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9 ORIGINAL ARTICLE / ARTÍCULO ORIGINAL

10 First report of *Capillaria Plica* Rudolphi, 1819 (Nematoda: Enoplida: Capillariidae)

11 infection in a Puma, *Puma concolor*

12 Linnaeus, 1771 (Carnivora: Felidae) from southern Brazil

13

14 Primer registro de *Capillaria Plica* Rudolphi, 1819 (Nematoda, Enoplida: Capillariidae)

15 en Puma, *Puma concolor* Linnaeus, 1771 (Carnivora: Felidae) del sur de Brasil

16

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31 Running Head: *Capillaria plica* Rudolphi, 1819 in *Puma concolor* Linnaeus, 1771.

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33

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42 **ABSTRACT**

43 *Capillaria plica* Rudolphi, 1819, a parasitic nematode, infects the urinary tract of both
44 domestic and wild carnivores. Despite its global presence, clinical reports are rare in
45 certain regions, including Brazil. This study documents the first occurrence of *C. plica* in
46 a *Puma concolor* Linnaeus, 1771 (Puma) from Santa Catarina, southern Brazil, marking
47 the first report of this parasite in the southern part of the country. Necropsy and urinary
48 sediment analysis revealed the presence of *C. plica* eggs and adult nematodes. The
49 parasite's diagnosis is challenging due to its subclinical nature and the difficulty in
50 visualizing eggs and adults, especially in wild felids. This case highlights the importance
51 of urinalysis in wild carnivores for diagnosing urinary capillariasis and emphasizes the
52 need for heightened awareness of this parasitic disease, particularly in under-researched
53 regions.

54 **Keywords:** urinary capillariasis – wild carnivores – nematode infection – South Brazil –
55 veterinary diagnosis – parasitology

56 **RESUMEN**

57 *Capillaria plica* Rudolphi, 1819, es un parásito nematodo que infecta el tracto urinario de
58 carnívoros domésticos y silvestres. A pesar de su presencia global, los informes clínicos
59 son raros en ciertas regiones, incluido Brasil. Este estudio documenta la primera
60 ocurrencia de *C. plica* en un *Puma concolor* Linnaeus, 1771 (Puma) de Santa Catarina,
61 en el sur de Brasil, marcando el primer registro de este parásito en la parte sur del país.
62 La necropsia y el análisis del sedimento urinario revelaron la presencia de huevos y
63 nematodos adultos de *C. plica*. El diagnóstico del parásito es un desafío debido a su
64 naturaleza subclínica y la dificultad para visualizar huevos y adultos, especialmente en
65 felinos silvestres. Este caso resalta la importancia del análisis de orina en carnívoros
66 silvestres para diagnosticar la capilariasis urinaria y enfatiza la necesidad de una mayor
67 conciencia sobre esta enfermedad parasitaria, particularmente en regiones poco
68 estudiadas.

71 **Palabras clave:** Capillariasis urinaria – carnívoros silvestres – infección por nematodos
72 – sur de Brasil – diagnóstico veterinario – parasitología

73

74 **INTRODUCTION**

75 *Capillaria* Zeder, 1800 (Trichuroidea: Capillaridae) is a genus of parasitic
76 nematodes that infect mammals, impacting the respiratory, urinary, hepatic, and
77 intestinal systems. Of these, only two species are known to affect the urinary tract of both
78 domestic and wild animals: *Capillaria plica* Rudolphi, 1819 and *Capillaria feliscati*
79 Diesing, 1851 (Sréter *et al.*, 2003). The disease caused by these species is known as
80 urinary capillariasis.

81 Clinical cases of this disease have been reported in the United States, Canada,
82 and Mexico, as well as in several European countries, including the Netherlands,
83 Denmark, Germany, Italy, Switzerland, Slovakia, Serbia, Greece, and Poland (Senior *et*
84 *al.*, 1980; Kirkpatrick & Nelson, 1987; Van Veen, 2002; Saeed *et al.*, 2006; Callegari *et*
85 *al.*, 2010; Bork-Mimm & Rinder, 2011; Ventura-Morales *et al.*, 2012; Basso *et al.*, 2014;
86 Maurelli *et al.*, 2014; Alić *et al.*, 2015; Mariacher *et al.*, 2016; Mariacher *et al.*, 2018;
87 Petersen *et al.*, 2018; Aleksić *et al.*, 2020; Komorová *et al.*, 2020; Pelligra *et al.*, 2020;
88 Ilić *et al.*, 2021; Sioutas *et al.*, 2021; Miszczak *et al.*, 2022).

89 Among these two species, *C. plica* stands out as the more frequently
90 encountered, commonly parasitizing domestic dogs, red foxes, wolves, badgers
91 (Miszczak *et al.*, 2022), bears (Mariacher *et al.*, 2018), wild felids, and, less commonly,
92 domestic cats (Bédard *et al.*, 2002; Callegari *et al.*, 2010; Fernández-Aguilar, 2010; Bork-
93 Mimm & Rinder, 2011), as well as mustelids (Basso *et al.*, 2014).

94 The biological cycle of *C. plica* is indirect, involving intermediate hosts such as
95 earthworms. The eggs, which are found in the bladder lumen of definitive hosts, measure
96 55–67 × 26–29 µm, are elongated and barrel-shaped, with a thick shell and a polar plug
97 at each end (Sioutas *et al.*, 2021). These eggs are excreted in the urine into the external
98 environment, where they develop into first-stage larvae; when ingested by earthworms,

99 these larvae infect carnivores; after passing through the intestines, the larvae develop
100 into second-stage larvae, remaining in this stage for 8 to 10 days before migrating to the
101 bladder; and there, they evolve into third- and fourth-stage larvae, eventually maturing
102 into adults (Fernández-Aguilar, 2010; Rossi *et al.*, 2011).

103 Despite the indirect biological cycle of *C. plica* and its typical asymptomatic
104 nature, the parasite can lead to a range of clinical symptoms in animals with significant
105 parasitic loads (Otranto, 2015). Animals infected with a high parasitic load may present
106 urinary tract disease symptoms such as pollakiuria, dysuria, hematuria, urinary
107 incontinence resulting from inflammatory reactions and edema in the submucosa of the
108 urinary bladder, as well as pyelonephritis due to secondary bacterial infection (Bork-
109 Mimm & Rinder, 2011; Basso *et al.*, 2014). According to Callegari *et al.* (2010), the
110 parasite may contribute to glomerular amyloidosis. In rare cases, renal insufficiency,
111 painful bladder expansion, and urinary tract obstruction may be observed (Sioutas *et al.*,
112 2021). The subclinical nature and potential impact of *C. plica* on animal health should be
113 considered, and veterinarians should be aware of this disease, even if it is uncommon
114 (Dimzas *et al.*, 2024).

115 The prevalence of this parasitosis varies according to the hosts, geographical
116 region, and season (Davidson *et al.*, 2006). The diagnosis of *C. plica* is made by
117 observing the eggs in the urinary sediment, with urine collected via cystocentesis being
118 the best method to avoid contamination from feces containing Trichuroidea superfamily
119 eggs (Dantas *et al.*, 2008). Urinalysis may show mild proteinuria, hematuria, and a slight
120 increase in transitional epithelial cells (Rossi *et al.*, 2011). There are no approved
121 medications for the treatment for this (Sant *et al.*, 2020). In cases of clinical signs, the
122 recommended treatment is fenbendazole or ivermectin (Pagnoncelli *et al.*, 2011), and in
123 severe cases with bladder inflammation, milbemycin may be indicated (Sioutas *et al.*,
124 2021).

125 Given its prevalence in both wild and domestic carnivores across various regions,
126 veterinarians should remain vigilant in diagnosing this infection, even though it may not

127 always present with clear symptoms. The findings of this study, which report the
128 presence of *C. plica* in the urine of wild felid, underscore the importance of urinalysis in
129 these animals as a key diagnostic tool, further emphasizing the need for awareness and
130 proper management of this parasitic disease.

131

132 MATERIAL AND METHODS

133 An adult male Puma, *Puma concolor* (Linnaeus, 1771), in good body condition,
134 weighing approximately 32 kg, was found dead in May 2018 by the side of SC390
135 highway in the city of Bom Jardim da Serra, in the state of Santa Catarina, southern
136 Brazil (geographical coordinates: 28° 20' 13" S, 49° 37' 29" W) (Figure 1). The animal
137 was sent by the Environmental Police to the Zoology Laboratory of the Universidade do
138 Planalto Catarinense (UNIPLAC) for routine anatomopathological studies.

139 During the necropsy, a urine analysis was conducted as there was liquid present
140 in the bladder. The collected urine was centrifuged at 33.33 g/s, and the urinary sediment
141 was examined under an optical microscope (Primestar 3 Zeiss at 20x and 100x
142 magnification). For histopathological analysis, small fragments of the liver, heart, spleen,
143 intestine, and kidney were collected and fixed in a 10% formalin solution. The preparation
144 process involved dehydrating the samples in a series of alcohol solutions with increasing
145 concentrations (70%, 80%, 90%, and 100%), clearing with xylene, and embedding in
146 paraffin. The tissues were then sectioned using a microtome, producing slices 6 to 8 µm
147 thick. These sections were mounted on slides and stained with eosin-hematoxylin-eosin.
148 The tissues were mounted on slides, stained with eosin-hematoxylin-eosin, analyzed
149 using optical microscopy (Primestar 3 Zeiss at 20x and 100 x magnification). The process
150 for histopathological analyses was carried out in the Pathology Laboratory of the
151 UDESC, Lages, Santa Catarina, Brazil.

152 **Ethic aspects:** The report on the animal adhered to the institutional policy for animal
153 care.

154

155 **RESULTS**

156 The histopathological examination revealed no lesions in the bladder. However,
157 during the microscopic analysis of the urinary sediment, the presence of filiform
158 nematodes was observed (Figure 2a). It was noted that the species exhibited a very
159 narrow esophagus, occupying approximately one-third of the body length, with a simple,
160 unarmed mouth, key morphological traits that help differentiate *C. plica* from *C. feliscati*,
161 as described by Taylor *et al.* (2017). The eggs, although present in low numbers,
162 displayed bipolar opercula, which is consistent with *C. plica* (Figure 2b). The urine also
163 contained exfoliated cells and leukocytes, indicating signs of inflammation. In
164 histopathology, no significant alterations in the tissues were found.

165

166 **DISCUSSION**

167 The infection by *C. plica* in carnivores, especially in animals that inhabit urban
168 and rural areas, is still poorly documented, with reports limited to a few specific
169 publications (Sioutas *et al.*, 2021). In wild animals, the diagnosis is even rarer, with egg
170 detection in urinary sediment analysis often occurring incidentally, since the routine
171 necropsy of these animals rarely includes the examination of the urinary mucosa to
172 identify eggs or adult parasites (Senior *et al.*, 1980).

173 Several studies highlight the prevalence of *C. plica* in carnivore's species.
174 Fernández-Aguilar *et al.* (2010) diagnosed the infection in the arctic fox, *Vulpes lagopus*
175 (Linnaeus, 1758) associated with cystitis, and Bork-Mimm and Rinder (2011) observed
176 that the prevalence in foxes can exceed 98%. Basso *et al.* (2014) suggest that the
177 increase in this infection in foxes, coupled with their movement into urban areas, may
178 represent a growing risk for domestic dogs and cats, further emphasizing the importance
179 of including *C. plica* in the differential diagnosis of urinary disorders. Additionally, Takács
180 *et al.* (2014) diagnosed *C. plica* in 45% of the 20 necropsied Golden Jackal, *Canis aureus*
181 Linnaeus, 1758, highlighting the high infection rate among canids.

182 Although these nematodes are little known in Brazil, there are already reports of
183 *Capillaria* spp. in the urine of carnivores in the states of Rio de Janeiro (Guimarães et
184 al., 2020), Piauí (Silva et al., 2021), and Sergipe (Costa et al., 2023). These findings
185 suggest that, although knowledge about this parasitosis is still emerging in Brazil, *C. plica*
186 may already be present in various regions.

187 The diagnosis of urinary capillariasis in wild animals is challenging due to host
188 behavior and limitations in necropsy routines. For better detection, it is essential to
189 perform a detailed examination of the urinary bladder and collect urine samples for
190 parasitic diagnosis.

191 The Puma, *P. concolor*, one of the largest carnivore species in the Americas, with
192 a range from Canada to South America, is one of the most endangered due to hunting,
193 deforestation, and population decline (Labarge et al., 2022). In Brazil, its distribution is
194 restricted and uncommon, and these felids have been forced to search for food in
195 anthropogenic areas, increasing the risk of contact with parasites. Therefore, studies on
196 the parasitic fauna of these animals are crucial to understanding their health status and
197 the implications for domestic species.

198 This study represents the first record of *C. plica* in the state of Santa Catarina in
199 a *Puma concolor* host, as well as the first report of this parasite in southern Brazil, a
200 region where data on this infection are still scarce. The detection of *C. plica* is
201 challenging, not only due to the difficulty in visualizing its eggs but also in identifying adult
202 parasites, especially in wild felids. Therefore, it is essential that, during post-mortem
203 examinations, veterinary professionals perform a thorough inspection of the urinary
204 bladder and collect urine samples to enable the diagnosis of this parasitosis and
205 contribute to the monitoring of these animals' health, as well as improving the
206 understanding of the distribution of *C. plica* in Brazil.

207

208 **Author contributions: CRediT (Contributor Roles Taxonomy)**

209 RMQ = Rosiléia marinho de Quadros

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211 **BVM** = Beatriz Valgas Marques
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213 **MSS** = Madja Schvan Schmitz
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216 **RBS** = Ricardo Bassini-Silva
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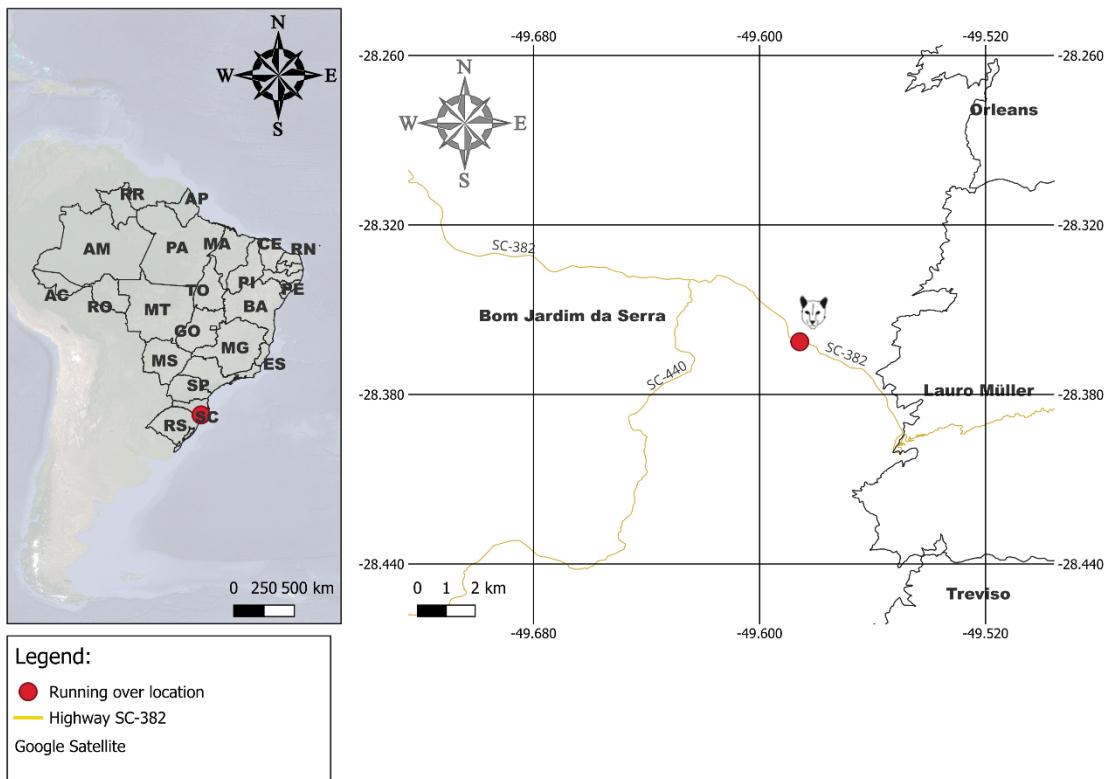
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320

321 Figure 1. Location of the rescue of the feline along the SC-390 highway, in the city of
 322 Bom Jardim da Serra, in the state of Santa Catarina, southern Brazil.

323



324

325 Figure 2. Presence of *Capillaria plica* in urine (a). Bioperculate egg of *Capillaria plica* in
 326 the urinary sediment (b).