

## Parasites in the Digestive Tract of Lion (*Panthera leo*) at the Safari Park of Gurun Putih Lestari Jantho, Aceh Besar

### Parasit pada Saluran Pencernaan Singa (*Panthera leo*) di Taman Safari Gurun Putih Lestari Jantho, Aceh Besar

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#### ABSTRACT

**Background:** Parasitic infections in wild animals housed in captivity pose a threat to the success of animal conservation endeavors. Factors such as environmental contamination, abrupt alterations in ecological circumstances, and spatial constraints render captive animals vulnerable to stress, consequently compromising their immune systems. These infections have a notable impact on the host. Furthermore, not only captive animals but also personnel, visitors, and the public at large can be at risk of contracting parasites that have the potential to infect various species. **Purpose:** This study seeks to ascertain the occurrence of gastrointestinal endoparasitic infections in lions housed at the Gurun Putih Lestari Safari Park in Jantho, district of Aceh Besar. **Method:** Samples were collected from five lions housed at Gurun Putih Lestari Safari Park in Jantho, district of Aceh Besar. The sampling procedure involved direct collection of lion feces, which were then placed in containers with 10% formalin and labelled accordingly. Sampling was conducted three times at 15-day intervals. The fecal samples were analyzed using four methods: flotation, sedimentation, formol ether, and modified Ziehl-Neelsen method. **Results:** The analysis of fecal samples revealed that three adult lions were found to be infected with *Toxocara cati*, *Toxascaris leonina*, *Ancylostoma spp.*, and *Cystoisospora rivolta*, while two lion cubs were solely infected with *Toxocara cati* and *Ancylostoma spp.* Notably, the modified Ziehl-Neelsen technique did not detect any gastrointestinal endoparasites. **Conclusion:** The findings suggest that lions housed at Gurun Putih Lestari Safari Park in Jantho, district of Aceh Besar are infected by gastrointestinal parasites. It is recommended to administer deworming treatment to lions as a preventive measure against a surge in parasitic infection among the lion population.

#### ABSTRAK

**Latar Belakang:** Infeksi parasit pada satwa liar yang berada di penangkaran merupakan ancaman bagi keberhasilan upaya konservasi satwa. Faktor-faktor seperti kontaminasi lingkungan, perubahan keadaan ekologi yang tiba-tiba, dan keterbatasan ruang membuat satwa di penangkaran rentan terhadap stres, yang pada akhirnya mengganggu sistem kekebalan tubuh mereka. Sistem kekebalan tubuh yang terganggu meningkatkan kerentanan hewan terhadap infeksi parasit. Infeksi ini memiliki dampak yang besar pada inang. Selain itu, tidak hanya satwa di dalam kurungan, tetapi juga petugas, pengunjung, dan masyarakat umum juga berisiko tertular parasit yang berpotensi menginfeksi berbagai spesies. **Tujuan:** Untuk mengetahui kejadian infeksi endoparasit gastrointestinal pada singa yang dipelihara di Taman Safari Gurun Putih Lestari, Jantho, Kabupaten Aceh Besar. **Metode:** Sampel dikumpulkan dari lima ekor singa yang berada di Taman Safari Gurun Putih Lestari di Jantho, Kabupaten Aceh Besar. Prosedur pengambilan sampel dilakukan dengan mengambil langsung kotoran singa, yang kemudian dimasukkan ke dalam wadah yang telah diberi formalin 10% dan diberi label. Pengambilan sampel dilakukan tiga kali dengan interval 15 hari. Sampel tinja dianalisis dengan menggunakan empat metode: pengapungan, sedimentasi, formol eter, dan metode Ziehl-Neelsen yang dimodifikasi. **Hasil:** Analisis sampel feses menunjukkan bahwa tiga ekor singa dewasa terinfeksi *Toxocara cati*, *Toxascaris leonina*, *Ancylostoma spp.*, dan *Cystoisospora rivolta*, sementara dua anak singa hanya terinfeksi *Toxocara cati* dan *Ancylostoma spp.* Khususnya, teknik Ziehl-Neelsen yang telah dimodifikasi tidak mendeteksi adanya endoparasit pencernaan. **Kesimpulan:** Temuan ini menunjukkan bahwa singa yang ditempatkan di Taman Safari Gurun Putih Lestari di Jantho, Kabupaten Aceh Besar terinfeksi oleh parasit gastrointestinal. Disarankan untuk memberikan pengobatan cacing pada singa sebagai tindakan pencegahan terhadap lonjakan infeksi parasit di antara populasi singa.

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## INTRODUCTION

Ex-situ captive breeding is a vital conservation strategy that allows animals to reproduce outside their natural habitats. However, the threat of disease can undermine these conservation efforts. Keeping animals in confined spaces, such as zoos, can lead to high levels of environmental contamination and exert significant pressure on the animals due to limited space and altered living conditions, resulting in chronic stress in captivity. This stress can make the animals' immune systems more susceptible to parasitic infections, weakening their overall health (Mir, et al., 2016). According to Dashe and Berhanu (2020), factors such as diet, environmental conditions, husbandry practices, temperature, and humidity significantly influence the vulnerability of captive wildlife to parasitic infections. Additionally, poor sanitation and inadequate feeding systems can lead to contamination, further exposing captive animals to these infections.

Creating environmental conditions that precisely replicate an animal's native habitat—such as extreme temperatures, humidity levels, breeding periods, and spatial requirements for zoo management—is nearly impossible (Ghazali, et al., 2020). Exposure to different environments alters the biology of wild animals that are brought from their natural habitats into captivity, causing stress and leading to gastrointestinal parasitic infections, which are the most prevalent infections in captive wildlife (Ferdous, et al., 2023). Helminth infections significantly impact host survival (Ganager, et al., 2019). In addition to being detrimental to wildlife, gastrointestinal parasites can also threaten the health of zoo staff, tourists, and the general public, as they can infect multiple host species (Nath, et al., 2021; Rahman, et al., 2023).

According to Batista, et al., (2021), many gastrointestinal parasites can infect carnivorous animals, including *Ancylostoma* sp., *Toxocara* sp., *Hymenolepis* spp., *Taenia* spp., and *Bartiella* spp. In contrast, research conducted by Dawet, et al., (2022) identified the most common gastrointestinal parasites in lions as *Isospora* spp., *Spirometra* sp., and *Sarcocystis* spp. The likelihood of trematode and cestode infections in captive animals is lower due to the indirect life cycles of these worms. Additionally, *Toxascaris leonina* and *Entamoeba* sp. have been found to infect lions Santos, et al., (2022). This study aims to determine the types of gastrointestinal helminths and protozoa that infect lions (*Panthera leo*) kept in Gurun Putih Lestari Safari Park Jantho, district of Aceh Besar.

## MATERIAL and METHOD

### Material

The sampling procedure included the utilization of sterile plastic containers, masks, gloves, and cooler boxes. Sample examination necessitated lion feces samples, distilled water, sheather sugar, methanol, ether, iodine, formalin 10%, carbol fuchsin 1%, acid alcohol 3%, methylene blue 0.1%, immersion oil, cover glass, object glass, microscope, dropper pipette, digital scale, tea strainer, centrifuge tube, mortar, test tube, and tube rack. Furthermore, documentation was facilitated using the Optilab Advance microscope camera.

Fecal samples were collected from three adult lions and two cubs at Gurun Putih Lestari Safari Park in Jantho, district of Aceh Besar. The fecal sampling was conducted directly; the feces expelled by the lions were placed into labeled plastic sample containers containing 10% formalin. The samples were then transported to the Parasitology Laboratory of the Faculty of Veterinary Medicine at Syiah Kuala University for examination. Sampling was performed three times, with an interval of 15 days between each collection. The collected fecal samples were analyzed using four methods: flotation, sedimentation, formol-ether, and modified Ziehl-Neelsen method. For the flotation method, fecal sample was homogenized with saturated sugar, filtered and placed into a centrifuge tube until the surface became convex. An object glass was then attached to the top for observation. For the sedimentation method, fecal sample was homogenized with water, filtered and transferred into a beaker. After which the supernatant was discarded and the sedimentation method repeated. The remaining mixture was placed on an object glass for observation. For the formol-ether method, fecal samples were homogenized with distilled water, filtered into a centrifuge tube, and then centrifuged. Formalin and ether were added and the mixture centrifuged again. The preparation was then dripped onto a glass slide. For the modified Ziehl-Neelsen method, the stool sample was crushed and homogenized with distilled water. The sediment was spread as thinly as possible on a glass slide, fixed with methanol, treated with carbol fuchsin, and rinsed with acid-alcohol. If the slide was dry, methylene blue was applied, followed by rinsing and allowing it to dry before observation.

### Analysis Data

Data on the types of gastrointestinal endoparasites obtained based on flotation, sedimentation, formol-ether, and modified Ziehl-Neelsen methods were tabulated and then analyzed descriptively.

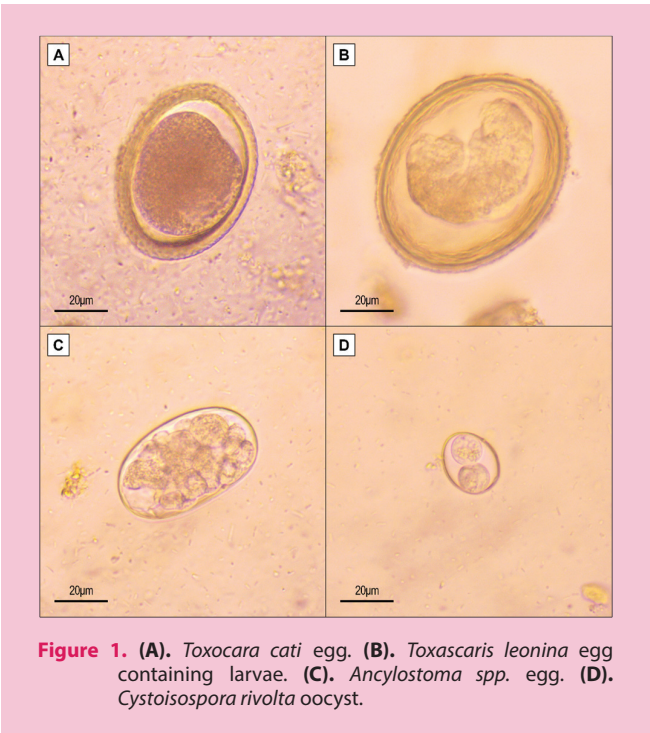
## RESULTS

### Fecal Examination

The results of the examination of lion fecal samples consisting of three adult African lions and two cubs kept at Gurun Putih Lestari Safari Park in Jantho, district of Aceh Besar using flotation, sedimentation, formol-ether, and modified Ziehl-Neelsen method are presented in Table 1. Table 1. presents the average length and width of eggs from gastrointestinal helminths and protozoa that infect lions. Kim, et al., (2020) reported that *Toxocara canis* eggs measure between 71.6-91.2  $\mu\text{m} \times 63.3$ -79.0  $\mu\text{m}$ , *T. cati* eggs measured 63.7-88.1  $\mu\text{m} \times 55.3$ -73.3  $\mu\text{m}$  while Soegiarto, et al., (2022) mentioned the range of *T. cati* egg size values ranging from 61-80  $\mu\text{m} \times 50$ -70  $\mu\text{m}$ , which allows for the differentiation of *T. canis* eggs from those of *T. cati*. In Figure 1, the *T. cati* egg is depicted as a dark, round single-cell embryo encased in a thick shell wall. Moudgil, et al., (2017) reported the presence of *T. leonina* in Asian lions (*Panthera leo persica*), with an average egg size of 74.8-86.2  $\mu\text{m} \times 57.0$ -71.2  $\mu\text{m}$ . *Toxascaris leonina* eggs are characterized by thick outer shell walls that contain light-colored single-cell embryos, and the surface of the shell wall

**Table 1.** Type and Average Size Of Worm Eggs and Gastrointestinal Protozoa Identified In Lions (*Panthera leo*)

Sample	Methods				Type of Parasite	Average	
	Flotation	Sedimentation	Formol Ether	Modified Ziehl-Neelsen		Length (µm)	Width (µm)
Lion-01 (Male)	<i>T. cati</i>						
	<i>T. leonina</i>						
	<i>Ancylostoma</i> spp.	<i>T. cati</i>	<i>T. cati</i>	-	<i>T. cati</i>	68.17 (65.63±76.70)	53.94 (47.40±62.19)
Lion-02 (Male)	<i>C. rivolta</i>						
	<i>T. cati</i>						
	<i>T. leonina</i>	<i>T. cati</i>	<i>T. cati</i>	-	<i>T. leonina</i>	75.55 (68.17±88.65)	65.57 (63.49±74.90)
Lion-03 (Female)	<i>Ancylostoma</i> spp.						
	<i>C. rivolta</i>	<i>T. cati</i>	<i>T. cati</i>				
	<i>T. leonina</i>	<i>T. leonina</i>	<i>T. leonina</i>	-	<i>Ancylostoma</i> spp.	56.22 (47.60±59.36)	35.03 (32.92±37.43)
Cubs-04 (Male)	<i>T. cati</i>						
	<i>Ancylostoma</i> spp.	<i>T. cati</i>	<i>T. cati</i>	-			
Cubs-05 (Male)	<i>T. cati</i>						
	<i>Ancylostoma</i> spp.	<i>T. cati</i>	<i>T. cati</i>	-	<i>C. rivolta</i>	23.56 (20.96±24.30)	20.08 (17.38±21.78)



appears rough or bumpy due to the vitelline membrane. *Ancylostoma* spp. are identified by their oval-shaped eggs, which have thin, smooth shells enclosing a single morula containing 8 to 16 cells, measuring approximately measuring approximately 55-75 × 30-40 µm (Beugnet, et al., 2018). Nagamori, et al., (2021) stated that *Cystoisospora felis* oocysts measuring 38-51 µm × 27-39 µm are slightly larger than *C. rivolta* oocysts measuring 17-27 µm × 15-24 µm and are characterized by thin, tear-like cyst walls containing cysts that are either non-sporulated or sporulated.

DISCUSSION

Captive-bred animals in zoos and safari parks are susceptible to various parasites. This study identified *Toxocara cati* as the most prevalent parasite infecting captive lions. Although *T.*

*cati* is less common than *T. canis*, its zoonotic potential warrants careful consideration, particularly in captive wildlife. *Toxocara cati* eggs can persist in the environment for extended periods. Adult *T. cati* lay eggs in the intestinal lumen of the host, which are subsequently expelled into the environment through defecation. These eggs undergo developmental arrest under optimal soil and climatic conditions. When embryonated eggs are ingested by another host, larvae emerge, invade the intestinal mucosa, and subsequently migrate to the lungs, liver, and kidneys. Transplacental and transmammary transmission are significant routes of infection (Eslahi, et al., 2020). Although toxocarosis does not typically present with respiratory symptoms, as it primarily develops in the small intestine, severe infections can be fatal due to the potential for *Toxocara*’s migration. Additionally, severe infections may lead to intestinal obstruction, resulting in bloating, intestinal wall lacerations, and peritonitis (Beugnet, et al., 2018).

*Toxascaris leonina* also infects captive lions; however, it is less pathogenic in young animals. This species exhibits limited migration into the intestinal wall and has a longer gestation period (48-72 days) compared to *Toxocara canis* (20-35 days) and *Toxocara cati* (25-42 days). As a result, young animals have the opportunity to grow and develop before experiencing the health effects associated with adult worms (Rostami, et al., 2020). The prevalence of *Toxascaris leonina* in definitive hosts varies significantly based on climatic factors, environmental conditions, host age, and season. *Toxascaris leonina* eggs remain viable for up to 40 days at -15°C and can complete their development to the infective stage when returned to 25°C (Okulewicz, et al., 2012). *Toxascaris leonina* can cause gastrointestinal distress and may lead to intestinal obstruction if the infection is severe. It is important to note that *T. leonina* is not more pathogenic than *T. canis* or *T. cati*. Unlike other *Toxocara* species, *T. leonina* larvae do not migrate to the heart, lungs, trachea, or esophagus; instead, they enter the intestinal wall but subsequently return to the lumen to develop into adults. This limited migration to the



intestinal wall results in the absence of transplacental and transmammary transmission.

*Ancylostoma* species infecting lions should also be taken into consideration since growth and development of *Ancylostoma* spp. larvae take place in humid soil environments, such as grassy soil and warm temperatures (Beugnet, et al., 2018). Sangpeng, et al., (2023) have noted that reservoir animals often release zoonotic parasites into the environment in the form of oocysts, eggs, and larvae through feces. The severity of hookworm infection correlates with the amount of blood loss in the animal. While estimating the prevalence of hookworm infection is challenging, common complications include iron deficiency anemia and adverse birth outcomes like low birth weight (Loukas, et al., 2016). Severe *Ancylostoma* spp. infection can also weaken the immune system of the animal, potentially leading to infections by other microorganisms.

The gastrointestinal protozoan *C. rivolta* is a coccidial protozoan parasite that commonly infects members of the Felidae family, particularly captive lions, leading to diarrhea. *Cystoisospora rivolta* typically enters the host through the ingestion of tissues or feces from intermediate hosts and subsequently develops within the enterocytes of the small intestine. Symptoms may include diarrhea, bloody colitis, and vomiting. Infection with *Cystoisospora* is particularly prevalent among animals with compromised immune systems (Petry, et al., 2011). Infection with *Toxocara cati* and *Ancylostoma* species in the two cubs may have occurred because the cubs and adult lions shared the same isolation cage for an extended period before the cubs were moved to new cages. During this time, horizontal transmission likely took place, as parasites present in the feces of the adult lions could have infected the cubs. Following the death of the mother lion due to dystocia, both cubs were fed formula milk, which significantly reduced the likelihood of vertical transmission of parasites through mammary secretions. The direct interaction and close contact between the lions facilitated the spread of parasitic infections. The high incidence of parasitic infections of the same species among captive lions indicates a significant spread of parasites between animals. The presence of visitors and the close proximity of humans to the animals facilitate the exchange of parasites between humans and captive animals. Unsanitary conditions maintained by caretakers can also contribute to the transmission of parasites to the lions. Furthermore, parasites can be transmitted from captive animals to domestic pets and humans (Dashe and Berhanu, 2020).

According to information provided by the lion keeper, the cages are cleaned only once a week, which may exacerbate the spread of parasites among the lions. Additionally, the lack of antiparasitic medications allows parasites to continue to thrive within the lions' bodies, as evidenced by the results of fecal examinations. The prevalence of parasites in captive animals can vary based on husbandry practices, disease prevention measures, parasite-host interactions, and treatment protocols (Avirupola, et al., 2016).

## CONCLUSION

The research found that three adult lions at Gurun Putih Lestari Safari Park were infected with gastrointestinal endoparasites, including *Toxocara cati*, *Toxascaris leonina*, *Ancylostoma* spp., and *Cystoisospora rivolta* while the two cubs were only infected with *Toxocara cati* and *Ancylostoma* spp.

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## CONFLICT of INTEREST

The authors declare that this research was conducted without any commercial or financial relationships that could be construed as a potential conflict of interest.

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## ETHICAL APPROVAL

This research activity did not require ethical review.

## AUTHORS' CONTRIBUTIONS

Conceptualize and design research: FD, RD, AS, YF. Collecting data: FD. Analyzing and interpreting data: FD, RD, AS, YF. Drafting the manuscript: FD, RD, AS, YF. Revising: FD, RD, AS, YF.

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