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ORIGINAL ARTICLE / ARTÍCULO ORIGINAL

LONGIBUCCA CATESBEIANAE (NEMATODA: CYLINDROCORPORIDAE) OF THE BULLFROG, *LITHOBATES CATESBEIANUS* (ANURA: AMPHIBIA) FROM FROG FARMS IN THE STATE OF SÃO PAULO, BRAZIL

LONGIBUCCA CATESBEIANAE (NEMATODA: CYLINDROCORPORIDAE) EN LA RANA-TORO *LITHOBATES CATESBEIANUS* (ANURA: AMPHIBIA) DE LAS GRANJAS DE RANAS EN EL ESTADO DE SÃO PAULO, BRASIL

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Abstract

Most parasites represent a serious socioeconomic problem, because they affect pets, commercially raised, wild, and zoonotic animals. Weight loss, growth retardation, predisposition to other diseases and death are symptoms presented by the parasitized animals. The aim of this study was to evaluate the helminth fauna in bull-frog *Lithobates catesbeianus* raised for sale. We worked with five frog farms in the Vale do Paraíba, São Paulo, Brazil, and examined a total of 185 animals. The autopsies were performed, and all bodies that may have parasites were examined. Diagnostic parasitology tests were also performed. We found one species of nematode, *Longibucca catesbeianae*, with a prevalence of 1.7%, mean abundance of 14.16 and an average intensity of 850 (with a range of 729 to 1014). Given that most studies of parasitology in amphibians is conducted in free-living animals, more research on frogs in captivity should be performed to better understand these pests and prevent future problems in the operations frog farm.

Key words: Bullfrog – infection - *Lithobates catesbeianus* – Nematode - parasite - *Rana catesbeiana.*

Resumen

La mayoría de los parásitos representan un problema socioeconómico grave, porque afectan a las mascotas, a los animales criados para la venta y a los animales salvajes, y algunos también son zoonóticos. La pérdida de peso, retraso del crecimiento, la predisposición a otras enfermedades y la muerte son los síntomas presentados por los animales parasitados. El objetivo de este estudio fue evaluar la helmintofauna en la rana-toro *Lithobates catesbeianus* criada para la venta. Se trabajó con cinco granjas de rana en el Vale do Paraíba, São Paulo, Brasil, y se examinaron un total de 185 animales. Las necropsias se realizaron, y todos los órganos que pueden tener parásitos fueron examinados. Exámenes de diagnóstico coproparasitológico también fueron realizados. Se encontró una sola espécie de nematodo y se identificó como *Longibucca catesbeianae*, con una prevalencia de 1,7%, abundancia media de 14,16 y una intensidad media de 850 (con un rango de 729 a 1.014). Teniendo en cuenta que la mayoría de los estudios de parasitología en los anfibios se llevan a cabo en animales de vida libre, más investigación en ranas en cautiverio se debe realizar con el fin de comprender mejor a estos parásitos y evitar futuros problemas en las operaciones comerciales de granjas de rana.

Palabras clave: Infección – *Lithobates catesbeianus* - Nematodos - parasitos, rana-toro - *Rana catesbeiana*.

INTRODUCTION

The American bullfrog (*Rana catesbeiana* Shaw, 1802), recently reclassified as *Lithobates catesbeianus* Frost *et al*., 2006, was introduced in Brazil in the 1930s from specimens brought from Canada and was established in a frog farm known as Ranário Aurora in the state of Rio de Janeiro (Ferreira *et al*., 2002). Until the 1970s, the frog rearing and fattening tanks were called multiple tanks, in which various types of food were offered, such as slaughterhouse leftovers to attract insects (flies) with developing larvae for attracting frogs. However, the unpleasant odor caused a negative impact on production that culminated in the development of several other systems of rearing and fattening. Notwithstanding the existence of a full range of rearing structures, Brazilian producers typically modify or mix systems so as to adapt these structures for a variety of purposes, giving rise to the so-called hybrid systems (Teixeira *et al.*, 2001; Ferreira *et al.*, 2002). Currently, frog farming in Brazil constitutes an alternative agroindustrial enterprise, particularly to producers with restricted area availability, due to its minimal space requirements (Dias *et al.*, 2008, 2009).

Large concentrations of animals constitute a factor favoring the emergence of disease through the inherent creation of a favorable environment for epizootic outbreaks due to the presence of different pathogens that, under natural conditions, would have a minimal presence (Pavanelli *et al.*, 2002). According to Hipolito (2004), most parasites do not cause the death of the animal; however, they can seriously compromise the host's development. Parasitism includes actions in which one organism, the parasite, is metabolically dependent on another, the host, and depending on numerous factors, may or may not cause damage leading to the emergence of parasitic disease.

Many protozoa and metazoa found in amphibians are not associated with disease unless the host is stressed or immunocompromised. Amphibians most susceptible to parasitism are those that are newly captured, transported or maintained under poor hygienic conditions and outside of their optimal thermal zone of activity (Wright & Whitaker, 2008). According to these authors, the parasites with an indirect life cycle tend to die if the amphibian used as an intermediate or definitive host is not in its native environment. With respect to parasites with a direct life cycle, the infections can be intensified in closed environments.

The main helminths with socioeconomic interest, due to their high prevalence in domestic and wild animals, belong to the phylum Nemathelminthes, which includes nematodes, and to the phylum Platyhelminthes, which comprises cestodea, trematodea and monogenea (Almeida & Ayres, 1996). According to Fagonde Costa (2005), most nematodes produce adverse effects that are dependent on the parasite load in the animal. Thus, the mere presence of some worms does not indicate that the hosts are being harmed. The pathological action of the parasite depends on the affected organ, perforating injuries, parasitism intensity and secondary bacterial contamination (Hipolito, 2004).

The study of parasitic diseases that affect amphibians in captivity is still incipient. Within the specialized literature, articles are mainly restricted to case references and surveys of the prevalence of parasites, with few studies on the parasite-host relationship. A review by Hipolito (2004) catalogued the occurrence of infestation by crustaceans (*Lernaea cyprinacea* Linnaeu,1758), insects (myiasis caused by the *Notochaeta* sp. fly larva), acanthocephala and nematodes, such as *Longibucca catesbeianae* Souza Junior, Artigas & Martins, 1993, in Brazil.

In the international literature, rare cases of infestation by nematodes have been referenced, and include infestations by *Eustrongylides* sp, which is considered a zoonotic parasite and has been found encysted in the muscles of farmed bullfrogs in Cuitzeo Lake, Mexico (Lezama & Sarabia, 2001).

The aim of this study was to understand the possible parasitoses suffered by commercially raised bullfrogs in the State of São Paulo and to assess the sanitary conditions of farms.

MATERIALS AND METHODS

The helminthofauna survey was performed in five frog farms in different municipalities in the region of Vale do Paraíba, São Paulo, Brazil. These frog farms were designated A, B, C, D and E, and this region was chosen for having the largest number of active frog farms in the state.

We collected 36 apparently healthy bullfrogs (*L. catesbeianus*) from each property, for a total of 180 individuals, between March and June 2008. The post-metamorphosis animals were collected randomly and redistributed in the laboratory into three groups of 12 animals each, according to their stage of development within the frog farm rearing (Lima & Agostinho, 1992).

Five wild bullfrogs (invaders) were also collected from the dependencies of frog farm B (dam) that inhabited this site in the absence of any husbandry, sanitation or food control.

The frogs were taken to the Pathology Laboratory of Aquatic Organisms of the Fisheries Institute in São Paulo, where they remained in vivaria adapted for bullfrogs (Bueno-Guimarães, 1999) for a maximum of four days. The water was changed daily, and the animals were force-fed with extruded feed containing 40% crude protein.

Prior to sacrifice, the animals were anesthetized by hypothermia and inspected to detect any injury or ectoparasites.

Subsequently, they were desensitized and sacrificed by medullar section and bleeding at the level of the atlas vertebra in the cervical region, using methods described by Cornel University (2005) that eliminate suffering.

Necropsies were performed on newly sacrificed animals under a stereoscopic microscope, via an incision made with scissors starting at the cloaca, exposing the entire coelomic cavity and viscera. All organs likely to be parasitized were removed and observed separately in Petri dishes. The oral cavity and tongue were also observed.

The gastric and intestinal contents were isolated for further analysis, and a portion of the final third of the intestine was used for copro-parasitological examination. This examination followed the Ritchie method for detection of helminth eggs and possible protozoa (Figueira de Mello, 1973).

Additionally, we used another method to detect helminth eggs with lighter densities, using a sucrose-saturated solution (density=1.203) instead of water. In this centrifugal flotation method, similar to that of Ritchie, ether was not used (Figueira de Mello, 1973).

The gastric mucosa and the anterior portion of the intestine were scraped with a spatula and the scrapings placed on a glass slide with a cover slip for detection of parasites by light microscopy (Souza Junior *et al.*, 1993).

The helminths found were collected and fixed in 4% formaldehyde heated to approximately 65°C, according to Eiras *et al.* (2006), and taken to the NUPELIA Ictioparasitology Laboratory (Research Nucleus in Limnology, Ichthyology and Aquaculture) at Maringá State University - UEM (Universidade Estadual de Maringá) for identification. The helminths were dehydrated in a series of alcohol baths and cleared with beech wood creosote, and the slide was mounted with Canada balsam. The identification of parasite specimens was performed using an optical microscope at a magnification of 1000x and by comparing what was observed with figures presented in the literature (Souza Junior *et al.*, 1993).

Parasite counting was performed on the McMaster chamber used in clinical laboratories for the quantification of helminth eggs (Nascimento *et al.*, 2009). Given that each reading field of the camera carries a volume of 0.15 mL and that the camera has two fields, the number of helminths found could be approximated. The liquid containing the parasites was homogenized, and three different readings of the same host were made, recording parasite counts and chamber volume. The total volume of formaldehyde with helminths was determined using a measuring cylinder. With this total volume and using a simple rule of three, the total volume of formaldehyde in which the helminths were fixed was used to estimate the total number of parasites. Calculations of amplitude, incidence, prevalence and parasite abundance followed the protocols proposed by Bush *et al.* (1997).

RESULTS

The only helminth found was the nematode *Longibucca catesbeianae* described by Souza Junior *et al.* (1993). This is a small nematode measuring 520-707 µm that is usually located in the gastric mucosa, although it can also be found in the intestinal mucosa.

Among the five frog farms examined, only two presented parasitized animals, and even then, not all individuals were infested (low prevalence). Only one frog originating from frog farm D and two frogs from frog farm E exhibited infestation by *L. catesbeianae* in the stomach, indicating a prevalence of 1.66%, a mean abundance of 14.16%, an average intensity of 850 parasites and an amplitude of 729-1014 parasites for all captive animals studied.

The infected animal from frog farm D weighed 357.1 g, making it the largest in the group, and according to farm reports, it was an animal from an older lot, above the normal age and weight for slaughter. In frog farm E, parasitized frogs weighed 201.4 g and 167.7 g, the former being the largest of the group and the latter of intermediate weight; both were within the normal weight and age for the frog fattening sector of commercial frog farming (Ferreira *et al.*, 2002).

The three parasitized animals presented worms only in the stomach that were adhered to the gastric mucosa. The other viscera and cavities did not show any lesions or structures suggestive of parasite fixation or migration, indicating that the animals were kept under appropriate husbandry conditions (Fontanello *et al.*, 1993).

The coproparasitology methods used did not reveal helminth eggs or protozoan cysts in the fecal samples analyzed.

DISCUSSION

The fact that larger animals are more highly parasitized may be due to a cumulative effect because these are, theoretically, the older animals such that they have had more opportunities to acquire parasites.

According to Measures (1994), the genus *Longibucca* was described in the 1930s in four species of bats in North America. The same study proposed that two of the nematode species, namely, *Longibucca eptesica* and *Longibucca lasiura*, are synonymous. The genus *Longibucca* has also been described in South American snakes, such as the Muçuranas, *Pseudoboa* *cloelia* and *Clelia clelia* Daudin, 1803, according to Souza Junior *et al.* (1993). The presence of *L. catesbeianae* causes gastric and intestinal lesions, bleeding and apathy in the hosts (Hipolito, 2004). However, this was not observed in this study.

The invading individuals showed a higher prevalence of parasites than did animals in farms D and E, which can be explained by the both the adoption of appropriate sanitary and feed management in farmed animals (Fontanello *et al.*,1993) and by the greater possibilities for parasite cycle completion in the natural environment.

Campos *et al.* (2007) and Tavares-Dias *et al.*, (2001), found no significant differences between the total parasite count and the count performed using a McMaster chamber, indicating the efficiency of this technique. These procedures to identify parasites, eggs and cysts in live animals need to be standardized and validated for aquatic organisms to avoid the sacrifice of animals and facilitate the collection of material.

Few studies in Brazil have described parasites affecting the commercially bred bullfrog, *L. catesbeianus*. One of these studies was the re-description of the nematode *Gyrinicola chabaudi* Araujo & Artigas, 1982, collected in the gastrointestinal tract of tadpoles from a nursery (Souza Junior & Martins, 1996). Another study was conducted in Mexico on skeletal muscle injuries of bullfrogs commercially raised in the Cuitzeo Lake, and reported the presence of the zoonotic nematode *Eustrongylides* sp, whose larvae not only cause injury but can also encyst (Lezama & Sarabia, 2001). These authors corroborate the relevance of the objective of this study of parasites in frogs with commercial importance that are primarily destined for human consumption, given that these can harbor zoonotic parasites such as those mentioned above.

Most reports on amphibian parasites are performed on wild animals that have been collected from ecological reserves, parks and other locations (McAlpine & Burt, 1998; Bursey & William, 1998). Invading frog farm amphibians in close proximity to the farmed bullfrogs were captured and analyzed for the observation of existing parasites (Souza Junior & Martins, 1996; Souza Junior *et al*., 1993).

McAlpine & Burt (1998), while conducting a field study in New Brunswick, Canada, collected *Rana catesbeiana, R. clamitans* Latreille, 1801 and *R. pipiens* Schreber,1782 and observed four species of cestodes, 14 species of digeneans, five species of nematodes and two species of Acanthocephala. Similar work performed by Brusey & Willian (1998) in Ohio, USA, described, in bullfrogs, the digenean *Haematoloechus longiplexus* Stafford, 1902 and nematode *Rhabdias ranae* Walton,1929 in the lung, the cestode *Ophiotaenia gracilis* Jones, Cheng and Gillespie, 1958 in the small intestine, the nematode *Cosmocercoides variabilis* Harwood, 1930 in the large intestine, *Physaloptera* sp Rudolphi, 1919 nematode larvae in the stomach and the nematode *Gyrinicola batrachiensis* Walton, 1929 in bullfrog tadpole intestines.

Pryor & Bjorndal (2005) hatched bullfrog eggs in the laboratory for subsequent studies of the effects of the nematode *Gyrinicola batrachiensis* on the development of tadpoles after experimental infestation. They concluded that the relationship between these animals is one of mutualism because intestinal development and fermentation are favored in the presence of nematodes.

In a survey of wild bullfrog parasites, Marcogliese *et al*., (2000) described the occurrence of digeneans of the genus *Diplostomum* sp Nordmann, 1832 in bullfrog tadpoles inhabiting the St. Lawrence River in the region of Quebec, Canada. Studies were also performed to assess intra-specific variation of the digenean *Haematoloechus floedae* Harwood, 1832, collected from the lungs of wild amphibians of the genus *Rana* in North and Central America (León-Règagnon *et al*., 2005). Yildirimhan *et al*., (2006) referenced the infestation of the urinary bladder of *Rana macrocnemis* Boulenger, 1885 by monogenea *Polystoma macrocnemis* Vaucher, 1990 and also described the presence of digenea, acanthocephala and nematoda in these frogs. All these studies show that *L. catesbeianus* are susceptible to infestation by helminths, especially in the wild, because the completion of the parasite cycle can occur under these conditions.

Distinct biological and ecological characteristics between hosts generate differences in the exposure and rates of infection/infestation, as well as differences in the degree of post-infection compatibility and susceptibility (Guidelli *et al*., 2006). In most cases, the host possesses genetic and immunological mechanisms against helminths because most of the immunological processes are under genetic control. Antibodies mainly act against helminths located on the mucosa of the digestive tract. With the evolution of relationships between parasites and hosts, helminths have evolved mechanisms to evade the immune system that are essential for their survival and proliferation (Araujo & Madruga, 2001).

The host animal has several immunological mechanisms against pathogens, infectious agents and parasites. Natural protective barriers, such as the skin, secretions and various biologically active substances, must be overcome by these agents. Helminths can overcome all these barriers (Soares, 2001).

Hipolito (2004) reported that in a survey of producers, most stated that they had adopted preventive measures. In the questionnaire administered to producers in this study, they stated that such measures, such as invader control and daily bay cleaning, were performed. However, the use of medication as a preventive measure was inappropriately reported and observed in three of the frog farms visited. At two of the facilities, water quality control was not observed and crossed other locations before arriving at the farm. In all frog farms, domestic animals had access to the bullfrog-fattening enclosure, and there was no site for cleaning of employee shoes and clothes. Only one farm used an effluent settling pond before effluent was released into the stream.

Studies should be conducted to clarify the *L. catesbeianae* life cycle because the invading animals, which lacked sanitation or food control, were much more highly parasitized. The parasite/host relationship must also be better elucidated because one can assume that bullfrogs, an exotic species in Brazil, are resistant to most existing parasites, as is the case for other introduced and farmed animals.

Even though the prophylactic and preventive measures were not ideal for biosecurity in the frog farms assessed, these measures proved effective because there was a low incidence of infections by helminths and other parasites. Only one nematode species was found to be infecting the bullfrogs, and this infection occurred with low prevalence.

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BIBLIOGRAPHIC REFERENCES

Almeida, MA, & Ayres, MC. 1996. *Considerações gerais sobre anti-helminticos*. In HS Spinosa, SL Górniak & MM Bernardi (eds). *Farmacologia Aplicada à Medicina Veterinária*. São Paulo, SP.

Araújo, FR & Madruga, CR. 2001. *Imunidade contra helmintos*. In CR Madruga, FR Araújo & FR Soares (eds). *Imunodiagnóstico em Medicina Veterinária*. Campo Grande, MS.

Bueno-Guimarães, HM. 1999. *Avaliação da resposta da Rana catesbeiana frente às variações ambientais: determinação das condições ideais de manutenção em biotério e da resposta aos poluentes aquáticos*. Tesis, Faculdade de Medicina da Universidade de São Paulo, São Paulo, SP.

Bursey, CR & William, FD. 1998. *Helminths of the Frogs*, Rana catesbeiana, Rana clamitans, *and* Rana palustris, *from Coshocton Country, Ohio*. Ohio Journal Science. vol. 98, pp. 28- 29.

Bush, AO, Lafferty, KD, Lotz, JM & Shostak, AW. 1997. *Parasitology meets ecology on its own terms: Margolis et al. Revisited*. The Journal of Parasitology, vol. 83, pp. 575 – 583.

Campos, CM, Moraes, FR & Pereira, GT. 2007. *Método estimativo para amostragem quantitativa de* Rondonia rondoni *(Nematoda, Atractidae) parasito de Peixes*. Revista Brasileira de Parasitologia Veterinaria, vol. 16, pp. 174-176.

Cornel University 2005. *Fish and Amphibian Euthanasia*. Cornel Center for Animal Resources and Education. Available from: http://www.research.cornell.edu/care/documents/SOPs/CARE306.pdf

Dias, DC, Stéfani, MV, Ferreira, CM & França, FM. 2008. *Uso de Probiótico em ração de Rã-touro* (Rana catesbeiana)*: Desempenho produtivo*. Archivos de Zootecnia, vol. 57, pp. 449-455.

Eiras, JC, Takemoto, RM & Pavanelli, GC (eds) 2006. *Métodos de estudo e técnicas laboratoriais em parasitologia de peixes*. University Maringá State, Maringá.

Fagonde Costa, C. A. 2005. Anti-helmintico. In Farmacologia Aplicada à Avicultura *Edited by* J. Palermo Neto. Roca. São Paulo, SP. pp. 192-203.

Figueira de Mello, EB. 1973. *Ensaio crítico de método de Mello e Campos para coleta de helmintos em infestações naturais de cães como critério de adequação de técnicas coproscópicas no diagnóstico dessas infestações*. Tesis. Faculdade de Medicina Veterinária da Universidade de São Paulo, SP.

Ferreira, CM, Pimenta, AGC & Paiva – Neto, JS. 2002. *Introdução à Ranicultura*. Boletim Tecnico do Instituto de Pesca, Vol. 33, pp. 1 – 15.

Fontanello, D, Wirz, RR, Arruda Soares, H, Campos, BES, Freitas, EAN & Ferreira, CM. 1993. *Comparação de quatro sistemas de engorda de rã-touro (*Rana catesbeiana *Shaw, 1802): Tanque-Ilha, Confinamento, Anfigranja e gaiolas. 1- Desenvolvimento ponderal; 2 – Custo operacional*. Boletim do Instituto de Pesca, vol. 20, pp. 43-58.

Frost, DR, Grant, T, Faivovich, J, Bain, RH, Haas, A, Haddad, CFB, De Sá, R, Channing, A, Wilkinson, M, Donnelan, SC, Raxworthy, CJ, Campbell, JA, Blotto, BL, Moler, P, Drewes, RC, Nussbaum, RA, Lynch, JD, Green, DM & Wheeler, WC. 2006. *The Amphibian tree of Life*. Bulletin of the American Museum of Natural History, vol. 297, pp. 1-370.

Guidelli, G, Taveschio, WLG, Takemoto, RM & Pavanelli, GC. 2006. *Fauna parasitária de* Leporinus lacustris *e* Leporinus friderici *(Characiformes, Anostomidae) da planície de inundação do alto rio Paraná, Brasil*. Acta Scientiarum Biological Science, vol. 28, pp. 281-290.

Hipolito, M. 2004. *Manejo Sanitário no Cultivo de Rã*. In MJT Ranzani-Paiva, RM Takemoto & MAP Lizama (eds). *Sanidade de Organismos Aquáticos*. São Paulo, SP.

León-Règagnon, V, Guillén-Hernández, S & Arizmendi-Espinosa, MA. 2005. *Intraespecfic variation of* Haematoloechus floedae *Harwood, 1932 (Digenea: Plagiorchidae), from* Rana *sp. in North and Central America*. The Journal of Parasitology, vol. 91,pp. 915-921.

Lezama, JR. & Sarabia, DO. 2001. *Lesiones histológicas em músculo esquelético, causadas por larvas de* Eustrongylides *sp (Nematoda: Dictophymatidae) em ranas comestibles del Lago Cuitzeo, Michoacán, México*. Veterinaria México, vol. 33,pp. 335- 341.

Marcogliese, DJ, Rodrigues, J, Oullet, M & Champoux, L. 2000. *Natural Occurence of* Diplostomum *sp. (Digenea: Diplosmatidae) in adult mudpuppies and Bullfrog tadpoles from the St. Lawrence River, Québec*. Comparative Parasitology, vol. 67, pp. 26- 31.

McAlpine, DF & Burt, MDB. 1998. *Helminthes of Bullfrogs,* Rana catesbeianae, *Green frogs,* Rana clamitans *and Leopard Frogs,* Rana pipiens *in New Brunswick*. The Canadian Field-Naturalist, vol. 112, pp. 50-68.

Measures, LN. 1994. *Synonymy of* Longibucca eptesica *with* Longibucca lasiura *(Nematoda: Rhabditoidea) and new host and geographic records*. The Journal of Parasitology, vol. 80, pp. 486-489.

Nascimento, EM, Furlong, J, Pimenta, DS. & Prata, MCA. 2009. *Efeito anti-helmintico do hidrolato de* Mentha villosa *HUBS (Lamiacea) em nematóides gástricos de bovinos*. Ciência Rural, vol. 6, pp. 13-19.

Pavanelli, GC, Eiras, JC, Takemoto, RM (eds). 2002. *Doença de Peixes; Profilaxia, diagnóstico e tratamento*. Universidade Estadual de Maringá, Maringá.

Pryor, GS & Bjorndal, KA. 2005. *Effects of Nematoda* Gyrinicola batrachiensis *on development, gut morphology, and fermentation in Bullfrog tadpoles (*Rana catesbeiana*): A novel mutualism*. The Journal of Experimental Zoology, vol. 303, pp. 704–712.

Soares, CO. 2001. *Imunodiagnóstico em Medicina Veterinária*. In Madruga, CR, Araújo, FR & Soares, CO (eds). *Imunodiagnóstico em Medicina Veterinária*. Embrapa Gado de Corte, Campo Grande, MS.

Souza Junior, FL, Artigas, PT & Martins, ML. 1993. Longibucca catesbeianae *n. sp. (Nematoda: Cylindrocorporidae), a gastrointestinal parasite of the bullfrog* Rana catesbeiana *Shaw, 1802 in Brazil*. Research and Reviews in Parasitology, vol. 53, pp. 97-102.

Souza Junior, FL & Martins, LM. 1996. A *redescription of* Gyrinicola chabaudi *Araujo and Artigas, 1982 (Nematoda: Pharygodonidae), a gastrintestinal parasite of tadpoles*. Revista Brasileira de Biologia, vol. 56, pp. 19–25.

Teixeira, RD, Mello, SCRP & Lima dos Santos, CAM. 2001. *The world market for frog legs*. Globefish versi, Vol *68***,**1–44. Food and Agriculture Organization (FAO), Rome.

Vazzoler, AEAM (ed). 1996. *Biologia da reprodução de peixes teleósteos: teoria e prática*. Universidade Estadual de Maringá, Maringá.

Yildirimhan, HS, Bursey, CR & Goldberg, SR. 2006. *Helminthes parasites of the Taurus Frog,* Rana holtzi, *and the Uludag Frog,* Rana macrocnemis, *with remarks on the helminth community of Turkish anurans*. Comparative Parasitology, vol. 73, pp. 237-248.

Wright, KN & Whitaker, BR. 2008. *Anfíbios*. In Kahn, CM & Line, S (eds). *Manual Merck de Veterinária*. Roca, São Paulo.

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