

REDISCOVERY OF *POLYSTOMA STELLAI* PÉREZ VIGUERAS, 1955 (PLATYHELMINTHES, MONOGENEA) IN CUBA REDESCUBRIMIENTO DE *POLYSTOMA STELLAI* PÉREZ VIGUERAS, 1955 (PLATYHELMINTHES, MONOGENEA) EN CUBA

✉ YANDER L. DIEZ^{1,2}, ✉ ALEJANDRO CATALÁ³, ✉ ANDREAS SCHMIDT-RHAESA¹

1. Museum of Nature Hamburg - Zoology, Leibniz Institute for the Analysis of Biodiversity Change (LIB), Martin-Luther-King-Platz 3, D-20146, Hamburg, Germany.
2. Hasselt University, Centre for Environmental Sciences, Research Group Zoology: Biodiversity and Toxicology, Universitaire Campus Gebouw D, B-3590 Diepenbeek, Belgium.
3. Universidad de Oriente, Biology & Geography Department, Ave. Patricio Lumumba s/n, CP 90500, Santiago de Cuba, Cuba.

ABSTRACT: The family Polystomatidae (Platyhelminthes, Monogenea) comprises parasitic flatworms with an affinity for infecting amphibians and reptiles. Among them, *Polystoma stellai* stands out as a particularly rare species documented in both Cuba and Florida (USA). Originally described in 1955 based on a single specimen, only one additional specimen has been collected since, and there exists no type or reference material for this species, resulting in a significantly limited understanding of its characteristics. Recently, a specimen of *P. stellai* was collected during a dissection of *Osteopilus septentrionalis* (Anura) by Biology students at Universidad de Oriente, Cuba. The studied material was collected more than 800 km from the area where the species was described. The newly collected specimen aligns with the original description of *P. stellai*; however, notable differences are observed, particularly in the reduced size of the body and certain structures compared to previously documented ones. We provide the first photos and detailed description of the hamulus, characterised by unequal hooks, one of which exhibits a distal flap. Given the available data, there arises a suspicion that the specimen previously recorded in Florida may belong to a distinct species of *Polystoma*. Furthermore, the position of *P. stellai* within *Polystoma* remains tentative due to some diagnostic characteristic of representatives of this genus have not been corroborated in this species.

KEYWORDS: Anura, Neodermata, *Osteopilus septentrionalis*, parasitic flatworms, Polystomatidae.

RESUMEN: La familia Polystomatidae (Platyhelminthes, Monogenea) incluye platelmintos parásitos con afinidad por infectar anfibios y reptiles. Entre ellos, *Polystoma stellai* resalta como especie particularmente rara documentada en Cuba y Florida (EUA). Originalmente descrita en 1955 en base a un único espécimen, solo un ejemplar adicional ha sido recolectado desde entonces y no existe material tipo o de referencia de la especie, resultando significativamente en el conocimiento limitado de sus características. Recientemente, un ejemplar de *P. stellai* fue recolectado durante la disección de *Osteopilus septentrionalis* (Anura) por estudiantes de Biología de la Universidad de Oriente, Cuba. El material estudiado fue recolectado a más de 800 km del área de donde la especie fue descrita. El nuevo ejemplar recolectado concuerda con la descripción original de *P. stellai*, no obstante diferencias notables son observadas, particularmente en el tamaño reducido del cuerpo y ciertas estructuras comparadas con las documentadas previamente. Se proveen las primeras fotografías y descripción detallada del hamulus, caracterizado por ganchos desiguales, uno de los cuales exhibe una proyección distal. Considerando los datos disponibles surge la sospecha de que el ejemplar previamente registrado de Florida podría pertenecer a otra especie de *Polystoma*. Además, la posición de *P. stellai* dentro de *Polystoma* permanece tentativa debido a que algunos caracteres diagnósticos de los representantes de este género no han sido corroborados aun en esta especie.

PALABRAS CLAVE: Anura, Neodermata, *Osteopilus septentrionalis*, platelmintos parásitos, Polystomatidae.

✉ Yander L. Diez
yanderluis87@gmail.com

Received: April 15, 2024

Accepted: July 22, 2024



This is an open-access article distributed under the terms of the Creative Commons Attribution License



<https://cu-id.com/2403/n515e05>

INTRODUCTION

The family Polystomatidae, which includes parasitic flatworms, is represented by two species in Cuba: *Neopolystoma orbiculare* (Stunkard 1916) and *Polystoma stellai* Pérez Viguera 1955 (see Pérez Viguera, 1955; Coy & Lorenzo, 1982). *Neopolystoma orbiculare* parasitizes the freshwater turtle *Trachemys decussata* (Gray 1831), while *P. stellai* is known to parasitize the tree frog *Osteopilus septentrionalis* (Duméril & Bibron 1841). In total, about 253 parasitic flatworms have been recorded from Cuba, comprising 72 cestodes, 34 monogeneans, and 146 trematodes, distributed across marine, freshwater, and terrestrial environments (Coy & Lorenzo, 1982; Mendoza-Franco *et al.*, 2006; Fernández *et al.*, 2015).

The genus *Polystoma*, a species-rich taxon within Monogenea, encompasses 67 species (WoRMS, 2024). Generally, polystomatids are regarded as cryptic taxa with challenging morphological identification (du Preez *et al.*, 2007). *Polystoma stellai* exemplifies such enigmatic characteristics, being known only from two specimens - the first collected in La Habana, Cuba (Pérez Viguera, 1955), and the second in Florida, USA (Stunkard, 1959). These specimens were not stored in any known collection, and the species was even omitted from the list of helminthic parasites of Cuban vertebrates (see Coy & Lorenzo, 1982); hence, knowledge of the species remains quite limited.

Recently, a second specimen of *P. stellai* from Cuba was collected, infecting its type host *O. septentrionalis*. This description offers a

comparison between the newly collected specimen and the two previously recorded from La Habana and Florida, contributing valuable morphological data to enhance understanding of the species.

MATERIAL AND METHODS

Ten specimens of the anuran *O. septentrionalis* were collected by Biology students of Universidad de Oriente, during dissecting practices. During the examination of the animals, one parasitic flatworm was detected in one frog collected in the locality of Santa María (Fig. 1), Santiago de Cuba, Cuba (February 22, 2022). This flatworm was isolated and studied alive under a microscope. Finally, the specimen was preserved as a whole mount with lactophenol. Hard structures of the whole mounted specimen were examined with a Leica DM 2500 LED microscope, using Nomarski interference contrast. The identification of the host frog was made on the base of description provide by Díaz and Cádiz (2008). Drawings were built on Inkscape 1.2.2. Measurements were taken along the central axis of the measured object. The voucher specimen was stored in the Museum of Nature Hamburg (ZMH).

RESULTS AND DISCUSSION

POLYSTOMA STELLAI PÉREZ VIGUERAS, 1955

KNOWN DISTRIBUTION

La Habana, Cuba (Pérez Viguera, 1955) and Florida, USA (Stunkard, 1959).

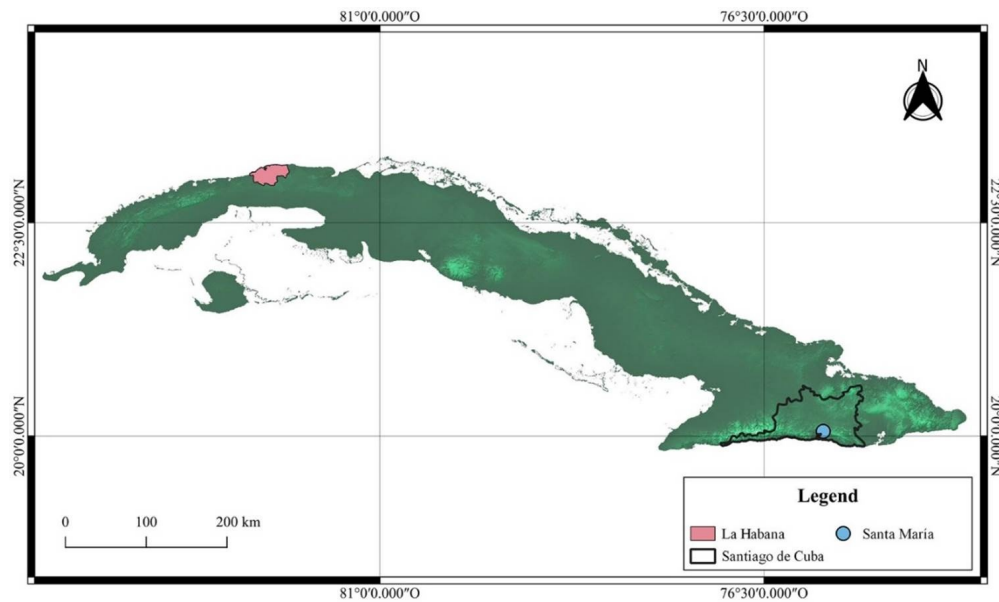


FIGURE 1. Map of the known distribution of *Polystoma stellai* in Cuba.

FIGURA 1. Mapa de la distribución conocida de *Polystoma stellai* en Cuba.

STUDIED MATERIAL

One adult specimen collected in Santa María (20.059248, -75.807753), Santiago de Cuba, Cuba (February 22, 2022), studied alive and whole mounted (ZMH V13666).

HOST

Osteopilus septentrionalis (Duméril & Bibron 1841) (Amphibia, Anura, Hylidae); female specimen; urinary bladder.

MORPHOLOGICAL DESCRIPTION

Specimen 3.94 mm long and 1.88 mm at its widest part (Fig. 2A). Pharynx (Figs. 2A-B: ph) 53 μ m long and 43 μ m at its posterior widest point. Digestive tract diverticulated with prehaptorial anastomoses. Haptor (Figs. 2A & 2C: h) 1,008 μ m long and 1452 μ m wide. Six haptorial suckers (Figs. 2A & 2C: s) measuring 257-286 μ m in diameter (\bar{x} = 274 μ m; n = 6). Anchor hooks (hamulus) (Figs. 2A & 2C: ha, 2D-E) unequal, 407-435 μ m long and 217-287 μ m at the widest proximal part. One of the hooks with a distal flap (Figs. 2D-E: f) 50 μ m long and 35 μ m wide. No marginal hooklets present.

Male genital system not observed. Ovary (Figs. 2A-B: ov) 344 μ m long and 124 μ m at its widest point, J-shaped. Vitellaria (Figs. 2A-B: vi) lie along the body sides and is better distinguishable in the anterior part of the animal. Paired vagina, only visible on the live specimen, open on each body side anterior to the ovary (Fig. 2: va). Other structures of the female system were not observed.

REMARKS

This represents the second documented specimen of *P. stellai* since its original description in 1955. Despite a lack of studies on the infection prevalence within this species, our suspicion of a very low infection rate is based on the scarcity of specimens collected over the 68 years following its description. Within our collected samples, a solitary frog exhibited infection (10%); however, since the frogs were collected from various locations, this infection rate cannot be generalised. The recently collected specimen was found approximately 800 km away from the original collection site in La Habana (Pérez Viguera, 1955), with Stunkard (1959) reporting the sole other sighting of the species in Florida.

Due to the limited number of known specimens for this species, its morphology remains somewhat mysterious. Specifically, the descriptions by Pérez Viguera (1955) and Stunkard (1959) lack detailed information regarding the morphology of the anchor hooks. In fact, Stunkard (1959) only provided measurements of specimen length and width, along

with sucker diameter (Table 1). Consequently, we present, for the first time, detailed photographs depicting the morphology of these structures. Although the male reproductive system was not observed in the most recent specimen, its well-developed ovary and vitellaria suggest adulthood. Probably, the used fixation method, typically used for free-living microturbellarians, damaged these structures; unfortunately, no other fixation method was available at that moment. Notably, the specimen from Santiago de Cuba is the smallest, yet the proportion of its body length to width resembles that of the specimen from La Habana (L/W = 2.10 vs. 2.73, respectively). Conversely, the specimen from Florida is notably more slender (L/W = 4.53). Given the scant description provided by Stunkard (1959) and the significant size disparity of their specimen, we suspect the Florida population may represent a distinct species of *Polystoma* or other related genera.

Remarkable differences in ovary length are observed between the specimens from La Habana (880 μ m) and Santiago de Cuba (344 μ m). However, this variance, along with differences in haptor and sucker sizes, may be attributed to the smaller body size of the latter specimen. Although the anchor hooks exhibit similar sizes between the two Cuban specimens (Table 1), we identified two previously unmentioned characteristics for these structures: one hook in the eastern Cuban specimen is larger and features a distal flap. It is important to note that du Preez *et al.* (2024: Fig. 4.111) purportedly presented a depiction of *P. stellai* drawn over the original illustration by Pérez Viguera (1955). However, upon examination, it becomes evident that the new drawing does not match the one provided in the original description but rather resembles the illustration by Stunkard (1959). This discrepancy, unfortunately, adds further complexity to the comprehension of this species. Therefore, we believe it is pertinent this clarification.

Some diagnostic traits observed in representatives of *Polystoma*, as detailed by Price (1939) and Schell (1970), such as the presence of numerous post-ovarian testes (currently consider a single follicular testis) and a short, preovarial uterus, have not been confirmed in *P. stellai*. Du Preez *et al.* (2024) presented a more comprehensive depiction of representatives of *Polystoma*, highlighting additional pertinent characteristics: a pyriform ovary; the uterus containing few operculated eggs; and a follicular, medial, posteriorly located testis. Based on previous findings and the current limited data available for *P. stellai*, it is not definitive to assert that this species unequivocally belongs to *Polystoma*. However, until further morphological and/or molecular evidence becomes accessible, the most cautious approach is to maintain the current classification of the species and regard both Cuban populations as part of the same species.

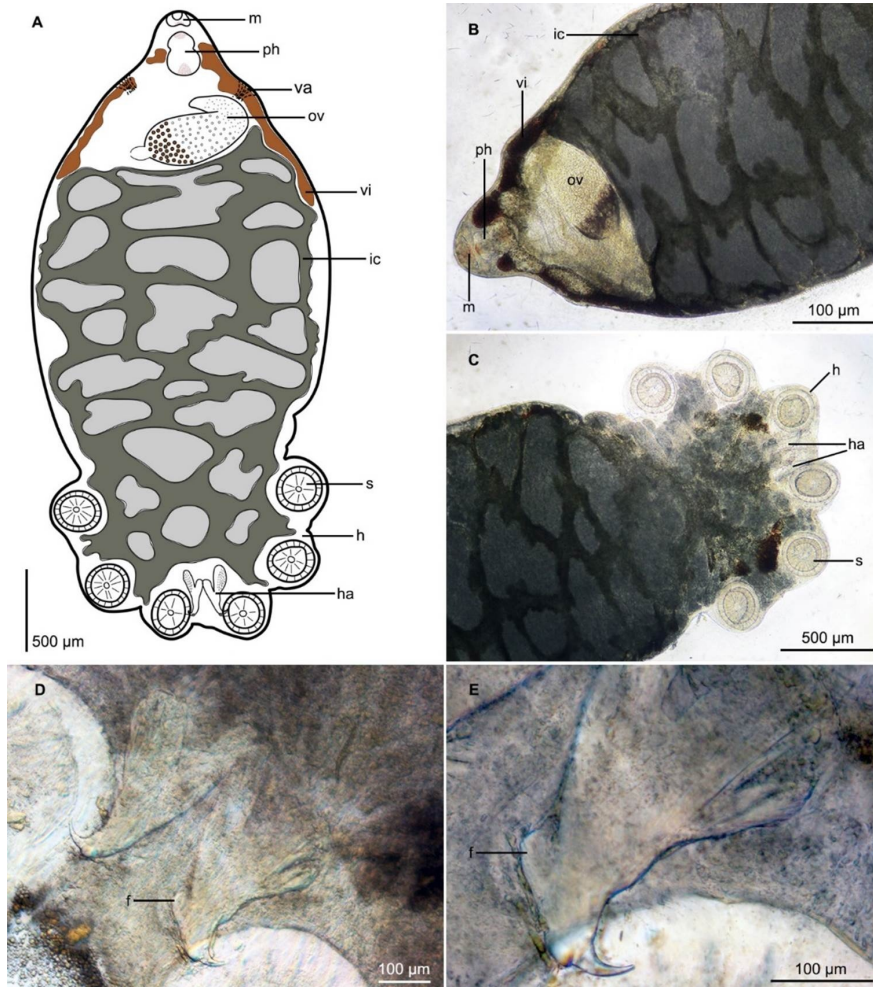


FIGURE 2. Morphology of the specimen of *Polystoma stellai* collected in Santiago de Cuba, Cuba, in 2022. Abbreviations: h, haptor; ha, hamulus; f, flap; ic, intestinal caecum; m, mouth; ov, ovary; ph, pharynx; s, sucker; vi, vitellaria.

FIGURA 2. Morfología del espécimen de *Polystoma stellai* recolectado en Santiago de Cuba en el año 2022. Abreviaturas: h, gancho; ha, hamulus; f, aletas; ic, ciego intestinal; m, boca; ov, ovario; ph, faringe; s, ventosa; vi, vitellaria.

TABLE 1. Morphological comparison among the three know specimens of *Polystoma stellai*. D, diameter; L, length; and W, wide.

TABLA 1. Comparación morfológica entre los tres especímenes conocidos de *Polystoma stellai*. D, diámetro; L, longitud; y W, ancho.

Characteristics	La Habana	Santiago de Cuba	Florida
Body L (mm)	7.10	3.94	4.30
Body W (mm)	2.60	1.88	0.95
Body L/W	2.73	2.10	4.53
Haptor L (µm)	1400	1008	-
Haptor W (µm)	2100	1452	-
Hamulus L (µm)	480	407-435	-
Suckers D (µm)	350-380	257-286	290
Ovary L (µm)	880	344	-
Ovary W (µm)	530	124	-
Cirrus spines L (µm)	21	-	-

In accordance with the International Code of Zoological Nomenclature, we refrain from designating our specimen as a neotype (see Articles 75.3.5 and 75.3.6). Our discoveries contribute to a better understanding of the distribution and morphology of *P. stellai*. However, further investigations are essential to confirm the intraspecific variability of the species or ascertain whether multiple species of *Polystoma* inhabit Cuba.

ACKNOWLEDGEMENTS. We thank the students of the Bachelor in Biology at Universidad de Oriente, Cuba, whom helped collecting the frogs in 2022, especially Ricet Vera. We also thank Prof. Dr Alejandro Barro (Universidad de La Habana, Cuba) for providing the original paper of Pérez Viguera (1955). YLD is supported by a Georg Forster Research Fellowship (Alexander von Humboldt Foundation, Germany, grant number 3.2 - CUB - 1226121 - GF-P).

REFERENCES

- Coy, A. and Lorenzo, N. (1982). Lista de los helmintos parásitos de los vertebrados silvestres cubanos. *Poeyana*, 235, 1-57.
- Díaz, L.M. and Cádiz, A. (2008). Guía taxonómica de los anfibios de Cuba. *Abc Taxa*.
- du Preez, L.H., Verneau, O. and Gross, T.S. (2007). *Polystoma floridana* n. sp. (Monogenea: Polystomatidae) a parasite in the green tree frog, *Hyla cinerea* (Schneider), of North America. *Zootaxa*, 1663, 33-45.
- du Preez, L.H., Landman, W.J. and Verneau O. (2024). *Polystomatid Flatworms. State of Knowledge and Future Trends*. Springer.
- Fernández, R.A., Corrada, R.I. and Armenteros, M. (2015). Variaciones de la infracomunidad parásita durante la ontogenia de *Chromis cyanea* (Perciformes: Pomacentridae) en la costa norte de La Habana, Cuba. *Revista de Biología Tropical*, 63(3), 717-726.
- Mendoza-Franco, E., Vidal-Martínez, V., Cruz-Quintana, Y. and Prats, F. (2006). Monogeneans on native and introduced freshwater fishes from Cuba with the description of a new species of *Salsuginus* Beverley-Burton, 1984 from *Limia vittata* (Poeciliidae). *Systematic Parasitology*, 64, 181-190.
- Pérez Viguera, I. (1955). Contribución al conocimiento de la fauna helmintológica cubana. *Memorias de la Sociedad Cubana de Historia Natural "Felipe Poey"*, 22, 21-71.
- Price, E.W. (1939). North American monogenetic trematodes. IV. The family Polystomatidae (Polystomatoidea). *Proceedings of the Helminthological Society of Washington*, 6(2), 80-92.
- Schell, S.C. (1970). *The Trematodes*. W.M. C. Brown Company Publishers.
- Stunkard, H.W. (1959). Induced gametogenesis in a monogenetic trematode, *Polystoma stellai* Viguera, 1955. *The Journal of Parasitology*, 45(4), 389-394.
- WoRMS (February 8, 2024). *Polystoma* Zeder, 1800. <https://www.marinespecies.org/aphia.php?p=tax-details&id=1287285>

Conflict of interest: The authors declare that there is no conflict of interest.

Author contribution. Conceptualization: Y. L. Diez. **Field samplign:** Y. L. Diez, A. Catalá. **Methodology and formal analysis:** Y. L. Diez, A. Catalá, A. Schmidt-Rhaesa. **The first draft of the manuscript was written by** Y. L. Diez and all authors commented on previous versions of the manuscript. **All authors read and approved the final manuscript.**