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Detection of *Ancylostoma ceylanicum* and *Toxascaris leonina* in Bengal Tigers (*Panthera tigris*) and Asian Black Bears (*Ursus thibetanus*) Captured at Hanoi Wildlife Rescue Center, Vietnam

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ABSTRACT

The Hanoi Wildlife Rescue Center (WRC) plays a crucial role in the rescue, care, and release of wild animals. The health of these animals was meticulously evaluated during their care and before their reintroduction into their natural habitat. An annual surveillance study of parasitic infections in wildlife at the Hanoi WRC was conducted to establish a scientific basis for developing preventive measures to manage the health of these animals. A total of 46 fresh fecal samples were opportunistically collected immediately following defecation using a shovel. These samples were obtained from 25 tigers (age: 10.8±3.6 years) and 21 bears (age: 11.2±4.6 years) captured at the Hanoi WRC in November 2024. Fecal samples were examined using sedimentation and centrifugal flotation methods to identify the presence of parasites. Subsequently, parasite eggs were collected using a micropipette for DNA extraction. Polymerase chain reaction and sequencing techniques were used to determine the parasite species. It was found that 20.0% of tigers (5 out of 25) and 23.8% of bears (5 out of 21) were infected with parasitic worms. The identification of these infections was achieved through the examination of the morphological characteristics of the eggs. This analysis identified the presence of trematode eggs, nematode eggs, including *Strongyle* type, and *Toxascaris leonina.* Molecular analysis further identified *Strongyle* eggs as belonging to *Ancylostoma ceylanicum*, which accounted for 12.0% of infections in tigers and 23.8% in bears. Additionally, *Toxascaris leonina* was detected exclusively in tigers, representing 12.0% of infections. Trematode eggs were found solely in tiger feces; however, molecular amplification was unsuccessful due to the insufficient number of eggs detected. The discovery of two zoonotic nematodes, *A. ceylanicum* and *To. leonina* in tigers and bears at the Hanoi (WRC) underscores the potential risk of nematode transmission from wildlife to humans and domestic animals in this area and its surrounding areas. The findings of this study will aid in the development of a prevention program aimed at controling gastrointestinal helminths in wild animals within the study region.

Keywords: *Ancylostoma ceylanicum,* Asian black bear, Bengal tiger, *Toxascaris leonina,* Wildlife Rescue Center

INTRODUCTION

Wildlife Rescue Centers play a significant role in the rescue of wild animals from illegal hunting and trafficking, subsequently releasing them back into their natural habitats. These activities not only guard endangered species from extinction but also contribute to the preservation of biodiversity and enhancement of ecosystems in Vietnam (Decision No. 1495/QD-UBND, dated March 15, 2025, issued by the Hanoi People's Committee). However, in a confined area, parasitic infection is a major problem due to the high risk of parasitic exposure in animals. It can directly affect animal health, cause severe diarrhea, and even be fatal. It also carries the risk of transmitting parasites from wild animals to domestic animals and even to humans (Mackenstedt et al., 2016). The Asian black bear (*Ursus thibetanus*) and Bengal tigers (*Panthera tigris*) are now critically endangered species on the list of animals that need to be preserved in the world (IUCN, 2015; Goodrich et al., 2015; IUCN, 2020; Garshelis and Steinmetz, 2020) and Vietnam's Red Data Book. However, worldwide studies on parasitic infections in these animals, as well as in Vietnam, have been undertaken.

Several studies have assessed the prevalence of gastrointestinal parasites in wild mammals that are housed in zoological gardens. The findings indicated a high incidence of parasitic infections among these animals, with prevalence rates ranging from 18% to 100% (Liza et al., 2020a; b; Chiu et al., 2021; Dibakou et al., 2021; Maharjan et al., 2025). In Chitwan National Park, Nepal, the prevalence of helminth infections among wild animals was documented to be 85.7% (Maharjan et al., 2025). Additionally, 90% of Bengal tigers in Bangabandhu Sheikh Mujib Safari Park, Gazipur, Dhaka, were found to be affected by gastrointestinal parasites (Liza et al., 2020b). Furthermore, the prevalence of helminth infections in Andean bears in Chingaza (Massif, Colombia) ranges from 5% to 62%, depending on the specific parasites involved (Quintero et al., 2023). Alaska brown bears captured in restricted areas of the United States exhibited an overall prevalence rate of 14.4% (Haynes et al., 2023), whereas in brown bears inhabiting the Cantabrian Mountains (north-

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western Spain), the rate was 64.1% to 79.3% (Cano et al., 2024; Remesar et al., 2024). The composition of parasites varied, including trematodes, cestodes, and nematodes, with nematodes being the most prevalent helminth type. For Bengal tigers, the nematodes identified were *Toxocara* spp., *Toxascaris leonina*, *Ascaris* spp., hookworms (*Strongyle* spp.), *Strongylus* spp., *Trichuris* spp., *Capillaria* spp., *Nematodirus* spp., and *Strongyloides* spp. (Liza et al., 2020a; b; Chiu et al., 2021; Maharjan et al., 2025). Additionally, trematodes such as *Paramphistomun* spp. and *Paragonimus* spp. were detected at a low prevalence (Ram et al., 2020; Maharjan et al., 2025). Furthermore, cestodes such as *Hymelolepsis* spp., *Diphyllobothrium* spp., *Spirometra* spp., and *Taenia* spp. have been identified in Bengal tigers (Liza et al., 2020b). For Asiatic black bears, besides nematodes such as *Toxocara* spp., *Ascaris* spp., *Trichuris* spp., and hookworms, *Bayliascaris transfuga*, *Oesophagostomum* sp., *Trichostrongylus* sp. were detected (Liza et al., 2020a; Hwang et al., 2021; Kim et al., 2025). The composition of cestodes parasitizing bears was similar to that observed in tigers, but no trematodes were detected in this species (Liza et al., 2020a). Most of these parasites were identified only at the genus level using fecal examination techniques or coprological analysis.

During an annual surveillance study of parasitic infections in wildlife at the Hanoi Wildlife Rescue Center (WRC), fecal samples were collected from Bengal tigers and Asian black bears. Fecal examination techniques and molecular methods were then employed to identify the prevalent parasites. Thus, the aim of this study is to determine the composition of parasites in tigers and bears at the Hanoi WRC, then to provide a scientific basis for establishing preventive measures to manage the health of these animals.

MATERIALS AND METHODS

Ethical approval

The study was conducted by the 2024 disease prevention plan for wild animals, which was approved by the Board of Directors of the Hanoi Wildlife Rescue Center (WRC) and authorized by the Hanoi Department of Agriculture and Rural Development.

Study area

The Hanoi Wildlife Rescue Center, formerly designated the Wildlife Rescue Center and Forest Protection Technology, was established in 1996. The center occupies an area of approximately 10,000 m^2, situated in the Soc Son district, 30 km North of Hanoi's urban center, Vietnam. The annual temperature in this area ranges from 14°C to 34°C and is divided into two distinct seasons. The hot season extends from April to October and is characterized by an average daily temperature exceeding 31°C. Conversely, the cool season spans November to March, with an average daily temperature below 22°C. The mean annual rainfall is recorded at 94.0±75.2 mm. This location constitutes the sole forested area in the capital city, encompassing 6,630 hectares. The center fulfills multiple functions, including coordinating rescue operations, implementing conservation efforts, conducting captive breeding programs, initiating scientific research, and facilitating educational visits. Additionally, it maintains domestic and international relations for the research, conservation, exchange, and provision of subsequent wildlife generations (F2, second generation of offspring). Furthermore, the center supports the management of wildlife rescue operations within the purview of the Department of Agriculture and Rural Development.

Fecal sample collection

A total of 46 fecal samples were collected from 25 tigers and 21 bears captured at the Hanoi Wildlife Rescue Center in November 2024. Fresh fecal samples were opportunistically collected immediately following defecation using a shovel and were subsequently placed in plastic bags labeled with name, age, and gender. As each animal was housed in its cage, repeated fecal collection was avoided. The feces were stored at 4 °C and sent to the Department of Veterinary Medicine, Vietnam National University of Agriculture. Five grams of feces of each animal were weighed and examined by two techniques, including sedimentation (Thawait et al., 2014) and centrifugal flotation methods (Dryden et al., 2005). Helminth eggs were examined under a light microscope at 10X and 40X magnification using an Olympus light microscope (Olympus, SZX7, Japan). The ova of different parasites were identified based on their morphologies (Soulsby, 1982). The eggs were then classified, counted to evaluate the intensity of infection based on the number of eggs per gram of feces. The eggs were then picked up using a micropipette under the microscope and kept in 1.5 ml Eppendorf tubes for further analysis.

DNA extraction and amplification by polymerase chain reaction assay

Nematode DNA was extracted from eggs using a commercial DNA extraction kit (Genomic DNA Pre Kit, BioFact, Korea) designed for animal tissue samples, following the manufacturer's protocol with slight modifications. Specifically, during the initial incubation phase, samples were incubated for 2 h at 56 °C, deviating from the standard 10-minute protocol. The final DNA elution was conducted in 50 µl buffer.

The conserved internal transcribed spacer (ITS) sequences of nematodes were amplified using the universal primer sets described by Zhu et al. (2002). The primer sequences were NC5-forward: 5'-GTA GGT GAA CCT GCG GAA GGA TCA TT-3' and NC2-reverse: 5'-TTA GTT TCT TTT CCT CCG CT-3' (Zhu et al., 2002). The polymerase chain reaction (PCR) reaction contained 15 µl Master Mix 2X (Phusa Biochem, Vietnam), 0.75 µl of each primer (final primer concentration of 0.4 mM), and 3 µl template DNA. The amplification conditions for the PCR assay were 94°C for 5 min for initial denaturation, followed by 35 cycles of denaturation at 94°C for 30 s, annealing at 60°C for 30 s, extension at 72°C for 30 s, and a final extension at 72°C for 5 min (Zhu et al., 2002). Verification of the gene amplification process was conducted through electrophoresis on a 1% agarose gel supplemented with GelRed ® DNA stain (Biotium, Fremont, CA, USA). This procedure was performed at 135 V for 30 min. To determine the size of the PCR products, a 100 bp ladder (BioFact, Daejeon, Republic of Korea) was used as a reference.

DNA sequencing and phylogenetic tree construction

The PCR products were sent and sequenced for both strands using the primer sets in the PCR assay. DNA sequencing was conducted using 1st BASE services (Apical Scientific, Sdn. Bhd, Selangor, Malaysia). In case of the appearance of a non-specific band in the PCR product, DNA gel extraction of the specific band was performed before the sequencing step. This procedure was also carried out by 1st BASE services (Apical Scientific, Sdn. Bhd, Selangor, Malaysia). A phylogenetic tree was generated using MEGA XI software (Tamura et al., 2021) by applying the maximum likelihood algorithm with 1000 bootstrap replicates. The sequences obtained from this study were aligned and compared with those from GenBank using the BLAST search tool (NCBI, 2024).

Statistical analysis

Parasite prevalence was determined using descriptive statistics with 95% confidence intervals (SPSS version 24). The standard deviation represents the variations in the intensity of infection and the age of animals.

RESULTS

The captured tigers and bears

Upon arrival at the center, rescued animals were cared for and raised in accordance with the Rescue Technical Process, as stipulated by Decision No. 31/2022/QD-UBND, dated September 7, 2022, issued by the Hanoi People's Committee. This decision delineated the processes and technical economic norms for the care and rescue of wild animals in Hanoi. As of November 2024, the center houses 25 tigers and 21 bears, all of which have been rescued from illegal trafficking and transportation by the authorities, as well as from individuals who voluntarily surrendered and donated them. In the context of tigers, the smallest individual weighed approximately 1.5 kg at around two weeks of age and required milk feeding. Some cubs were rescued in a condition of severe obesity, with joint deformities resulting from the enforced weight gain. Furthermore, some individuals (non-public data) were anesthetized during transport, which led to a decline in their health, whereas others were confined to restricted enclosures, limiting their movement. Regarding bears, the smallest individual weighed approximately 2 kg at approximately one month of age and was similarly fed with milk. These bears were rescued from inadequate welfare conditions, predominantly from honey farms where they displayed emaciated bodies and damaged paw pads due to ingrown claws, a consequence of being kept in confined spaces without sufficient care. The entirety of tigers and bears were maintained and nurtured at the rescue center rather than being released into their natural habitats. Detailed information regarding the age and sex of the animals is shown in Table 1.

Table 1. The number of tigers and bears captured at Hanoi Wild Rescue Center, Vietnam, from June to May 2024

	Tiger (*Panthera tigris*) (No. of individuals)	**Bear (*Ursus thibetanus*) (No. of individuals)**
Age (year) (mean±SD)	10.8±3.6	11.2±4.6
< 10	9	9
From 10 to 20	16	10
≥ 20	0	2
Gender		
Male	15	10
Female	10	11

No: Number; Mean: Average value; SD: Standard deviation

The prevalence of intestinal helminths

Among the 46 tigers and bears housed at the rescue center, five out of 25 tigers and five out of 21 bears were found to be infected with parasitic worms, representing 20.0% and 23.8%, respectively (Table 2). Based on the morphological characteristics (Soulsby, 1982), it can be preliminarily concluded that these animals were infected with trematode eggs (*Fasciola*-like eggs) (2/46 infected animals, accounting for 4.4%), *Toxascaris leonina (Ta. leonina)* eggs (3/46 infected animals, accounting for 6.5%), and *Strongyle* eggs (8/46 infected animals, accounting for 17.4%) (Table 2; Figure 1.). Notably, tigers were infected with all three types of helminths, whereas bears were infected only with *Strongyle* roundworms. Regarding trematode eggs, only two tigers exhibited very low infection intensity (0.2 ± 0.0 EPG). Because of the low intensity of infection, it was not succeeded to identify this fluke egg using molecular techniques. The intensity of roundworm egg infection was higher, particularly for *Ta. leonina* eggs (7.7 ± 6.5 EPG). The infected individuals were distributed across different cages. Both the infected and uninfected individuals were detected in the same cage. Two tigers were found to be co-infected with both trematode and *Strongyle* eggs, as well as *Strongyle* and *Ta. leonina* eggs.

Table 2. Prevalence of intestinal helminths in Bengal tigers (*Panthera tigris*) and Asian black bears (*Ursus thibetanus*) at Hanoi WRC, Vietnam in November 2024

Items	**Tiger (*Panthera tigris*) (n=25)**	**Bear (*Ursus thibetanus*) (n=21)**	**Total**
Fluke	2/25 (8.0%)	0/21 (0.0%)	2/46 (4.4%)
Rate of infection (%)	(95%, CI: 1.0-26.0)		(95%, CI: 0.0-10.2)
Intensity of infection (EPG) (mean ± SD)	0.2±0.0	0	
Strongyle type	3/25 (12.0%)	5/21 (23.8%)	8/46 (17.4%)
Rate of infection (%)	(95%, CI: 2.6-31.2)	(95%, CI: 8.8-47.2)	(95%, CI: 6.4-28.3)
Intensity of infection (EPG) (mean ± SD)	1.4±1.1	3.6±4.1	
Toxascaris leonina	3/25 (12.0%)	0/21 (0.0%)	3/46 (6.5%)
Rate of infection (%)	(95%, CI: 2.6-31.2)		(95%, CI: 0-13.7%)
Intensity of infection (EPG) (mean ± SD)	7.7±6.5	0	
Total	5/25 (20.0%) (95%, CI: 6.8-40.7)	5/21 (23.8%) (95%, CI: 8.8-47.2)	

n: The number of animals examined; EPG: Egg per gram; mean: Average value; SD: Standard deviation; CI: Confidence interval

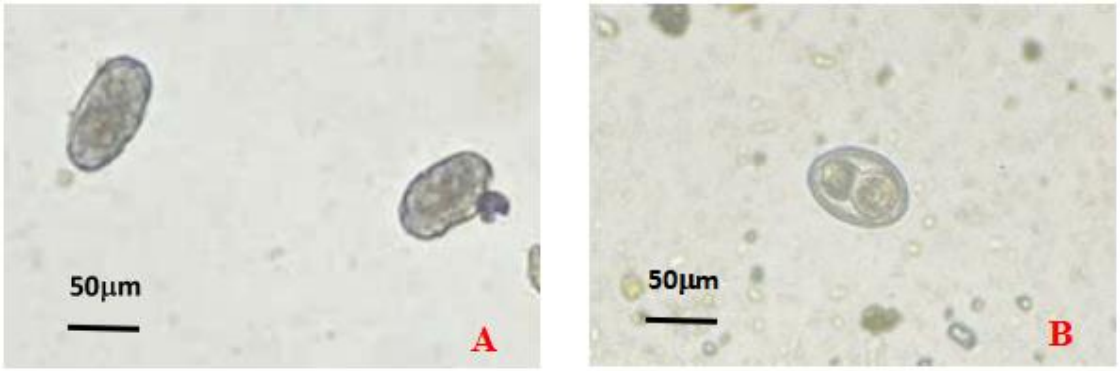

Figure 1. The eggs detected in the present study. **A:** Strongyle eggs, **B:** *Toxascaris leonina*.

Molecular analysis

Using a PCR assay, the internal transcript space (ITS) gene was successfully amplified, yielding a size of 850 base pairs for the *Strongyle* worm and approximately 900 base pairs for *Ta. leonina* (Figure 2). The gene sequencing process yielded two sequences of *Strongyle* worm, each comprising 806 nucleotides, and a sequence of *Ta. leonina* contains 825 nucleotides. *Strongyle* eggs were identified as *A. ceylanicum* by comparison with the reference sequences available in GenBank (Figure 3). The nucleotide similarity between the two *A. ceylanicum* sequences obtained in this study were 100%. Furthermore, the nucleotide similarity rate between these sequences and the reference sequences (LC036567, DQ381541, KM066110, PP527745, KF279132, KF279134, KF279135, and OR826944) ranged from 99.71% to 100%. Sequences exhibiting 100% identity with the sequences in this study included PP527745, which corresponds to *A. ceylanicum* isolated from humans, and KF279132 and OR826944, which correspond to *A. ceylanicum* isolated from wolves. The results of gene sequencing confirmed that the ascarid eggs isolated from tiger feces were identified as *Ta. leonine* (Figure 4). Nucleotide identity between *Ta. leonina* sequences in this study and those referenced from GenBank

ranged from 96.28% to 100%. A nucleotide identity of 100% was observed between *Ta. leonina* sequence in this study and sequences derived from wildlife animals, such as tigers (*Panthera tigris* spp.), lynx (*Lynx lynx*), and leopard (*Panthera leo*; GenBank accession numbers: MK309895, JF837179, JF837175, MK309905, KR999999, JF837177, MK381264).

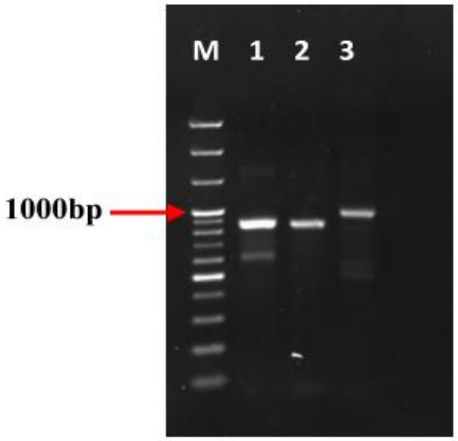

Figure 2. The image of PCR products amplified from the partial internal transcript space (ITS) gene. M: 100bp ladder; lane 1: PCR product amplified from Strongyle eggs isolated from tigers; lane 2: PCR product amplified from Strongyle eggs isolated from bears; lane 3: PCR product amplified from *Toxascaris leonina* eggs isolated from tigers.

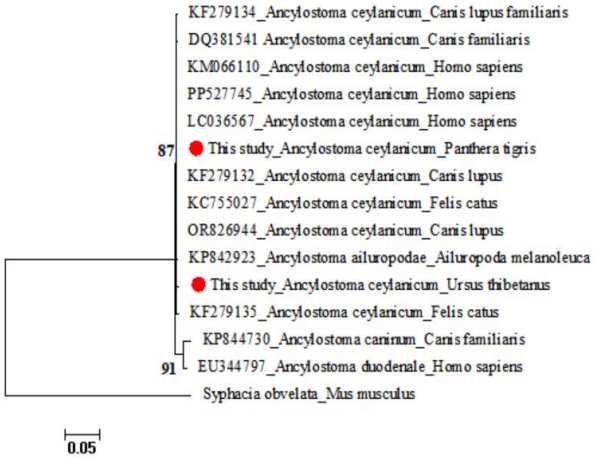

Figure 3. The phylogenetic analysis of *Ancylostoma ceylanicum* based on the partial ITS gene using the Maximum likelihood algorithm with 1,000 bootstrap replicates on MEGA XI. The *A. ceylanicum* isolates in this study were indicated by solid red circles.

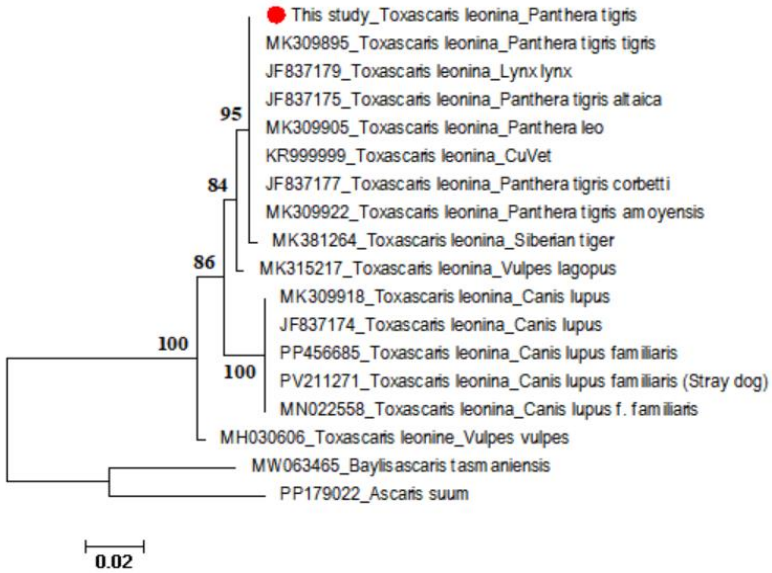

Figure 4. The phylogenetic analysis of *Toxascaris leonina* based on the partial ITS gene using the Maximum likelihood algorithm with 1,000 bootstrap replicates on MEGA XI. The *Toxascaris leonina* isolate in this study was indicated by solid red circles.

DISCUSSION

Wildlife species, including primates and carnivores, are vulnerable to gastrointestinal parasites, particularly when confined to cages or restricted areas, where inadequate sanitation, feeding management, and crowding can elevate the risk of parasite exposure and transmission (Malan et al., 1997). Consequently, these conditions can compromise the immune systems of animals, resulting in decreased resistance and increased mortality rates (Malan et al., 1997; Bista et al., 2017; Adhikari et al., 2023). Identifying parasitic infections in wildlife is crucial for developing preventive strategies to reduce the risk of reinfection, enhance animal health, and mitigate the transmission of pathogens to domestic animals and humans.

The diversity of nematode species was notable in Bengal tigers and Asian black bears, with *Ascaris* spp. being detected at the highest rate (57.14%), followed by hookworms (37.71%). Additionally, several other nematodes have been identified, including *Toxocara* spp., *Strongyloides* spp., *Toxascaris leonina*, *Trichuris* spp., *Capillaria* spp., *Strongylus* spp., and *Nematodirus* spp. except for *Ta. leonina*, other roundworms have been identified only at the genus level (Liza et al., 2020a; b; Chiu et al., 2021; Maharjan et al., 2025). In the present study, two types of nematodes were detected, including *Ta. leonina* in Bengal tigers and *Strongyle* spp. in both Bengal tigers and Asiatic black bears. *Ta. leonina* can be readily identified based on egg morphology (Soulsby, 1982). However, for *Strongyle* spp., molecular techniques are essential for species-level identification, and *A. ceylanicum* has been identified as a species that parasitizes these animals in this study.

Ancylostoma ceylanicum is commonly known as a canid hookworm that is highly endemic in the Asia-Pacific region (Colella et al., 2021). The prevalence of *A. ceylanicum* in dogs ranges from 4.7% to 96.5%, and approximately 50% in cats in countries in the Asia-Pacific region (Kladkempetch et al., 2020; Zendejas-Heredia et al., 2022; Tenorio et al., 2024). In Vietnam, the prevalence of *A. ceylanicum* in dogs is 25% in Dong Thap and Soc Trang provinces (Nguyen et al., 2016), and 67% in Dak Lak province (Ng-Nguyen et al., 2015), and no information on the detection of *A. ceylanicum* in cats has been reported. In addition, the detection rate of *A. ceylanicum* in humans has fluctuated from 5.3% in Thailand (O'Connell et al., 2018) to 95.5% in Timor Leste (Papaiakovou et al., 2017) and 52.1% in Long province, Vietnam (Bui et al., 2021). In addition to the detection of this hookworm in domestic dogs and cats, *A. ceylanicum* has been detected in some wild animals such as Asian golden cats (*Viverricula malaccensis*), leopard cats (*Felis bengalensis*), civets (*Felis temminchii*), coyotes (*Canis latrans*), and dingos (*Canis lupus dingo* (Smout et al., 2013; Zendejas-Heredia et al., 2024). However, no information has been reported on the detection of *A. ceylanicum* in wild animals such as Bengal tigers and Asiatic black bears, although some reports have documented the detection of other *Ancylostoma* species, such as *A. caninum, A. tubaeforme in* black bears (Crum et al., 1978; Foster et al., 2004); *A. malayanum, A. ceylanicum, A. caninum,* and *A. braziliense* in captive sloth bears in India (Baylis and Daubney, 1922). The most significant consequence of hookworm infection in both humans and animals is iron deficiency anemia (Pasricha et al., 2008; Bowman et al., 2010). In cases of acute heavy infection, blood loss exceeding 500 mL can occur within the first two weeks in dogs (Rep et al., 1971). Severe anemia may manifest in human patients due to depletion of erythrocytes, iron, and hemoproteins (Jourdan et al., 2018). *A. ceylanicum* is estimated to cause nearly 100 million cases in Southeast Asia and the Pacific region, making it the second most prevalent hookworm infecting humans in this area (Traub, 2013; Traub et al., 2021; Colella et al., 2021). These findings support the assertion that *A. ceylanicum* represents an emerging zoonotic health issue in the Asia-Pacific countries.

The *Ta. leonina* is classified as an ascarid nematode, alongside *Toxocara canis* and *T. cati*, affecting carnivorous species, including both canines and felines (Okukewicz et al., 2012). The global prevalence of *Ta. leonina* in domestic dogs and cats is estimated at 2.9% and 3.4%, respectively (Rostami et al., 2020). However, this prevalence is significantly higher in wild canid species, such as the red fox (*Vulpes vulpes*), jackal (*Canis aureus*), wolf (*Canis lupus*), raccoon dog (*Nyctereutes procyinides*), and arctic fox (*Alopex lagopus*), reaching up to 52.2% (Okulewicz et al., 2012), and in wild felid species, including lions (*Panthera leo*), bengal tigers (*Panthera tigris*), jaguars (*Panthera onca*), pumas (*Puma concolor*), and lynx (*Lynx* sp.), reaching 57.1% (Okulewicz et al., 2002). The intensity of *Ta. leonina* infection is notably high, with 27,150 eggs per gram (EPG) in South China tigers, China (Chiu et al., 2021) or 65 eggs per field of view in direct microscopic smear preparations in an Angolan lion in the Wroclaw Zoological Garden, Poland (Okulewicz et al., 2012). In the current study, *Ta. leonina* was detected in three out of 25 tiger fecal samples, accounting for 12.0%, with an intensity ranging from 8 to 135 eggs per 5 g of feces. Another study has reported the prevalence of *Ta. leonina* in dogs in Vietnam was 0.54% (Nguyen et al., 2020). These findings suggest that, *Ta. leonina* is more prevalent in wildlife than in domestic dogs or cats. Stray dogs and cats infected with *Ta. leonina* often/usually showed poor nutritional condition, leading to susceptibility to other infections (Rostami et al., 2020) or affected body weight and fur quality in heavily infected arctic foxes (Bauer et al., 2024). Although the transmission of *T. canis* and *T. cati* to humans is well documented globally, causing conditions such as ocular larval migrans, visceral migrans, and covert toxocariasis (Magnaval et al., 2001), there is limited information regarding *Ta. leonina* as a zoonotic pathogen. Although infrequently reported, *Ta. leonina* is considered a potential agent for hypereosinophilia in humans (Rausch and Fay, 2011).

In this study, both *A. ceylanicum* and *Ta. leonina* are classified as soil-transmitted helminths and are characterized by a direct life cycle that does not involve intermediate hosts. Additionally, *Ta. leonina* can invade and persist for extended periods in paratenic hosts, including laboratory animals and natural rodents, such as voles and mice, which may serve as a food source for carnivores (Okoshi and Usui, 1968; Okulewicz, 2008; Rausch and Fay, 2011). Following the fecal examination results, the animals in this study were administered a deworming drug (Sanpet 125 mg containing praziquantel 25 mg and pyrantel pamoate 100 mg, Hanvet company, Vietnam) following the manufacturer's instructions. Two weeks later, feces were collected for re-examination, and no eggs were detected. However, in zoological settings or

confined environments, eradication of *A. ceylanicum*, *Ta. leonina* and other soil-transmitted helminths present significant challenges because of their persistent presence in the surrounding environment, which facilitates reinfection in carnivores (Singh et al., 2006; Bartosik and Górski, 2010; Okulewicz et al., 2012).

CONCLUSION

This study represents the first report on the detection of *Ancylostoma ceylanicum* in Bengal tigers (*Panthera tigris*) (12.0%) and Asian black bears (*Ursus thibetanus;* 23.8%) within the Hanoi WRC, Vietnam. Alongside *Toxascaris leonina*, which infects 12.0% of tigers, have been identified as potential zoonotic pathogens. The findings of this study highlighted the risk of nematode transmission from wildlife to humans and domestic animals in the Hanoi WRC and its surrounding regions. Therefore, further research is needed to investigate gastrointestinal parasites in other animal species within the Hanoi WRC as well as in domestic animals in adjacent areas. The development of a prevention program for eliminating helminths in the Hanoi WRC and its neighboring areas can be considered in future studies.

DECLARATIONS

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Availability of data and materials

The data to support the findings of this study is available upon reasonable request from the corresponding author.

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Author's contributions

Yen Thi Hoang Nguyen Planned and designed the research, analyzed the data, wrote, edited, and revised the manuscript. Duc Minh Nguyen collected samples and provided significant information about Hanoi WRC for writing the manuscript. Thị Thu Hang Trinh collected samples and provided significant information about Hanoi WRC for writing the manuscript. All authors have read and approved the final edition of the submitted manuscript.

Competing interests

The authors declare that they have no conflict of interest.

Ethical considerations

This paper was originally written by the authors and has not been submitted or published elsewhere. The authors checked the text of the article for plagiarism index and confirmed that the text of the article is written based on their original scientific results

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