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EPIDEMIOLOGICAL ASPECTS AND RISK FACTORS OF TOXOCARIASIS IN A PEDIATRIC POPULATION IN SRI LANKA

SD Fernando¹, VP Wickramasinghe², GMG Kapilananda¹, RL Devasurendra¹,
JDMS Amarasooriya³ and HGAK Dayaratne³

¹Department of Parasitology, ²Department of Pediatrics, ³Faculty of Medicine,
University of Colombo, Sri Lanka

Abstract. This cross-sectional study, carried out over a period of 11 months, investigated the relationship between *Toxocara* seropositivity, socio-demographic and environmental variables in a pediatric population. Risk factors for *Toxocara* infection were assessed by direct interview of parent or guardian using a structured pre-tested questionnaire. Eosinophilia and presence of helminth eggs or protozoan cysts in a fecal smear were recorded. Diagnosis of *Toxocara* seropositivity in children was based on IgG *Toxocara* Microwell Serum Elisa Kits. The ELISA test was regarded as positive if the optical density was 0.3 units or above. Unadjusted and adjusted odds ratios were calculated to determine risk factors for disease. The proportion of children who were positive for *Toxocara* antibodies in the study population was 20%. Children being exposed to a puppy of less than 3 months at home, visiting a playground frequently, living in a poorly constructed house and dogs having access to playgrounds were significant risk factors on univariate analysis. Of these four variables, only the first three variables (OR 19, OR 4 and OR 3, respectively) remained significant risk factors on the multivariate model. Presence of eosinophilia in seropositive children was significantly higher than the seronegative group (77% vs 40%; $p < 0.001$). This study indicates that dogs contribute significantly to children being seropositive for toxocariasis in Sri Lanka. Implementation of public health programs specifically focused on anti-parasitic treatment of dogs is recommended.

INTRODUCTION

Human toxocariasis is caused by *Toxocara cati* and *Toxocara canis*, nematode parasites which live as adult worms in the proximal intestine of cats and dogs, respectively. Most human infections occur in young children and are classically associated with geophagia. The three clinical forms of toxocariasis are visceral larva migrans, ocular larva migrans and covert toxocariasis. Most

cases of visceral larva migrans (VLM) are characterized by frequent fever, hepatomegaly and eosinophilia. The ocular form can cause a spectrum of ocular disease and covert toxocariasis is associated with non specific symptoms (Coombs and Crompton, 1991; Gillespie, 1993).

The seroprevalence of toxocariasis in children living in different countries varies widely, as shown by different epidemiological studies. It is approximately 10% in temperate populations (Schantz, 1989) while in selected areas with low socio-economic conditions, higher prevalences of 47% and 83% have been documented (Thompson *et al*, 1986; Radman *et al*, 2000). In Sri Lanka, a positive

Correspondence: Dr SD Fernando, Department of Parasitology, Faculty of Medicine, PO Box 271, Kynsey Road, Colombo 8, Sri Lanka.

E-mail: deepfern@slt.lk

Tel: +94 11 2695300 ext 178; Fax: +94 11 2699284

antibody carriage of 43% for toxocariasis in the 1-12 year age group has been reported (Iddawela *et al*, 2003). Childhood infection is associated with several risk factors, such as pica, pet ownership, poor sanitation and lack of health awareness (Glickman *et al*, 1981; Thompson *et al*, 1986; Marmor *et al*, 1987; Iddawela *et al*, 2003).

This study provides information about the epidemiology and risk factors of toxocariasis in a pediatric population in Sri Lanka.

SUBJECTS AND METHODS

Study site

The study was carried out at the University Pediatric Unit of Lady Ridgeway Hospital, the only Children's hospital in Sri Lanka that serves as the National Referral Center for pediatric care.

Study design

A cross-sectional study was conducted from April 2006 to February 2007. The study population comprised 196 children age 5-12 years presenting to the hospital. One hundred of the children included in the study population had confirmed bronchial asthma. The other 96 children were children who presented to the hospital for routine check-ups for non-respiratory related medical conditions. Children were enrolled after obtaining written informed consent from their parents or guardians.

Children whose parents/guardians did not give consent and children with any illness that required long term management, such as those suffering from renal, cardiac, or endocrine problems or genetic diseases were excluded from the study.

Determination of risk factors for toxocariasis

Risk factors for *Toxocara* infection were assessed by direct interview of a parent or guardian using a structured pre-tested questionnaire. Exposure to domestic pets at home,

geophagia, weekly contact with a playground or field and the socio-economic status of the family were considered as the risk factors for acquiring toxocariasis. Information on worm treatment of pets and access of dogs to play areas were recorded.

Assessment of the socio-demographic status of the family

The socio-demographic status of the family was assessed by educational status of parents, monthly family income and house type. The educational status of the parents who were deceased was not recorded. The houses were classified as "good" or "poor" depending on the materials used for roof, floor and walls as previously defined (Mahawithanage, 2005). The area of residence was classified as urban or rural based on the classification made in a Census and Statistics report for 2001 (Department of Census and Statistics, 2004).

Measurement of seropositivity for toxocariasis and other hematological investigations

An aliquot of 2 ml of blood was collected under aseptic conditions by trained nurses and divided equally into EDTA (ethylenediaminetetraacetic acid) and plain tubes. The samples in the plain tubes were centrifuged at 800g for 5 minutes after which the sera were collected and stored at -20°C until used. Diagnosis of *Toxocara* seropositivity was based on IgG *Toxocara* Microwell Serum Elisa Kits (Diagnostic Automation Inc: Cat No. 8206-3). The ELISA test was regarded as positive if the optical density was 0.3 units or above (as indicated on the instructions sheet).

The sample collected into the EDTA tube was used to perform a white blood cell count using the hemocytometer and a differential count using a counting chamber. An eosinophil count over 400/mm³ was regarded as being high.

The technical officers performing the investigations were blind to the clinical status, age and sex of the child.

Stool examination for intestinal helminths

A sample of feces collected from each child was examined by a trained technician or consultant parasitologist for helminth ova and protozoan cysts by saline and iodine smears.

Data analysis

Data were analyzed with the statistical package SPSS version 15.0. The relationship between selected risk factors and presence of toxocariasis was determined using unadjusted odds ratios (ORs). The significant risk factors were included in a logistic regression multivariate model.

Ethical considerations

Ethical clearance was obtained from the Ethical Review Board of the Faculty of Medicine, University of Colombo, Sri Lanka. To protect the identity of the children, the blood samples and questionnaires were assigned a study number that was kept separate from patient identifying information.

RESULTS

Characteristics of the population

The socio-demographic and clinical characteristics of the study population are shown in Table 1. The proportion of children who were positive for *Toxocara* serology in the study population was 20% (39 out of 196 children). Of these 39 children, 29 (29%) had bronchial asthma and 10 (10.4%) were children without asthma. Twenty-two percent of male children and 17% of female children recruited for the study were positive for *Toxocara* serology. When the different age groups were compared, the seropositivity was similar in all three age groups. There was no significant difference in *Toxocara* seropositivity with regard to the sex or age of the child, whether the house was located in a rural or urban location or the socio-demographic characteristics.

Eosinophilia in peripheral blood was detected in a significantly higher number of se-

ropositive children ($\chi^2=15.78$; $p<0.001$). Approximately 65% of the parents returned a feces sample for the child. All samples tested negative for helminth ova and cysts

Risk factors for toxocariasis

Table 2 displays the unadjusted ORs determined through univariate analysis for toxocariasis and risk factors previously identified in published literature. Children who were exposed to puppies less than 3 months old at home were at 17 times higher risk of developing toxocariasis (OR, 17.51; 95% CI, 5.67-56.57). They also visited a playground at least once a week, and were thereby at a three times greater risk of being *Toxocara* seropositive compared to children who had visits at less regular intervals (OR, 3.36; 95% CI 1.50-7.63). A greater proportion of parents (53.8%) whose children were positive for toxocariasis indicated that dogs had free access to these same grounds, compared to 27% of parents of children who were *Toxocara* negative and indicated that dogs did not have access to the playground (OR, 3.09; 95% CI 1.42-6.77). No significant association was found between *Toxocara* seroprevalence and the other selected risk factors for the disease.

Of the four variables included in the logistic regression model, (namely house type, exposure to puppies less than 3 months of age, frequency of visiting the playground and access of dogs to the playground), access of dogs to playgrounds did not meet the significance level to enter into the final model. The adjusted ORs for exposure to puppies under 3 months of age, house type and frequency of visiting the playground were, 19.0 (95% CI, 6.2-58.1), 3.3 (95% CI 1.1-9.8) and 3.7 (95% CI 1.5-8.8), respectively.

DISCUSSION

This study describes the epidemiological and other risk factors for toxocariasis in a hospital population. The study population com-

Table 1
Characteristics of the study population.

	No. surveyed	<i>Toxocara</i> IgG ELISA positive (%)	χ^2	p
Socio-demographic characteristics				
Sex			0.82	0.367
Male	108	24 (22.2)		
Female	88	15 (17.0)		
Age in years			0.17	0.918
4-6	72	15 (20.8)		
7-9	76	14 (18.4)		
10-12	48	10 (20.8)		
Ethnic group ^a			0.21	0.645
Sinhalese	159	31 (19.5)		
Tamil	12	3 (25)		
Muslim	25	5 (20)		
Place of residence			2.44	0.118
Urban	79	20 (25.3)		
Rural	117	19 (16.2)		
Father literacy ^a			0.95	0.328
Completed O-levels	129	23 (17.8)		
Primary or secondary	63	15 (23.8)		
Mother literacy ^a			2.77	0.090
Completed O-levels	116	24 (20.7)		
Primary or secondary	78	9 (11.5)		
Monthly family income (SLR) ^b			0.93	0.334
> 10,000.00	97	22 (22.7)		
< 10,000.00	99	17 (17.2)		
House type			5.32	0.02
Good	172	30 (17.4)		
Poor	24	9 (37.5)		
Clinical characteristics				
Presence of physician diagnosed asthma			10.61	0.001
Yes	100	29 (29)		
No	96	10 (10.4)		
Eosinophilia			15.78	<0.001
Yes	95	30 (31.6)		
No	101	9 (8.9)		

^aTotal number of subjects who responded to maternal and paternal education status differs according to questionnaire response for each variable.

^b100 SLR= 1 US\$

prised 196 children age 5-12 years who presented to the University Pediatric Unit of Lady Ridgeway hospital for children, Colombo. The overall proportion of *Toxocara* serology posi-

tive children was 20% with a greater percentage being under 9 years of age. These figures are lower than reported in a previous study carried out in the Central province of Sri Lanka

Table 2
Risk factors for toxocariasis.

Risk factor	Seropositive for <i>Toxocara</i> n=39		Seronegative for <i>Toxocara</i> n= 157		OR	95% CI
	n	%	n	%		
Ethnic group						
Minority groups ^a	8	21.6	29	78.4	-	-
Sinhalese	31	19.5	128	80.5	1.14	0.43-2.93
Place						
Urban	20	25.3	59	74.7	-	-
Rural	19	16.2	98	83.8	0.57	0.27-1.23
Father literacy						
Completed O-levels	23	17.8	106	82.2	-	-
Primary or Secondary	15	23.8	48	76.2	1.44	0.65-3.19
Mother literacy						
Completed O-levels	24	20.7	92	79.3	-	-
Primary or Secondary	9	17.9	69	82.1	0.84	0.38-1.85
Monthly family income (SLR) ^b						
> 10,000.00	22	22.7	75	77.3	-	-
< 10,000.00	17	17.2	82	82.8	0.71	0.33-1.51
House type						
Good	30	17.4	142	82.6	-	-
Poor	9	37.5	15	62.5	2.84	1.03-7.73
Exposure to adult dogs						
No	33	19.1	140	80.9	-	-
Yes	6	26.1	17	73.9	1.50	0.48-4.45
Exposure to puppies (0-3 months old)						
No	23	13.2	151	86.8	-	-
Yes	16	72.7	6	27.3	17.51	5.67-56.57
Exposure to puppies (3-12 months old)						
No	36	18.8	155	81.2	-	-
Yes	3	60.0	2	40.0	6.46	0.84-57.74
Exposure to cats						
No	34	19.8	138	80.2	-	-
Yes	5	20.8	19	79.2	1.07	0.32-3.33
Exposure to kittens						
No	37	19.6	152	80.4	-	-
Yes	2	28.6	5	71.4	1.64	0.21-10.12
Regular worming of pets						
Yes	7	28.0	18	72.0	-	-
No	32	18.7	139	81.3	0.59	0.21-1.72
Playground play						
Not weekly	12	11.3	94	88.7	-	-
At least once a week	27	30.0	63	70.0	3.36	1.50-7.63
Access of dogs to playground						
No	18	13.6	114	86.4	-	-
Yes	21	32.8	43	67.2	3.09	1.42-6.77
Geophagia						
No	29	18.0	132	82.0	-	-
Yes	10	28.6	25	71.4	1.82	0.73-4.51
Hand washing						
Yes	29	23.6	94	76.4	-	-
No	10	13.7	63	86.3	0.51	0.22-1.20

^aMinority ethnic groups include children belonging to Muslim and Tamil ethnic groups, ^b100 SLR= 1 US\$. Total number of subjects who responded to maternal and paternal education status differed according to questionnaire response for each variable.

by Iddawella *et al* (2003) where a prevalence of 43% was reported in a community-based study. Younger children are at higher risk for developing both infection and disease because of their habits of play and placing fingers in their mouths and being less careful about hygienic practices.

Among the risk factors investigated, the exposure to a puppy less than 3 months old, visits to a playground at least once a week, dogs having regular access to these playgrounds and living in a poorly constructed house were significantly associated with presence of *Toxocara* antibodies on univariate analysis. *Toxocara canis* is an important causative agent of human disease and puppies less than 10 weeks of age (infected transplacentally) and lactating bitches are the typical reservoir of this parasite (Shoop, 1991; Jacob, 2000). Adult female *T. canis* worms can excrete up to 200,000 eggs per day and the eggs become embryonated 2-5 weeks after being passed by the animal. Human beings, especially children, are infected after contact with contaminated soil containing the embryonated eggs (Josephson, 1988). As most puppies are born with worms, deworming should commence as young as 2 weeks and should continue every 2 weeks until the pup is 12 weeks old (Anonymous, 2007). The deworming of pets in Sri Lanka depends on individual families that own pets and the advice they receive from the veterinary surgeons.

Children who visit a playground at least once a week and those who live in poorly constructed houses were approximately at a three times greater risk of being seropositive for *Toxocara*. Playgrounds in Sri Lanka are not surrounded by protective fences and are thereby freely accessible to stray dogs as indicated by the majority of parents (58%) whose children visit a playground regularly. Most schools in Sri Lanka have sandy play areas. Similarly poorly constructed houses in Sri Lanka are generally not enclosed by bound-

ary walls. Stray dogs which roam the streets have free access to these premises. Those areas inaccessible by dogs may still be readily accessible by cats. The prevalence of soil contamination with *T. canis* eggs has been recorded as 47% of public places in the Kelaniya Secretariat division in the suburbs of Colombo, with a higher prevalence being recorded in places covered with grass which is typically seen in playgrounds (Ransiri *et al*, 2005).

When all the factors were analyzed using backward logistic regression, exposure to puppies under 3 months of age, weekly visits to a playground and residence in a poorly constructed house remained significant in the final model. Dogs having access to playgrounds not being significant in the final model may be due to the fact that this variable is related to the other 3 significant variables. However, the increase in the odds ratio from 17 (unadjusted) to 19 (adjusted) for exposure to puppies less than 3 months indicates the significant role played by puppies in the development of *Toxocara* seropositivity in children. Similar patterns were observed in regard to the frequency of visits to a playground and residing in a poorly constructed house.

Seventy-seven percent of children had a high eosinophil count. Like many other tissue helminths, *T. canis* larvae induce a Th2 type CD4+ cellular immune response characterized by eosinophilia and IgE production, which is likely to be induced by parasite excretory-secretory antigens (Del Prete *et al*, 1991). In murine models of infection with *T. canis* the dominant cytokine produced is interleukin-5 (Takamoto *et al*, 1995), which plays a major role in the induction of local tissue and peripheral eosinophilia (Parsons *et al*, 1993; Kusama *et al*, 1995). Absence of other soil transmitted helminth or protozoan infections in children whose feces were examined for ova and cysts is probably due to regular administration of anthelmintics by the parents (Fernando *et al*, 2007), but which were not

effective against *Toxocara* species.

The high proportion of children positive for toxocariasis in this study population (20%) indicates that prevention and control methods against *Toxocara* infection should be reinforced in the community. *T. canis* is common in dog populations in the tropics, especially where unleashed dogs roam and forage freely and receive little veterinary attention, a common sight in Sri Lanka. Human toxocariasis can be minimized by public health measures to prevent dog feces containing *T. canis* eggs from contaminating the environment and thereby preventing accidental ingestion of embryonated eggs. Contamination can be decreased in public areas by restrictions on uncontrolled dogs and cats, collection of feces by dog owners, regular deworming of pets at home and preventing animal access to public places, such as children's playgrounds. Such control measures for canine toxocariasis are lacking in developing countries, such as Sri Lanka, where toxocariasis represents an ongoing and poorly recognized parasitic infection.

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