

## ORIGINAL ARTICLE / ARTÍCULO ORIGINAL

*POLYSTOMA CUVIERI* (MONOGENEA, POLYSTOMATIDAE) IN *PHYSALAEMUS CUVIERI*  
(ANURA, LEIUPERIDAE) IN SOUTHERN BRAZIL

*POLYSTOMA CUVIERI* (MONOGENEA, POLYSTOMATIDAE) EN *PHYSALAEMUS CUVIERI*  
(ANURA, LEIUPERIDAE) EN EL SUR DE BRAZIL

Viviane G. T. Santos<sup>1,2</sup> & Suzana B. Amato<sup>1,3</sup>

<sup>1</sup>Departamento de Zoologia, Instituto de Biociências, Universidade Federal do Rio Grande do Sul, Porto Alegre,  
91501-970 Porto Alegre, Rio Grande do Sul, Brasil;

<sup>2</sup>CAPES Doctoral Fellowship; <sup>3</sup>CNPq Productivity Scholar.  
E-mail: santosvgt@gmail.com; sbamato@ufrgs.br

Suggested citation: Dos Santos, VGT & Amato, SB. 2012. *Polystoma cuvieri* (Monogenea, Polystomatidae) in *Physalaemus cuvieri* (Anura, Leiuperidae) in Southern Brazil. *Neotropical Helminthology*, vol. 6, nº 1, pp. 1-7.

### Abstract

*Physalaemus cuvieri* Fitzinger, 1823 occurs in Brazil, Paraguay and Argentina. In order to describe the helminth fauna of these anurans in southern Brazil, 68 specimens were captured in Campo Belo do Sul and Anita Garibaldi, State of Santa Catarina, and in Bom Jesus, State of Rio Grande do Sul, between May 2009 and January 2010. The anurans were taken alive to the laboratory where they were anesthetized and necropsied. Monogeneans identified as *Polystoma cuvieri* Vaucher, 1990 were found in the urinary bladder with a prevalence of 31.86% and a mean intensity of 2.06 helminths/host. In the same location of this study, other anuran species were captured and examined for helminths, but *P. cuvieri* was found exclusively in *Ph. cuvieri*. This result may indicate a high parasite/host specificity.

**Key words:** anuran - monogenean - urinary bladder - host/parasite specificity - southern Brazil.

### Resumen

*Physalaemus cuvieri* Fitzinger, 1823 está presente en Brasil, Paraguay y Argentina. Con el objetivo de conocer la helmintofauna de estos anuros, 68 especímenes fueron colectados en Campo Belo do Sul y Anita Garibaldi, Estado de Santa Catarina y Bom Jesus, Estado de Rio Grande do Sul, en el sur de Brasil, entre mayo de 2009 y enero de 2010. Los anuros fueron llevados vivos al laboratorio, anestesiados y necropsiados. Monogéneos indentificados como *Polystoma cuvieri* Vaucher, 1990, fueron encontrados en la vejiga urinaria con una prevalencia de 31,86% y una intensidad média de 2,06 helmintos/hospedador. En la misma área de estudio, se han capturado otras espécies de anuros y sus helmintos fueron identificados, pero *P. cuvieri* se encontró exclusivamente en *Ph. cuvieri*. Este resultado puede indicar una alta especificidad parásito/hospedador.

**Palabras clave:** anuros, monogéneos, vejiga urinaria, especificidad parásito/hospedador, sur de Brasil.

## INTRODUCTION

*Polystoma* Zender, 1800 is the most diverse among the 20 recognized genera of Polystomatidae sensu Sinnappah *et al.* (2001) with 63 species, about one third of the total number of described species. Species of *Polystoma* are characterized by their strict host specificity. They have monoxenous life cycles and one larval stage, the oncomiracidium, which infests the host as tadpoles. The development initiates and can be completed in the tadpole gills, or when they reach the urinary bladder at the end of host's metamorphosis (Combes, 1968; Maeder, 1973; Kok & Du Preez, 1989; Badets & Verneau, 2009). *Polystoma* species have been recorded in different parts of the world, mainly in Africa, but there are no records in Australia (Bentz *et al.* 2001). In South America eight species of *Polystoma* have been registered, and they are known in Argentina, Ecuador and Paraguay (Combes & Laurent, 1974; 1978, 1979; Vaucher, 1987, 1990) (Table 1). According to Bentz *et al.* (2001), the genus originated in South American hylids, after South America and Africa broke apart, and the colonization of North America occurred during the Paleocene, and Eurasia during the Cenozoic Period, with the dispersion of the bufonid and hylid ancestrals. Meanwhile, the African continent appears to have been colonized only recently.

*Physalaemus cuvieri* Fitzinger, 1826 (Leiuperidae), barker-frog or frog-dog, is found in open areas in southern Brazil and in the eastern region of Paraguay and Argentina (Misiones and Corrientes). They breed in small ponds and dams, or in temporary water sources such as puddles. Their tadpoles live in the water bottom and feed on organic suspensions (Kwet & Di-Bernardo, 1999).

New taxonomic characters of *Polystoma cuvieri* Vaucher, 1990 are elucidated, and specimens collected from *Ph. cuvieri* from southern Brazil described.

## MATERIAL AND METHODS

Ninety-one specimens of *Ph. cuvieri* (41 females, 50 males) were captured with permission of SISBIO ("Sistema de Autorização e Informação em Biodiversidade", "Ministério do Meio

Ambiente", Brazil) (N° 19937-1) between May 2009 and January 2011, in Campo Belo do Sul (27°53'58.77"S, 50°45'31.97"W) and Anita Garibaldi (26° 59'57.33"S, 51°24'32.94"W) State of Santa Catarina, and Bom Jesus (28°40'01.48"S, 50°26'12.42"W), State of Rio Grande do Sul, southern Brazil. The frogs were captured by hand, taken alive to the laboratory, and killed with Lidocaine Geyer® 2% (Di Bernardo, personal communication) which was applied to the ventral surface to be absorbed by the skin. They were weighed and measured (snout to cloaca). The sex was determined by the gular region (in males this region is dark). The monogeneans were collected and placed in 0.65% saline physiologic solution, fixed in AFA (93 parts of ethanol 70 °GL, 5 parts of commercial formalin, 2 parts of glacial acetic acid) under slight pressure, stained with Delafield's hematoxylin (Humason, 1972), cleared in cedar oil and mounted in Canada balsam (Amato & Amato 2010). A few monogenean specimens were mounted in De Faure's medium (Romeis, 1958) to better visualize the hamuli. Morphologic and biometric studies were done with an Axiolab Zeiss light microscope. Measurements are in micrometers (µm) unless otherwise indicated; ranges for each character are followed inside parentheses, by mean, standard deviation and the number of specimens measured. Voucher specimens were deposited in the Helminthological Collection of the "Instituto Oswaldo Cruz" (CHIOC), Rio de Janeiro, RJ and in the Helminthological Collection (CHDZ), "Departamento de Zoologia, Universidade Federal do Rio Grande do Sul" (UFRGS), Porto Alegre, RS. All frogs examined were deposited in the Amphibian Collection, "Laboratório de Herpetologia, Departamento Zoologia, UFRGS, Porto Alegre, RS", Brazil.

## RESULTS

*Polystoma cuvieri* Vaucher, 1990 (Figs. 1-3)

Description. Based on 17 specimens mounted *in toto*, 16 measured and three hamuli pairs mounted in Faure. Polystomatidae. Body elongated while alive, whitish with brown caeca (Fig. 1). Total body length 2 to 4.8 mm (3.1; 0.94; 16); body width 0.63 to 2.21 mm (1.24; 0.48; 16). Ventral,

subterminal mouth; oral sucker 40 to 210 (182; 29; 10) long; pharynx 150 to 270 (188; 33; 13) long, 145 to 280 (191; 40; 13) wide. Bifurcate and anastomosed intestine with numerous external digitations, extending to the haptor (Fig. 2). Haptor 766 to 1718 (1179; 288; 16) long; 780 to 2271 (62; 459; 16) wide, with six similar suckers; anterior suckers 175 to 500 (320; 92; 15) long, 213 to 510 (334; 100; 15) wide; median suckers 175 to 470 (319; 73; 16) long, 180 to 450 (294; 67; 16) wide; posterior suckers 200 to 430 (293; 67; 15) long, 215 to 470 (314; 73; 16) wide; hamuli length to the tip of the handle (X) 210 to 345 (285; 45; 15), hamuli length to the tip of the guard (Y) 210 to 325 (260; 36; 15), ratio X/Y 0.9 to 1.2 (1.1; 0.1; 15) (Fig. 3), hamuli hook length 48 to 68 (59; 6; 15).

Testis single and follicular in middle of body, post-ovarian, hidden by vitelline follicles. Genital bulb posterior to intestine, 8 genital spines 13 to 28 (18; 6; 8) long. Ovary sinistral, anterior, curved, 400 to 960 (637; 204; 8) long, 100 to 514 (292; 145; 8) wide. Vitellarium follicular, dorsal and ventrally occupying most of body from pharynx to hamuli. Uterus reaching the genital pore through several loops. Lateral vagina near anterior extremity of ovary. Only two mounted specimens had eggs; each specimen with only one egg, 160 to 280 (220; 85; 2) long, and 95 to 100 (98; 4; 2) wide.

Infection site: urinary bladder.

Collection site: Municipalities of Campo Belo do Sul and Anita Garibaldi, State of Santa Catarina, and Bom Jesus, State of Rio Grande do Sul, Brazil. Prevalence: 31.86%.

Mean intensity of infection: 2.06 helminths/host.

Mean abundance of infection: 0.65 helminths/host.

Range intensities of infection: 1-11 helminths/host.

Deposited specimens: CHIOC 35805. CHIOC 37771. CHIOC 37772. CHDZ 3027-1-2. CHDZ 3042-1-1. CHDZ 3055-1-1.

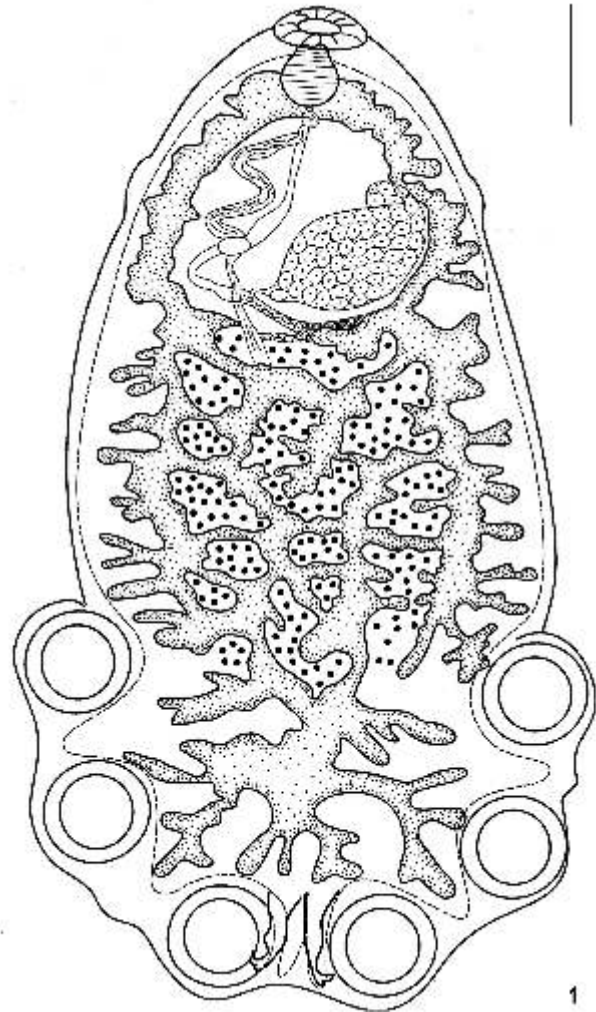
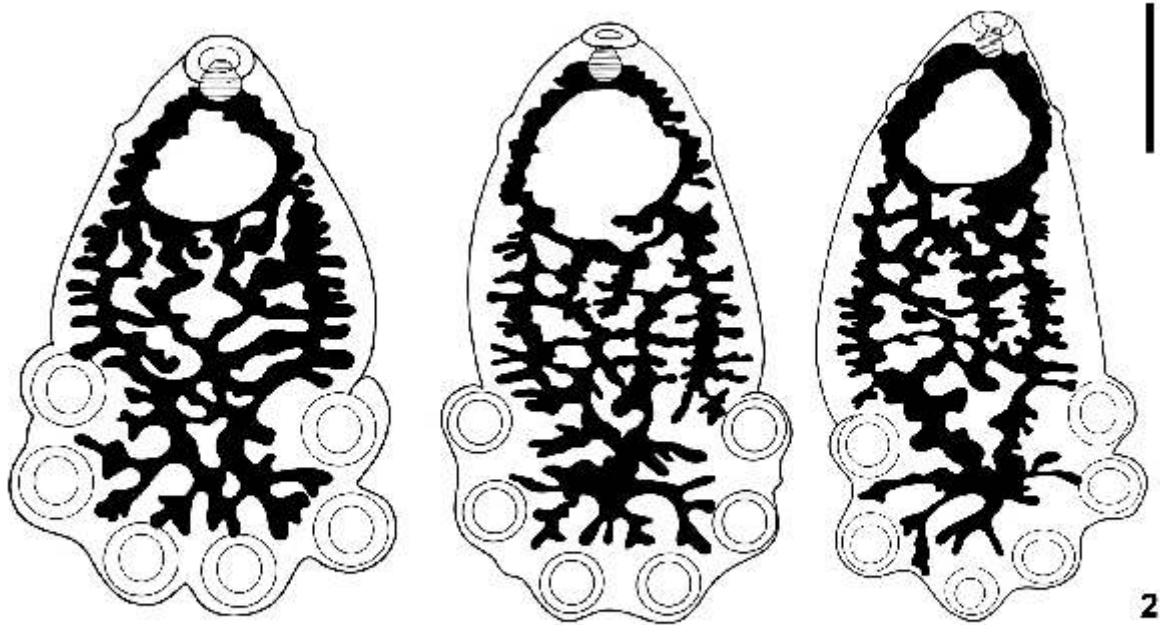


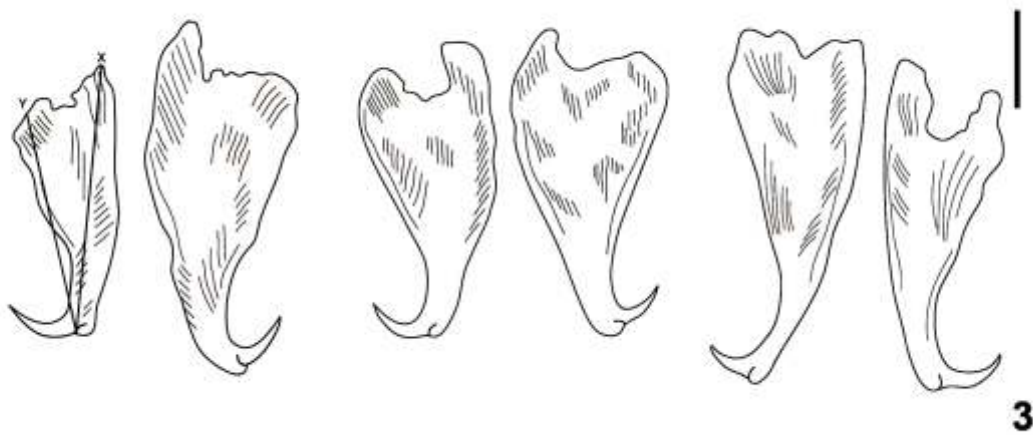
Figure 1. *Polystoma cuvieri* ventral view. Bar = 400  $\mu$ m.

## DISCUSSION

*Polystoma cuvieri* was highly specific to *Ph. cuvieri*, seven other anuran species (60 *Rhinella icterica*; 48 *Leptodactylus latrans*; 45 *Scinax fuscovarius*; 29 *Melanophryniscus simplex*, 11 *Ischnocnema henselii*; six *Elachistocleis bicolor* and four *L. plaumanni*) were captured along with 91 *Ph. cuvieri*, and only *Ph. cuvieri* hosted *P. cuvieri*. Vaucher (1990) examined 15 species of anurans and only *Ph. cuvieri* and *Physalaemus biligonigerus* (Cope, 1861) hosted *P. cuvieri*. Since *Ph. biligonigerus* had only one specimen in its



**Figure 2.** *Polystoma cuvieri* general ventral view showing caeca variations. Bar = 500  $\mu$ m.



**Figure 3.** Hamuli from three specimens of *Polystoma cuvieri*. Bar = 100  $\mu$ m.

**Table 1.** *Polystoma* species recorded for South America.

Species	Body length (mm)	Hamuli length (µm)	Host	Locality	References
<i>P. borellii</i> Combes & Laurent, 1974	4.2 - 5.6	350 - 530	<i>Pleurodema borelli</i> (Peracca, 1895)	Argentina	Combes & Laurent (1974)
<i>P. praecox</i> Combes & Laurent, 1978	3.0 - 6.4	350 - 377	<i>Telmatobius oxycephalus</i> Vellard, 1946	Argentina	Combes & Laurent (1978)
<i>P. andinum</i> Combes & Laurent, 1978	4.9 - 8.0	370-480	<i>Melanophryniscus rubriventris</i> (Vellard, 1947)	Argentina	Combes & Laurent (1978)
<i>P. guevarai</i> Combes & Laurent, 1979	6.79 - 7.88	298 - 348	<i>Hyla pulchella</i> Duméril & Bibron, 1841 (= <i>Hypsiboas pulchellus</i> (Duméril & Bibron, 1841))	Argentina	Combes & Laurent (1979)
<i>P. lopezromani</i> Combes & Laurent, 1979	6.99 - 8.16	544 - 606	<i>Phrynohias venulosa</i> (Laurenti, 1768)	Argentina	Combes & Laurent (1979)
<i>P. diptychi</i> Vaucher, 1986	8.3	970 - 980	<i>Bufo diptychus</i> Cope, 1862 (= <i>Rhinella diptycha</i> (Cope, 1862))	Paraguay	Vaucher (1986)
<i>P. napoensis</i> Vaucher, 1987	2.68-3.47	286-368	<i>Osteocephalus lepreurii</i> (Duméril & Bibron, 1841)	Ecuador	Vaucher(1987)
			<i>Osteocephalus taurinus</i> Steindachner, 1862	Ecuador	Vaucher(1987)
<i>P. touzei</i> Vaucher, 1987	4.18	315-319	<i>Gastrotheca tobamba</i> (Fowler, 1913)	Ecuador	Vaucher(1987)
<i>P. cuvieri</i> Vaucher, 1990	2.44 - 4.23	278 - 413	<i>Physalaemus cuvieri</i> and <i>Ph. biligonigerus</i>	Paraguay	Vaucher (1990)
<i>P. cuvieri</i>	2.37 - 4.79	210 - 330	<i>Physalaemus cuvieri</i>	Brazil	Present study

urinary bladder, Vaucher (1990) considered it as an accidental host and that the infestation was promoted by the ecological conditions. The prevalence found in this study was relatively high and evidenced of high parasite/host specificity.

Measurements of the specimens collected from the Brazilian *Ph. cuvieri* are similar to the ones in the original description done by Vaucher (1990), but differed from the other seven species registered in South America (Table 1). For the first time measurements of ovary, hamuli length to the tip of the handle and to the tip of the guard, hamuli hook length, and genital spines are given for *P. cuvieri*. This is the first record of *P. cuvieri* in Brazil.

### ACKNOWLEDGMENTS

To the herpetologists, Rafael Balestrin, Patrick Colombo, Martin Schossler, Daniel Bühler and Simone Leonardi for helping with the field work, and to Mariana Gliesh and Etiele Senna for helping with the laboratory work. Phillip J. Scholl, for kindly reviewing the English version; and Marta Fabián and Cynthia González for reviewing the Spanish abstract.

### BIBLIOGRAPHIC REFERENCES

- Amato, JFR & Amato, SB. 2010. *Técnicas gerais para coleta y preparação de helmintos endoparasitos de aves*. In: Von Matter, S, Straube, FC, Accordi, I, Piacentini, V, Cândido - Jr., JF (eds.). *Ornitologia e Conservação: Ciência Aplicada, Técnicas de Pesquisa e Levantamento*, Technical Books Editora, Rio de Janeiro, 518 pp.
- Badets, M & Verneau, O. 2009. *Origin and evolution of alternative developmental strategies in amphibious sarcopterygian parasites (Platyhelminthes, Monogenea, Polystomatidae)*. *Organisms, Diversity & Evolution*, vol. 9, pp. 155-164.
- Bentz, S, Leroy, S, Du Preez, L, Mariaux, J, Vaucher, C & Verneau, O. 2001. *Origin and evolution of African Polystoma (Monogenea: Polystomatidae) assessed by molecular methods*. *International Journal for Parasitology*, vol. 31, pp. 697-705.
- Combes, C. 1968. *Biologie, écologie des cycles et biogéographie de digènes et monogènes d'amphibiens dans l'est des Pyrénées*. *Memoires du Museum National d' Histoire Naturelle, Serie A, Zoologie*, vol. 1, pp. 1-195.
- Combes, C & Laurent, RF. 1974. *Polystoma borelli n. sp. (Monogenea, Leptodactylidae) parasite de Pleurodema borellii Peracca (Anura, Leptodactylidae) en Republique Argentine*. *Acta Zoológica Lilloana*, vol. 31, pp. 57-64.
- Combes, C & Laurent, RF. 1978. *Deux nouveaux Polystomatidae (Monogenea) de Republique Argentine*. *Acta Zoologica Lilloana*, vol. 33, pp. 85-91.
- Combes, C & Laurent, RF. 1979. *Les monogènes Polystomatidae de Republique Argentine: Description de deux nouvelles espèces et essai de synthèse*. *Revista Ibérica de Parasitologia*, vol. 79, pp. 545-557.
- Humason, GL. 1972. *Animal tissue techniques*. São Francisco, W.H. Freeman and Company, 641p.
- Kwet, A & Di-Bernardo, M. 1999. *Pró-Mata – Anfíbios*. EDIPUCRS, Porto Alegre, 107p.
- Kok, DJ & Du Preez, LH. 1989. *Polystoma australis (Monogenea): Developmental and reproduction in neotenic parasites*. *South African Journal of Zoology*, vol. 24, pp. 225-230.
- Maeder, AM. 1973. *Monogènes et Trématodes parasites d' Amphibiens en Côte d' Ivoire*. *Revue Suisse de Zoologie*, vol. 80, pp. 267-322.
- Romeis, B. 1948. *Mikroskopische Technik*. Munchen, Leibniz, XI+695p.
- Sinnappah, ND, Lim, LHS, Rohde, K, Tinsley, R, Combes, C. & Verneau, O. 2001. *A paedomorphic parasite associated with a neotenic amphibian host: phylogenetic evidence suggests a revised systematic position for Sphyrnanuridae within anuran and turtle polystomatoineans*. *Molecular*

Phylogenetics and Evolution, vol. 18, pp. 189-201.

Vaucher, C. 1990. *Polystoma cuvieri n. sp.* (*Monogenea: Polystomatidae*), a parasite of the urinary bladder of the leptodactylid frog *Physalaemus cuvieri* in *Paraguay*. *The Journal of Parasitology*, vol. 76, pp. 501-504.

Vaucher, C. 1987. *Polystomes D'Équateur*, avec description de deux nouvelles espèces. *Muséum D'Histoire Naturelle de Genève*, vol. 110, pp. 45-56.

Received December 20, 2011.  
Accepted February 15, 2012.

\*Author for Correspondence / Autor para correspondencia:

Viviane G. T. Dos Santos  
Departamento de Zoologia, Instituto de  
Biociências, Universidade Federal do Rio Grande  
do Sul, Porto Alegre, 91501-970 Porto Alegre,  
Rio Grande do Sul, Brasil.

E-mail/correo electrónico:  
santosvgt@gmail.com