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Zoonotic risks and conservation challenges: Gastrointestinal parasites in wild mammals of Chitwan National Park, Nepal

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ABSTRACT

Gastrointestinal parasites (GIPs) pose a significant threat to wildlife health and biodiversity, impacting reproductive activities, behavior, survival, and population dynamics. Identifying parasitic infections in wild animals can help to mitigate extinction risk and support conservation efforts. This study investigates the prevalence, diversity, and zoonotic risks of GIPs in six large wild mammals in Chitwan National Park, Nepal. Fresh fecal samples were collected between December 2022 and April 2023 and examined using direct wet mount and concentration methods. By analyzing 63 fecal samples: Royal Bengal Tiger (*Panthera tigris*) (n = 7), Asian elephant (*Elephus maximus*) (n = 9), One-horned rhinoceros (*Rhinoceros unicornis*) (n = 10), Sloth bear (*Melursus ursinus*) (n = 9), Spotted deer (*Axis axis*) (n = 25), and Rhesus Monkey (*Macaca mullata*) (n = 3), we identified 19 GIP types: 3 protozoan species (*Balantioides coli, Isospora* spp., and coccidia) and 16 helminth species, revealing an 85.7% infection rate. Helminths had a higher prevalence (85.7%) than protozoans (22%). Among helminths, nematodes were the most prevalent (69.8%) followed by trematodes (38.0%) and cestodes (17.4%). Eleven types of nematodes, three types of cestodes, and two types of trematodes were recorded. Multiple infections were more common than single infections. The high prevalence of GIPs indicates a major health issue that could affect species survival and conservation efforts in Chitwan National Park, highlighting the need for proactive conservation and health monitoring strategies for conservation.

1. Introduction

Gastrointestinal parasites (GIPs) are a major concern for wildlife, affecting health, reproductive success, behavior, survival, and population dynamics (Albon et al., 2002; Hudson, 1986, 2001; Hudson et al., 1998; Stien et al., 2002). These parasites include protozoa, nematodes, cestodes, and trematodes that infect the host's gut through ingestion of infective cysts, oocyst, egg, and larva leading to malnutrition and increased disease susceptibility (Soulsby, 2012). GIPs are a common health issue in wild animals in Nepal (Shahi and Gairhe, 2019; Shrestha & Maharjan, 2017) and globally (Kobbekaduwa et al., 2017; Zazay et al., 2023).

Understanding the epidemiology of GIPs; the parasite interaction between wildlife, livestock and human health, referred as the one health framework is critical in developing effective management strategies and ensuring a healthy ecosystem for conservation of wildlife and livestock productivity and public health (Obanda et al., 2019). Wildlife often serves as reservoirs for parasites that can infect domestic animals or humans (Shirbhate, 2007). Additionally, wild animals may act as an intermediate or temporary host, spreading and amplifying various infections over time and space, or become victims of disease spillover from domestic animals and or humans (Kock, 2014). The prevalence of parasites in wildlife varies by species, habitat, environmental conditions, and host immunity (Brearley et al., 2013; Maganga et al., 2023; Sharma and Achhami, 2022).

Gastrointestinal parasites posed significant challenges to wildlife in protected areas of Nepal, impacting both animal health and ecosystem dynamics (Aryal et al., 2022; Sharma and Achhami, 2022; Subedi et al., 2023). Wildlife species, such as primates, ungulates, and carnivores are particularly susceptible to GIPs, due to factors like diet, social behaviours and environmental exposure that increase parasite transmission and leading to reduced fitness and increased mortality rates (Adhikari et al., 2023; Bista et al., 2017; Subedi et al., 2023). Identifying parasitic infections in wildlife is essential for reducing extinction risk and

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supporting conservation (Pedersen et al., 2007).

Previous studies in Nepal on GIPs are sparse and often species-specific. Studies have identified *Paramphistomum* spp., *Fasciola* spp., *Strongyle* spp., *Strongyoides* spp., *Trichuris* spp. and *Moniezia* spp., in deer species (Sapkota et al., 2021); *Eimeria* spp., *Anoplocephala* spp., *Fasciola* spp., *Paramphistomum* spp., *Nematodirus* spp., *Trichostrongylus* spp., *Chabertia* spp., *Bunostomum* spp., *Oxyuris* spp., *Dromeostongylus* spp., *Haemonchus* spp., *Ascaris* spp., *Strongyloides* spp., *Schistosoma* spp., *Moniezia* spp., *Toxocara* spp., *Trichostrongylus* spp. in rhinoceros (Paudel et al., 2022; Shahi and Gairhe, 2019); *Strongylus* spp., *Trichostrongylus* spp., *Fasciola* spp., *Paramphistomum* spp., *Ascaris* spp., *Anoplocephala* spp., *Schistosoma* spp., *Dicrocoelium* spp., *Moniezia* spp., *Oesophagostomum* spp., *Chabertia* spp. in elephant (Shahi and Gairhe, 2019; Dahal et al., 2023). However, studies on GIPs in wild animals in CNP remain very limited. There is not any report on GIPs of the protected species like: tiger, and sloth bear.

These findings emphasize the significance of GIPs as an important health concern in wild animals and their zoonotic importance and need of proper monitoring and managing parasitic infection to protect wildlife populations and prevent cross-species transmission. The proximity of wildlife habitats to human settlements in Chitwan intensifies the risk of zoonotic transmissions, particularly from parasites such as *Strongyloides* spp. and *Toxocara* spp (Shahi and Gairhe, 2019; Sapkota et al., 2021; Paudel et al., 2022; Dahal et al., 2023). *Strongyloides* spp poses risks to local communities through contaminated water sources (Echazú et al., 2015). Thus, the study aimed to assess the prevalence, diversity, and richness of GIPs in six large wild animals in Chitwan National Park, Nepal including IUCN red listed wildlife: *Panthera tigris* (Bengal tiger, EN), *Melursus ursinus* (sloth bear, VU). *Elephas maximus* (Asian elephant, EN) and *Rhinoceros unicornis* (Greater one-horned rhino, VU) along with *Macaca mullata* (Rhesus monkey) and *Axis axis* (Spotted deer) as baseline study. The six selected mammalian species include endangered and ecologically significant species representing habitat diversity and conservation priorities.

2. Materials and methods

2.1. Study area

The Chitwan National Park (CNP) is the first national park of Nepal established in 1973 AD and listed as a UNESCO World Heritage Site. It extends over three provinces (Bagmati, Gandaki, and Madhesh) and four districts (Chitwan, Nawalpur, Parsa, and Makwanpur) with an area of 952 Km^2 an additional 729 Km^2 buffer zone (Fig. 1). It lies between 27° 16′ 56″ to 27° 42′14″ N and 83° 50′ 23″ to 84° 46′ 25″ E with an altitude ranging from 150 to 815 masl. CNP harbors over 70 mammal species, 541 bird species and 49 amphibians and reptiles, 120 species of fishes (DNPWC, 2013). It consists of seven different types of habitats including sal forest, mixed forest, short grassland, tall grassland, riverine forest, pine mixed forest, and sal mixed forest (Dahal, 2022).

It is home to endangered species like tiger (*Panthera tigris*), one horned rhinoceros (*Rhinocerous unicornis*) and Asian elephant (*Elephus maximus*) The major carnivore species in CNP are leopard (Panthera pardus), wild dog (Cuon alpinus), jungle cat (Felis chaus), fishing cat (Prionailurus viverrinus), Asian palm civet (Paradoxurus hermaphroditus), and Jackal (Canis aureus). The major prey species in CNP are spotted deer (*Axis axis*), sambar deer (*Rusa unicolor*), hog deer (*Axis porcinus*), barking deer (*Muntiacus muntjak*), blue bull (*Boselaphus tragocamelus*), four-horned antelope (*Tetracerus quadricornis*), wild boar (*Sus scrofa*), gaur (*Bos grunniens*), rhesus macaque (*Macaca mulatta*) and langur (*Semnopithecus entellus*).

2.2. Fecal analysis

2.2.1. Sample collection

Fresh fecal samples of targeted wildlife species were collected between 7:00 a.m. and 10:00 a.m. from the roadside, trails, waterways, and grasslands in the CNP. The protocal of sample collection was approved by research and scientific committee of Department of National Park and Wildlife Conservation and Chitwan National Park [Code number of approval letters 420/078/79/DNPWC and 3651/078/79/CNP]. Fecal identification was conducted based on direct observation of defecation, following identification keys mentioned by Chame (2003) and further confirmation with expert nature guides of CNP. Furthermore, the feces of tiger and sloth bear were identified based on size, appearance, signs, and tracks (Biswas and Sankar, 2002). To minimize pseudo-replication of individual samples, minimum distance 500 m was applied between two samples. And the samples from monkeys and spotted deer were collected under direct observation. The freshness of feces was confirmed by observation of defecation, its high moisture content, the presence of mucus, and the absence of insect activity (Cox et al., 2005; Hewavithana et al., 2022). Approximately 5–10 g of fecal matter was collected in a 50 ml dry sterilized plastic vial containing Potassium dichromate (3:1 ratio) as preservative and transported then stored at 4^0c at the Department of Zoology, Amrit Campus, Tribhuvan University for further analysis.

2.3. Microscopic examination

2.3.1. Direct wet mount method

Fecal samples were examined using saline and iodine wet mount methods. A small amount of feces (1 g) was mixed well with a drop of saline and iodine separately in a dry, grease-free glass slide. Debris, plant particles, and solid wastes were removed and the slide was covered with a coverslip. It was then observed under a compound microscope, initially at 10X and subsequently at 40× magnification. Parasitic cysts, oocysts, eggs, and larvae were identified based on literature (Soulsby, 2012; Zajac and Conboy, 2012), and their microphotographs were captured using a camera model of a Samsung F22 model.

2.4. Concentration method

2.4.1. Flotation method

Approximately 2–3 g of fecal sample was mixed well with Zinc Sulphate solution (specific gravity 1.35) and centrifuged at 2000 rpm for 10 min then the tube was further kept in a test tube stand, and more flotation solution was added to the tube to form an upper meniscus, covered with a coverslip on the top of the tube, and left it for 5–10 min, it was examined microscopically at 10X and 40× magnification (Soulsby, 2012; Zajac and Conboy, 2012).

2.5. Sedimentation method

Formalin-ethyl acetate sedimentation technique was used to process the fecal samples. First, 1 g of fecal sample was mixed with 10 ml of normal saline and filtered through mesh gauze into a centrifuge tube. The mixture was centrifuged at 2000 rpm for 10 min. After centrifugation, the supernatant was carefully decanted, and the sediment was washed again with 10 ml of normal saline solution followed by another round of centrifugation. The washing process was repeated until the supernatant became clear.

For fixation, after the final wash, the supernatant was decanted, and 10 ml of 10% formalin was added to the sediment, mixed thoroughly, and allowed to stand for 5 min. Next, 1–2 ml of ethyl acetate was added, and the mixture was centrifuged at 1500 rpm for 10 min. After configuration, the tube was placed in a test tube stand, revealing four distinct layers: the top ethyl acetate layer, a plug of debris, a formalin layer, and sediment at the bottom. The top three layers were carefully decanted, and the remaining sediment was pipetted onto glass slides. A drop of saline and a drop of iodine solution were added separately to the sediment on each side of a glass slide, mixed well, and covered with coverslips. The samples were then examined under a microscope at 10X and

40× magnifications.

Identification of eggs, cysts, oocyst, and larva was performed using the descriptions and key provided by (Soulsby, 2012; Zajac and Conboy, 2012) based on morphological and morphometric characters. Parasites from Strongyloidea group were identified based on their egg morphology and morphometric measurements. Generally, they are large in size, with thin outer shell, a transparent space between shell and morula. Moreover, the distinction between Strongyloidea and Strongyloides was made based on the comparatively small size egg containing developing larvae in case of Strongyloides, and the larger egg with morula inside in case of other Strongyloidea eggs. Additionally, based on revised nomenclature *Balantidium coli* was changed to *Balatioides coli* (Li et al., 2020; Ponce-Gordo and Garcia-Rodriguez, 2021). The size of all identified parasite eggs was measured using a calibrated ocular micrometer under 400× magnification. Parasite richness was calculated as the total number of distinct parasite species found in a single host (Poulin, 1998). The study acknowledges the limitation of samples size and not assessing intensity of infection with parasite egg density per gram of feces. Since most of the identified helminth genera exihibit poor morphological charateristics, accurate identification presents significant challenges. Therefore, further molecular analysis of fecal samples is recommended for more accurate analysis.

3. Results

3.1. Prevalence of GIPs in wild mammals of CNP

A total of 63 fecal samples were collected from six wild mammals in Chitwan National Park: tiger (*Panthera tigris*) (n = 7), Asian elephant (*Elephus maximus*) (n = 9), one-horned rhinoceros (*Rhinoceros unicornis*) (n = 10), sloth bear (*Melursus ursinus*) (n = 9), spotted deer (*Axis axis*) (n = 25), monkeys (*Macaca mullata*) (n = 3). Among them 54 (85.7%) of samples tested positive for one or more GIPs, with 19 distinct parasite species: Three (22%) protozoans (*Balantioides coli, Isospora,* and coccidia) and 16 (85.7%) helminths (Table 1, Fig. 2).

Nematodes were the most common type of helminths (69.8%), followed by trematodes (38.0%) and cestodes (17.4%). In total, eleven types of nematodes (*Strongyle, Strongyloides* spp*, Ascarid, Mullerius* spp*, Trichuris* spp*, Physaloptera* spp*, Protostrongylus* spp*, Nematodirus* spp*, Toxocara* spp*, Capilariid,* and one unidentified species), three types of cestodes (*Moniezia* spp*, Spirometra* spp*., Taeniid* spp), and two types of trematodes (*Fasciola* spp. and *Paramphistomum* spp.) were identified.

3.2. Distribution of GIPs in wild mammals of CNP

The distribution of GIPs varied among the six mammalian species (Table 1). The protozoan *Balantioides coli* was detected only in monkeys, *Isospora* spp exclusively in tigers, and coccidia was recorded in four species: one-horned rhinoceros, elephant, sloth bear, and spotteddeer. Among the helminths, *Strongylus* spp. was recorded in all six species, while *Strongyloides* spp was recorded in five. Some parasites showed host-specific distribution: *Trichuris* spp and *Physaloptera* spp were confined to monkeys only, while *Protostrongylus* spp*, Nematodirus* spp*., Toxocara* spp*.*, *Capillaria* spp*.*, *Taenia* spp*.* , and *Spirometra* spp. were detected only in tigers. *Fasciola* spp. was found in One-horned rhinoceros and potted deer, while *Mullerius* sp. larva was found only in spotted deer, *Paramphistomum* spp. was recorded only in one-horned rhinoceros.

3.3. Parasite richness in wild mammals of CNP

Parasite richness was recorded as the highest diversity in tiger, harboring 12 different species (63.15% N = 19), followed by the one-horned rhinoceros (7 species), spotted deer (5 species), monkey (6 species), sloth bear (5 species), elephant (3 species) (Table 2).

Table 1
Prevalence of protozoan and helminth parasites in six wild animals in Chitwan National Park, Nepal.

Parasites	Host animals	Prevalence (n = 63)	Life cycle
Protozoans			
Balantioides coli	Monkey	4.7% (3)	Direct, human pathogen
Isospora spp.	Tiger,	1.5% (1)	Direct, human pathogen
Coccidia	Greater one horned rhinoceros, Elephant, Sloth bear, Spotted deer	16.6% (10)	Direct, human pathogen
Helminths			
Nematodes			
Strongylus spp.	Tiger, Greater one-horned rhinoceros, Elephant, Sloth bear, Monkey, Spotted deer,	33.3% (21)	Direct, human pathogen
Strongyloides spp.	Tiger, Greater one-horned rhinoceros, Elephant, monkey, Spotted deer,	50.7% (32)	Direct/ Indirect, human pathogen
Ascarid spp.	tiger, Greater one-horned rhinoceros, Sloth bear, Monkey,	17.4% (11)	Direct
Protostrongylus spp.	Tiger	1.5% (1)	Indirect,
Nematodirus spp.	Tiger	1.5% (1)	Direct
Toxocara spp.	Tiger	9.5% (6)	Direct, human pathogen
Trichuris spp.	Tiger, Monkey	3.1% (2)	Direct, human pathogen
Capillaria spp.	Tiger	3.1% (2)	Direct/ indirect, human pathogen
Physaloptera spp.	Monkey	4.7% (3)	Indirect, human pathogen
Mullerius sp.	Spotted deer	7.9% (5)	Indirect
Cestodes			
Taenia spp.	Tiger	1.5% (1)	Indirect, human pathogen
Moniezia spp.	Greater one horned rhinoceros and Sloth bear	7.9% (5)	Indirect
Spirometra spp.	Tiger	7.9% (5)	Indirect, human pathogen
Trematodes			
Fasciola spp.	Greater one-horned rhinoceros and Spotted deer	33.3% (21)	Indirect
Paramphistomum spp.	Greater one-horned rhinoceros	4.7% (3)	Indirect
Unknown Parasite	Tiger	1.5% (1)	

4. Discussions

4.1. Prevalence and geographic comparison

Nepal has very few studies on GIPs of wild animals such as red panda (Bista et al., 2017; Sharma and Achhami, 2022; Shrestha & Maharjan, 2017), one-horned rhinoceros (Paudel et al., 2022; Shahi and Gairhe, 2019), Asian elephant (Dahal et al., 2023; Shahi and Gairhe, 2019), monkey (Adhikari et al., 2023; Bhattarai et al., 2019; Dhakal et al., 2018; Jain & Maharjan, 2023; Pokhrel & Maharjan, 2015; Sapkota et al., 2020; Tandan et al., 2023), ghoral (Adhikari et al., 2021), wild water buffalo (Aryal et al., 2022), spotted deer (Baral et al., 2019; Sapkota et al., 2021), musk deer (Achhami et al., 2016), black buck (Chaudhary & Maharjan, 2017), Himalayan tahr (Thapa & Maharjan, 2015), barking deer (Thapa & Maharjan, 2015), and bat (Adhikari et al., 2020). This

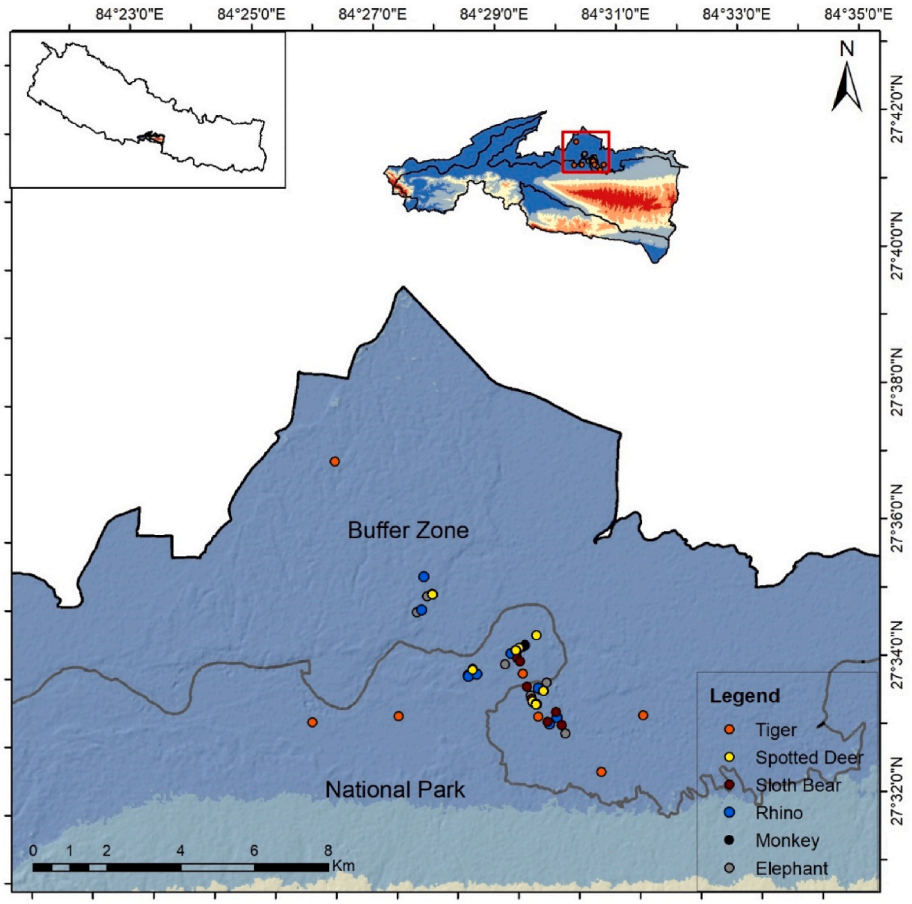

Fig. 1. The map of the study area showing Chitwan National Park, and its buffer zone with the respective locations of sample collection.

study is among the first to document the prevalence of gastrointestinal parasites (GIPs) in free-ranging tigers and sloth bears in Nepal, with significant findings for other globally and nationally threatened species, including the one-horned rhinoceros and Asian elephant.

The prevalence in Nepal (85.7%) aligns with Sri Lanka (86%) (Hewavithana et al., 2022; Sepalage and Rajakaruna, 2020) but exceeds (.) Bangladesh (65.3%) (Ferdous et al., 2023) and 52.7% Brazil (Sprenger et al., 2018), highlighting regional differences in host-parasite dynamics (Fagiolini et al., 2010).

Out of nineteen parasite species identified, nematode showed higher prevalence. Most of these nematodes have a direct life cycle, except *Protostrongylus* spp. and *Mullerius* spp., which are transmitted via contamination of grazing land, food and water. The identified nematodes include *Strongylus* spp., *Strongyloides* spp., *Protostongylus* spp., *Nematodirus* spp., *Ascarid* spp., *Trichuris* spp., *Capillaria* spp., and *Toxocara* spp. Contamination of grazing land with the infective stages of parasites plays significant role in transmission (Wani et al., 2024). Additionally, shared feeding grounds and overlapping habitat use further facilitate parasite transmission, presenting significant challenges for controlling their spread (Ofori et al., 2024). In Chitwan National Park, the shared habitats herbivores facilitates the transmission of parasites through contamination of the environment, including food and water sources. Additionally, visitors may inadvertently contribute to the mechanical spread of parasites, further increasing their prevalence. Furthermore, *Toxocara* spp. was uniquely observed in tigers, suggesting ecological or dietary influences.

4.2. Habitat overlap and cross-species transmission

The high rate of mixed infections observed in the present study (67%) is similar to findings from Sri Lanka (Aviruppola et al., 2016; Farooq et al., 2012; Hewavithana et al., 2022) and India (Zazay et al., 2023). Multiple infections often more damaging than single infections, can weaken the immune system and make animal more susceptible to additional diseases (Wang et al., 2006). Many of the host species in CNP share habitats and resources, such as water and grazing areas, which likely promote parasite transmission. For instance, the proximity of rhinoceros latrine to deer bedding sites suggests significant habitat overlap, increasing the risk of cross-species parasites transmission and emphasizing the importance of further epidemiologic studies (Wani et al., 2024). Therefore, habitat management, livestock regulation and parasite monitoring programs should be regularly conducted for conservation of wild animal. Regular monitoring of parasite prevalence in grazing areas is an essential measure to identify and manage high-risk zones effectively. This includes conducting routine fecal examinations to detect parasite eggs and larvae, mapping grazing patterns to pinpoint heavily contaminated areas, and implementing targeted deworming programs based on the parasite burden in specific regions. Educating local communities and stakeholders about proper grazing practices and hygiene can also enhance the effectiveness of these measures to mitigate gastrointestinal parasite infections in the studied species.

4.3. Helminth dominance

Helminth infections were more prevalent than protozoan infections in this study, consistent with findings from Bangladesh (Ferdous et al., 2023), Sri Lanka (Hewavithana et al., 2022) and India (Zazay et al., 2023). This could be attributed to the resilience of helminth eggs, which can survive and hatch over extended periods in the environment, as well as favorable environmental conditions that support transmission and

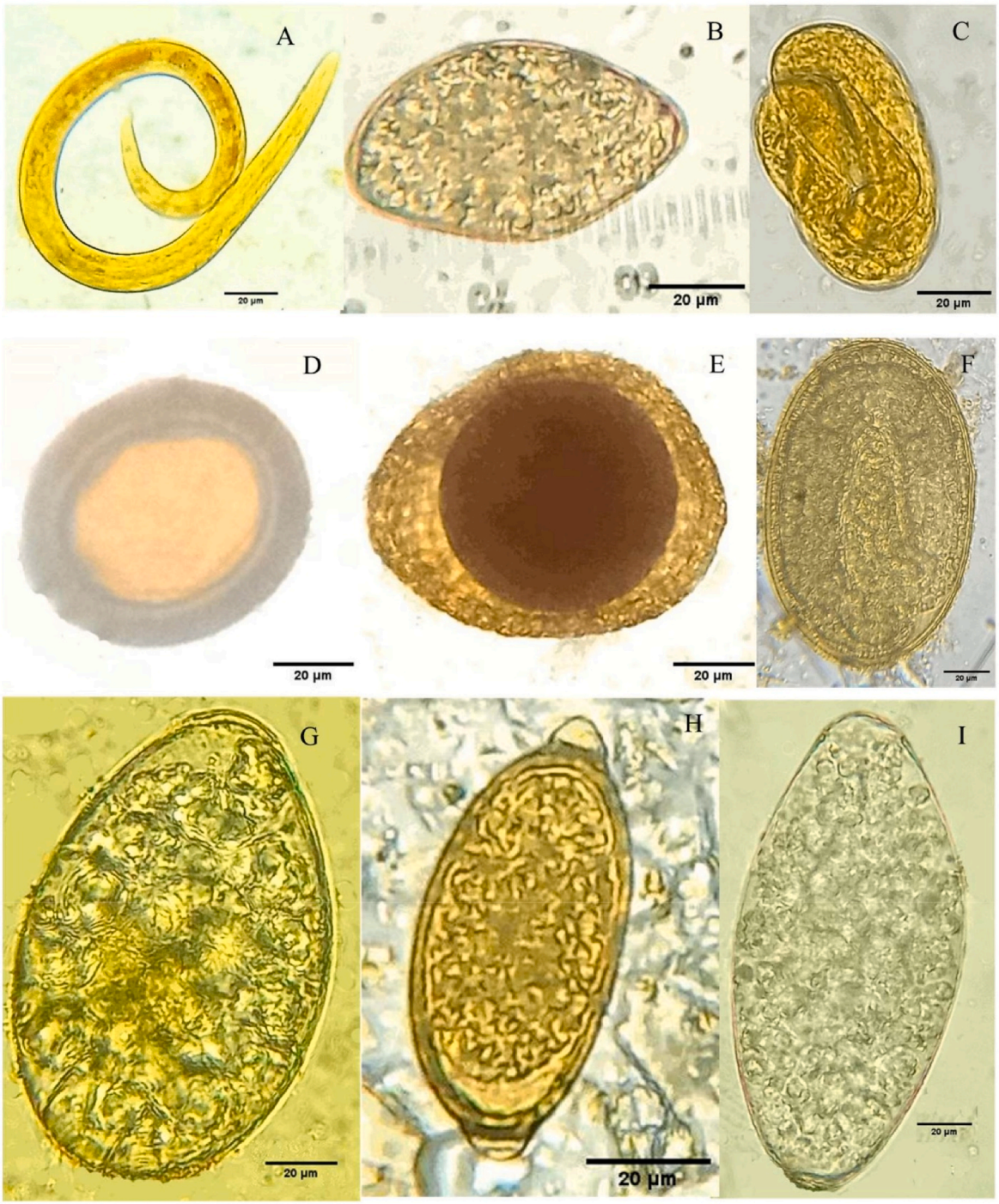

Fig. 2. Egg of: *Protostrongylus* spp in tiger (58x32) [A], Spirometra spp. in Tiger(58x32) [B], *Strongyloides* spp. in Tiger(52x33) [C], *Taenia* spp. in Tiger(48x32) [D], *Toxocara* spp. in Tiger(72x60) [E], Unknown parasite. in Tiger(108x62) [F], *Fasciola* spp. in Rhino(120x80) [G], *Callaria* spp. in Tiger (60x28) [H], *Paramphistomum* spp. in Rhino(142x70) [I].

survival in the park areas (Rahman et al., 2014). Additionally, nematode prevalence exceeds that of trematode and cestodes, likely due to their direct life cycle which completes within a single host with free-living larva stages outside the host. Their infective stages are easily transmitted via contaminated food, water, and soil whereas trematodes and cestodes require one or more invertebrate or vertebrate hosts respectively, to complete their life cycle (Soulsby, 2012; Walker and Morgan, 2014).

Among nematodes, *Strongyloides* had the highest prevalence (50.7%) being detected in all sampled animals. This is likely due to the capacity to undergo trans-mammary transmission. *Strongyloides* is a gut-parasitic nematode that infects a wide range of vertebrates, including amphibians, reptiles, birds and mammals, and humans (Viney and Lok, 2015). It is zoonotic and causes Strongyloidiasis, a usually asymptomatic and self-limiting disease. However, a higher infection rate may lead to diarrhea, severe enteritis, bronchopneumonia, intestinal nodules, skin lesions, and fecal mucus (Thamsborg et al., 2017).

Toxocara sp. was exclusively observed in tiger. It is cosmopolitan and zoonotic, with infections commonly found in both wild and captive felids (Moreira et al., 2023; Uribe et al., 2021). Infection occurs through

Table 2
Parasite richness in studied wildlife in Chitwan National Park, Nepal.

Host animal	Sample size	Parasites			
		Protozoans	Helminths		
			Nematodes	Cestodes	Trematodes
Spotted deer (*Axis axis*)	25	*Coccidia*	*Strongylus* spp., *Strongyloides* spp., *Mullerius* sp.		*Fasciola* spp.
Asian elephant (*Elephus maximus*)	9	*Coccidia*	*Strongylus* spp., *Strongyloides* spp.,		
Greater one horned rhinoceros (*Rhinoceros unicornis*)	10	*Coccidia*	*Strongylus* spp., *Strongyloides* spp., *Ascaria* spp.	*Moniezia* spp.	*Fasciola* spp. *Paramphistomum* spp.
Sloth bear (*Melursus ursinus*)	9	*Coccidia*	*Strongylus* spp., *Strongyloides* spp., *Ascaria* spp.	*Moniezia* spp.	
Monkey (*Macaca mullata*)	3	*Balantioides coli*	*Strongylus* spp., *Strongyloides* spp., *Ascaria* spp., *Trichuris* spp., *Physaloptera* spp.		
Tiger (*Panthera tigris*)	7	*Isospora* spp.	*Strongylus* spp., *Strongyloides* spp., *Protostongylus* spp., *Nematodirus* spp., *Ascarid* spp., *Trichuris* spp., *Capillaria* spp., *Toxocara* spp., Unknown parasite	*Taenia* spp., *Spirometra* spp.	

contaminated environments where eggs are dispersed by infected animals (Moreira et al., 2023). Similarly, The cestode, *Moniezia* spp was reported from one horned rhinoceros and sloth bear. It is a common herbivore parasite, typically transmitted via ingestion of intermediate hosts (oribatid mites) during grazing. In sloth bears, *Moniezia* spp may have been ingested accidently (Sepalage and Rajakaruna, 2020). This cestode has also been reported from red pandas (Shrestha & Maharjan, 2017) and also from blue sheep (Khanyari et al., 2021). Two additional cestodes, *Taenia* sp., and *Spirometra* sp., were found only in tiger, both of which are zoonotic.

Two trematodes *Fasciola* and *Paramphistomum* were identified in this study. *Fasciola* had a high prevalence (42.8%) and was found in one-horned rhinoceros and spotted deer, aligning with the previous studies (Ferdous et al., 2023; Sepalage and Rajakaruna, 2020). This suggests the presence of molluscan intermediate hosts and nearby water bodies. *Paramphistomum,* a common trematode of domestic animals that cause Paramphistomiasis and significant economic loss, was found in wild animals, indicating possible cross-transmission between domestic and wild animals due to habitat overlap (Banerjee et al., 2005; Khanyari et al., 2021; Shrestha & Maharjan, 2017).

4.4. Zoonotic risk

Wildlife arerecongnized as an important source of human infections, including those caused by parasites. Zoonotic parasites of wildlife origin include nematodes, cestodes, trematodes and protozoa (Polley, 2005). Human acquire these parasites through contaminated environments. The infective stages (eggs/larvae) of these parasites can survive in environment under favorable condition for extended periods, thereby increasing the chances of transmission (Soulsby, 2012). Similarly, wild carnivores play significant role in transmission of zoonotic nematodes to human through contaminated food, water, and soil (Otranto and Deplazes, 2019).

In the present study, eleven of the nineteen parasite species identifies are zoonotic; posing potential threats to humans, especially in areas where wildlife and humans interact closely. Zoonotic parasites such as *Strongyloides* spp., and *Toxocara* spp. are particularly concerning, as they can infect both humans and wildlife, potentially leading to disease outbreaks (Moreira et al., 2023). *Strongyloides* spp.can infects a wide range of vertebrates, including humans (Viney and Lok, 2015),. *Toxocara* spp. is well recognized as a nematode parasite of dogs and cats, transmitted to human through infective eggs in soil and fur (Merigueti et al., 2022; Wolfe and Wright, 2003). However, recent studies have showed that wild animals canserve as potential host of this parasite (Holland, 2023), posing a significant threat to human and public health. Numereous cases of human toxocariasis have been reported from various parts of the world (Wolfe and Wright, 2003), but the route of transmission from wild animal has been poorly studied.

Human activities such as livestock raising, and ecotourism can increase the risk of parasite transmission to communities living near protected areas. Therefore, effective management of these zoonotic nematodes require an integrated approach that combines conservation efforts, public health initiatives and interdisciplinary collaboration with epidemiologist, ecologist, biologist and medical and veterinary professionals to address the complex interactions between wildlife, domestic animals and human.

5. Conclusions

This study provides crucial baseline data on the prevalence, richness and distribution of gastrointestinal parasites in six wild mammals of Chitwan National Park, emphasizing the need for continued surveillance and management to prevent cross-species transmission, particularly of zoonotic parasites. The findings highlight the complexity of parasitic infections in wildlife and their potential public health implications. The presence of multiple GIPs, especially those shared between wildlife and domestic animals, highlights the need for comprehensive parasite management strategies in and around Chitwan National Park. Effective management of pasture contamination and minimizing habitat overlap between wild and domestic species could reduce parasite transmission. Further epidemiological studies under one earth approach is essential to protect environment, animal and human health.

CRediT authorship contribution statement

Babita Maharjan: Writing – review & editing, Writing – original draft, Visualization, Validation, Resources, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Payal Jain:** Writing – original draft, Visualization, Validation, Resources, Investigation, Formal analysis, Data curation. **Narayan Prasad Koju:** Writing – review & editing, Writing – original draft, Validation, Supervision, Investigation, Formal analysis, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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