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# ORIGINAL ARTICLES /ARTÍCULOS ORIGINALES

## NEMATODE PARASITES OF THE NEOTROPICAL RATTLESNAKE, *CROTALUS DURISSUS* LINNAEUS, 1758 (OPHIDIA, VIPERIDAE) FROM BRAZIL: NEW RECORDS AND GENERAL ASPECTS

## NEMATODOS PARÁSITOS DE LA CASCABEL NEOTROPICAL, *CROTALUS DURISSUS* LINNAEUS, 1758 (OPHIDIA, VIPERIDAE) EN BRASIL: NUEVOS REGISTROS Y ASPECTOS GENERALES

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# Abstract

Forty-six samples of nematodes recovered from Brazilian specimens of *Crotalus durissus* Linnaeus, 1758, commonly named rattlesnake were studied. Snakes either died in captivity, under which they were kept for production of serum, or were found dead in nature. Other samples of nematodes from this host and preserved in the Helminthological Collection of the Oswaldo Cruz Institute (CHIOC) were also analyzed. The most prevalent species were those of the genus *Ophidascaris* Baylis, 1920: *O. arndti* Sprehn, 1929, *O. durissus* Panizzutti, Santos, Vicente, Muniz-Pereira & Pinto, 2003, *O. sicki* Freitas, 1951, *O. tuberculatum* Siqueira, Panizzutti, Muniz-Pereira & Pinto, 2005 and *Ophidascaris* sp., followed by specimens allocated in *Hexametra* Travassos, 1920: *H. boddaertii* (Baird, 1860) Kreis, 1944, and *Kalicephalus* Molin, 1861: *K. inermis macrovulvus* Caballero, 1954, *Kalicephalus* sp. Also, specimens representing the genera *Hastospiculum* Skrjabin, 1923: *Hastospiculum* sp., *Physaloptera* Rud, 1819: *Physaloptera* sp. and *Travassosascaris* Sprent, 1978: *T. araujoi* (Schneider, 1866) Sprent, 1978 were identified, as well. The species *Ophidascaris arndti*, *O. sick*, *O. tuberculatum*, *K. inermis macrovulvus* and *Physalopera* sp., represent new occurrences in this host. Species are briefly presented and figured, with emphasis in the cross sections of buccal structures.

Key words: Brazil - Crotalus durissus - nematodes - rattlesnake.

# Resumen

Cuarenta y seis muestras de nemátodos recuperados en especímenes brasileños de *Crotalus durissus* Linnaeus, 1758, comúnmente conocida como cascabel fueron estudiadas. Las serpientes o se sacrificaban en cautiverio para la producción de suero o fueron encontradas muertas en la naturaleza. Otras muestras de nemátodos de este huésped preservadas en la Colección Helmintológica del Instituto Oswaldo Cruz (CHIOC) fueron también analizadas. Los nematodos mas prevalentes fueron aquellos del género *Ophidascaris* Baylis, 1920: *O. arndti* Sprehn, 1929, *O. durissus* Panizzutti, Santos, Vicente, Muniz-Pereira & Pinto, 2003, *O. O. sicki* Freitas, 1951, *O. tuberculatum* Siqueira, Panizzutti, Muniz-Pereira & Pinto, 2005 y *Ophidascaris* sp., seguidas por otros incluidos en *Hexametra* Travassos, 1920: *H. boddaertii* (Baird, 1860) Kreis, 1944, y *Kalicephalus* Molin, 1861: *K. inermis macrovulvus* Caballero, 1954 y *Kalicephalus* sp. Además, fueron identificados especímenes representados en los géneros *Hastospiculum* Skrjabin, 1923: *Hastospiculum* sp., *Physaloptera* Rud, 1819: *Physaloptera* sp. y *Travassosascaris* Sprent, 1978: *T. araujoi* (Schneider, 1866) Sprent, 1978. Las especies *Ophidascaris arndti*, *O. sicki*, *O. tuberculatum*, *K. inermis macrovulvus* y *Physaloptera* sp., representan nuevos registros en este huésped. Las especies son brevemente presentadas y ilustradas con énfasis en las estructuras bucales, por medio de cortes transversales.

Palabras clave: Brasil - cascabel - Crotalus durissus - nemátodos.

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## INTRODUCTION

The first report of helminths from Brazilian reptiles occurred far back in the XVII century, when some parasites of iguanas were referred by Piso & Marcgrave (1648). From this date on, studies of the parasitism affecting reptiles in Brazil have been relatively few and scarce. However, in the last century, complete surveys of nematodes recovered from Brazilian vertebrates have been made available by Vicente & Pinto (1999) and Vicente *et al.* (1985, 1991, 1993, 1995, 1997).

Taking into account that the catalogue of nematodes from Brazilian reptiles (Vicente et al. 1993) permits an easier identification of the species parasitizing snakes, the present investigation aimed at the updating of data related to the nematodes collected in specimens of Crotalus durissus Linnaeus, 1758 presently captured in different Brazilian localities together with other nematode samples previously recovered from the same host and deposited in the Helminthological Collection of the Oswaldo Cruz Institute (CHIOC). The present findings also complement those already reported by Araujo et al. (1969 a-b), Araújo et al (1999), Silva et al. (2001, 2007), Panizzutti et al. (2003), Dias et al. (2004) and Panizzutti (2005), specifically related to the nematodes from C. durissus in Brazil, together with data after Rossellini (2007), listing the helminths parasitizing Brazilian snakes.

# MATERIALS AND METHODS

Forty-six samples of nematodes recovered from specimens of *C. durissus* captured in the States of Mato Grosso, Minas Gerais, Pará, Paraná, Rio de Janeiro and São Paulo were studied. Parasites were either collected from animals found dead in nature, or from those captured during the flood of the Represa Itaipu Binacional (Binational Itaipu Dam), in the State of Paraná and that died before the donation for institutional maintenance for research and/or anti-ophidic serum production; some samples derived from contaminated feces or from regurgitation of some specimens that have died under captivity in the Instituto de Biologia do Exercito, IBEx (Army Institute of Biology) in the city of Rio de Janeiro, that is an institution registered and licensed by the IBAMA, the Instituto Brasileiro do Meio Ambiente e dos Recursos Naturais Renováveis (Brazilian Institute of the Environment and Natural Renewable Resources) to develop specific scientific research activities and to produce hyper immune equine plasma. Also, other samples recovered from rattlesnakes and previously deposited in the Helminthological Collection of the Oswaldo Cruz Institute (CHIOC) were examined.

Captivity conditions at the IBEx: snakes are maintained to supply the Fundação Nacional de Saúde (National Health Foundation) with antiophidic sera. At the time they come from nature, snakes are submitted to a quarantine period to be evaluated for sex, coprological examinations and other procedures to determine their health conditions to improve their survival under captivity.

Concomitantly, the contamination with parasites and other pathogens from the other already maintained specimens is avoided. Snakes are kept in plastic or polypropylene boxes with corrugated paper bedding and are weekly fed on mice (Mus musculus Linnaeus, 1758) in an amount that represents 10% of the animal weight and water is available ad libidum. In despite of the taken actions, some specimens do not adapt to the new conditions and die, to be further submitted to necropsy and investigated for parasites. Nematodes studied here were already fixed and kept as wet material in different solutions: 70° ethanol GL, AFA (93% ethanol 70° GL; 5% formaldehyde, 2% glacial acetic acid), 10% formalin or ethanol with glycerin. Nematodes were dehydrated in a crescent alcoholic series (70°GL-100° GL), clarified in acetic acid and phenol and preserved as whole mounts in balsam and beechwood creosote, 1:3, respectively.Some specimens were clarified in glycerin for the en face cross sections, in accordance with the methodology of Anderson (1968). Depending on the necessity, some females were dissected on Petri dishes with paraffin, according to Travassos (1950) for the counting of uterine branches.

Figures were obtained with a camera lucida connected to bright-field Zeiss or Olympus microscopes. Measurements are in millimeters (mm) and range appears in parentheses. The studied samples were deposited in the CHIOC.

# RESULTS

Investigated snakes were parasitized only with nematodes. A total amount of 46 samples were studied. Approximately, 80% were represented by ascaridoids distributed among the Ophidascaris Baylis, 1920 (50%), Hexametra Travassos, 1920 (30%) and Travassosascaris Sprent, 1978 (20%). The remaining 20% of the total amount of samples consisted of specimens included in Hastospiculum Skrjabin, 1923, Kalicephalus Molin, 1861 and Physalopera Rudolphi, 1819, with 6.6%, each. At first sight, ascaridoids from snakes are very much alike; curiously, their prior identification on the basis of the nature of the hosts only, in the case the ophidians, has frequently been adopted by those not so well acquainted with the morphology of nematodes. This fact alone, has most of the times, induced to the general and erroneous nomination of Ophidascaris sensu lato for different species, included in Hexametra, Ophidascaris, Polydelphis Baylis, 1920 and Travassosascaris; thus, efforts have to be made so that apical views of buccal structures of the ascaridoid nematodes can be obtained, whenever possible, together with the analysis of other morphological distinguishing characters, aiming at an accurate generic/specific diagnosis of the worms.

#### Ascaridoidea

Ascarididae Baird, 1853 *Ophidascaris arndti* Sprehn, 1929 (= *Ophidascaris travassosi* Vaz, 1938, *O. sprenti* Araujo, 1969) (Figs 1-7)

Morphometrics based on sixteen specimens, nine males, seven females. Males 56.6 (47-64) long, 0.63 (0.52-0.75) wide; females 68.9 (64-77) long, 0.62 (0.50-0.81) wide. *General*: mouth provided with three quadrangular lips with thin dentigerous ridges on the inner border; interlabia present. A large papilla together with another small, are observed on each ventro-lateral lip; on the dorsal lip there are two large papillae. Esophagus clubshaped, esophageal ventriculus and intestinal cecum absent. *Males*: with subequal spicules with short alae, 2.1 (1.89-2.38) long, gubernaculum absent. Caudal alae reduced; terminal caudal mucron present. Thirty-six pairs of pre-cloacal

papillae, an unpaired papilla near the cloacal opening; the number of post-cloacal papillae varies from five to six pairs. *Females*: didelphic, opistodelphic, oviparous. Vulvar opening located at the middle-third of the body, frequently anterior to the half of its total length. From the vulva, the ovijector (*vagina vera*) reaches an undivided portion of the uterus from which two straight uterine branches arise. Ovaries sinuous, not reaching the vulvar level. Eggs sub-spherical, 0.061-0.076 long, 0.057-0.051 wide. Tail long, conical, without mucron.

Sites of infection: esophagus, stomach; localities: Itaipu Binational Reserve, Foz do Iguaçu, Brazil-Paraguay border, State of Paraná (24°05'45"S, 54°35'07"W), Valença, State of Rio de Janeiro, (22°14'44"S, 43°42'01"W), São João Del Rey, State of Minas Gerais (21°08'00"S, 44°15'40"W); deposited samples: CHIOC: 35326, 35336, 35337, 35333, 35343, 35344, 36487 a-e, 36488 (a-c), 36489, 36490 (a-c), 36492 a-e, 36493 (a-b).

*Remarks*: The species *Ophidascaris travassosi* Vaz, 1938 and *O. sprenti* Araujo, 1969, were considered synonyms of *O. arndti* by Sprent (1988). This proposition was overlooked by Vicente *et al.* (1993).

Ophidascaris durissus Panizzutti, Santos, Vicente, Muniz-Pereira & Pinto, 2003 (Figs 8-14)

Morphometrics based on eighteen specimens, nine males, nine females. Males 60.8 (52-68) long, 0.67 (0.61-0.76) wide; females 67.8 (56-93) long, 0.70 (0.56-0.83) wide. *General*: mouth with three lips with dentigerous ridges extending to the inner border; the dorsal lip presents two large papillae and the latero-ventral possess one papilla each. Interlabia absent. Esophagus club-shaped. Esophageal ventriculus and intestinal cecum absent. Males: with subequal spicules, alate, 2.53 (2.48-2.90) long. Gubernaculum absent. Caudal alae narrow. Seventy pairs of pre-cloacal papillae, one pair ad-cloacal and six pairs post-cloacal. Females: didelphic, opistodelphic and oviparous. Vulvar opening situated between the 1<sup>st</sup> and 2<sup>nd</sup> third of middle body. From the vulva a short ovijector joins an undivided portion of the uterus, from which two uterine branches arise. Eggs subspherical, 0.05-0.07 long, 0.068-0.084 wide. Tail conical.





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**Figures 1-7.** *Ophidascaris arndti.* Fig. 1. Anterior of male, dorsal view. Fig. 2. Head, *en face* view. Fig. 3. Vulva, vagina, ovijector and uterine bifurcation, lateral view. Fig. 4, Female genitalia (partial), frontal view. Fig. 5. Posterior of male, lateral view. Fig. 6. Posterior of male, ventral view. Fig. 7. Posterior of female, lateral view. Bar (common to Figs 1-7) = 0.04 in Fig. 2; 0.4 in Figs 1, 3; 0.5 in Figs 4, 5; 0.2 in Figs 6, 7 (in mm).

Site of infection: stomach; locality: Itaipu Binational Reserve, Foz do Iguaçu, Brazil-Paraguay border State of Paraná; deposited samples: CHIOC: 34687, 34937, 34937 b-f.

*Remarks*: Considering that *O. durissus* is the only Brazilian species of the genus devoid of interlabia, the status of this species should be further reevaluated, taking into account that the absence/presence of interlabia is a generic character and may not be regarded as an isolate specific variation among the representatives of *Ophidascaris*.

> Ophidascaris sicki Freitas, 1951 (Figs 15-21)

Morphometrics based on seven specimens, four males and three females. Males 63.25 (55-78) long, 0.71 (0.67-0.75) wide; females 70 (52-86) long,

**Figures 8-14.** *Ophidascaris durissus*. Fig. 8. Anterior of male, lateral view. Fig. 9. Head, *en face* view. Fig. 10. Posterior of male, ventral view. Fig. 11. Vulva, vagina, ovijector and uterine bifurcation, lateral view. Fig. 12. Posterior of male, ventral view. Fig. 13. Posterior of male, lateral view. Fig. 14. Male, posterior extremity, lateral view. Bar (common to Figs 8-14) = 0.7 in Figs 8, 11, 12; 0.08 in Fig. 9; 0.5 in Figs 10, 12, 13 (in mm).

0.76 (0.62-0.93) wide. General: mouth with three quadrangular lips, 0.15 long, 0.17 wide showing dentigerous ridges on the inner border; interlabia short. One prominent labial papilla is present on each dorso-lateral lip; dorsal lip with two papillae. Esophagus club-shaped. Ventriculus and intestinal cecum absent. Males: spicules equal, with narrow alae, 0.76 (0.48-0.91) long. Gubernaculum and caudal alae absent, tail with mucron, presenting up to forty pairs of pre-cloacal papillae and three postcloacal. Females: didelphic, opistodelphic and oviparous. Vulva slit-shaped, located at the posterior third of the body. The ovijector joins the undivided portion of the uterus, from which arise two uterine branches that run straight to the origin of coiled ovaries not reaching the vulvar region. Eggs sub-spherical, 0.07-0.084 long, 0.056-0.07 long. Tail short and blunt. Mucron absent.

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**Figures 15-21.** *Ophidascaris sicki.* Fig. 15. Anterior of male, ventral view. Fig. 16. Head, *en face* view. Fig. 17. Female, anterior extremity, ventral view. Fig. 18. Posterior of male, ventral view. Fig. 20. Male, anterior extremity, dorsal view. Fig. 21. Female, posterior extremity, ventral view. Bar (common to Figs 15-21) = 0.5 in Fig. 15; 0.04 in Fig. 16; 0.4 in Figs 17, 20; 0.2 in Figs 18, 19, 21 (in mm).

**Figures 22-28.** *Ophidascaris tuberculatum.* Fig. 22. Anterior of female, dorsal view. Fig. 23. Vulva, vagina, ovijector and uterine bifurcation, lateral view. Fig. 24. Male, anterior extremity, dorsal view. Fig. 25. Female, posterior extremity, lateral view. Fig. 26. Male, posterior extremity, ventral view. Fig. 27. Posterior of male, lateral view. Fig. 28. Detail of male posterior extremity, ventral view. Bar (common to Figs 22-28) = 0.3 in Figs 22, 23; 0.09 in Fig. 24; 0.4 in Figs 25, 26, 27; 0.25 in Fig. 28 (in mm).

Site of infection: esophagus; locality: Itaipu Binational Reserve, Foz do Iguaçu, Brazil-Paraguay border, State of Paraná); deposited sample: CHIOC: 34.779 a-f.

Type material of *O. sicki*, recovered from *Waglerophis merremii* (Wagler, 1824) [=*Xenodon merremii*] captured in Xavantina, State of Mato Grosso (14°40'09"S, 52°20'09"W), previously deposited was revised (CHIOC no. 19120, 19121, 19122, 19123).

*Remarks*: This is the first occurrence of the species parasitizing specimens of *Crotalus durissus*, since,

as informed, *O. sicki* had been reported only from the Wagler's snake in Brazil.

*Ophidascaris tuberculatum* Siqueira, Panizzutti, Muniz-Pereira & Pinto, 2005 (Figs 22-28)

Morphometrics based on eleven specimens, four males and seven females. Males 62 (40-70) long, 0.85 (0.77-0.94) wide. Females 87 (74-132) long, 1.37 (0.94-1.53) wide. *General*: mouth with three slightly quadrangular lips, with well developed dentigerous ridges along the internal border and interlabia short, reaching about 1/3 of the lips in length. Esophagus club-shaped. Ventriculus and



**Figures 29-34.** *Hexametra boddaertii*. Fig. 29. Anterior of male, showing the intestinal cecum (arrow), ventral view. Fig. 30. Head, *en face* view. Fig. 31. Vulva, vagina, ovijector and the six uterine branches, frontal view. Fig. 32. Esophageal-intestine junction, with the intestinal cecum (arrow), dorsal view. Fig. 33. Posterior of male, ventral view. Fig. 34. Male, posterior extremity, lateral view. Bar (common to Figs 36-41) = 0.7 in Fig. 29; 0.05 in Fig. 30; 0.3 in Figs 31, 32; 0.2 in Figs 33, 34 (in mm).

intestinal cecum absent. *Males*: with subequal spicules and narrow alae, 2.85 (2.63- 2.87) long. Gubernaculum and caudal alae absent. Twenty-six pairs of pre-cloacal papillae, one pair ad-cloacal, five pairs post-cloacal and one umpaired papilla. Tail long, conical, presenting a conspicuous mucron. *Females*: didelphic, opitodelphic, oviparous. Vulva slit-shaped, either in the middle third or in the anterior third of the body. From the vulva, extends a sinuous ovijector to join the undivided uterine portion, from which arise two uterine branches to reach the coiled ovaries. Eggs sub-spherical, 0.085 long, 0.070 wide. Tail with a prominent muscular post-anal protuberance, easily observed that appears as a tubercle.

Site of infection: stomach; locality: State of Minas Gerais (Municipality not informed); samples deposited: CHIOC: 8132, 36494 (a-d).



**Figures 35-39.** *Kalicephalus inermis macrovulvus.* Fig. 35. Dorsal ray of the copulatory bursa, ventral view. Fig. 36. Anterior of female, ventral view. Fig. 37. Female, posterior extremity, lateral view. Fig. 38. Posterior of male, lateral view. Fig. 39. Vulvar region, lateral view. Bar (common to Figs 35-39) = 0.08 in Fig. 35; 0.1 in Figs 36-39 (in mm)

*Remarks: Crotalus durissus* is a new host record for the species previously described by Siqueira *et al.* (2005) parasitizing specimens of *Bothrops jararaca* Wied, 1832.

#### Ophidascaris sp.

Sites of infection: esophagus, stomach, body cavity (?), feces; localities: Itaipu Binational Reserve, Foz do Iguaçu, Brazil-Paraguay border State of Paraná, PR, Valença, State of Rio de Janeiro, Três Corações, State of Minas Gerais (21°58'10"S, 45°22'20"W); deposited samples: CHIOC: 34894, 34897, 35330, 35334, 35338.

## Hexametra boddaertii (Baird, 1860) Kreis, 1944 (Figs 29-34)

Morphometrics based on thirty-six specimens, thirteen males, twenty-three females. Males 86

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(66-108) long, 1.0 (0.71-1.51) wide; females 123 (72-173) long, 1.17 (0.79-2.1) wide. General: mouth with three lips, dentigerous ridges extending along anterior border of lip and round anterior angles of lip to level of double papillae; interlabia absent. Labial papillae consisting of double papillae on each ventro-lateral lip and two on the dorsal lip. Esophagus club-shaped. Ventriculus absent, intestinal cecum present or absent. Males: with subequal spicules, with narrow alae, 1.29 (0.98-1.58) long. Tail almost rounded, ending in a tip presenting an inconspicuous mucron. A variable number of pre-cloacal papillae, one pair ad-cloacal and two to three pairs post-cloacal. Females: opistodelphic, oviparous. Vulvar aperture slit-like, at the end of the middle third or at the posterior third of the body From the vulva, extends a sinuous ovijector to join the undivided uterine portion, from which arise six uterine branches to reach the coiled ovaries that extend to the vulvar region. Eggs subspherical, 0.080-0.086 long, 0.070-0.080 wide. Sites of infection: intestine, body cavity, stomach; localities: Foz do Iguaçu, State of Paraná (municipality not informed), State of São Paulo (municipality not informed), Valença, State of Rio de Janeiro; deposited samples CHIOC: 8179, 34685, 34780 a-e, 34781 a-g, 34810, 34895, 34896, 34899, 35327, 35329, 35331, 36192, 36194 a-b, 36191 a-b.

### *Travassosascaris araujoi* ((Schneider, 1866) Sprent, 1978 (Figs 44, 45)

Morphometrics based on a single immature female specimen. Body 110 long, 1.06 wide. Mouth with three quadrangular lips, shallow interlabia present; dentigerous ridge extending around whole border of lips to posterior corner. Vulva without salient lips, vagina short, extending posteriorly to join undivided uterus, leading to four uterine brnches. Tail with conical tip, without mucron, phasmids near tip.

Site of infection: stomach; locality: Valença, State of Rio de Janeiro; deposited sample CHIOC: 36491.

*Remarks*: Data reported by Sprent (1978) and Araújo (1969 a, b) on the material previously studied by Schneider (1866), reinforce the statement that ascaridid females that occur in specimens of *Crotalus durissus* and that present more than two uterine branches are included in the genera *Hexametra* Travassos, 1920 (6 uterine branches and absence of interlabia) *Polydelphis* Dujardin, 1845 (4 uterine branches and absence of interlabia) and *Travassosascaris* Sprent, 1978 (4 uterine branches and presence of interlabia).

Diaphanocephaloidea Diaphanocephalidae Travassos, 1920 *Kalicephalus inermis macrovulvus* Caballero, 1954 (Figs 35-39)

Morphometrics based on five specimens, two males and three females. Males 12.94 long, 0.32 wide; females 13.94 (12.40-16.32) long, 0.39 (0.33-0.49) wide. General: anterior extremity slightly bent. Buccal capsule 0.22 (0.16-0.23) long, 0.25 (0.17-0.31) wide. Esophagus club-shaped with a posterior bulb, 0.42 long, 0.22 wide. Teeth absent. Males: with equal and slender spicules, 0.84 (0.81-0.86) long. Gubernaculum present. Copulatory bursa broad and oblique. Dorsal rays somewhat close to each other and splitting at the extremities ("V" pattern). The externo-lateral rays are short and with rounded tips. Females: amphidelphic, vulva with salient lips, followed by divergent muscular ovijectors. Eggs 0.065-0.072 long, 0.036-0.039 wide. Tail elongate with rounded tip.

Site of infection: stomach; locality: Peixoto de Azevedo, State of Mato Grosso (10°13'12"S, 54°58'08"W); deposited samples: CHIOC: 35341, 35342.

Remarks: According to Schad (1962), in the revision of the genus *Kalicephalus* Molin, 1861, there is a great intra-specific variability and, conversely, also an inter-specific homogeneity among the species of this complex group. In order to properly identify the samples presently studied as K. inermis Molin, 1861 some reliable and well defined characters have been considered, such as the absence of the corona radiata and the buccal teeth, amphidelphic females, males with equal and long spicules and the pattern of rays of the caudal bursa. In despite of the existence of morphological differences among the representatives of the "inermis group", K. inermis inermis Molin, 1861, K. inermis coronellae Ortlep, 1923 e K. inermis macrovulvus Caballero, 1954, that were considered only as geographic groups by Schad (1962), the samples studied here were identified to K. inermis

*macrovulvus*. The identification was mainly based on the "V" pattern of bursal rays, on the aspect of the female tail that is narrow, elongate, with round tip, and also on the anterior portion of the buccal capsule, varying in shape from pointed to straight.

#### Kalicephalus sp.

Infection site: stomach; locality: Peixoto de Azevedo, State of Mato Grosso; deposited samples: CHIOC: 35339, 35340.

## Physalopteroidea Physalopteridae Leiper, 1908 *Physaloptera* sp. (Figs 40-42)

Morphometrics based on two female specimens. Body 106.2 (104.6-108.9) long, 0.55 wide. Mouth with two lateral, simple triangular lips, each bearing either apically or internally, a variable number of teeth and external papillae. Cuticle generally bent over the lips, forming a huge cephalic collar. Esophagus with an anterior muscular portion, followed by a posterior glandular, 6.75 (5.9-7.6) in total length and 0.5 in width. Vulva opening at the posterior portion of the body. Tail slender.

Site of infection: not informed [frequently in the stomach]; locality: Foz do Iguaçu, State of Paraná; deposited samples: CHIOC: 35100, 36195 a-d.

*Remarks*: This is the first report of a physalopterid worm in C. terrificus in Brazil. Probably, this species could be identified to Physaloptera obtusissima Molin, 1860, recovered from specimens of *Bothrops jararaca* (Wied, 1824), the common jararaca, and Cloelia sp., known as mussurana in Brazil. The other species of the genus that occur in Brazilian snakes are: P. monodens from the stomach of Constrictor constrictor (L., 1758), the red-tailed boa, P. liophis Vicente & Santos, 1974, from the stomach of specimens of Liophis miliaris (L., 1758), the common water snake, and Physaloptera sp. from the stomach of specimens of Wagleropsis merremii (Wagler, 1824) [=Xenodon merremii] the Wagler's snake, and Phylodrias sp., the green snake. Considering that females of *Physaloptera* share many common characters, the unavailability of male specimens did not permit a reliable specific diagnosis of the presently investigated samples.



Figures 40-45. *Physaloptera* sp. Fig. 40. Female, anterior extremity, ventral view. Fig. 41. Head, *en face* view. Fig. 42. Female, posterior extremity, lateral view. *Hastospiculum* sp. Fig. 43. Anterior extremity of female, with vulvar aperture (arrow), lateral view. *Travassosascaris araujoi*. Fig. 44. Female, posterior extremity, lateral view. Fig. 45. Vulva, vagina, ovijector and uterine bifurcation, lateral view. Bar (common to Figs 40-45) = 0.4 in Fig. 40; 0.01 in Figs 41, 42, 44; 1.1 in Fig. 43; 0.15 in Fig. 45 (in mm).

Diplotriaenoidea Diplotriaenidae Anderson, 1958 *Hastospiculum* sp. (Fig. 43)

Morphometrics unavailable. An overall description, based on broken females, refers to the mouth with two outstanding lip-like chitinous processes in each side of the aperture. Esophagus clearly divided into two muscular portions, the anterior short and narrow, the posterior broad and much longer. Vulva inconspicuous, located near the end of the anterior esophageal portion. Ovejictor stout and wide, ending in a single portion of the uterus that further bifurcates into two branches. Amphidelphic, oviparous, with embryonated eggs in utero. Anus terminal.

Site of infection: connective tissue; locality: Cachimbo, State of Pará (09°20'11"'S, 54°57'36"W); deposited samples: CHIOC: 21647, 36495.

*Remarks*: Considering that no males were available, only the generic diagnosis was obtained. Two other species of the genus are referred in Brazil: *Hastospiculum digiticaudum* Freitas, 1956, from the body cavity of specimens of *Philodrias aestivus* Dumeril, Bibron & Dumeril, 1854 captured in the State of Rio de Janeiro and *H. onchocercum* Chitwood, 1932, from *Crotalus durissus terrificus* (Laurenti, 1768), in Recife, State of Pernambuco (Vicente *et al.* 1993).

Final remarks: Other nematode species not found in the present study, and referred by Vicente et al. (1993), parasitizing Brazilian specimens of C. durissus are Ascaridia flexuosa (Schneider, 1866) Railliet & Henry, 1914, Capillaria crotali (Rudolphi, 1819) Travassos, 1915, Hastospiculum onchocercum Chitwood, 1932, Kalicephalus costatus costatus (Rudolphi, 1819) Yorke & Maplestone, 1926, K. inermis inermis (Molin, 1861) and Ophidascaris trichuriformis Vaz, 1935. Except for the pentastomid Cephalobaena tetrapoda Heimans, 1922 cited by Rego, (1983) from the lungs of Crotalus terrificus (Laurenti, 1758) and the digenetic trematode Ophiodiplostomum spectabile Dubois, 1936, from the intestine of the same host, referred by Rosselini (2007), there are no reports of other metazoan endoparasites infecting rattlesnakes in Brazil so far.

## DISCUSSION

The major restricting aspect of the present study was mainly related to the source and amount of investigated rattlesnakes. Considering that animals were taken from nature into captivity by donators, the tracking of the proper origin of the specimens was sometimes unsuccessful. Also, the number of samples was not statistically standardized taking into account that rattlesnakes were captured randomly mostly by the farmers during sowing and harvest seasons.

Specimens of *C. durissus* are animals that assumed great importance circa 1901, when Vital Brazil began with the studies related to ophidic venoms, already referring to the higher toxicity of the crotalic venom, when compared to others and the necessity in obtaining specific sera.

According to Cardoso (2003), from 1950 to 1980, when systematical investigations regarding ophidism were undertaken, mainly in the Brazilian southeastern region, the official institutes, namely Butantan, Vital Brazil and Ezequiel Dias and the private laboratory Syntex do Brasil, were the only to produce anti-ophidic sera. Until 1985, those sera were not considered a priority by the National Immunization Program of the Health Ministry. Thus, when the private facility finished with its production in 1983, the public institutions had no capacity to support the demand for sera in the country. Following the collapse of sera distribution, press releases informed the population about the occurrence of several deaths due to the lack of sera supplies. Further administrative efforts were made aiming at the support of the already existing and at the establishment of new ophidiology centers to promote the capture, maintenance, reception of the snakes, training of staff and the identification of snakes responsible for human accidents. Therefore, it was necessary to improve the knowledge of the physiology, behavior and pathologies related to snakes, taking into account that the production of sera depends on the venom extracted from these reptiles maintained in serpentariums and/or animal houses.

Captive specimens of *C. durissus* have a remarkable importance in the development of pharmacological studies aiming at the isolation of venom peptides; the required quality and volume of this venom depend on the health and adaptation of the animals submitted to the extraction procedure.

It is widely known that under captivity the stress to which animals are submitted can induce the settlement of several pathologies, mainly those related to parasites (Leinz et al. 1989, Santos et al. 2008, Siqueira et al. 2009). The former authors describe the "Poor Adaptation Syndrome" to the captivity, as the most important cause of mortality of snakes in the two first years of imprisonment, considering that animals develop remarkable susceptibility to pathogenic agents in this period of adaptation. Thus, it is necessary to provide additional data on the helminth fauna of Brazilian reptiles, in order to permit, among other approaches, the establishment of prophylactic procedures, adequate handling and effective treatment to properly reduce the mortality of captive animals.

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