First report of five nematode species in *Phasianus colchicus* Linnaeus (Aves, Galliformes, Phasianidae) in Brazil

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ABSTRACT. This paper deals with the first report of the nematodes *Dispharynx nasuta* (Rudolphi, 1819) Stiles & Hassall, 1920, *Gongylonema ingluvicola* Ransom, 1904, *Oxyspirura mansoni* (Cobbold, 1879) Ransom, 1904, *Subulura brumpti* (Lopez-Neyra, 1922) Cram, 1923 and *Tetrameres fissipina* (Diesing, 1860) Travassos, 1914, parasitizing the ring-necked pheasant, *Phasianus colchicus* Linnaeus, 1758 in Brazil. Morphological data on the parasites as well as on the prevalence, mean intensity, mean abundance and range of intensity of the nematodes in the infected birds are provided. KEY WORDS. Birds, ecology, helminths, morphology.

RESUMO. Primeiro registro de cinco espécies de nematóides em Phasianus colchicus Linnaeus (Aves, Galliformes, Phasianidae) no Brasil. O presente trabalho diz respeito à primeira referência dos nematóides Dispharynx nasuta (Rudolphi, 1819) Stiles & Hassall, 1920, Gongylonema ingluvicola Ransom, 1904, Oxyspirura mansoni (Cobbold, 1879) Ransom, 1904, Subulura brumpti (Lopez-Neyra, 1922) Cram, 1923 e Tetrameres fissipina (Diesing, 1860) Travassos, 1914, parasitando o faisão-coleira, Phasianus colchicus Linnaeus, 1758 no Brasil. São fornecidos dados morfológicos sobre os parasitos, bem como sobre a prevalência, intensidade média, abundância média e amplitude de variação da intensidade de infecção dos nematóides nas aves parasitadas.
PALAVRAS CHAVE. Ecologia, helmintos, morfologia.

The ring-necked pheasants (*Phasianus colchicus* Linnaeus, 1758, Galliformes, Phasianidae) are birds of Asian origin and worldwide distributed. Their raising is a profitable activity, since they can be commercialized for food and alive either as chicks or adults as game or ornamental birds; moreover the feathers, alone, can potentially be traded for ornamentation purposes (OLIVEIRA & ROSSI 2000).

More recently, in Brazil, the so called alternative or organic aviculture has increased on what concerns the raising of ring-necked pheasants and other domestic Galliformes, such as the common chickens and the guinea fowls in extensive or semi-confinement systems. This activity is a good option for extra profits for small producers, since the costs with chow are low, the market for natural food is expanding and meat and eggs produced are higher priced than those industrially obtained (Costa 2000).

The adopted systems in the alternative or organic aviculture permit the birds to be in contact with the ground and thus, also with the intermediate hosts for several helminth species; this condition promotes frequent cases of helminthosis that are among the most prevalent diseases that affect these

animals, with heavy economic losses in the production of meat and eggs, deceleration in the development, rejection of the viscera during the slaughtering and death (Mendonça 1953, Permin *et al.* 1999, Sage *et al.* 2002, Menezes *et al.* 2001, 2003a, b). Among the helminths infecting the ring-necked pheasants, the nematodes are the most outstanding group considering number of species and induced pathology (Ruff 1999).

According to Costa et al. (1986), Vicente et al. (1995) and Gomes et al. (2004) seven helminth species, nematodes only, were referred in Brazilian ring-necked pheasants; this is a very few amount if compared to the 50 and 18 helminth species reported in domestic chickens (Gallus gallus Linnaeus, 1758) and turkeys (Meleagris gallopavo Linnaeus, 1758), respectively. Considering the economic importance of the pheasants and the harm provoked by helminths in these hosts, the present paper reports to data on the morphology, prevalence, mean intensity, mean abundance and range of infection of nematodes recovered from ring-necked pheasants from backyard flocks in the State of Rio de Janeiro, Brazil, with the establishment of this bird as a new host for the nematode species considered herein.

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MATERIAL AND METHODS

Fifty adult specimens of ring-necked pheasants (*Phasianus colchicus* Linnaeus, 1758), 25 males, 25 females, weigh 200 – 1750 g from backyard flocks of 11 localities of the State of Rio de Janeiro, Brazil, were investigated (Fig. 1). Localities and number of examined hosts are, respectively: Niterói (22°53′S, 43°06′W): 9; Rio de Janeiro (22°54′S, 43°12′W): 8; Tanguá (22°73′S, 42°71′W): 9; São Francisco do Itabapoana (21°28′S, 41°08′W): 8; Santo Antônio de Pádua (21°54′S, 42°18′W): 2; Areal (22°14′S, 43°65′W): 3; Petrópolis (22°30′S, 43°10′W): 2; São José do Vale do Rio Preto (22°09′S, 42°55′W): 2; Rio Bonito (22°43′S, 42°37′W): 4; Engenheiro Paulo de Frontin (22°32′S, 43°40′W): 2; Laje do Muriaé (21°12′S, 42°07′W): 1. After individual clinical evaluation birds were killed and submitted to necropsy, according to the technique of Zander *et al.* (1997).

During the necropsy the eyes were examined for helminths and after, all organs of the digestive and respiratory tract were removed and opened with the aid of a scissor, in separated Petri dishes containing 0.85% NaCl solution. Nematodes were collected with the aid of thin brushes, (n° 00), rinsed in the same solution, fixed with hot A.F.A (alcohol 70° GL, 93 ml; formaldehyde, 5 ml; acetic acid, 2ml) and counted under a stereoscope microscope. Values referring to prevalence, mean intensity, mean abundance and range of infection are in accordance with Bush *et al.* (1997).

Nematodes were clarified in acetic acid and phenol, and some were stained with alcoholic chloride Langeron's carmine by the regressive process, according to Amato (1985). Some specimens were preserved in Canada balsam and other maintained as wet material and deposited in the Helminthological Collection of the Oswaldo Cruz Institute (CHIOC). Photomicrographs derived from a Axiophot Zeiss micrographic system in a Differential Interference Contrast (DIC) apparatus. Measurements are in millimeters (mm) unless otherwise indicated. Means are in parenthesis. Classification of the nematodes to the specific level is in accordance with Vicente et al. (1995).

The development of this research has been authorized by the Committee of Ethics for the Use of Animals (CEUA/Fiocruz) no. P0095-01.

RESULTS

Thelazioidea, Thelazidae

Oxyspirura mansoni (Cobbold, 1879) Ransom, 1904 Figs 2-6

Morphometrics based on ten adult specimens, five males and five females.

Males: total length 11.016-12.244 (11.880), maximum width 0.238-0.272 (0.258). Buccal capsule divided in two parts: the anterior 0.018-0.0216 (0.019) long, 0.0215-0.0324 (0.026) wide, the posterior 0.0180-0.0252 (0.023) long, 0.0216-0.0324 (0.026) wide (Fig. 2). Muscular and glandular esophagus, 0.238-

0.248 (0.242) and 0.966-1.078 (1.024) long, respectively. Nerve ring and excretory pore 0.238-0.248 (0.243) and 0.317-0.425 (0.361) from the anterior end, respectively. Spicules unequal and dissimilar (Fig. 6). The longer, 3.248-3.822 (3.600), the shorter, 0.210-0.244 (0.220) in length. Spicular ratio: 1: 15.5-17.1 (1: 16.7). Gubernaculum V-shaped in ventral view. Nine pairs of caudal papillae: four pairs pre-cloacal, one pair ad-cloacal and four pairs post-cloacal. Cloacal aperture 0.280-0.350 (0.308) from the posterior extremity.

Females: total length 11.594-16.922 (14.618), maximum width 0.238-0.340 (0.286). Buccal capsule divided in two parts: the anterior 0.018-0.0216 (0.019) long, 0.0252-0.0324 (0.0288) wide. Muscular and glandular esophagus, 0.224-0.287 (0.263) and 0.798-1.120 (0.980) long, respectively. Nerve ring and excretory pore 0.238-0.258 (0.248) and 0.287-0.399 (0. 350) from the anterior end, respectively. Opisthodelphic, tending to prodelphy. Vulvar opening in the posterior region of the body, near the anal aperture, 0.742-1.176 (1.008) from the posterior extremity (Fig. 3). Eggs 0.0396-0.0432 (0.040) long, 0.0252-0.0288 (0.026) wide, larvate, when oviposition occurs (Fig. 4). Anal aperture 0.280-0.420 (0.384) from the posterior extremity.

Site of infection: eyes (conjunctival sacs, under the nictitant membrane and nasolacrymal ducts) (Fig. 5).

Localities: Niterói, Rio de Janeiro, São Francisco do Itabapoana, State of Rio de Janeiro, Brazil.

Specimens deposited: CHIOC 36312 a-d, 36313, 36314 a-b, 36315 a-b, 36316, 36317 (whole mounts): 35163-35195 (wet material).

Acuarioidea, Acuariidae

Dispharynx nasuta (Rudolphi, 1819) Stiles & Hassall, 1920 Figs 7-11

Morphometrics based on ten adult specimens, five males and five females.

Males: total length 4.216-6.086 (5.182), maximum width 0.204-0.272 (0.238). Buccal capsule (vestibulum) 0.0884-0.1080 (0.095) long, 0.0180-0.0252 (0.0223) wide (Fig. 7). Cephalic cordons, recurrent, not anastomosed (Figs 7-8), ending at 0.216-0.420 (0.284) from the anterior extremity. Muscular and glandular esophagus 0.420-0.560 (0.508) and 1.008-1.932 (1.574) long. Nerve ring 0.184-0.237 (0.222) from the anterior end. Spicules unequal and dissimilar (Fig. 9). The longer, 0.322-0.454 (0.373), the shorter 0.140-0.158 (0.148) in length. Spicular ratio: 1: 2.5-2.9 (1: 2.5). Gubernaculum absent. Cloacal aperture 0.196-0.280 (0.232) from the posterior extremity.

Females: total length 5.134-6.902 (5.705), maximum width 0.306-0.578 (0.401). Buccal capsule (vestibulum) 0.097-0.119 (0.109) long, 0.018-0.216 (0.021) wide. Cephalic cordons as described for the males, ending at 0.115-0.252 (0.191) from the anterior extremity. Muscular and glandular esophagus, 0.476-0.700 (0.588) and 1.302-2.030 (1.646) long, respectively. Nerve ring 0.254-0.349 (0.292) from the anterior end. Vulvar



Figure 1. Map of the State of Rio de Janeiro, Brazil, showing the localities that were the source of the investigated ring-necked pheasants.

opening, 0.966-1.386 (1.170) from the posterior extremity (Fig. 10). Ovijector 0.1044-0.1908 (0.150) long (Fig. 10). Eggs 0.0288-0.0324 (0.0324) long, 0.0180-0.0216 (0.021) wide, larvate, when oviposition occurs (Fig. 11). Anal aperture 0.0828-0.1260 (0.1044) from the posterior extremity.

Site of infection: proventriculus (most often partially inserted in the mucosa and also free in the lumen).

Localities: Niterói, São Francisco do Itabapoana, Rio Bonito, State of Rio de Janeiro, Brazil.

Specimens deposited: CHIOC 36318, 36319 a-e, 36320 a-b, 36321, 36322 (whole mounts); 35216-35218 (wet material).

Subuluroidea, Subuluridae

Subulura brumpti (Lopez-Neyra, 1922) Cram, 1926 Figs 12-15

Morphometrics based on ten adult specimens, five males and five females.

Males: total length 7.174-9.758 (7.813), maximum width 0.204-0.340 (0.265). Buccal capsule (vestibulum) 0.032-0.043 (0.039) long, 0.029-0.036 (0.035) wide. Esophagus 0.700-1.022 (0.907) with a posterior bulb separated from the rest of the esophagus by a well marked constriction (Fig. 12). Bulb 0.168-0.196 (0.187) long, 0.154-0.210 (0.176) wide. Cervical alae extending beyond the esophagean bulb. Nerve ring and excre-

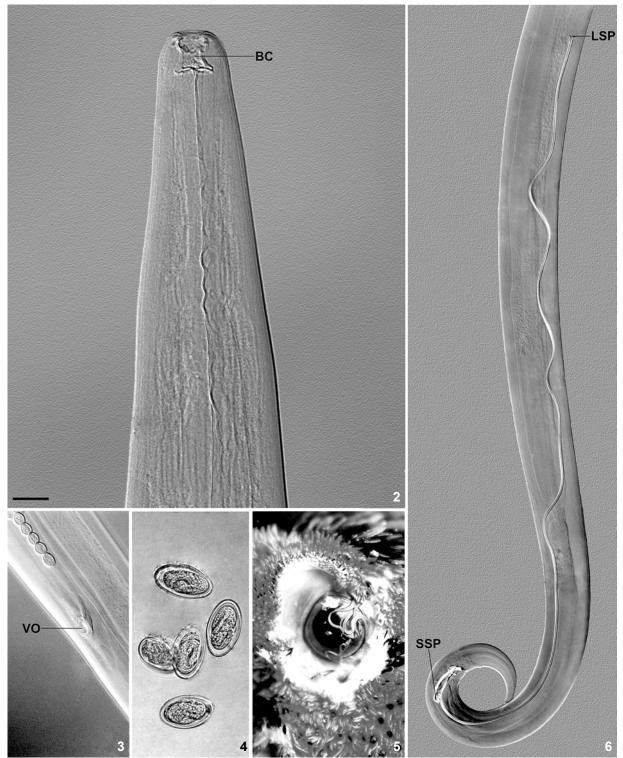
tory pore 0.101-0.280 (0.207) and 0.252-0.420 (0.336) from the anterior end, respectively. Ventral sucker 0.140-0.210 (0.182) long, 0.324-0.560 (0.462) from the posterior extremity and 0.252-0.322 (0.297) from the cloacal aperture (Fig. 13). Spicules equal, similar, 0.812-1.190 (0.994) long (Fig. 13). Gubernaculum 0.108-0.440 (0.132) long. Caudal alae narrow. Ten pairs of caudal papillae: three pairs pre-cloacal, two pairs adcloacal and five pairs post-cloacal. Cloacal aperture 0.182-0.238 (0.208) from the posterior extremity.

Females: total length 7.140-13.872 (9.948), maximum width 0.272-0.374 (0.313). Buccal capsule (vestibulum) 0.043-0.058 (0.052) long, 0.036-0.040 (0.038) wide. Esophagus as described for the males, 0.952-1.078 (0.094) long. Bulb 0.196-0.210 (0.202) long, 0.168-0.238 (0.199) wide. Nerve ring and excretory pore 0.191-0.288 (0.226) and 0.223-0.288 (0.266) from the anterior end, respectively. Cervical alae as described for the males. Vulvar opening 3.264-3.774 (3.513) from the anterior extremity (Fig. 15). Eggs 0.0576-0.0792 (0.0674) long, 0.0468-0.0576 (0.0504) wide, larvate when oviposition occurs (Fig. 14). Anal aperture 0.490-0.812 (0.627) from posterior extremity.

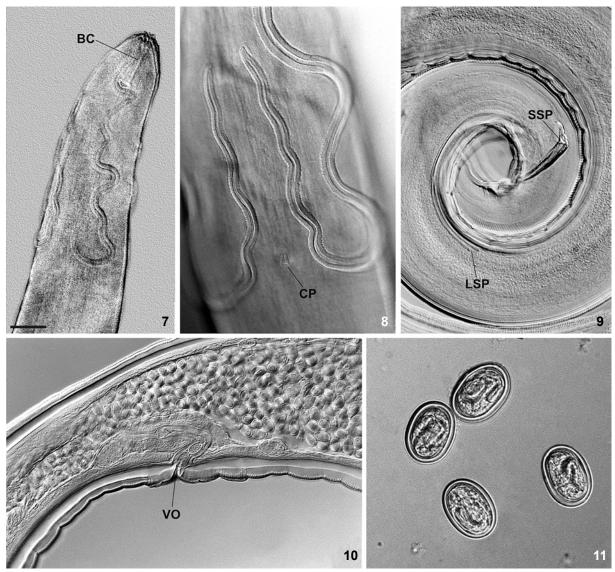
Site of infection: cecal lumen.

Locality: São Francisco do Itabapoana, State of Rio de Janeiro, Brazil.

Specimens deposited: CHIOC 36323 a-d, 36324 a-e, 36325 (whole mounts); 35219-35221 (wet material).



Figures 2-6. Oxyspirura mansoni. (2) Anterior region showing buccal capsule (VC) (vestibulum); (3) vulvar opening (VO). (4) eggs; (5) worms in the eye of a ring-necked pheasant specimen; (6) male posterior region showing small (SSP) and large (LSP) spicules. Bar of Fig. 2 common to Figs 3-6 = 0.05 in Fig. 2, 0.10 in Fig. 3, 0.02 in Fig. 4, 2.5 in Fig. 5, 0.15 in Fig. 6.



Figures 7-11. *Dispharynx nasuta*. (7) Anterior region showing buccal capsule (VC) (vestibulum) and cephalic cordons; (8) region of the terminal portion of cephalic cordons, with the cephalic papilla (CP); (9) male posterior region showing small (SSP) and large (LSP) spicules; (10) vulvar region with the vulvar opening (VO); (11) eggs. Bar of Fig. 7 common to Figs 8-11 = 0.10 in Fig. 7, 0.04 in Fig. 8, 0.05 in Figs 9, 10, 0.01 in Fig. 11.

Spiruroidea, Gongylonematidae Gongylonema ingluvicola Ransom, 1904 Figs 16-17

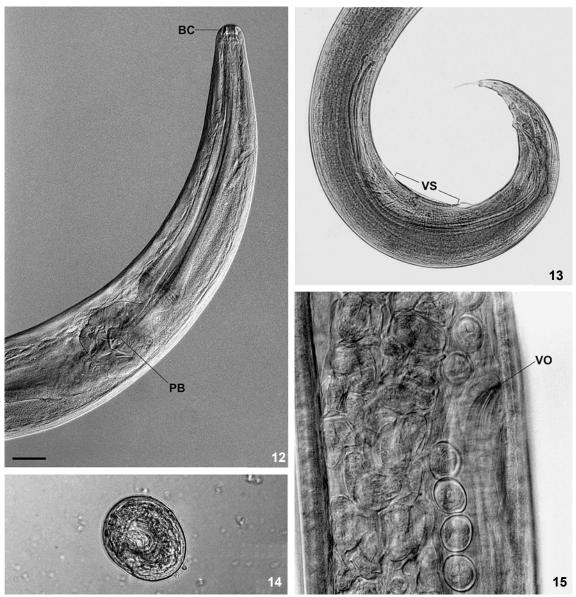
Morphometrics based on two immature female specimens. Total length 6.560-10.720 (8.640), maximum width 0.120-0.200 (0.160). Cephalic end and esophageal region ornamented with cuticular plaques irregularly arranged in longitudinal rows on dorsal and ventral aspects; lateral cuticular flanges on anterior part of the body (Fig. 16). Cephalic portion 0.300-0.340

(0.320) long. Buccal capsule (vestibulum) 0.018-0.021 (0.019) long, 0.006 (0.006) wide. Nerve ring 0.285 from the anterior extremity. Muscular and glandular esophagus 0.290-0.300 (0.290) and 1.440-2.040 (1.740) long, respectively, with the ratio $1\colon4.8\text{-}7.03$ $(1\colon1.59)$. Anal aperture 0.126-0.144 (0.135) from the posterior end. Posterior extremity abruptly pointed (Fig. 17).

Site of infection: inserted in the mucosa of the crop.

Locality: São Francisco do Itabapoana, State of Rio de Janeiro, Brazil.

Specimens deposited: CHIOC 36326 a-b (whole mounts).



Figures 12-15. Subulura brumpti. (12) Anterior region showing buccal capsule (VC) (vestibulum) and esophagus with a posterior bulb (PB) separated from the rest of the esophagus by a well marked constriction; (13) male posterior region showing ventral sucker (VS) and spicules; (14) egg; (15) vulvar region with the vulvar opening (VO). Bar of Fig. 12 common to Figs 13-15 = 0.08 in Fig. 12, 0.10 in Fig. 13, 0.03 in Fig. 14, 0.05 in Fig 15.

Habronematoidea, Tetrameridae Tetrameres fissipina (Diesing, 1860) Travassos, 1914 Figs 18-21

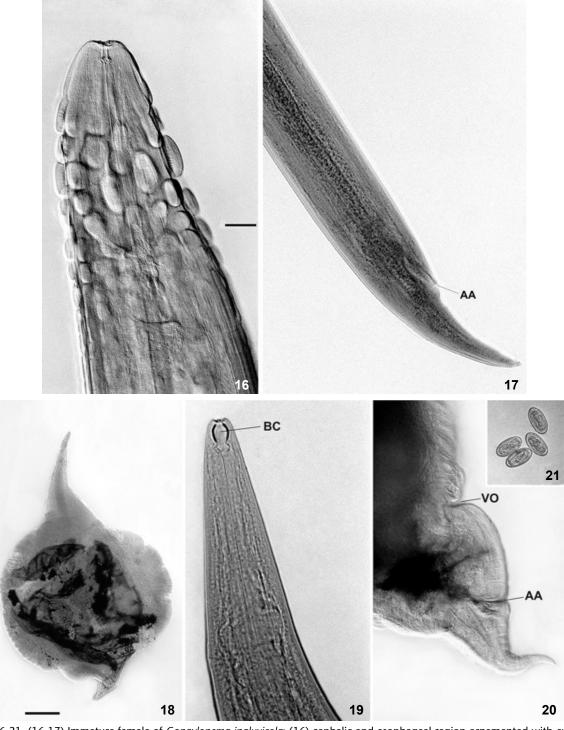
Morphometrics based on two adult females.

Body enlarged, with transversal striations in the middle region, with fusiform extremities, 3.600-3.910 (3.750) long, 2.550 (2.550) wide (Fig. 18). Buccal capsule (vestibulum) 0.0216

long, 0.0180 wide (Fig. 19). Nerve ring 0.18 from the anterior end. Vulvar aperture in the posterior portion of the body, 0.590 from the posterior extremity (Fig. 20). Anal aperture 0.322-0.406 from the posterior end (Fig. 20). Eggs elongate, thin shelled, larvate when oviposition occurs, 0.050-0.058 (0.052) long, 0.022-0.029 (0.026) wide (Fig. 21).

Site of infection: proventricular glands.

Localities: Rio de Janeiro and Rio Bonito, Rio de Janeiro



Figures 16-21. (16-17) Immature female of *Gongylonema ingluvicola*: (16) cephalic and esophageal region ornamented with cuticular plaques irregularly arranged in longitudinal rows; (17) posterior extremity, showing anal aperture (AA). Bar of Fig. 16 common to Fig. 17 = 0.03 in Fig. 16, 0.05 in Fig. 17; (18-21) *Tetrameres fissipina*: (18) Body enlarged, with transversal striations and fusiform extremities; (19) anterior region showing buccal capsule (VC) (vestibulum); (20) female posterior region showing vulvar opening (VO) and anal aperture (AA); (21) eggs. Bar of Fig. 18 common to Figs 19-21 = 0.5 in Fig. 18, 0.02 in Fig. 19, 0.16 in Fig. 20, 0.05 in Fig. 21.

State, Brazil.

Specimens deposited: CHIOC 35222 e 35223 (wet material).

Data on prevalence, mean intensity, mean abundance and range of infection of the nematodes reported are presented in table I.

DISCUSSION

The nematode species reported herein are referred for the first time in ring-necked pheasants from Brazilian backyard flocks. The studied nematodes present a wide distribution in domestic birds, mostly Galliformes and have been referred in chickens, turkeys, guinea fowls and peacocks in Brazil (Duarte & Dórea 1987, Vicente et al. 1995, Menezes et al. 2001). Some of these nematodes species are less specific to the host, such as *Dispharynx nasuta*, that was found in the domestic pigeon together with *Tetrameres fissipina* that also occurs in ducks and teals.

Data show a great possibility of cross infections with the nematodes recovered from the ring-necked pheasants and those from other domestic birds. This condition is particularly severe, taking into account that strains of these parasites, adapted to domestic birds inducing mild pathologies to these hosts, can provoke severe lesions and death in the ring-necked pheasants (Clapham 1961, Menezes *et al.* 2003b). Thus, is strongly recommended, as a prophylactic procedure, that mixed or neighboring raisings of pheasants with other domestic birds must be avoided, since the nematode species presently studied have a heteroxenic life cycle; birds naturally feeding on suitable intermediate hosts, that are frequently present where domestic flocks are kept, can maintain and transmit the parasite infections.

References of the occurrence and prevalence of the studied nematode species in the ring-necked pheasants are very few worldwide. The prevalence and mean intensity of *D. nasuta* presently observed were low and in accordance with data after Madsen (1941) in Denmark and Goble & Kutz (1945) in the USA. Nevertheless, in disagreement with the present results, the former authors reported this nematode species only occurring in chicks but not in adult birds.

In Brazil, Menezes *et al.* (2001) in guinea fowls and Grisi & Carvalho (1974) in domestic chickens, referred to prevalences of 44.0% and 4.70%, of *D. nasuta*, respectively; in both hosts, the

parasite burdens were significantly higher than those recovered from the ring-necked pheasants, suggesting that specimens of this host, mainly the adults, are not easily infected by *D. nasuta*.

The nematode Oxyspirura mansoni was reported from ringnecked pheasants from Hawaii by Schwartz & Schwartz (1951) and Lewin & Holmes (1971) with no data on prevalences. Another species of the genus, O. petrowi Skrjabin, 1929, has already been recovered from these birds in USA, with a prevalence of 3.3% (Pence et al. 1980). In Brazil, the prevalence and intensity of infection of O. mansoni were significantly higher than those observed in guinea fowls and domestic chickens from backyard flocks in the State of Rio de Janeiro (Grisi & Carvalho 1974, Menezes et al. 2001). Nevertheless, values were lower than those reported for this nematode in domestic chickens from other localities in Brazil Costa et al. (1975) and Machado et al. (1980). The present results show that the ring-necked pheasants are very susceptible to infections with O. mansoni yielding very high worm burdens reaching almost the maximum value of 200 eye worms/bird according to Schwabe (1950).

The species S. brumpti, that in accordance with BARUS (1969) is synonymous of S. suctoria (Molin, 1860) Railliet & Henry, 1912, has already been referred in Hawaiian ring-necked pheasants by Schwartz & Schwartz (1951) and Lewin & Holmes (1971). The first report of this nematode in Brazil was in domestic chickens, turkeys and guinea fowls from the State of Piauí, by Costa & Freitas (1962), with no data on prevalences. In the present investigation, values referring to the prevalence and mean intensity of infection were very low in comparison with the observed by Menezes et al. (2003b) in ring-necked pheasants parasitized with another cecal nematode, Heterakis gallinarum, recovered from birds in the State of Rio de Janeiro. This fact does not indicate that the pheasants are partially permissive to S. brumpti, taking into account that BARUS (1969) showed that the ring-necked pheasants are highly susceptible to experimental infections with this nematode species.

The prevalences of *Tetrameres fissipina* and *Gongylonema ingluvicola* in the ring-necked pheasants were lower than those reported for those species infecting Brazilian domestic chickens by Grisi & Carvalho (1974), Costa *et al.* (1975), Machado *et al.* (1980) and by Mukaratirwa *et al.* (2001), Magwisha *et al.* (2002), Permin *et al.* (2002) during avian helminth surveys in

Table I. Prevalence, mean intensity, mean abundance and range of infection of nematodes parasitizing ring-necked pheasants from backyard flocks of State of Rio de Janeiro, Brazil.

Helminths	Prevalence (%)	Mean intensity	Mean abundance	Range of intensity
Oxyspirura mansoni	24	29.1	6.98	1-121
Dispharynx nasuta	10	3.20	0.32	1-6
Subulura brumpti	8	19.5	1.56	1-37
Tetrameres fissipina	4	1.00	0.04	1
Gongylonema ingluvicola	2	2.00	0.04	2

Africa. The present results show that either the ring-necked pheasant is a semi-permissive host for these nematodes or that the intermediate hosts are not properly ingested by the birds in the local the flocks are kept.

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