Article

Not peer-reviewed version

Morphometry and Molecular Detection of *Spirometra mansoni* in Domestic Dogs from Rural Areas of Ecuador, and Its Clinical, Epidemiological and Public Health Implications

Roberto D. Coello Peralta *, Zully Baquerizo Orrala , Aldo Rubén Andrada , Davis Calle Atariguana , Geraldine Ramallo , Alicia Rojas

Posted Date: 18 May 2026

doi: 10.20944/preprints202605.1151.v1

Keywords: *Spirometra mansoni*; coproparasitic techniques; morphometry; PCR; domestic dogs; epidemiology; public health

 doi:10.20944/preprints202605.1151.v1

Article

Morphometry and Molecular Detection of *Spirometra mansoni* in Domestic Dogs from Rural Areas of Ecuador, and Its Clinical, Epidemiological and Public Health Implications

Roberto D. Coello Peralta [1,2,*], Zully Baquerizo Orrala [3], Aldo Rubén Andrada [4], Davis Calle Atariguana [1], Geraldine Ramallo [5] and Alicia Rojas [6]

[1] Department of Microbiology, Faculty of Veterinary Medicine and Zootechnics, Universidad de Guayaquil, Guayaquil 090514, Ecuador
[2] Instituto de Genética y Microbiología, Fundación Miguel Lillo, San Miguel de Tucumán T4000JFE, Argentina
[3] Consultorio Zu Veterinaria, comuna San Pablo, calle 9 de octubre y Abdón Calderón, Santa Elena 241702, Ecuador
[4] Instituto de Invertebrados, Fundación Miguel Lillo, San Miguel de Tucumán T4000JFE, Argentina
[5] Laboratory of Helminthology, Centro de Investigación en Enfermedades Tropicales, Universidad de Costa Rica, San José 2060, Costa Rica
* Correspondence: coelloroberto1@gmail.com; Tel.: +593969092172

Simple Summary

Spirometra mansoni causes sparganosis, a parasitic disease affecting human tissues, and spirometrosis in dogs and cats, which can cause gastrointestinal disorders; both pathologies are an animal and public health concern. Freshwater fish and amphibians play a crucial role in the spread of this parasite in the environment. However, there is no information on infection in dogs in Ecuador. In this study, we examined fecal samples from 402 dogs from riverside areas along the Daule River (Ecuador) to confirm the presence of the parasite. We used several coproparasitological techniques as screening methods, followed by morphometry and PCR for confirmation, indicating exposure to this tapeworm. 17% of the dogs tested showed evidence of infestation, suggesting that the parasite circulates among dogs in these areas. We also analyzed clinical and epidemiological characteristics of spirometrosis in dogs and the risk of sparganosis in humans. Our findings provide new information on the presence of this parasite in dogs living in rural environments in Ecuador and can contribute to improving disease surveillance and control strategies to reduce the risk of infection in pets and humans.

Abstract

Sparganosis is a zoonotic parasitosis associated with freshwater aquatic environments, prevalent in tropical and subtropical regions of the world. *Spirometra (S.) mansoni* causes sparganosis in humans and spirometrosis in domestic dogs, which is transmitted through the consumption of raw or undercooked meat from fish, frogs or paratenic animals, producing subcutaneous and tissue infections in humans, whereas dogs or cats develop gastrointestinal infections. The purpose of this investigation was to identify *S. mansoni* in domestic dogs from riverine sectors of the Daule River in Ecuador, using coproparasitological methods: direct examination, flotation and sedimentation with centrifugation using saline solution (as screening); and for confirmation, morphometric methods and PCR were used. Through a descriptive, prospective and cross-sectional study, 402 domestic dogs were analyzed, and *Spirometra mansoni* were determined in 17% of the collected samples. Clinical and epidemiological characteristics of spirometrosis in dogs and the risk of sparganosis in humans were determined, revealing a profound lack of information and knowledge about the infection;

 doi:10.20944/preprints202605.1151.v1

consequently, there is a possibility that cases will spread in pets and that humans will develop sparganosis.

Keywords: *Spirometra mansoni*; coproparasitic techniques; morphometry; PCR; domestic dogs; epidemiology; public health

1. Introduction

Spirometra is a genus of pseudophyllid cestodes that cause an anthropozoonotic disease known as sparganosis. The biological life cycle of this parasite begins when adult *Spirometra mansoni* located in the small intestine of their definitive hosts, usually domestic cats and dogs, lay eggs that reach fresh water [1]. The eggs mature and release the coracidia (ciliated mobile larvae), which are then ingested by copepod first intermediate hosts of the genus *Cyclops,* where procercoid larvae develop. Second intermediate hosts, such as fish or amphibians, ingest infected copepods and develop plerocercoid larvae. The latter are the infective forms for definitive hosts, accidental hosts (such as humans) and paratenic hosts (such as reptiles, birds and mammals including rodents, bears, pigs and monkeys) [2]. In the final definitive hosts, plerocercoids larvae reach the digestive tract, and adult parasite develops in the small intestine, producing spirometrosis [1,2], whereas in accidental hosts (humans) larval development is halted in hosts and sparganosis develops.

Sparganosis is a metacestodiasis or larval cestodiasis caused by *Spirometra mansoni* infection [1]. This tapeworm is transmitted to humans by the ingestion of copepods infected with procercoid larvae in natural water; the consumption of fish and amphibians (like tadpoles and frogs); or the consumption of undercooked meat, viscera or blood from paratenic animals containing plerocercoid larvae [4]. These larvae can reach several centimeters and live for up to 20 years in humans. The lesions can give rise to granulomas that can transform into abscesses and produce clinical manifestations depending on the place where they are finally located [2]. Plerocercoid larvae in humans have been described with greater frequency in subcutaneous tissues as palpable, fixed or migrating painless masses, associated with redness or itching. In addition, plerocercoid larvae can be found in other anatomical sites, such as the abdominal wall, chest wall, lower limbs, scrotum, pleural cavity, lungs, eye, orbit, abdominal viscera and nervous system [1,2]. The most serious manifestation is cerebral sparganosis, which is accompanied with headache, hemiparesis, hemianopia, and seizures [2].

There are more than 64 described species of *Spirometra*, and the most relevant include *Spirometra mansoni, Spirometra mansonoides, Spirometra erinaceieuropaei, Spirometra decipiens, Spirometra pretoriensis, Spirometra theileri, Spirometra ranarum* and *Spirometra proliferum* [1,2]. However, the species with the greatest epidemiological importance is *S. mansoni*, which is distributed on several continents, including Europe, Asia, South Africa and Australia [2], with the global incidence of infection between 0.089% and 0.313% [3]. In addition, human cases of sparganosis for *Spirometra mansoni, S. erinacei S. ranarum, S. proliferum* have been reported in China, Japan, Taiwan, Korea, Vietnam, Thailand and other countries in Southeast Asia and the Western Hemisphere after the consumption of intermediate hosts infected with procercoid or plerocercoid larvae [2].

Spirometra mansoni infection in domestic dogs and cats is usually asymptomatic but can lead to diarrhea, vomiting, weight loss, and enteritis [5]. *S. mansoni* has been reported in Canada [6], the United States [2], Mexico [6], Puerto Rico, Costa Rica [7], Honduras, Belize, Cayman Islands [6], Cuba [7], Grenada, the Netherlands Antilles, Venezuela, Colombia [6], Ecuador [8], Brazil [9], Bolivia [10], Paraguay [2], Uruguay and Argentina [10].

In Ecuador, a natural infection of *S. mansoni* was described in 1974 in a domestic cat determined by morphometry of the adult parasite [8], and in 1990, nodulation located in the left scapular region was identified in a human, which, after its extirpation, revealed the presence of a cestode classified morphometrically as *Spirometra* sp. [11]. Therefore, this study aims to describe this parasite in domestic dogs through stool analysis and analyze its prevalence in a riverine zone of Ecuador (Fig.

 doi:10.20944/preprints202605.1151.v1

1), the clinical characteristics associated with the animals, its epidemiology and its implications for public health.

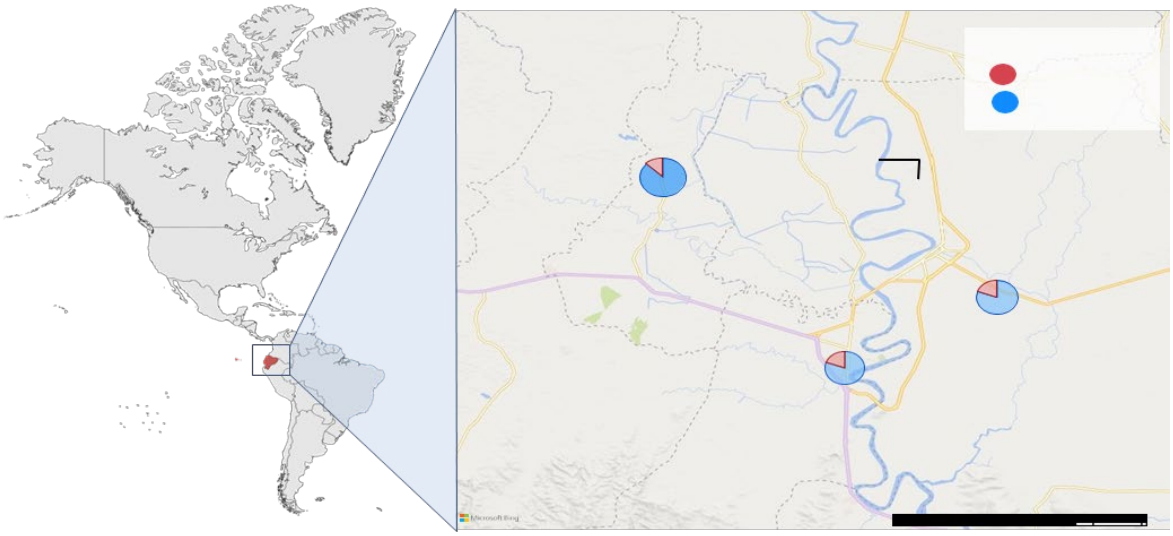

Figure 1. Map with sites of collection and results of *Spirometra mansoni* presence in domestic dogs. Bubble size is proportional to the number of collected samples per region.

2. Materials and Methods

2.1. Collection of Samples and Physical Analysis of Participants

Fecal samples of domestic dogs were collected by non-probabilistic convenience sampling in riverside sectors of the Daule River, including Loma Larga in the city of Nobol; Santa Rosa in the city of Daule; and Las Cañas in the city Lomas de Sargentillo; all of which are located in the province of Guayas on the Ecuadorian coast. The temperatures in these geographical regions range between 20 °C and 37 °C, with a tropical savanna climate.

An average of 5 domestic dogs per household (3,650 dogs) live in this area. With this information sample size was calculated using the online program Qualtrics [XM], with a confidence level of 95%, a sampling error of 5% and a population size of 3,650 domestic dogs. A sample size of 348 dogs was estimated, and in this study, a total of 402 domestic dogs were analyzed [12].

With the help of residents of the sector, recognition of the areas was carried out, and the importance of the investigation and the risk of acquiring sparganosis to the inhabitants were explained. In addition, a targeted survey was conducted with the individuals who decided to participate in the study prior informed consent. These questions included questions such as knowledge of sparganosis, knowledge of cases, consumption of exotic foods (fish, frogs, tadpoles, copepods, snakes, birds and pigs) and water without or little heating, consumption of food and water by dogs, contact with the river, possession of domestic dogs, deworming of animals, habitat of dogs and veterinary care.

The physical verification of domestic dogs was carried out, and the following parameters were evaluated: body temperature; body weight (estimated with the naked eye on the following ordinal scale: 2–4 kg, 5–7 kg, 8–10 kg, 11–13 kg, and 14–16 kg); body condition (scale: overweight, good, fair and poor) following the guidelines established by the World Association of Small Animal Veterinarians [WSAVA]); and muscle mass (scale: 1: severe muscle wasting; 2: mild to moderate muscle wasting; 3: normal muscle); conditions of the skin, coat, ocular mucosa; and signs of disease [13]. To obtain fecal samples from domestic dogs (from March 1 to September 30, 2023), sterile jars were used, and the following data were recorded: address, owner name, and telephone contact. In cases where the owner of the animal could not collect a sample from his pet, a technical team carried out it following the procedures of Dubie et al. [14].

 doi:10.20944/preprints202605.1151.v1

Additionally, physical observation of dog owners was carried out in search of a granuloma or abscess in any part of the body; similarly, if they had sparganosis or if they knew of anyone in their surroundings that did, it was recorded [15].

2.2. Transportation and Analysis of Samples From Domestic Dogs

Dog fecal samples were stored between 4 and 8 °C and transported to the laboratory of Besito Vet Pet Lab in the city of Guayaquil (Ecuador) [16]. All samples were immediately analyzed via the following coproparasitic methods: direct examination, flotation (Willis), and sedimentation with centrifugation using saline solution (to increase the sensitivity and specificity of the screening). The samples were observed via Lugol light microscopy at magnifications of 10 and 40X [17,18].

Additional stool and blood samples were collected from those animals confirmed to be infected with *Spirometra mansoni*. Immunochromatography tests will be carried out: parasitological (antigens for *Babesia* sp.), bacterial (antigens for *Ehrlichia can*is, *Anaplasma* sp., *Leishmania* sp. and *Dirofilaria immitis*.), viral (antigens for *Parvovirus* and *Canine Distemper Virus*) and fungal (tinction of AFB and lactophenol) to rule out the presence of another infectious disease.

2.3. Diagnostic Criteria and Morphometry

For the identification of *Spirometra mansoni* eggs the following criteria described by Bowman [18] and Alvarado et al. [7] were considered: eggs capped at a distal end of the egg with a flattened shape, equatorial bulge, brown in color, between 60 and 70 µm long and 30–40 µm wide; elliptical, rounded or convex in shape and the presence of morulated structures inside.

2.4. DNA Extraction and PCR

The samples that were positive by the screening methods were processed with a DNA/RNA shield and 0.5 g of sample in 500 µL of solution in the Besito Vet Pet Lab laboratory. Then, they were transported at −20°C to the Institute of Genetics and Microbiology of the Miguel Lillo Foundation (San Miguel de Tucumán-Argentina), where the eggs were lysed following the procedures of Coello et al [19]. DNA extraction was subsequently performed via the Qiagen DNeasy blood & tissue kit. The sequences of the ITS2 intron were amplified via the specific primers Sman-F (5′- CGC CTA ATA AAA CAG CCG GC-3′) and R (5′- GTT CAG CGG GTA ATC TCG ACT-3′). A sample generously provided by Dr. Marcos Javier Butti of the Laboratory of Human Parasitosis and Zoonosis Parasitarias of the National University of La Plata (Argentina) was used as a positive control.

The PCRs were conducted in a final volume of 25 µL, which included: 2 µL of genomic DNA template (100 ng/µL), 0.3 µL of 2U TAQ T-plus High Way polymerase, 2.5 µL of Taq 10 × High Way Buffer, 1.5 µL of 25 mM $MgCl_2$ from Thermo Scientific, 0.5 µL of primers, 1 µL of 0.5 mM dNTPs, and 16.7 µL of ddH_2O. Amplification was performed in a Nyxtecnik® thermocycler using the following PCR profile: 1 cycle at 94 ºC for 5 min, followed by 36 cycles of 94 ºC for 40 s, 46 ºC for 40 s, and 72 ºC for 1 min and 10 s, with a final step of 1 cycle at 72 ºC for 5 min and a hold at 4 ºC. The PCR products were analyzed by electrophoresis on 1.5% agarose gels stained with GelRed Nucleic Acid Gel Stain (10,000X) and visualized under UV light.

2.5. Statistical Analysis

Data obtained herein were tabulated and presented in tables generated using the Microsoft Excel 2010 program. Furthermore, the prevalence of *Spirometra mansoni* in dogs and the variables studied were analyzed via inferential statistical analysis (t student test and chi-square test) performed with the SPSS statistical software.

 doi:10.20944/preprints202605.1151.v1

3. Results and Discussion

3.1. Microscopic identifications Using Coproparasitological Screening Techniques and Morphometry

During coprological analysis of dog fecal samples, brown capped eggs that were 60–70 µm long and 30–40 µm wide were observed. The eggs had equatorial bulges and the operculum was located at a distal end of the egg with a flattened shape. Some eggs were elliptical, whereas others were more rounded or convex. Additionally, morulated structures were observed inside the eggs as described by Alvarado et al. [7] (Fig. 2). These characteristics corresponded to those reported for *Spirometra mansoni* [7,18]. Importantly, other intestinal parasites, such as *Cystoisospora* sp., *Ancylostoma* spp., *Uncinaria stenocephala, Toxocara canis*, *Trichuris* sp., *Taenia* sp., *Strongyloides* sp., and *Capillaria* sp., were not detected in dogs.

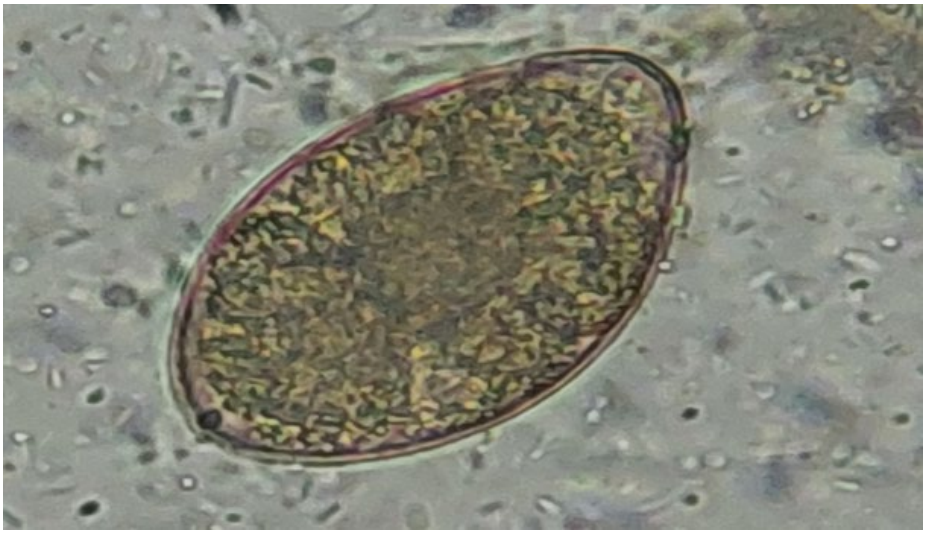

Figure 2. The eggs of *Spirometra mansoni* in the feces of domestic dogs were identified through sedimentation by centrifugation in saline solution, and the eggs were observed by optical microscopy at 40× and stained/not with Lugol.

Although mature proglottids and adult parasites could not be recovered, the species was confirmed by egg morphometry and molecular detection of the parasite's DNA by PCR from fecal samples, identifying canine infections by *S. mansoni.* This species has been reported in frogs and domestic cats in the United States [6], domestic cats in Mexico [20], coyotes, domestic dogs and cats from Costa Rica [7], crab foxes (*Cerdocyon thousand*) in Colombia [21] and domestic cats in Ecuador [8]. Moreover, *S. decipiens* has also been reported in the Americas, but in the Southern latitudes including a case from a human in Venezuela, snakes and field fox from Brazil [6], a Bolivian fox *S. decipiens* has been reported in Bolivian fox [6]. a domestic cat from Chile and wild felids from Argentina [21]; *S. proliferum* has been reported in humans from Venezuela [22], and Other *Spirometra* spp. have not been reported in the Americas.

Ecuador is a tropical country located in Western South America, and it has 4 regions with a variety of climates (Coastal, Andean, Amazon and Insular) [23]. Neglected freshwater zoonotic diseases such as opisthorchiasis, diphyllobothriasis, alariasis, gnathostomiasis and sparganosis may occur in coastal territories [24]. This is the first report on the presence of *Spirometra mansoni* in domestic dogs of riverside sectors of the Daule river in Ecuador and its clinical, epidemiological and public health implications. Moreover, in Ecuador, *S. mansoni* has been previously described in domestic cats [8] and in humans [11], suggesting that the parasite has been present in the country for an indefinite period of time.

 doi:10.20944/preprints202605.1151.v1

3.2. Epidemiology and Prevalence of Spirometra Mansoni in Domestic Dogs from Riverine Sectors of the Daule River

The general frequency of this parasite in domestic dogs has been reported 17%. The prevalence varied between the sampled areas (Table 1 and Fig. 1), since frequencies of 19.49%, 19.35%, and 12.50% were detected in the samples collected from dogs located in Loma Larga, Santa Rosa, and Las Cañas, respectively (Table 1), with significant differences ($p < 0.05$ and t student test). These values are above the prevalence rates reported worldwide (0.089 and 0.313%) and those in Asia (0.696%), Africa (0.224%) and Oceania (0.203%) [3]. However, in China, prevalences ranging from 0–27.5% to 77.9% have been reported in domestic dogs [4]. These findings suggest that the animals sampled in this study are highly exposed to paratenic and second intermediate hosts carrying larval stages of *Spirometra mansoni* and thus, become infected with the parasite, either by feeding or hunting habits.

Table 1. Prevalence of *Spirometra mansoni* in domestic dogs from Riverine Sectors of the Daule River, Ecuador.

Riparian sectors of the Daule river	No. of animals studied	Positive samples	Prevalence
Loma Larga	118	23	19.49%
Santa Rosa	124	24	19.35%
Las Cañas	160	20	12.50%
Total	402	67	17%

3.3. Molecular Identification of Spirometra mansoni in Fecal Samples from Domestic Dogs using the PCR Technique

Regarding the PCR products, 67 of the 67 samples were obtained using specific primers. All DNA samples yielded a PCR product of approximately 120 bp. Although there are few studies on the molecular identification of *Spirometra mansoni* in domestic dogs using the ITS2 molecular marker, but Zendejas-Heredia et al [25] described it in 15 dogs from Cambodia. However, it is different from other studies that use the molecular marker COX1, described by Alvarado et al. [7] in Costa Rica, Salazar et al. [20] in Mexico and Yamasaki et al. [26] in Japan.

3.4. Determination of Clinical Characteristics in Domestic Dogs from the Studied Sectors

All domestic dogs studied were of mixed breed, aged between 1 and 10 years, with males (60%) and females (40%). Yamasaki et al. [26] and Muñoz [2] described that any domestic dog of any age can be susceptible to spirometrosis.

Physical examination of the 402 domestic dogs revealed that 385 (96%) had normal body temperatures, whereas and 17 (4%) had high body temperatures. In addition, 90% had body weights between 2–4 kg and 10% had body weights between 5–7 kg; 60% of the dogs had poor body conditions, 30% had fair body conditions and 10% had good body conditions, with significant differences ($p < 0.05$ and t student test). Regarding muscle mass, 60% of the dogs had severe muscle wasting, 30% had mild to moderate muscle wasting, and 10% had normal muscle mass. Ninety percent of the animals presented dull skin and coat conditions with pale ocular mucous membranes. Finally, 17 animals (4%) presented vomiting, diarrhea, decay, weakness, severe weight loss, poor body condition and pale mucous membranes, which may be consequence of *Spirometra mansoni* infection or other vector-borne pathogens distributed in the region such as *Ehrlichia canis, Anaplasma* spp., *Leishmania* spp. or *Dirofilaria immitis.* The manifestations reported here differ from those reported by Coello et al. [23], but they are similar to those explained by Bowman [18] and Alvarado et al. [7]. However, it is important to note that there is little reference on this subject, as infection rates in dogs vary from 1% to 33% [27]. Other reports have shown that infected animals can be asymptomatic, whereas others can present with enteritis, diarrhea, vomiting and weight loss [5,18,28]. In this sense, further studies analyzing blood samples from dogs should be conducted to

 doi:10.20944/preprints202605.1151.v1

determine the true morbitidy associated with *Spirometra mansoni* infections, as well as to decipher the prevalence of these other pathogens in dogs from endemic regions.

3.5. Determination of Clinical Characteristics in People from the Studied Sectors

People interviewed (177 persons) were unaware about the infection caused by *Spirometra* sp. and about any sparganosis infections that may had occurred in the last five years. Forty percent said they consumed some type of exotic animal (fish, frogs, tadpoles, copepods, snakes, birds or pigs) with little or no cooking, 60% consumed water with little or no heating, and each family owned between 6 and 7 domestic dogs. Moreover, the owners of the dogs stated that all animals consumed untreated food and water. In addition, all people and dogs have contact with the river, and none of the dogs are dewormed or receive veterinary care and give birth either in the home or near the home. Moreover, all studied sites were rural, where there is no sewerage facilities or treatment plants to purify drinking water. These results, revealed socioepidemiological variables that increase the risk of parasitic infection of 17% or higher E, are similar to the those of Coello et al. [23] Badri et al. [3] and Kavana et al. [29], but differ from the findings of Hong et al. [4], where a prevalence of socioepidemiological variables that increase the risk of parasitic infection of 27.5% and lower was described. These results demonstrate the susceptibility of the human population to infection by this parasite and the widespread ignorance of the infection.

No cases of subcutaneous lesions suggestive of sparganosis were recorded during the physical examination of subjects. However, Muñoz [2], Lin et al. [27], Muigg et al. [30] and Yim et al. [31] described sparganosis in the abdominal wall, chest wall, lower limbs, scrotum, pleural cavity, lungs, eye, orbit, abdominal viscera and nervous system of human patients. Therefore, sparganosis-associated lesions may not have been properly identified. For this purpose, it would be ideal to carry out serological tests that evaluate the antigens of the parasite in individuals, providing insights into their contact with dogs infected with the parasite. Similarly, bacterial, fungal or viral infections were ruled out in domestic dogs, resulting in negative results in cultures, stains and serological tests for parvovirus and canine distemper.

5. Conclusions

This is the first report of *Spirometra mansoni* in domestic dogs of Ecuador with a prevalence of 17%. The parasites were confirmed by morphometry and PCR. Clinical and epidemiological implications were obtained by means of a survey to determine whether there is a risk of human sparganosis and *Spirometra* infection in dogs. Through a physical examination of domestic dogs, clinical characteristics, such as vomiting, diarrhea, decay, weakness, severe weight loss, poor body condition and pale mucous membranes were detected, all associated with *Spirometra mansoni* infection. In humans, no cases of sparganosis were detected. The results of this research highlight the risk of zoonotic transmission to humans due to the high prevalence of *Spirometra mansoni* in definitive hosts of the area, the great contact between humans and animals, and the unawareness of the inhabitants regarding this parasitic infection. Therefore, it is important to educate the population about this parasite from the One Health perspective, take sanitary measures, periodically deworm animals and avoid the consumption of raw or undercooked meat from fish and exotic animals to reduce the risk of infection.

Author Contributions: Conceptualization, R.D.C.P. and A.R.; methodology, R.D.C.P, A.R. and G.R.; software, A.R.A., D.C.; validation, R.D.C.P., A.R.A, G.R. and A.R.; formal analysis, R.D.C.P., A.R.A. and A.R.; investigation, R.D.C.P., Z.B.O., D.C., A.R.A. and A.R.; resources, R.D.C.P., D.C. and Z.B.O.; data curation, Z.B.O.; writing—original draft preparation, R.D.C.P.; writing—review and editing, R.D.C.P., G.R., and A.R.; visualization, R.D.C.P.; supervision, R.D.C.P. and Z.B.O.; project administration, R.D.C.P.; funding acquisition, R.D.C.P. All authors have read and agreed to the published version of the manuscript.

 doi:10.20944/preprints202605.1151.v1

Funding: This work was funded by the University of Guayaquil, Dean of Research, Graduate Studies and Internationalization, research project FCI-029-2022.

Institutional Review Board Statement: This research was approved by the Institutional Animal Care and Use Committee (IACUC) of the Faculty of Veterinary Medicine and Zootechnics (FMVZ) of the University of Guayaquil, the Research Department of the University of Guayaquil (DIUG) and the Ethics Committee of the "Luis Vernaza" Hospital of Guayaquil (Ecuador) (03-EI- CEISH-HLV-2023).

Informed Consent Statement: If aplicable.

Data Availability Statement: The data presented in this study are available on request from the corresponding author.

Acknowledgments: We want to thank Denny Borbor Prudente for his help in the statistical analysis and the University of Guayaquil and Mr. Roberto Coello Fuentes, Owner Manager of the "Don Robert" Commercial Store, for their financial support.

Conflicts of Interest: No conflicts of interest are declared by any of the authors.

References

1. Kim, J.G.; Ahn, C.S.; Sohn, W.M.; Nawa, Y.; Kong, Y. Human sparganosis in Korea. *J. Korean Med. Sci*. 2018, 33, e273.
2. Muñoz, P. The sparganosis. *Rev. Chilena Infectol*. 2015, 32, 457.
3. Badri, M.; Olfatifar, M.; Karimi, V; Zaki, V; Carvalho, M.M.; Fasihi, M. The global prevalence of *Spirometra* parasites in snakes, frogs, dogs, and cats: A systematic review and meta-analysis. *Vet. Med. Sci*. 2022, 8, 2785–2805.
4. Hong, Q.; Feng, J.; Liu, H.; Li, X.; Gong, L.; Yang, Z.; Yang, W.; Liang, X.; Zheng, R.; Cui, Z.; Wang, W.; Chen, D. Prevalence of *Spirometra mansoni* in dogs, cats, and frogs and its medical relevance in Guangzhou, China. *Int. J. Infect. Dis*. 2016, 53, 41–45.
5. Adolph, C.; Peregrini, A. Greene's infectious diseases of the dog and cat, in: J.E. Sykes (Ed.), *Spirometra Masonoides*, 5th ed; Saunders, California, United States, 2024; pp. 1455–1484.
6. Kuchta, R.; Phillips, A.J.; Scholz T. Diversity and biology of *Spirometra* tapeworms (*Cestoda: Diphyllobothriidea*), zoonotic parasites of wildlife: A review. *Int. J. Parasitol. Parasites Wildl*. 2024, 24, 100947.
7. Alvarado, I.; Campos, J.; Arguedas, Y.; Romero, L.; Alfaro, A.; Anchia, G. Molecular, morphological and histopathological evidence of *Spirometra mansoni* in wild and domestic animals from Costa Rica. *Vet. Parasitol. Reg. Stud. Rep*. 2024, 51, 101030.
8. Mueller, J.F.; Fróes, O.M.; Fernández, T. On the occurrence of *Spirometra mansonoides* in South America. *J. Parasitol*. 1975, 61, 774–775.
9. Silva, W.I.; Lima, E.F.; Silva, J.O.; Alves, M.M.; Alves, C.L.P.; Silva, A.L.P.; Lima, J.A.; Feitosa, T.F.; Vilela, V.L.R. Endoparasites in domestic cats (*Felis catus*) in the semi-arid region of Northeast Brazil. *Rev. Bras. Parasitol. Vet*. 2023, 32, e012123.
10. Vettorazzi, R.; Norbis, W.; Martorelli, S.R.; Garcia, G.; Rios, N. First report of *Spirometra (Eucestoda; Diphyllobothriidae*) naturally occurring in a fish host. *Folia Parasitol*. 2023, 70, 1–7.
11. Guderian, R.; Roldan, J.; Guevara, A.; Chico, M. Human sparganosis in Ecuador: Report of a case in the province of Esmeraldas. *Rev. Soc. Bras. Med. Trop*. 1990, 23, 115–116.
12. QualtricsXM, How to calculate sample size: Ensure sampling is correct. Available online: https://www.qualtrics.com/es/gestion-de-la-experiencia/investigacion/calcular-tomano-muestra/. (accessed on13 March 2026).
13. Milstein, M.S.; Shaffer, C.A.; Suse, P.; Marawanaru, A.; Heinrich, D.A.; Larsen, P.A.; Wolf, T.M. A mixed-methods approach to understanding domestic dog health and disease transmission risk in an indigenous reserve in Guyana, South America. *PLoS Negl. Trop. Dis*. 2022, 16, e0010469.
14. Dubie, T.; Sire, S.; Fentahun, G.; Bizuayehu, F. Prevalence of gastrointestinal helminths of dogs and associated factors in Hawassa city of Sidama region, Ethiopia. *J. Parasitol. Res.* 2023, 6155741.

 doi:10.20944/preprints202605.1151.v1

15. Liu, W.; Gong, T.; Chen, S.; Liu, Q.; Zhou, H.; He, J.; Wu, Y.; Li, F.; Liu, Y. Epidemiology, diagnosis, and prevention of sparganosis in Asia. *Animals*. 2022, 12, 1578.
16. Giraldo, M.I.; García, N.L.; Castaño, J.C. Prevalence of intestinal helminths in dogs from Quindío province, *Biomedica*. 2005, 25, 346–352.
17. Girard, R. Parasitology Manual, Methods for Primary Health Care Laboratories, 3rd ed.; PAHO/WHO/ National Autonomous University of Honduras: Tegucigalpa, Honduras, 2014; pp. 103-167.
18. Bowman, G.D. Parasitology for Veterinarians, 11 th ed.: Elsevier, New York, United States, 2024; pp. 135-244.
19. Coello, R.D.; Andrada, A.R.; Vinueza, R.L.; Pazmiño, B.J., Valencia, E.D.; Rodríguez, E.X.; Salazar, M.L.; Ramallo, G. Identification of *Ancylostoma caninum* in Domestic Dogs from Ecuador via Various Techniques. *Med Sci Monit*. 2025, 31, e947069.
20. Salazar, E.; Rodriguez, R.I.; Bolio, M.E.; Romero, D.; Ramos, R.; Solano, A.; Rojas, A. First morphological and molecular characterisation of *Spirometra mansoni (Cestoda, Diphyllobothriidae)* in a domestic cat from Veracruz, Mexico. *Vet. Parasitol. Reg. Stud. Rep*. 2025, 57, 101189.
21. Brabec, J.; Uribe, M.; Chaparro, J.J.; Hermosilla, C. Presence of *Spirometra mansoni*, causative agent of sparganosis, in South America. *Emerg. Infect. Dis*. 2022, 28, 2347–2350.
22. Fredes, F.; Mercado, R.; Salas, I.P.; Sugiyama, H.; Kobayashi, H.; Yamasaki, H. Morphological observation and molecular phylogeny of *Spirometra* decipiens complex 1 *(Cestoda: Diphyllobothriidae*) found in cat from Chile. *Parasitol. Int*. 2022, 87, 102493.
23. Coello, R.; Pazmiño, B.; Salazar, M.; Parra, S.; Vinueza, R.; Rodas, J. Ecoepidemiology of *Ancylostoma spp*. in urban-marginal and rural sectors of the ecuadorian coast and prevalence of cutaneous larvae migrans. *Med. Sci. Monit*. 2024, 30, e943931.
24. Menconi, V.; Lazzaro, V E.; Bertola, M.; Guardone, L.; Mazzucato, V; Prearo, M.; Bilska-Zajac, E.; Cortinovis, L.; Manfrin, A.; Arcangeli, G.; Angeloni, G. The occurrence of freshwater fish-borne zoonotic helminths in Italy and neighbouring countries: A systematic review. *Animals*. 2023, 13, 3793.
25. Zendejas-Heredia, P.; Colella, V.; Huggins, L; Schaper, R.; Schunack, B.; Traub, R. An Integrated Coproscopic and Molecular Method Provides Insights into the Epidemiology of Zoonotic Intestinal Helminths of Dogs across Cambodia. *Transbound. Emerg. Dis*. 2023, 2001871, 1-15.
26. Yamasaki, H.; Sugiyama, H.; Morishima, Y.; Sako, Y. Molecular identification of Spirometra infections in companion animals and wildlife in Japan. *J Vet Med Sci*. 2024, 86, 409-412.
27. Rojas, A.; Salazar, E.; Beck, R. *Spirometra mansoni. Trends Parasitol*. 2026. 42, 426-427.
28. Lin, Q.; Ouyang, J.S.; Li, J.M.; Yang, L.; Li, Y.P.; Chen, C.S. Eosinophilic pleural effusion due to *Spirometra mansoni* spargana: A case report and review of the literature. *Int. J. Infect. Dis*. 2015, 34, 96–98. .
29. Kavana N, Sonaimuthu P, Kasanga C, Kassuku A, Al-Mekhlafi HM, Fong MY, et al. Seroprevalence of Sparganosis in Rural Communities of Northern Tanzania. *Am J Trop Med Hyg*. 2016, 95, 874-876. .
30. Muigg, V.; Ruf, M.T.; Schwarzkopf, S.; Huang, S.; Denisjuk, N.; Stürmann, A.; Ritzler, M.; Wampfler, R.; Poppert, S.; Neumayr, A. Case report: Human subcutaneous sparganosis in a Thai migrant. *Am. J. Trop. Med. Hyg*. 2019, 101, 1170–1173. .
31. Yim, J.; Kim, Y.A.; Park, J.H.; Park, H.E.; Song, H.B.; Kim, J.E. Revisiting human sparganosis: a pathologic review from a single institution. *J Pathol Transl Med*. 2026, 60, 83-91. .