Veterinary Parasitology 244 (2017) 39–43

Contents lists available at ScienceDirect

Veterinary Parasitology

journal homepage: www.elsevier.com/locate/vetpar

Research paper

Protective and risk factors associated with the presence of *Toxocara* spp. eggs in dog hair

Ysllla Fernanda Fitz Balo Merigueti[a], Vamilton Alvares Santarém[a,*], Lívia Magosso Ramires[a], Aline da Silveira Batista[a], Layron Vinícus da Costa Beserra[b], Amábyle Lopes Nuci[b], Talita Mirella de Paula Esposte[b]

[a] Post-Graduate Program in Animal Science, Laboratory of Veterinary Parasitology, Veterinary Teaching-Hospital (UNOESTE), Presidente Prudente, São Paulo, Brazil
[b] Laboratory of Veterinary Parasitology, Veterinary Teaching-Hospital, Universidade do Oeste Paulista (UNOESTE), Presidente Prudente, São Paulo, Brazil

ARTICLE INFO

Key-words:
Dog's hair
Toxocariasis
Larva migrans
Zoonosis

ABSTRACT

Toxocariasis is one of the most prevalent parasitic zoonoses in the world. The disease is principally caused by the nematode *Toxocara canis*, whose definitive host is the dog. The transmission of toxocariasis to humans is mainly caused by accidental ingestion of embryonated eggs of the parasite, present in the soil. Studies have shown that dog hair has the capacity to harbor eggs of the parasite and represents a risk for transmission of the zoonosis. The objective of the present study was to evaluate the frequency and factors associated with the contamination of dog hair by *Toxocara* spp. of animals attended and/or abandoned at a Veterinary-Teaching Hospital in Southeast Brazil. The hair samples were collected from the perineal region, and upper and lower tail regions. For analysis of the samples and recovery of *Toxocara* spp., the material was washed in Tween 20 and then filtered through sieves of 300 μm, 212 μm, and 38 μm. Hair samples from 165 dogs were analyzed. Of the analyzed samples, 59 (35.8%) were from puppies and 106 (64.2%) from adult animals. In the sample evaluation, 6.7% of the dogs (11/165) were contaminated, with a mean of 12.2 eggs per animal (1–70 eggs/animal) and 57.5 eggs/gram of hair. All the recovered eggs were not embryonated. There was an influence of age (puppies), breed (without defined breed), and origin (stray) of the dogs. On the other hand, deworming was a protective factor. Our results show that the risk of transmission of toxocariasis by direct contact, mainly in well-cared dogs, is low. Thus, prophylactic anthelmintic treatment and correct care regarding the hygiene of animals, especially puppies, should be recommended to reduce any risk of transmission of toxocariasis.

1. Introduction

The relation between humans and domestic animals, especially dogs, is becoming ever closer, more frequent, and important, particularly with regard to the physical and psychological benefits (Scheibeck et al., 2011). Although this relationship brings broad benefits to humans, there is a danger of transmission of pathogens associated with the lack of knowledge of the zoonotic risks that these animals represent to the owners (Stull et al., 2012).

The WHO (2017) estimates that in Latin America, 100 out of every 100,000 inhabitants are affected by at least one zoonosis of a parasitic nature. Zoonoses caused by parasitic agents are among the most frequent (Molyneux, 2011) and are usually associated with the presence of animals in environments frequented by humans (Marques et al., 2012).

Among the helminthic zoonoses, toxocariasis is one of the most prevalent in both industrialized and developing countries (Magnaval et al., 2001); however, it is included in the list of neglected diseases (Zinsstag et al., 2007).

Human infection is principally caused by the nematode *Toxocara canis*, whose definitive host is the dog (Barriga, 1991). The disease can lead to the erratic migration of larvae through the tissues, and as humans are not the usual hosts, the parasite cannot complete the evolutionary cycle. These larvae may remain viable for prolonged periods and cause various clinical manifestations (Fu et al., 2014), depending on the affected organ and number of infective larvae (Magnaval et al., 2001).

The clinical spectrum of toxocariasis in humans, ranging from asymptomatic infection to severe organ injury, including respiratory (Yoshikawa et al., 2010; Demirci et al., 2012; Kang et al., 2013; Mazur-Melewska et al., 2015), hepatic (Park et al., 2012), cardiac (Kim et al.,

* Corresponding author at: Laboratório de Parasitologia Veterinária, Hospital Veterinário da Universidade do Oeste Paulista (UNOESTE), Raposo Tavares Km 572, Bairro Limoeiro, Presidente Prudente, São Paulo, CEP: 19067-175, Brazil.
E-mail address: vamilton@unoeste.br (V.A. Santarém).

http://dx.doi.org/10.1016/j.vetpar.2017.07.020
Received 11 April 2017; Received in revised form 18 July 2017; Accepted 19 July 2017

2012; Lemaire et al., 2013), dermatological (Kim et al., 2010; Qualizza et al., 2014), neurological (Park et al., 2012), and ophthalmic alterations (Ahn et al., 2013; Besirli and Elner, 2013). Toxocariasis is characterized by eosinophilia, and diagnostic is based on the detection of anti-*Toxocara* antibodies by ELISA indirect test (Rubinsky-Elefant et al., 2010).

Toxocariasis is primarily a geo-zoonosis, since the main route of transmission of *Toxocara* spp. is through accidental ingestion of larval eggs present in contaminated environments (Macpherson, 2013), notably public parks and squares.

Ingestion of *Toxocara* spp. eggs through direct contact with dogs has been considered an alternative way of transmission for toxocariasis to humans (Wolfe and Wright, 2003; Aydenizöz-Ozkayhan et al., 2008; Roddie et al., 2008; Amaral et al., 2010; Öge et al., 2014). On the other hand, some authors argue that the risk of transmission of toxocariasis by contact direct with dogs is likely to be negligible, due mainly to the very low number of embryonated eggs of *Toxocara* spp. detected on host hair (Overgaaw et al., 2009; Paoletti et al., 2015).

Several factors may contribute to the presence of *Toxocara* spp. eggs in the hair of dogs, among them the age of the animals, origin, breed, and hygiene care and deworming (Roddie et al., 2008; Tavassoli et al., 2012; Holland, 2015).

The present study was undertaken to evaluate the frequency and factors associated with contamination of dog hair by *Toxocara* spp. in a Veterinary School Hospital in Presidente Prudente, São Paulo, Brazil.

2. Materials and methods

2.1. Area, period, and population

The study was carried out at the Veterinary-Teaching Hospital of the Universidade do Oeste Paulista (HV Unoeste), Presidente Prudente, São Paulo, from September 2015 to September 2016.

In total, 165 dogs were included in the study, regardless of gender, breed, or age, accompanied by the owners/guardians (domiciled animal), either brought for attendance or those in a state of neglect at the Veterinary Hospital of the University of Oeste Paulista.

Information on the characteristics of the animals (sex, age, coat pattern, origin, breed, contact with soil, and characteristics of the feces) and origin were recorded on a specific form prepared by the researchers, based on data provided by the owner of the dog after giving consent for their animal's participation in the research.

The animals were classified by age, following the pattern adopted by Keegan and Holland (2010): puppies (0–12 months) and adults (over 12 months). Other variables, including origin (abandoned or domiciled), sex, breed, hair length, soil contact, feces consistency, anthelmintic (deworming), and hygiene (bath/grooming frequency) were evaluated.

2.2. Collection of samples

During the hospital and kennel care at the University, hair samples from the perineal region, and upper and lower tail regions of the dogs were collected, from lateral cuts of the hair, transversely sectioned and without injury to the skin, with the help of sterile scalpel blades, transferred to a first use plastic bag, weighed, and stored under refrigeration at 4 °C until processing (Roddie et al., 2008).

2.3. Hair contamination analysis

For recovery of *Toxocara* spp. eggs from the hair, the protocol described by Roddie et al. (2008) and Overgaauw et al. (2009) was adopted, with some modifications.

The hair samples were weighed on an analytical scale (Shimadzu-Model AY220) and those weighing less than 0.0025 g were excluded from the study, following the model adopted by Overgaauw et al. (2009). They were subsequently transferred to falcon tubes containing approximately 20 mL of distilled water and 0.2 mL of Tween 20, where they remained overnight. After this period, another 20 mL of distilled water was added to each hair sample, which was homogenized using a vortex for 3 min.A second wash was performed with two drops of Tween 20 in 40 mL of distilled water.

The material was filtered through 300 µm, 212 µm, and 38 µm sieves. After filtration, the hair was discarded, and the material obtained from the filtrates was centrifuged (800 x g for 5 min). After centrifugation, 100 µL of the sediment was transferred to a slide and evaluated under optical microscopy (10X and 40X objective) for counting and morphological evaluation of the eggs.

Parasites structures recovered from hair dogs were identified according to Bowman (2009). *Toxocara canis* eggs were classified according to the stage of development (Roddie et al., 2008): viable (intact egg with content), non-viable (egg not intact or with damaged wall), embryonating (egg with two or more cell divisions), and embryonated (egg containing larvae of the parasite).

2.4. Analysis of the results

Fisher's exact test was used to evaluate the association between the presence of eggs and the variables: animal origin, age, gender, breed, hair length, contact with soil, and deworming, considering values of $P < 0.05$.

Statistical analyzes were performed using BioEstat 5.0 Software (Ayres et al., 2007).

2.5. Ethical aspects

The research project was approved by the Commission for Ethics in the Use of Animals (CEUA) of Unoeste (Protocol number 2637).

3. Results

In this study, *Toxocara* spp. eggs were observed in the hair of 11 (6.67%) animals, with a mean of 12.2 eggs per positive animal (1–70 eggs).

Among the evaluated animals, 38 (23.03%) dogs were abandoned, and 127 (76.97%) came from the routine of the hospital. Of the 165 animals evaluated, 59 (35.76%) were puppies and 106 (64.24%) adults.

Table 1 summarizes the results of some studies performed to recover eggs of *Toxocara* spp. in the hair of dogs, including the present study.

Table 2 presents the association between the presence of *Toxocara*

Table 1
Frequency of contamination of dog hair by *Toxocara* spp. eggs, with emphasis on the findings in puppies.

Reference	Location of study	Animals evaluated	Positive animals (%)	Number and frequency in puppies (%)
Wolfe and Wright (2003)	United Kingdom	60	15 (25.0%)	4 (6.6%)
Aydenizöz-Ozkayhan et al. (2008)	Turkey	51	11 (21.6%)	9 (82.0%)
Roddie et al. (2008)	Ireland	100	67 (67.0%)	25 (25.0%)
Amaral et al. (2010)	Brazil	104	25 (24.0%)	22 (88.0%)
Tavassoli et al. (2012)	Iran	138	50 (36.2%)	41 (82.0%)
Present Study	Brazil	165	11 (6.67%)	9 (82.0%)