the owners the dog fecal samples were collected in the morning, at midday and in the afternoon, and placed in airtight containers and transported to the laboratory. If not analyzed on the day of collection, they were stored at 4°C.

Soil samples

Soil samples were collected from the courtyards of homes in communities from the San Juan and Cumanacoa parishes. These samples were obtained by superficially scraping the courtyards with a spatula, 5 cm from any faecal matter after a random selection of sites with bare soil. Soil samples were collected upto 10 cm deep and stored in labeled polyethylene bags at room temperature until processing (Leventhal & Cheadle, 1992).

Parasitological diagnosis

Small portions of all the stool samples were examined by direct microscopic examination after mixing with 0.85% physiological saline and Lugol's solution (Botero & Restrepo, 1998). In addition, concentration and enrichment techniques such as: spontaneous tube sedimentation and flotation by Willis-Malloy were applied (Quinn *et al.*, 1980; Camacho *et al.*, 2006). It was also carried out with soil samples taken in each parish. These examinations were also done on soil samples taken in each parish.

Statistical analysis

The results obtained were expressed as percentages and represented in tables and

graphs. The seroprevalence of T. canis infection was determined by applying the formula described by Gordis (2004). The Chi-square test (χ^2) was used to establish the association between the clinical-epidemiological aspects evaluated and the seroprevalence of T. canis in the inhabitants of San Juan and Cumanacoa parishes (Martínez, 2003). Data was analyzed using SPSS 18 software.

Ethics aspects: This research was approved by the IIBCAUDO Bioethics and Biosafety Committee, (CBBIIBCAUDO-2009-09-3) year 2009, September, project 3.

RESULTS

The seroprevalence of *T. canis* was evaluated using the *Toxocara* IgG commercial kit. Of the 172 serum samples analyzed from the parish of San Juan, 10 samples were discarded because they belonged to people who had *A. lumbricoides* eggs in their faeces. Of the remaining 162 sera, 146 (90.12%) were positive, 9 (5.56%) indeterminate and 7 (4.32%) negatives. The average sample index of the positive cases was 3.03 of the indeterminate 0.99 and of the negative 0.55 (table 1). From the 100 serum samples analyzed from the Cumanacoa parish, Montes municipality, 16 sera were discarded because they belonged to people who had *A. lumbricoides* eggs in their stool samples. Of the 84 remaining sera, 69 (82.14%) were positive, 6 (7.14%) indeterminate and 9 (10.17%) negatives, the average sample rate for positive cases was 2.18, for indeterminate cases 0.96 and for negative cases 0.60 (table 1).

Table 1. Seroprevalence and summary statistics of sample rates for IgG anti-*T. canis* antibodies in inhabitases of San Juan and Cumanacoa (Sucre and Montes municipality), Sucre state, Venezuela.

	Inhabitants of San Juan / Cumanacoa								
Samples	n.	%	X	SD	LL	UL			
Positive	146/69	90,12/82,14	3,03/2,18	1,22/0,94	1,20/1,13	7,74/5,04			
Indeterminate	9/6	5,56/7,14	0,99/0,96	0,08/0,04	0,91/0,92	1,10/1,02			

Negative 7/9 4,32/10,17 0,55/0,60 0,14/0,21 0,37/0,34 0,73/0,8	Negative	7/9	4,32/10,17	0,55/0,60	0,14/0,21	0,37/0,34	0,73/0,89
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n: number of cases, χ : mean, SD: standard deviation, LL: lower limit, UL: upper bound %: percentage.

To understand the epidemiology of toxocariasis in both parishes of Sucre municipality, Sucre state, Venezuela, the seroprevalence of anti-*T. canis* IgG was analyzed according to gender; it was found that females were more affected in both San Juan and Cumanacoa parishes. However, the chi-square test did not show a statistically significant association (table 2).

Table 2. Association between genders of inhabitants and seroprevalence of anti-*Toxocara canis* IgG in San Juan and Cumanacoa (Sucre and Montes municipality), Sucre state, Venezuela.

IgG Anti- <i>Toxocara canis</i> in Inhabitants of San Juan / Cumanacoa										
•	Po	ositives	Inde	terminate	N	egatives		Total		
Gender	n	%	n	%	n	%	n	%		
Females	102/45	69.86/65,22	7/3	77.80/50	2/6	28.57/66.67	111/54	68.52/,64.29		
Males	44/24	30.14/34,78	2/3	22.20/50	5/3	71.43/33.33	51/30	31.48/35.71		
Total	146/69	100/100	9/6	100/100	7/9	100/100	162/84	100/100		
		$\chi^2 = 5,66$	3 ns	$p = 0.06 / \chi^2 =$	= 0,58 n	s p= 0,75				

n: number of cases %: percentage x2: chi-square ns: not significant p: probability

Table 3 show the seroprevalence of anti-*T. canis* IgG and its distribution according to age group, with significant results in the Cumanacoa and San Juan parishes where the 0-10 age group was the most affected. Curiously, the chi-square test showed no significant association within this age group from the San Juan parish, which was also the most affected.

Table 3. Association between age and seroprevalence of anti-Toxocara canis IgG in inhabitants of San Juan and Cumanacoa (Sucre and Montes municipality), Sucre state, Venezuela.

	Positives		Inc	leterminate	N	Negatives	Т	otal
Age (years)	n	%	n	%	n	%	n	%
0-10	55/38	37,67/55,07	5/4	55,56/66,67	6/4	85,71/44,44	66/46	40,74/54,76
11-20	31/13	21,23/18,84	1/0	11,11/0	0/0	0/0	32/13	19,75/15,48
21-30	21/9	14,38/13,04	1/0	11,11/0	0/3	0/33,33	22/12	13,59/14,29
31-40	13/1	8,90/1,45	0/0	0/0	1/1	14,29/11,11	14/2	8,64/2,38
41-50	4/1	2,74/1,45	0/2	0/33,33	0/1	0/11,11	4/4	2,47/4,76
51-60	11/4	7,53/5,80	2/0	22,22/0	0/0	0/0	13/4	8,02/4,76
61-70	7/3	4,80/4,35	0/0	0/0	0/0	0/0	7/3	4,32/3,57
71-80	3/0	2,05/0	0	0/0	0/0	0/0	3/0	1,85/0
81-90	1/0	0,70/0	0	0/0	0/0	0/0	1/0	0,62/0
Total	146/69	100/100	9/6	100/100	7/9	100/100	162/84	100/100
				χ^2 = 12.80 ns	p= 0.69		7	

n: number of cases %: percentage x2: chi-square ns: not significant *s: significant

p: probability

Table 4 show the seroprevalence of anti-*T. canis* IgG and its distribution according to handwashing practices before eating, food preparations and contact with soil by individuals in both parishes. There was no statistically significant association with any of the variables assessed by the chi-square test. This was contrary to the self-declared food handling and handwashing practices. Majority of those testing positive for anti-*T. canis* IgG antibodies individuals declared washing their hands before eating (84.93% San Juan parish) and (92.75% Cumanacoa parish) or cleaning their food together with handwashing before consumptio (75.34% for San Juan parish, and 85.51% for Cumanacoa parish). We also disclose that at the time of the surveys many of the people, particularly children, had dirty fingernails. In addition, if they washed their hands or wash their food (fruits and vegetables), they did so using water from the rivers that transversed these parishes. The water used was neither treated nor boiled, further compounding to the environmental contamination of these uncooked foodstuffs with *T. canis* eggs and other parasites. The results from both parishes were comparable.

Table 4. Association between hand washing practices before eating, foodstuff handling parctices, contact with soil and seroprevalence of anti-Toxocara canis IgG in

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	Positives		In	ndeterminate	1	Negatives	Total	
	n	%	n	%	n	%	n	%
Hand washing before eating								
Yes	124/64	84,93/92,75	7/5	77,78/83,33	7/8	100/88,89	138/77	85,19/91,67
No	22/5	15,07/71,25	2/1	22,22/16,67	0/1	0/11,11	24/7	14,81/8,33
Total	146/69	100/100	9/6	100/100	7/9	100/100	162/84	100/100
		$\chi^2 = 1.62$	2 ns p=	$0,44 / \chi^2 = 0,74 \text{ ns}$	p = 0.69			
Foodstuff washing and cleaning								
Yes	110/59	75,34/85,51	7/4	77,78/66,67	5/7	71,43/77,78	122/70	75,31/83,33
No	36/10	24,66/14,49	2/2	22,22/66,67	2/2	28,57/22,22	40/14	24,69/16,67
Total	146/69	100/100	9/6	100/100	7/9	100/100	162/84	100/100
		$\chi^2 = 0.09$	9 ns p=	$0.96 / \chi^2 = 1.63 \text{ ns}$	p=0,44			
Contact with soil								
Yes	92/47	63,01/68,12	7/3	77,78/50	6/4	85,71/44,44	105/54	64,81/64,29
No	54/22	36,99/31,88	2/3	22,22/50	1/5	14,29/55,56	57/30	35,19/35,71
Total	146/69	100/100	9/6	100/100	7/9	100/100	162/84	100/100

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An analysis of anti-T. canis IgG seroprevalences and their distribution with regard to presence and contact with dogs by the participants from the two parishes is shown in table 5. The sero-conversion status was not associated with the contact with dogs.

Table 5. Association between the presence of dogs, contact with dogs and the seroprevalence of anti-Toxocara canis IgG in inhabitants of San Juan and Cumanacoa (Sucre and Montes municipality), Sucre state, Venezuela.

		IgG ANTI-To	oxocara co	anis in Inhabitar	ts of San .	Juan / Cumanacoa		
	Positives		Inde	Indeterminate		Negatives		Total
	n	%	n	%	n	%	n	%
Dog present								
Yes	99/23	67,81/33,33	8/3	66,89/50	4/6	57,14/66,67	111/32	68,52/38,10
No	47/46	32,19/66,67	1/3	11,11/50	3/3	42,86/33,33	51/52	31,48/61,90
Total	146/69	100/100	9/6	100/100	7/9	100/100	162/84	100/100
		χ^2	= 2,19 ns	$p=0.33 / \chi^2=4$	1,14 ns p	= 0.13		
Contact with dogs								
Yes	60/48	41,10/69,57	7/6	77,78/100	3/4	42,86/44,44	70/58	43,20/69,04

No	86/21	58,90/30,43	2/0	22,22/0	4/5	57,14/55,56	92/26	56,80/30,96
Total	146/69	100/100	9/6	100/100	7/9	100/100	162/84	100/100
		χ^{2} =	5,25 ns	p= 0,07				

n: number of cases %: percentage x2: chi-square ns: not significant p: probability

These tables show the seroprevalence of anti-*T. canis* IgG and its distribution according to age, deworming and time of deworming of pet dogs of the participants from in both parishes. However, there were no significant associations established (Table 6).

Table 6. Role of age, deworming status, and time of deworming of pet dogs in the anti-Toxocara canis IgG seroprevalence of inhabitants of San Juan and Cumanacoa (Sucre and Montes municipality), Sucre state, Venezuela.

		IgG ANTI-To.	xocara c	anis in Inhabitant	s of San Juan /	Cumanacoa					
Variables	P	ositives	Inc	leterminate	Ne	gatives	s Total				
	n	%	n	%	n	%	n	%			
Age of dog											
6 months-1 year	41/13	41,41/56,52	4/0	50/0	4/3	100/50	49/16	44,14/50			
Older than 1 year	58/10	58,59/43,48	4/3	50/100	0/3	0/50	62/16	55,86/50			
Total	99/23	100/100	8/3	100/100	4/6	100/100	111/32	100/100			
$\chi^2 = 5.47 \text{ ns}$ p= 0.06 / $\chi^2 = 3.39 \text{ ns}$ p= 0.18											
Received dewormer											
Yes	39/14	39,40/60,87	3/2	37,60/33,33	1/5	25/83,33	43/21	38,74/65,63			
No	60/9	60.60/39,13	5/1	62,50/66,67	3/1	75/16,67	68/11	61,26/34,37			
Total	99/23	100/100	8/3	100/100	4/6	100/100	111/32	100/100			
		$\chi^2 =$	= 0,34 ns	$p=0.84/\chi^2=1$,	07 ns p=0.59	9					
Deworming time											
At birth	10/0	10,10/0	1/0	12,50/0	0/0	0/0	11/0	9,91/0			
Monthly	11/0	11,11/0	0/0	0/0	0/0	0/0	11/0	9,91/0			
Every 6 months	11/6	11,11/26,09	1/0	12,50/0	1/3	25/50	13/9	11,71/28,13			
Annually	7/8	7,07/34,78	1/2	12,50/33,33	0/2	0/33,33	8/12	7,21/37,50			
Never	60/9	60,61/39,13	5/1	62,50/66,67	3/1	75/16,67	68/11	61,26/34,37			
Total	99/23	100/100	8/3	100/100	4/6	100/100	111/32	100/100			
		$\chi^2 =$	= 3,16 ns	$p=0.92 / \chi^2=3$	35 ns p=0.50	0					

n: number of cases %: percentage x2: chi-square ns: not significant p: probability

It was observed that puppy owners do not provide adequate deworming treatment, of which after 3- or 4-weeks puppies begin to shed *T. canis* eggs freely into the soil, where they become infective. Consulted databases did not produce any published work on this parasitosis

that considered these epidemiological variables. Both parishes had different patterns of behavior in relation to these items, but no statistically significant association was found in either parish.

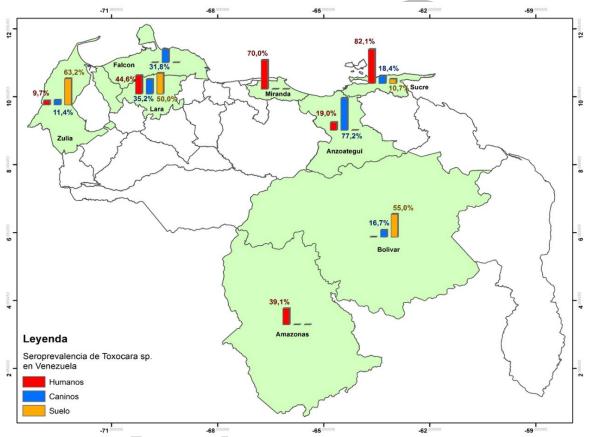
There was a statistically significant association with the collection of canine faeces by their owners in the parish of Cumanacoa, where 91.30% of the animals defecated in the yard and 78.26% of the owners did not collect the dog fecal matter. Thus, these faeces, together with the evolutionary forms of parasites in the contaminated fecal material can develop and be ready for infection (Table 7).

Table 7. Association between location of dog defecation, collection of dog faeces by dog owners, the frequency of dog faeces collection and disposal, and the seroprevalence of anti-*Toxocara canis* IgG in inhabitants of San Juan and Cumanacoa (Sucre and Montes municipality), Sucre state, Venezuela.

	IgG ANTI-Toxocara canis in Inhabitants of San Juan / Cumanacoa										
Variables	T	ositives	Ind	eterminate	Ne	gatives	-	Гotal			
variables	n	%	n	%	n	%	n	%			
Location of dog defecation											
Courtyard	81/21	81,82/91,30	7/2	87,50/66,67	4/5	100/83,3	92/28	82,88/87,50			
						3					
Thicket	17/0	17,17/0	0/0	0/0	0/0	0/	17/0	15,32/0			
House	1/2	1,01/8,70	1/1	12,50/33,33	0/1	0/16,67	2/4	1,80/12,50			
Total	99/23	100/100	8/3	100/100	4/6	100/100	111/32	100/100			
		$\chi^2 = 7,73 \text{ ns}$	p=0,1	$0/\chi^2 = 1,59 \text{ ns}$	p=0,45						
Collection of faeces											
Yes	51/5	51,52/21,74	6/1	75/33,33	4/5	100/66,6	61/26	54,96/81,26			
						7					
No	48/18	48,48/78,26	2/2	25/66,67	0/1	0/33,33	50/6	45,05/18,75			
Total	99/23	100/100	8/3	100/100	4/6	100/100	111/32	100/100			
		$\chi^2 = 5,05 \text{ ns}$	p=0,0	$8/\chi^2 = 8,01 \text{ ns}$	p=0.02						
Contact with the site											
Yes	51/20	51,52/86,96	6/2	75/66,67	4/4	100/83,3	61/11	54,96/34,37			
						3					
No	48/3	48,48/13,04	2/1	25/33,33	0/2	0/16,67	50/21	45,05/65,63			
Total	99/23	100/100	8/3	100/100	4/6	100/100	111/32	100/100			
		$\chi^2 = 5,05 \text{ ns}$	p= 0,0	$08/\chi^2 = 8.01 * s$	p= 0,02						

n: number of cases %: percentage χ2: chi-square ns: not significant *s: significant p: probability

In summary, we have established prevalence of the *Toxocara* infection in human subjects, dogs (pets and strays) and in soil samples obtained from the vicinity of the homes and playgrounds. Thus, we have identified two zoonotic foci of potential *Toxocara* sp. transmission and maintenance parasite life cycle in Sucre state. An epidemiological map was



charted comparing the situation in Sucre state with other states in Venezuela (Fig. 2).

Figure 2. Prevalence of *Toxocara* sp. in Sucre state compared to states in Venezuela with existing reports of *Toxocara* sp. studies.

DISCUSSION

The parish of San Juan, Sucre municipality, Sucre state, Venezuela, is a rural area

dedicated to agriculture and animal husbandry (pigs, cattle, and poultry) usage. These activities are both a source of income and food, as well as an area of regional tourism. The municipality of Montes carries out similar economic activities, especially agriculture and livestock farming with a special emphasis on sugar cane, coffee, vegetable crop production and cattle, poultry and pig rearing.

We found high seroprevalence of *T. canis* (Table 1), nonetheless, globally, the prevalences reported are lower than those of this study: in Colombia, a prevalence of 39% was reported, in Argentina 37% and in Brazil 28% (Ulloque-Badaracco *et al.*, 2023). However, a study in Paraguay (78%), but not in Turkey (7.6%) and Italy (31.87%) reports prevalences similar to those obtained in this study (Rivarola *et al.*, 2009; Akdemir, 2010; Qualizza *et al.*, 2011).

In Venezuela, most studies on *T. canis* infection in humans have been carried out in the central north, west and south of the country. In Amazonas, a seroprevalence of 19.66% was found (Oliart-Guzmán *et al.*, 2014), in preschool children in Aragua state, Venezuela, were detected in 29% (65/224) of children. The seroprevalence in the different preschools studied ranged between 4.2% and 60.6% (Martínez *et al.*, 2015). In Zulia state, García *et al.* (2004) found a prevalence of 9.72% while a study conducted in Barquisimeto, a seroprevalence of 44.60% was reported (Delgado *et al.*, 2009).

Few studies have been conducted on *T. canis* infection in eastern Venezuela. Gómez (2004), for instance, obtained a 77.14% prevalence in Sucre State, using the ELISA method, while Fernández *et al.* (2009), using the RIDASCREEN ELISA technique, reported a prevalence of 19.0% in Anzoátegui State. It is noteworthy that participants evaluated in this study, from both parishes, have high prevalence rates, a reflection of zoonotic foci present in these communities.

The seroprevalence of anti-T. canis IgG according to gender, was found that females were

more affected in both San Juan and Cumanacoa parishes (Table 2). Comparable results were obtained by Espinoza *et al.* (2003) and Gómez (2004), who found a higher seroprevalence in females with 57.80% and 61.11%, respectively. However, these findings differ from those by Martín *et al.* (2008), Delgado *et al.* (2009), Akdemir (2010) and Wisniewska *et al.* (2011) who report that 57.63%, 51.86%, 77.80% and 62.14%, respectively, were in males. Nevertheless, Roldán *et al.* (2010b) mentioned that the gender of the individual seems not to be an important predisposing factor for toxocariasis in human populations.

The results found in the age group of both municipalities coincide with those reported by various authors such as Aguiar-Santos *et al.* (2004) who reported a prevalence of 60% in children under 10 years of age and Gómez (2004) who reported a prevalence of 40.74% in children aged 2-4 years. Similarly, López *et al.* (2005) reported 67.70% in children aged 1-14 years while Fernández *et al.* (2009) recorded a prevalence of 22.50% in children aged 6 years and Rivarola *et al.* (2009) found a prevalence of 64.15% in children aged 3 to 9 years. However, none of these researchers found age-group specific differences in the occurrence of toxocariasis. García *et al.* (2004) suggested that the reduced risk of infection in older individuals might be due to improved hygienic habits and spending more time away from home thus distancing them from sources of infection. It was proposed that this parasitic infection was most likely to occur at home more than at school or at work, where sanitary and infrastructural conditions were better.

It is important to highlight that in the two parishes; the schools are close to rivers and surrounded by fields, where children carry out their recreational activities, and where dogs defecate openly consequently depositing parasite eggs. Additionally, many of these communities and schools lack piped water and thus draw water from the rivers for consumption by children and residents. Subsequently, residents of these communities, including school children, are exposed to contracting this parasitosis both at home and in their schools.

These results differ from those obtained by Acero *et al.* (2000), who found that 46.15% of children never washed their hands before eating and by Gómez (2004) who found that 62.96% of the *Toxocara* seropositive children did not wash their hands before eating, but 100% of them declared washing their food before consumption, particularly those to be eaten raw.

Most participants from San Juan (63,01%) and Cumanacoa (68,12%), had contact with *T. canis* soil contaminated, had parasitized dogs and did not wash their hands well before eating or ingesting food. These participants were predisposed to high risk of easily getting infected. These rates were comparable to results found by Acero *et al.* (2000) who reported that 53.85% of toxocara-infected children consumed soil, had poor hygiene practices, López *et al.* (2003) found that 30% of toxocariasis positive children had a history of geophagy whilst Delgado *et al.* (2009) noted that 96.3% of the infected children had a history of geophagy.

In both evaluated parishes, there were large tracts of land, contaminated with canine faeces, and consequently *T. canis* eggs. At the same time, these fields served as playgrounds for the children or fruit and vegetable gardens.

Concerning the presence of dogs in the homesteads, 67.81% of the seropositive individuals in the San Juan parish stated that they had dogs as pets, which differed from those of Cumanacoa parish (33.33%). However, it should be noted that majority of the inhabitants from both parishes declared coming in contact with other canines in the area due to the large and uncontrolled population of stray dogs found in both parishes. Gómez (2004) found that 52.26% of seropositive participants had dogs while Martín *et al.* (2008) reported that 74.57% of those seropositive for IgG anti-*T. canis* antibodies owned dogs. However, in a study evaluating playgrounds in Turkey, Akdemir (2010) found that only 7.60% of those seropositive had dogs. We note that having dogs as pets was not significantly associated with testing positive for the IgG anti-*T. canis* antibodies.

Fan et al. (2004) argued that toxocariasis is not related to dog breeding, but dog breeding

is a condition that in most cases contributed to the occurrence of infection, especially in low socio-economic societies. Similarly, Alonso *et al.* (2004) pointed out that neither direct contact with dogs, even puppies, nor the presence of the animals in the home are sufficient conditions for acquiring *T. canis* infection; other epidemiological circumstances had to coincide to favor the risk of an infection being acquired from a contaminated environment.

Some studies suggest that the likelihood of infection had more to do with individual susceptibility, and behavioral and hygienic habits rather than with the environmental factors present in the population. Considering this, we sought to understand if epidemiological variables such as age of the canines present, deworming and time of deworming of the individuals who indicated that they owned dogs and the seroprevalence of anti-*T. canis* IgG.

The majority of the seropositive individuals in the parish of San Juan stated that their dogs defecated in the yard (81.82%), 51.52% reported collecting dog faeces and frequenting the place of disposal, 27.27% disposed of these faeces in the thicket and 15.15% accumulated them until the waste collection day. This was indicative that these individuals also had frequent and direct contact with *T. canis* contaminated dog faeces in addition to being surrounded by animal waste subsequently increasing the possibility of parasite transmission and reinfection. In the databases consulted, no published studies on this parasitosis were found that took these epidemiological variables into account.

One of the main sources of *T. canis* infection found within the homesteads are gardens and yards heavily contaminated with dog faeces. Additionally, the ease with which unwormed dogs access homes greately contributes to this contamination and high likelihood of infection. It is estimated that one gram of faeces from an infected animal can harbour about 10.000 *T. canis* eggs, while a female parasite can shed up to 200.000 eggs daily into the environment. Larval eggs can survive up to 10 years in the environment if conditions are right, thanks to their high resistance (García *et al.*, 2004).

Taking into account the observations by García *et al.* (2004) and Alonso *et al.* (2004), geophagia, the high dog population, the manner in which dogs are kept without restrictions of space or specific places to defecate, and the lack of collection of excrement from stray dogs, the parishes of San Juan and Cumanacoa have become a favorable place for the transmission of this zoonosis. In these localities, the prevalent conditions expose the inhabitants, especially children, to the parasite-contaminated excrement.

The population sample studied corresponded to individuals from two different socioeconomic and cultural backgrounds, but with the same social deficiencies. This could explain why no major differences in risk factors association were found between serology-positive and serology-negative patients.

Additionally, a representative sample of canine faeces was also taken and evaluated for parasitological contamination in these communities. In the parish of San Juan, 93.87% (46 of 49) of the canine faecal samples were parasitized of which the prevalence of *T. canis* was 18.37%. Similarly, in the parish of Cumanacoa, 65.40% (53 of 81) of the samples were parasitized, of which 9.90% were the *T. canis* species. This is indicative of which shows the risk of infection with *T. canis* in both communities. These findings are similar to those of Ramírez-Barrios *et al.* (2004) and Devera *et al.* (2008) that showed a 11.4% and 16.7% prevalence of canine intestinal parasites in Venezuela, specifically in the city of Maracaibo and Ciudad Bolívar.

In addition to the faecal samples, soil samples were also analyzed. Of the 35 and 28 soil samples from San Juan and Cumanacoa parishes analyzed, 5.0% and 10.71% were positive for *Toxocara* sp. Eggs, respectively. We however declare that at the time of soil collection, the weather condition in the localities was unpredictable, with constant and torrential rains, such that the parasite eggs in the soil could have been washed by Surface run off into the rivers that flow through the communities. Despite the weather conditions, the presence of *Toxocara* sp.

eggs in the soil of both parishes in Sucre municipality, Sucre State, Venezuela, demonstrates the high risk of contact and possible accidental ingestion by children in these communities. This subsequently predisposes them to acquiring the Visceral or Ocular Larva Migrans syndrome, in addition to advancing high infection rates in dogs. Very heterogeneous result was reported by Laiño *et al.* (2024), (1.5%) and Antonio *et al.* (2020) (32.5%).

Comparing with other studies around Venezuela, from the two zoonotic foci evaluated in Sucre State demonstrates that this region has one of the highest seroprevalences of toxocariasis in humans. However, Anzoátegui State reports the highest prevalence of *Toxocara* sp. in canines while Zulia State has the highest rate of soil infected with *Toxocara* sp. eggs (Cazorla-Perfetti *et al.*, 2007; Tortolero *et al.*, 2008; Devera, 2008; Fernández *et al.*, 2009; Nieves *et al.*, 2012; Apóstol *et al.*, 2013;). We are of the opinion that the high seroprevalence is attributed to the rural condition of both parishes evaluated in Sucre State. It is further compounded by the lack of a Zoonosis department in the local government to manage the large number of stray dogs in the communities and promote civic education on disease, dynamics of transmission, consequences of infection.

As a zoonosis and geohelminthiasis, this parasitosis represents a public health problem for both parishes in Sucre State. Its epidemiology depends closely on maintaining the chain of infected domestic and/or stray dogs and cats, on human contact with soil contaminated with the infected and on the general habits and attitudes that tend to acquire the infection. It is crucial that the public health authorities implement a control and health education plan to raise awareness of the forms of contagion of this parasitosis and the most appropriate way to prevent it. Primary prevention should focus on veterinarial control of pets, culling of stray dog populations and improving habits and attitudes that tend to maintain the infection in both municipalities of Sucre State, Venezuela.

447 448 **EGM** = Erika Gomez-Martinez 449 **MVGV** = Márcia Viviana Gonçalves-Vallejos **MBBR** = María Betania Bravo-Rodríguez 450 **MTD** = Marcos Tulio-Díaz 451 452 **ZSG** = Zulay Simoni-Gonzalez **DVG** = Del Valle Guilarte 453 454 **EFS** = Elizangela Farias-da Silva 455 **ZMS** = Zeca Manuel-Salimo **LSX** = Luziana de Sousa-Xavier 456 457 **GOP** = Grafe Oliveira-Pontes Conceptualization: EGM, GOP, ZSG, DVG 458 Data curation: GOP, LSX, ZMS 459 460 Formal Analysis: EGM, DVG, ZSG, GOP 461 Funding acquisition: EGM, GOP, LSX, MBBR, MVGV, MTD, ZMS, ZSG, DVG, EFS 462 Investigation: EGM, DVG, ZSG, MTD, MBBR, EFS Methodology: EGM, DVG, ZSG 463 464 Project administration: EGM, DVG, ZSG 465 Resources: EGM, DVG, ZSG 466 Software: EGM, DVG, ZSG Supervision: EGM, ZMS, GOP 467 Validation: EGM, GOP 468 Visualization: EGM, GOP 469 Writing - original draft: EGM, GOP, LSX, MVGV, ZMS 470 Writing - review & editing: EGM, GOP, LSX, MBBR, MVGV, MTD, ZMS, ZSG, DVG, 471 472 **EFS** 473 474 **ACKNOWLEDGMENTS** 475 This work was financed by Instituto de Investigaciones Cientificas en Biomedicina y Ciencias 476 Aplicadas, "Susan Tai" IIBCAUDO, the Universidad de Oriente, estate Sucre, Venezuela. The 477 authors would like to thank all the participants and their families who accepted to participate in

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