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Case Report

Severe Parasite Co-Infection in a Captive Bactrian Camel: Case Report

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Abstract: The aim of this study was to document a case of parasite co-infection in a captive Bactrian camel and to supply morphometric data of the found pathogens. It concerned a 20-year-old male animal inhabiting Sofia Zoo, Bulgaria. A decreased appetite and gastrointestinal disorders were observed in it during the summer of 2022. Improvement in the animal's condition was achieved after the administration of antibiotics, sulfonamides, and other symptomatic medicines. However, two weeks after treatment, clinical symptoms reappeared. Then, a diarrheal fecal sample from the animal was subjected to parasitological examination by direct smear and flotation and sedimentation techniques. Multiple infections by helminths (*Trichostrongylus* sp., *Haemonchus* sp., *Oesophagostomum* sp., *Trichuris* sp., and *Dicrocoelium* sp.), ciliates (*Buxtonella cameli*), and protozoa (*Eimeria cameli*) were found, with *E. cameli* being reported for the first time in zoo conditions. Deworming led to the recovery of the general condition and appearance of the animal's feces, but two weeks later, it died suddenly. We considered that the parasitic infection was not the direct cause of the fatal outcome, and its presence, other health disorders, and the advanced age of the animal were among the contributing factors. This case reveals the need to combine planned preventive deworming with routine parasitological diagnostics to take timely and targeted actions to protect the health of animals inhabiting zoo facilities.

Keywords: *Camelus bactrianus*; gastrointestinal parasites; helminths; protozoa; ciliates; parasite morphometry

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1. Introduction

The one-humped camel, or dromedary camel (*Camelus dromedaries* Linnaeus, 1758), and the two-humped camel, or Bactrian camel (*Camelus bactrianus* Linnaeus, 1758) (Artiodactyla, Camelidae), are even-toed ungulates distributed mainly in North Africa and Asia, with the majority of their populations domesticated and raised as livestock. A small number of two-humped camels are left in the wild, standing out as a separate species (*Camelus ferus* Przew, 1878) that is critically endangered [1]. Camels are very hardy animals, well adapted anatomically and physiologically to harsh climatic conditions; nevertheless, they suffer from various parasitic diseases, which are major constraints to the improvement of their health [2].

Parasite infections in camels can lead to nutritional and immune inadequacy, stunted growth and delayed development, infertility problems, adverse effects on the quality of meat and milk, reduced working efficiency, and sometimes death [3,4]. The parasite influence increases significantly when the animals are kept at zoos and other closed facilities, where various factors cause wider distribution and difficult control of parasitoses [5]. In recent years, a variety of helminths, protozoa, and ciliates have been reported parasitizing livestock and zoo camels: *Trichostrongylus* spp., *Trichuris* spp., *Ascaris* spp., *Moniezia*

spp., and *Eimeria* spp. in Egypt [3,6]; *Ostertagia* spp., *Trichostrongylus* spp., *Haemonchus contortus*, *Nematodirus* spp., *Marshallagia* spp., *Trichuris* spp., *Chabertia ovina*, *Bunostomum* spp., *Strongyloides papillosus*, *Thysaniezia ovilla*, *Moniezia expansa, Dicrocoelium* spp., *Fasciola hepatica*, *Hasstilesia ovis*, and *Eimeria* spp. in China [4]; *Nematodirus* spp., *Trichostrongylus* spp., *Haemonchus* spp., *Trichuris* spp. *Marshallagia* spp., and *Eimeria cameli* in Iran [7], *Eimeria* sp. and *Cystoisospora* sp. in different parts of the world [8]; *Paramphistomum* sp., *Fasciola* sp., *Moniezia* sp., *Dicrocoelim* sp., *E. cameli*, *E. dromedarii*, *E. rajasthani, E. pellerdyi*, *Cryptospridium* sp., and *Balantidium coli* in Egypt [9]; *E. cameli*, *E. rajasthani*, and *E. pellerdyi* in Saudi Arabia [10]; *Trichuris* sp., Strongylidae, *Eimeria* sp., and *Buxtonella* sp. in the Ljubljana Zoo, Slovenia [11]; *Trichuris* sp. in Zoological Garden "Ogród Zoologiczny" in Warsaw, Poland [12]; *Trichostrongylus* sp., *Cooperia* sp., *Eimeria bactriani*, and *E. dromedarii* in two zoos located in southern Poland [13]; and *Trichuris* spp. in the Bioparco Zoological Garden of Rome, Italy [14].

Timely and accurate identification of parasites in zoos and breeding centers for rare animals can be decisive for the health of animals by helping with the parasite source recognition and its elimination [11,15]. Lifetime diagnosis of parasitic diseases in veterinary practice is most often performed through coprological investigations. Its correctness depends on the accuracy of the techniques and methods used and especially on the morphological identification of the parasite forms. In connection with the above, the purpose of this report was set, namely to document a case of parasite co-infection in a captive Bactrian camel and to supply morphometric data of the found pathogens.

2. Materials and Methods

2.1. Case History

It concerns a 20-year-old male Bactrian camel inhabiting Sofia Zoo (Bulgaria). The animal lived with Cameroonian goats in the same enclosure, which consisted of an outdoor part (main one, covered by soil and vegetation) and an indoor part (covered by concrete). All animals from the zoo, including this camel, were routinely dewormed according to the accepted preventive plan.

In the summer of 2022, the animal's health worsened—profuse diarrhea, lack of appetite, and weight loss were observed. During 20 days, several courses of treatment were carried out, in which antibiotics, sulfonamides, symptomatic antidiarrheal agents, and stabilizers of the normal intestinal flora were used. As a result of the treatment, the condition of the animal normalized, but two weeks later, diarrhea reappeared. Then, zoo officials approached us with a request to carry out a parasitological examination of the camel.

2.2. Laboratory Investigations

Fresh diarrheal feces were received in the laboratory. A part of the sample was examined immediately, and another part was stored at room temperature (22–25 °C) and was examined on the 10th and 17th days after its receipt. Microscopic examinations were carried out by common flotation (salt solution gravity = 1.18) and sedimentation techniques and direct smear [8]. Imaging and measurement of parasite forms were performed using a Motic Images Plus 3.0 camera connected to an Amplival microscope with accompanying software. Parasite forms were identified morphologically according to the descriptions by Kotelnikov [16], Yagoub [17], Thienpont et al. [18], Foreyt [19], and Abbas et al. [6]. The metric data were statistically analyzed using Microsoft® Excel software—version number 2013.

3. Results

Microscopic examinations of the fresh feces (on the first day after collection) by the flotation technique revealed four types of nematode eggs, three of which were strongylid ones. The strongylid eggs of the first type (Figure 1a) were asymmetrical, with dissimilar poles (one of which was more rounded than the other), dissimilar side walls, smooth shell

surface, and numerous blastomeres. The strongylid eggs of the second type (Figure 1b) were a regular ellipse in shape, with nearly similar wide poles, barrel-shaped side walls, smooth shell surface, and numerous hardly distinguishable blastomeres. The third type of strongylid eggs was similar to the second one, but their blastomeres were clearly distinguished (Figure 1c). Nematode eggs of the fourth type were lemon-shaped and brown-colored, with two protruding, transparent polar plugs, thick walls, and granular content (Figure 1d). Considering the morphological criteria for identification (shape, poles, walls, surface, and content) and the metric features (Table 1), the eggs observed were assigned to the *Trichostrongylus* sp. (asymmetrical, dissimilar poles, and 81–94 µm), *Haemonchus* sp. (elliptical, barrel-shaped side walls, hardly distinguishable blastomeres, and 72–90 µm), *Oesophagostomum* sp. (elliptical, clearly distinguished blastomeres, and 76–85 µm), and *Trichuris* sp. (lemon-shaped, two protruding polar plugs, and 62–72 µm), respectively.

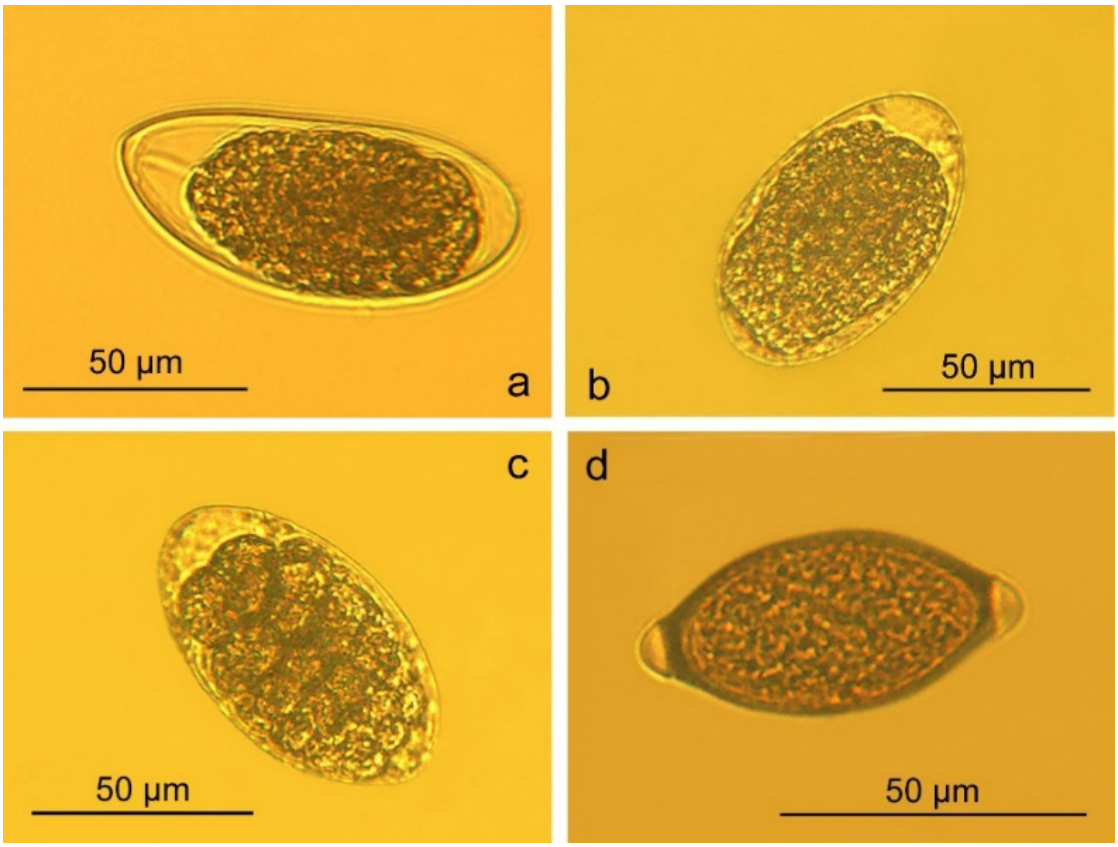

Figure 1. Nematode eggs found in the feces of a *Camelus bactrianus* with multiple parasite infections: (**a**) *Trichostrongylus* sp.; (**b**) *Haemonchus* sp.; (**c**) *Oesophagostomum* sp.; (**d**) *Trichuris* sp. Original pictures.

Examination of feces by the sedimentation technique revealed small (37–40 µm), asymmetrical, and dark-brown, with one operculum and two eye spots, trematode eggs (Figure 2), which were identified as *Dicrocoelium* sp. Oocysts with average sizes of 87/63 µm were also found through the sedimentation technique. They were ovoid shaped and brown to black in color, with a three-layer wall (outer and inner dark brown and middle yellowish) and a micropyle, which was not always visible (Figure 3). They were morphometrically identified as *Eimeria cameli* (Henry and Masson, 1932) Reichenow, 1952.

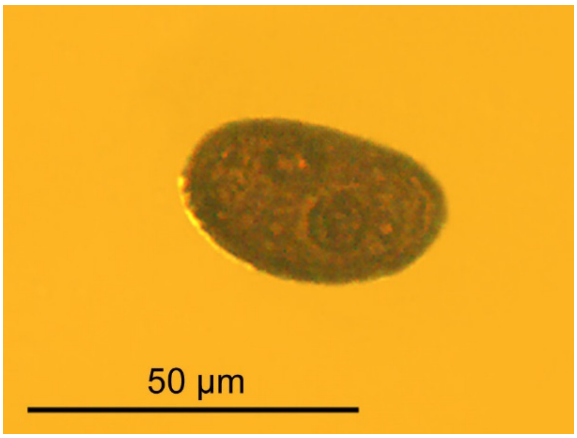

Figure 2. *Dicrocoelium* sp. egg found in the feces of a *Camelus bactrianus* with multiple parasite infection. Original picture.

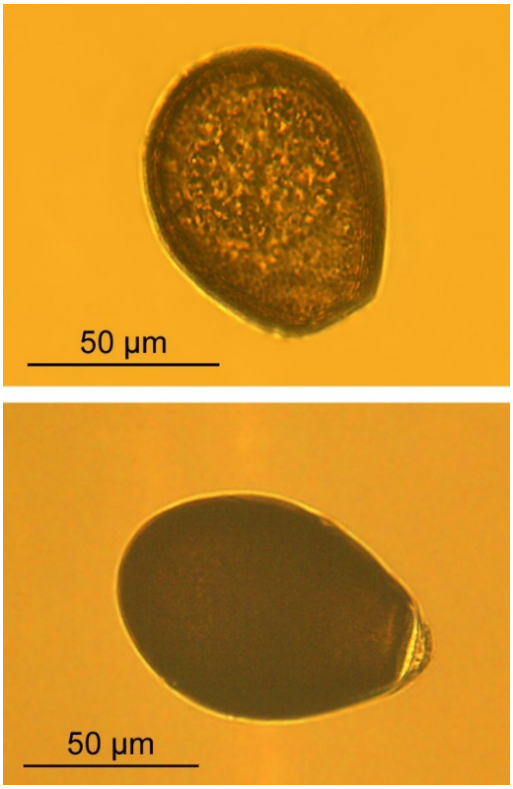

Figure 3. *Eimeria cameli* oocysts found in the feces of a *Camelus bactrianus* with multiple parasite infections. Original pictures.

Direct smear revealed motile unicellular forms with a morphology (Figure 4a) and dimensions (Table 1) resembling those of trophozoites of *Buxtonella cameli* (Boschenko 1925) Esteban-Sánchez et al. 2023. On the tenth day after fecal collection, active trophozoites were still observed. They were relatively less than those visualized in the fresh feces. Motile formations with the same morphology that were smaller in size and closely contacted side by side were also found (Figure 4b), with this probably being the trophozoites in the process of reproduction.

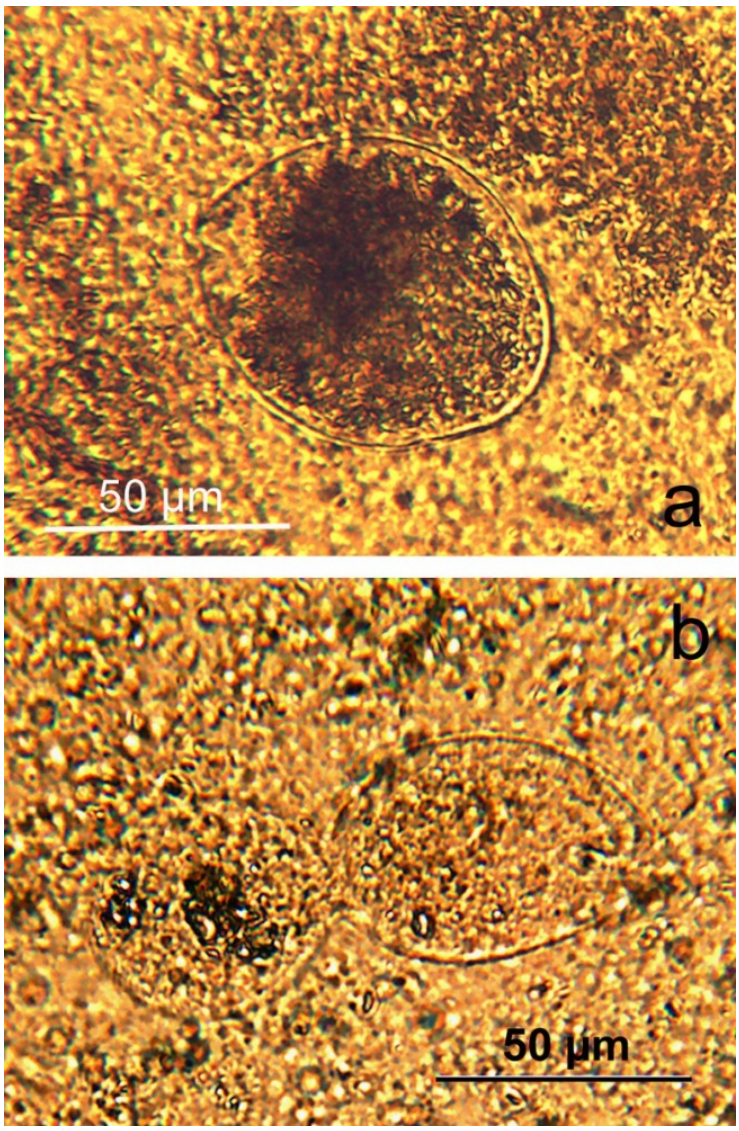

Figure 4. *Buxtonella cameli* trophozoites found in the feces of a *Camelus bactrianus* with multiple parasite infections: (**a**) On the first day after fecal collection. (**b**) On the tenth day after fecal collection. Original pictures.

Gastrointestinal strongylid larvae were found in the diarrheal feces (by direct smear) on day 17 of their collection. They were medium in size (Table 1), with the following morphological features: square head end; tail end with a small spike at the tip; presence of a sheath, which formed a short cone on the caudal larval end; and 16 triangular intestinal cells arranged in two rows, with sharp tips pointing forward and backward (Figure 5). The larvae observed were assigned to *Trichostrongylus* sp.

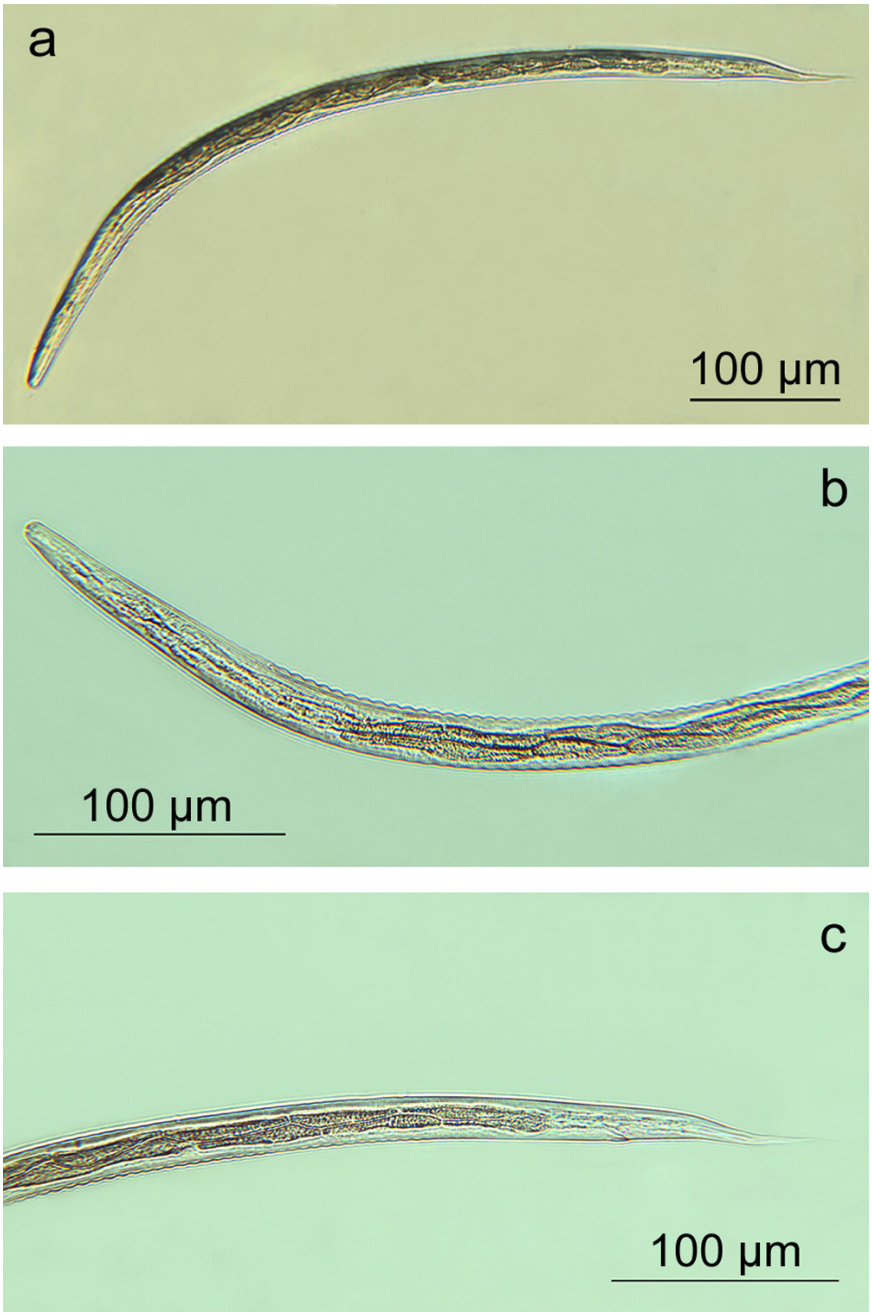

Figure 5. *Trichostrongylus* sp. larva found in the feces of a *Camelus bactrianus* with multiple parasite infections. (**a**) Whole larval body. (**b**) Anterior body end. (**c**) Posterior body end. Original pictures.

Table 1. Comparative measurements (in micrometers) of parasites from a *Camelus bactrianus* (present case) and other sources.

Parasite		Present Case		Other Sources		
		Range	Mean ± SD	Range	Hosts	References
Trichostrongylus sp. eggs	L	81–94	86.18 ± 4.96	70–125	Cattle, Sheep, Goats	Thienpont et al. [18] Foreyt [19]
	W	37–46	42.45 ± 2.62	30–55		
Haemonchus sp. eggs	L	72–90	83 ± 4.94	62–95	Cattle, Sheep, Goats	Thienpont et al. [18] Foreyt [19]
	W	44–50	47.33 ± 1.72	36–50		

Table 1. *Cont.*

Parasite		Present Case		Other Sources		
		Range	Mean ± SD	Range	Hosts	References
Oesophagostomum sp. eggs	L	76–85	81 ± 3.24	65–120	Cattle, Sheep, Goats	Thienpont et al. [18] Foreyt [19]
	W	41–48	44.22 ± 2.05	40–60		
Trichuris sp. eggs	L	62–72	66.8 ± 3.83	70–80	Cattle, Sheep, Goats	Thienpont et al. [18] Foreyt [19]
	W	27–32	30.2 ± 2.17	30–42		
Trichostrongylus sp. L1	L	480–676	602.33 ± 106.68	650–770	Ruminants	Kotelnikov [16]
	W	20–23	21.33 ± 1.53	-		
Dicrocoelium sp. eggs	L	37–40	38.33 ± 1.21	38–45	Cattle, Sheep	Thienpont et al. [18]
	W	23–24	23.33 ± 0.52	22–30		
Eimeria cameli oocysts	L	76–96	86.62 ± 6.05	70–100	Dromedary camels	Abbas et al. [6] Yagoub [17]
	W	56–70	63.10 ± 3.65	52.5–73.8		
Buxtonella cameli trophozoite	L	64–86	74.5 ± 6.29	30–150	Pigs	Foreyt [19]
	W	51–72	61.9 ± 5.15	-		

L1—first-stage larva; L—length; W—width.

Following the initial laboratory studies, the animal was consecutively treated with sulfaguanidine (0.1 g/kg q 24 h × 4 d PO) and fenbendazole (15 mg/kg q 24 h × 5 d PO), considering the recommendations for treating parasitic infections in camels [20]. On the first day after treatment, a fecal sample was obtained for a control test. The feces were fully formed and of normal consistency and color. Eggs of *Dicrocoelium* sp. and single ones of *Trichuris* sp. were observed microscopically. *Eimeria* and ciliates were not detected. Ten days later, the camel died suddenly, and abdominal distension and liver damage were detected. There were no other data from the autopsy, and subsequent parasitological investigations of the dead camel were not performed.

4. Discussion

In this case, we are dealing with a severe infection in a camel infested with numerous parasites from different taxa-nematodes, trematodes, coccidia, and ciliates. According to available research, gastrointestinal parasites are the most frequently registered pathogens in camels, and nematodes of orders Strongylida and Enoplida and protozoa of *Eimeria* sp. are the most common of them [3,4,6,7,9,10,17,21–23]. The present findings of *Trichostrongylus* sp., *Haemonchus* sp., *Oesophagostomum* sp., *Trichuris* sp., and *E. cameli* confirm this trend. Previous studies showed that camels were often co-infected with a large number of parasite species [7,9,24,25], reaching up to 14 different species in a single animal [4]. Having in mind favorable conditions for parasites spreading in zoos, the occurrence of this multiple infection in the present case is not a surprise. Infestation of camels with gastrointestinal strongylids and trichurids were also found in a number of European zoos [11–14,23,26–28], including the Sofia Zoo, where *Trichuris* sp. and *Nematodirus* sp. were registered more than 30 years ago [29].

Literature reports of *E. cameli* establishment are relatively few. This protozoan was found in livestock camels [6,10,17], but according to our best knowledge, there are no records in zoo animals. It should be borne in mind that the diagnosis of *E. cameli* could be missed, especially in captive camels. This could be for several reasons: Infected adult animals in a good general condition rarely excrete oocysts in feces, or excreted oocysts are in low numbers [8]; the excretion of oocysts does not begin immediately with the development of clinical symptoms but occurs later [22]; and false negative results could occur because of technical reasons—*E. cameli* oocysts are large and heavy [8] and may not be detected by flotation coproscopic methods using low-gravity solutions. Probably

because of the third reason, we did not observe oocysts by common flotation in the present case. Here, we found *E. cameli* oocysts only by the sedimentation technique. It is necessary to keep in mind that solutions of specific gravity > 1.28 are recommended for the floatation of this protozoan [22].

Trophozoites of the ciliate parasite that we observed in this case initially were thought to correspond to *Balantioides coli* [(Malmsten 1857) Stein 1863] Alexeieff 1931, as it is the most frequently cited ciliate (as *Balantidium coli*) in camels [9,30–33]. However, *Buxtonella* sp. has been also cited in camels [11,34]. Moreover, one month after the death of the camel, we received fecal samples from other healthy camels of the Sofia Zoo with a request for a prophylactic parasitological examination. These camels entered the zoo a year ago and lived separately from the dead camel. Parasitological tests of the new camels revealed ciliate cysts that were identified by genetic analysis as *B. cameli* [35]. This led us to consider that the ciliates found in the deceased camel likely corresponded to *B. cameli*.

The severe progression of the infection was influenced not only by its multiplicity but also by certain factors associated with zookeeping. The camel lived in an area with a natural covering of grass and bushes, which was difficult to disinfect and clear of parasites. A study at Dublin Zoo, for example, showed that strongylid eggs and larvae persist in soil and grass of camel enclosures, with 1500 *Trichostrongylus* larvae found in 1 kg of grass [36]. The joint keeping of the camel with Cameroonian goats probably contributed to the increased parasite load on the environment and hence the animals. Such a positive correlation between cross-species animal contact and parasite infestation was found in two zoos in Poland, where the level of parasite infection was higher in camels kept in the same enclosure with alpacas and Shetland ponies than those kept separately [13].

Clinical symptoms, such as anorexia, weight loss, and diarrhea observed in the present case, are usual in camels with gastrointestinal parasite infestation [30,37–39]. However, the treatment of such infections has yielded varied results. For example, severe trichurid infection was successfully cured in young 3-year-old camels, but regardless of the same treatment, it ended fatally in an adult 13-year-old animal [37]. In the present case, the advanced age of the animal and the concurrent infestation, factors determining the severe course of gastrointestinal infections [40], probably contributed to the fatal outcome, despite the treatment undertaken.

Morphometric features of parasites found, as a whole, corresponded to those from other sources (Table 1). Only the dimensions of *Trichuris* sp. eggs and first-stage larvae of *Trichostrongylus* sp. were smaller or closer to the lower limits than indicated in manuals [16,18,19]. Certain varieties in color and morphological structures of *E. cameli* oocysts were shown [9,17]. Some authors even distinguished four different oocyst morphotypes within the species [6]. In this case, we observed oocysts corresponding to the first and fourth morphotypes described by Abbas et al. [6]. Thus, our data confirmed the trend of the morphological diversity of *E. cameli* oocysts; furthermore, they were collected from only one animal.

5. Conclusions

Despite the high endurance of camels in adverse conditions, the current case of multiple parasite infestation in a camel was severe. Keeping the animal in a zoo, together with other herbivores, probably contributed to the complication of the infection. The parasitic infection was probably not a direct cause of the camel's death, but its presence, other health disorders, and advanced age were among the factors that led to the fatal outcome of the animal. This case reveals the need to combine preventive planned deworming in zoos with routine parasitological diagnostics in order to take timely and targeted actions to protect the health of the animals.

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