

Short note

Relationship of the parasite *Rhadinorhynchus plagioscionis* (Acanthocephala) with *Plagioscion squamosissimus* host and its specificity of occurrence in the Brazilian Amazon

Lincoln Lima CORRÊA¹, Darlison Chagas de SOUZA², Edson Aparecido ADRIANO^{3,4}

¹Instituto de Ciências e Tecnologia das Águas - ICTA, Universidade Federal do Oeste do Pará (UFOPA), Avenida Mendonça Furtado, nº 2946, Fátima, CEP 68040-470, Santarém, PA, Brazil

²Programa de Pós-Graduação em Biodiversidade Tropical da Universidade Federal do Amapá (UNIFAP), Rod. Juscelino Kubitschek, s/n, Jardim Marco Zero, CEP 68903-419 Macapá, AP, Brazil

³Departamento de Ciências Biológicas, Universidade Federal de São Paulo (UNIFESP), Rua Professor Artur Riedel, 275, Jardim Eldorado, CEP 09972-270, Diadema, SP, Brazil

⁴Programa de Pós-Graduação em Biologia Animal da Universidade Estadual de Campinas (UNICAMP). Prédio da PG-IB, Bloco O, CP. 6109, Avenida Bertrand Russel, s/n, 13083-865, Campinas, São Paulo, Brazil

Corresponding Author: Lincoln Lima Corrêa; e-mail: lincorre@gmail.com

ABSTRACT. Objective was the relationship of the parasite *Rhadinorhynchus plagioscionis* with its host *Plagioscion squamosissimus*, providing morphological and morphometric data to prove the specificity of this parasite with its host, with reports occurring only in rivers in the Brazilian Amazon. *Rhadinorhynchus plagioscionis* infected *P. squamosissimus* captured in the municipal region of Santarém, in the state of Pará, in the eastern Brazilian Amazon. The specimens were analyzed by light and scanning electron microscopy. Morphological data obtained confirmed conspecificity with *R. plagioscionis* and supplemental information was added to the morphological concept of this species. The body of *R. plagioscionis* is cylindrical and milky-coloured with dorsal curvature, a long proboscis displaying ventral curvature with 12 longitudinal rows containing 22 to 24 hooks, which are larger and more prominent towards the end of the ventral curvature, and decrease in size in the posterior extremity of the proboscis. The present study describes, in addition to the morphological and morphometric characteristics of *R. plagioscionis*, which have host specificity for *P. squamosissimus*, a relationship being reported exclusively in the Brazilian Amazon.

Keywords: freshwater fish, morphology, Acanthocephala, similarity

Introduction

Plagioscion squamosissimus Heckel, 1840 (Sciaenidae), a fish known as croaker or white hake, is endemic to South America and distributed in the Magdalena, Amazon and Orinoco river basins. This fish was introduced into the São Francisco River, Paraná River and reservoirs in northeastern Brazil [1].

The genus *Rhadinorhynchus* Lühe, 1911 (Rhadinorhynchidae) harbors 47 species of Acanthocephala, which infect a wide range of hosts around

the world [1,2]. Among those, *Rhadinorhynchus plagioscionis* Thatcher, 1980 has been described parasitizing specimens of *P. squamosissimus* obtained in markets in Manaus, in the western Brazilian Amazon [3]. Furthermore, *R. plagioscionis* has been found infecting the same host from the Negro River [4] and from the Amazonas and Tapajós rivers [5]. The host fish, *P. squamosissimus*, is popularly known as “corvina” or “pescada branca” in Portuguese and South American silver croaker in English [6,7].

Despite the description of *R. plagioscionis* 40

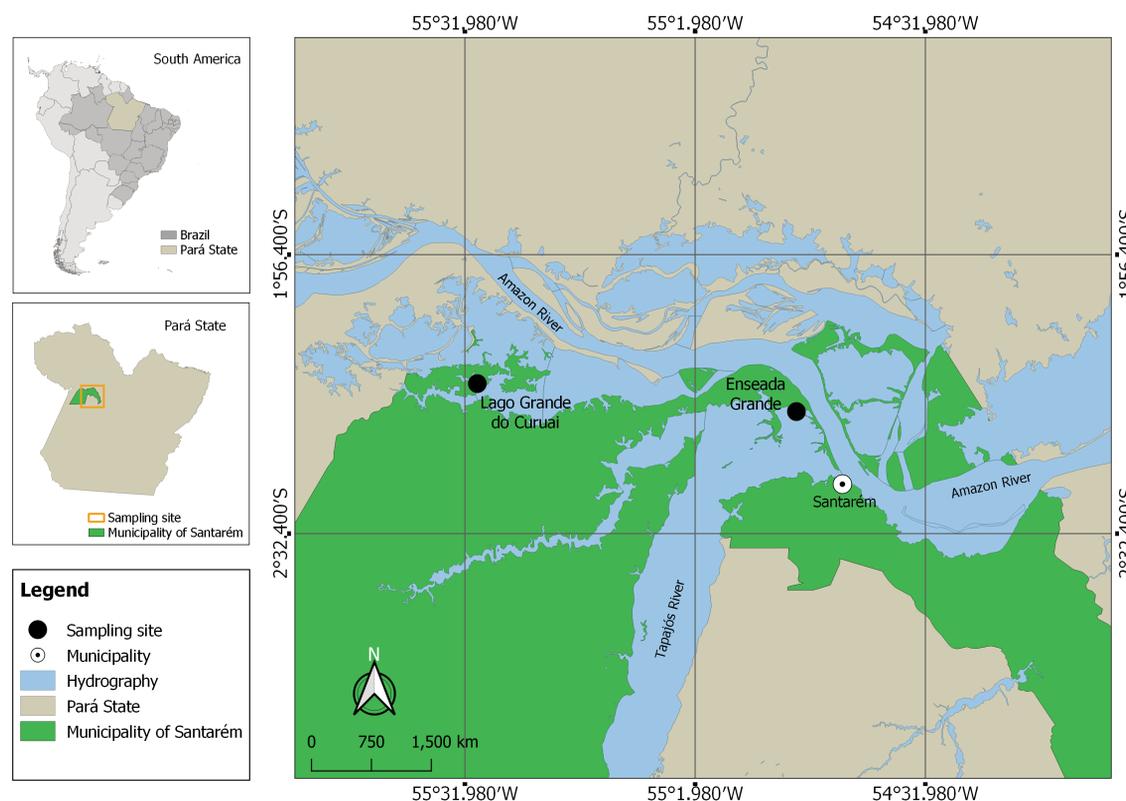


Figure 1. Geographic location of the collection points of *Plagioscion squamosissimus* captured from the region of the Municipality of Santarém, State of Pará, Brazil. Highlights are the collection points (Lake Grande do Curuai on the Amazon river) and (Enseada Grande on the Tapajós River)

years ago, data related to its morphology are still limited. The present study describes, in addition to the morphological and morphometric characteristics of *R. plagioscionis* parasites, which have host specificity for *P. squamosissimus*, a relationship being reported exclusively in fish from Brazilian Amazon.

Materials and Methods

The present study used light and electron scanning microscopy to provide supplementary morphologic information for *R. plagioscionis* (n=75) found infecting *P. squamosissimus* caught in the municipal region of Santarém, in the state of Pará, in August 2017, in the Lago Grande do Curuai (Grande do Curuai Lake) (Amazon river bank) 2°13'1.81"S 55°30'21.74"W and in March 2018 in the mouth of the Tapajós River 2°16'38.57"S 54°48'47.37"W (Fig. 1).

Once caught with fishing nets, the specimens of *P. squamosissimus* were taken alive to the field laboratory on a boat, where they were euthanized by the medullar section method. Their total length (cm) and weight (g) were then measured. Capture and

access to genetic heritage was authorized by the Brazilian Ministry of the Environment (SISBIO # 62384-2 SISGEN # A0A0058), and the methodology of the present study was approved by the Research Ethics Committee of the Universidade Federal do Oeste do Pará (the Federal University of Western Pará, or UFOPA) (Protocol # 0420180027). The parasitological search was carried out *in loco* following the method proposed by Eiras, Takemoto and Pavanelli [8].

The acanthocephalans recovered were fixed in 100% ethanol and 2.5% glutaraldehyde and taken to UFOPA, where they were photographed and measured (μm) using a Zeiss Axioplan optical light microscope equipped with an Axiocam ERc 5s camera and connected to a computer with the Blue Zen edition 2 software package, according to Amin and Heckmann [9]. The following measurements were taken: total length of male; total length of female; length of proboscis; length of proboscis receptacle; length and width of the anterior and posterior testis. The illustrations were made with the aid of a drawing tube attached to a microscope (Leica DM 2500) with differential interference contrast and phase contrast optics. Subsequently, the

Table 1. Morphometric measurements of representatives of the genus *Rhadinorhynchus*

Measures (mm)	<i>R. polynemus</i>	<i>R. seriolae</i>	<i>R. bififormis</i>	<i>R. pichelinae</i>	<i>R. polydactyli</i>	<i>R. plagioscionis</i>	<i>R. plagioscionis*</i>
ML min	4.5	4.5	4.0	2.0	–	8.8	8.6
ML max	9.0	9.0	0	12.0	0.5	11.0	10.0
FL min	15.0	15.0	11.0	11.0	14.0	14.5	14.2
FL max	17.0	30.0	15.0	18.0	35.0	16.4	15.9
PL min	0.8	1.1	0.8	0.1	–	1.5	1.5
PL max	1.3	2.0	0.9	1.3	1.3	1.1	1.0
PRL min	1.7	1.2	1.2	0.3	2.4	–	–
PRL max	3.4	2.7	1.7	4.5	2.8	–	–
TAL min	0.3	0.5	–	0.6	0.6	–	–
TAL max	0.6	1.2	0.2	1.0	1.2	0.4	0.4
TAW min	0.1	0.2	–	0.2	0.2	–	–
TAW max	0.4	0.3	0.2	0.4	0.3	0.9	0.8
TPL min	0.3	0.5	–	0.6	0.5	–	–
TPL max	0.68	1.5	0.2	1.0	1.1	0.3	0.2
TPW min	0.1	0.2	–	0.8	0.3	–	–
TPW max	0.4	0.3	0.1	1.8	0.2	0.6	0.6

* Present study; MA: Male length; FL: Female length; PL: Proboscis length; TAL: Anterior testis length; TAW: Anterior testis width; TPL: Posterior testis length; TPW: Posterior testis width

illustrations of the body and appendages of the parasites were prepared in CorelDRAW 2019 software and treated using Adobe Photoshop CS6 software.

For scanning electron microscopy (SEM), specimens previously fixed in 2.5% glutaraldehyde with 0.1 M cacodylate buffer (pH 7.4), washed in the same buffer and post-fixed with osmium tetroxide, were used. All procedures were performed at 4°C. Two specimens (n=2) being one male and one female were subsequently washed and dehydrated in an ascending ethanol series, and dried in hexamethyldisilazane (HMDS) (5 min.) [10]. Finally, the samples were covered with metallic gold and examined under a Leo Stereoscan S-440 microscope at the Universidade Federal de São Paulo (UNIFESP).

The ecological terms as prevalence, mean intensity and mean abundance were used according to [11]. These parameters were calculated using Quantitative Parasitology 3.0 software [12]. The morphometric measurements of the parasites were compared with six species of the genus

Rhadinorhynchus described by Thatcher and Smales [2–4] for the morphometric comparison of similarity between species. We also use the Bay-Curtis dissimilarity index from the PAST Software version 4.0.3

Results and Discussion

Of the 75 *P. squamosissimus* specimens (Fig. 2) caught from the Amazon/Tapajos rivers, only two specimens from the Tapajos River (mean abundance 2.6%) were parasitized by acanthocephalans. Morphological and morphometric characterization using light microscopy was based on eight specimens (n=8) four males and four females, and their characters were compatible with *Rhadinorhynchus*.

Taxonomic summary

Rhadinorhynchidae Travassos, 1923

Rhadinorhynchinae Lühe, 1911

Rhadinorhynchus Lühe 1911

Rhadinorhynchus plagioscionis (Thatcher, 1980) (Tab. 1)



Figure 2. Fish of *Plagioscion squamosissimus* species collected in the Brazilian Amazon, in the municipal region of Santarém, State of Pará, Brazil

Host: *Plagioscion squamosissimus* (Heckel, 1840)
 Type locality: Manaus, state of Amazonas, Brazil
 (Thatcher, 1980)
 Additional locality: Tapajós River (2°16'38.57"S

54°48'47.37"W), municipal region of Santarém,
 state of Pará, Brazil

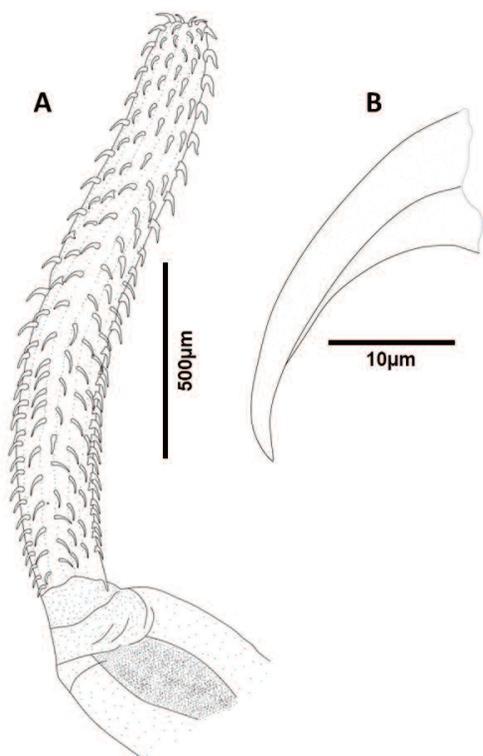


Figure 3. Proboscis (A) and dorsal hook of the proboscis (B) of *Rhadinorhynchus plagioscionis*

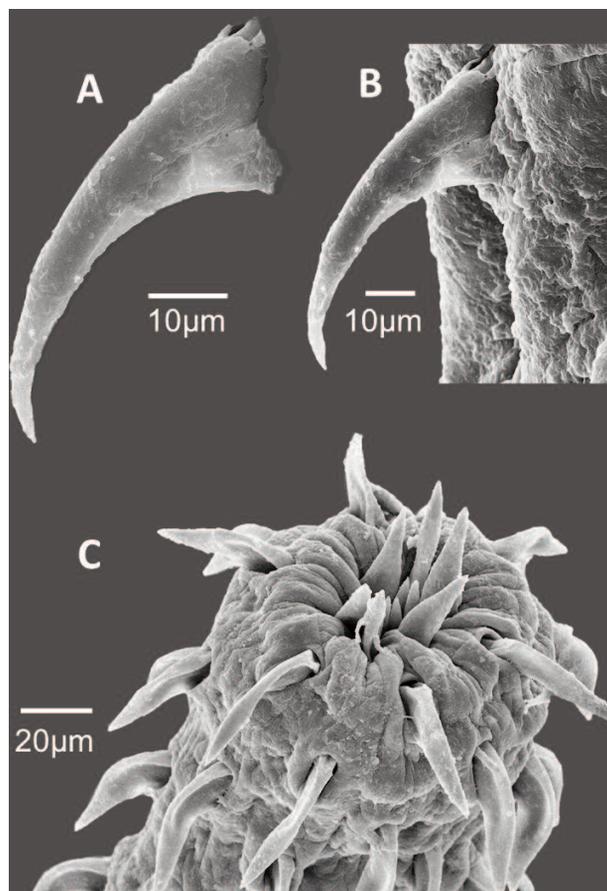


Figure 4. Hook from the median region of the proboscis (A and B) detail of distal region of the proboscis (C) of *Rhadinorhynchus plagioscionis*

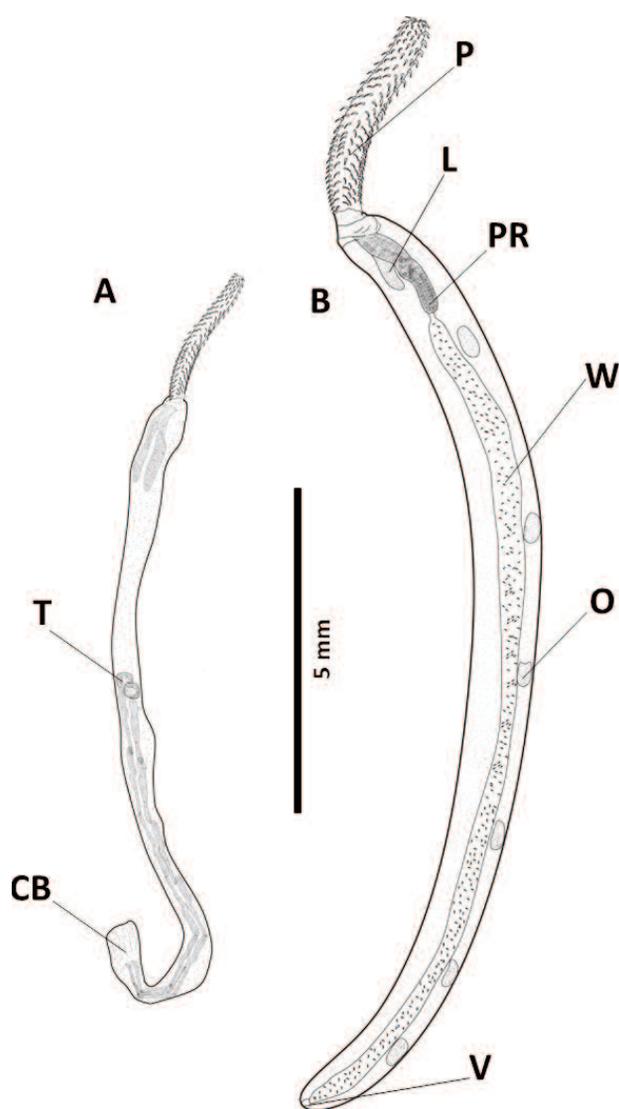


Figure 5. *Rhadinorhynchus plagioscionis*. Male (A). Female (B). P: Proboscis; L: Lemnisci; PR: Proboscis receptacle; T: Testicles, CB: Copulatory pouch, W: Uterus; O: Ovary and V: Vagina

Bilky-coloured cylindrical body with dorsal curvature, long proboscis exhibiting ventral curvature with 12 longitudinal rows containing 22 to 24 hooks, larger and more prominent in the ventral curvature, and decreasing in size at the posterior end of the proboscis (Fig. 3 and 4). Females larger than males, with a long uterus extending along the posterior region ending in the vagina. Easily observed ovaries. Male with globular testicles and highly evident copulatory pouch (Fig. 5). These characteristics are consistent with those of the original descriptions [11]. However, in this study, the use of scanning electron microscopy allowed the presence of a longitudinal fissure in the

median region of the larger ventral hooks to be added to Thatcher's original description. The apical structures of the proboscis were also examined, allowing the identification of six pairs of lanceolate structures which varied in size and shape, with two pairs of underdeveloped structures visible in the central region ($n=4$), with an average length of $9.8 \mu\text{m}$ (min: 95.0 , max: $10.18 \mu\text{m}$) and an average width of $3.7 \mu\text{m}$ (min: 3.5 , max: $3.9 \mu\text{m}$), bordered by four more developed pairs ($n=8$), with an average length of $72.3 \mu\text{m}$ (min: 5.47 , max: $9.06 \mu\text{m}$) and an average width of $24.8 \mu\text{m}$ (min: 12.63 , max 32.68) (Fig. 3A and B), a characteristic present in both male and female specimens.

The Bray-Curtis index showed 93.38% similarity of *R. plagioscionis* with data described for this species by Thatcher [4], showing greater specificity of morphological characterizations for this study (Tab. 2 and Fig. 6).

This study is not an attempt to revise the description of *R. plagioscionis*, which was well described by Thatcher [4] not least because this author provided a much more detailed description and measured many more resources. So much so that, throughout the study, we confirmed that the parasites analyzed and studied here are conspecific with those of Thatcher [4].

Despite *P. squamosissimus* being introduced into other South American watersheds, such as the San Francisco and Paraná basins [7], there is no report of *R. plagioscionis* in these basins, which may suggest that this parasite was not introduced along together with this host, or did not find its invertebrate hosts necessary to complete its biologic cycle in this new environment.

The prevalence of *R. plagioscionis* in *P. squamosissimus* was low, with abundance only in hosts from one of the localities (Enseada Grande). *Plagioscion squamosissimus* is a carnivorous fish species that feeds on crustaceans, molluscs and small fish [6,7,12]. It can therefore be assumed that the low levels of *R. plagioscionis* are most probably related to the availability and diversity of seasonal infective forms in the environment studied. Furthermore, *R. plagioscionis* displays host specificity for *P. squamosissimus* [6] and its low levels may also be related to the biology of this intermediate host and the diet of the definitive host, factors that may influence infection levels [15]. Furthermore, as the collections were made in specific periods, one in the dry season and the other in the rainy season, it is possible that the occurrence

Table 2. Morphometric similarity contingency values of *Rhadinorhynchus plagioscionis* infecting *Plagioscion squamosissimus* in Santarém, western Pará State, Brazil, and *Rhadinorhynchus* species already described

	<i>R. polynemus</i>	<i>R. seriolae</i>	<i>R. biformis</i>	<i>R. pichelinae</i>	<i>R. polydactyli</i>	<i>R. plagioscionis</i>	<i>R. plagioscionis</i> *
<i>R. polynemus</i>	1						
<i>R. seriolae</i>	80.00						
<i>R. biformis</i>	69.71	63.88	1				
<i>R. pichelinae</i>	74.40	74.63	48.98	1			
<i>R. polydactyli</i>	80.74	83.20	56.94	70.93	1		
<i>R. plagioscionis</i>	47.46	43.81	50.05	43.16	31.27	1	
<i>R. plagioscionis</i> *	44.85	41.57	50.54	40.24	28.90	93.38	1

* Present study

of *R. plagioscionis* in *P. squamosissimus* in the Amazon region is seasonal. Further studies are therefore needed on the seasonal dynamics of *R. plagioscionis* and other factors that may be involved in the occurrence of this acanthocephalan in *P. squamosissimus*.

Acknowledgements

The author Lincoln L. Corrêa would like to express his gratitude for the guaranteed and financial support granted by the CAPES/FAPESPA project N. 06/2015 – Process n° 88881.160660 /2017-01. Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) for the studies scholarship granted to Darlison C. de Souza. Edson A. Adriano

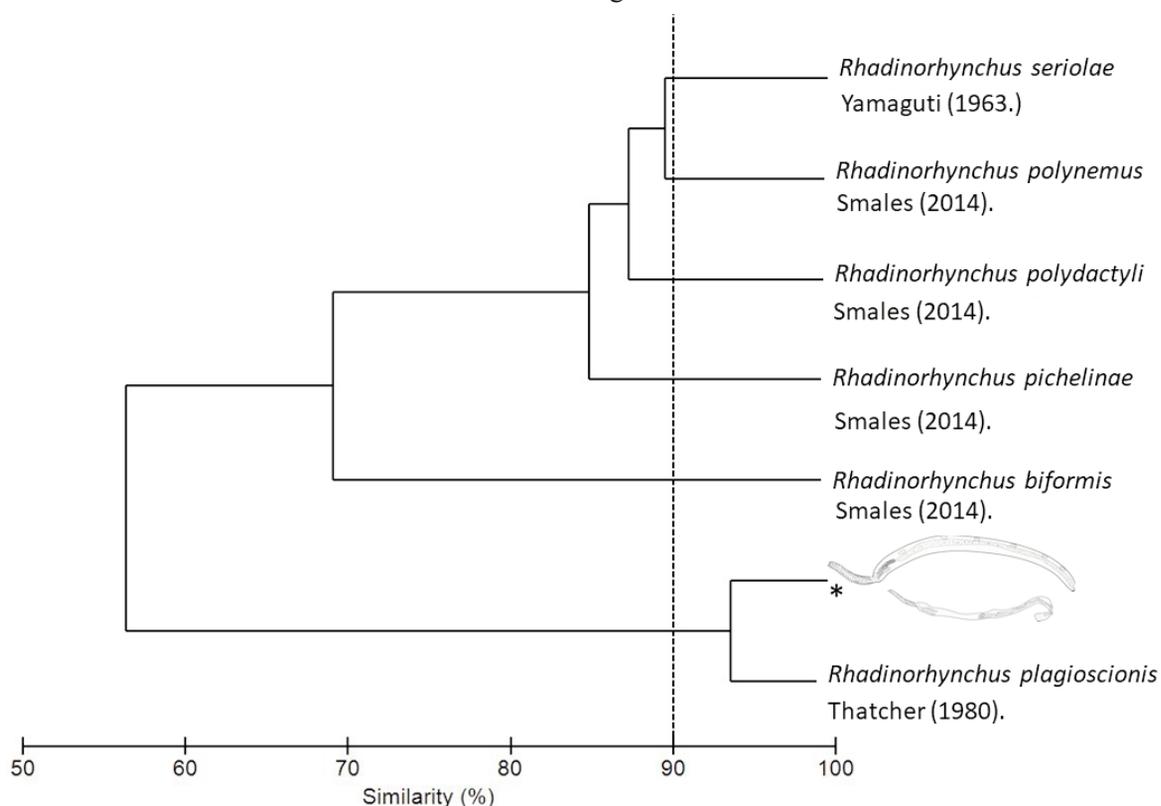


Figure 6. Dendrogram of morphometric similarity by the Bray-Curtis method for *Rhadinorhynchus plagioscionis* infecting *Plagioscion squamosissimus* in Santarém, western Pará State, Brazil, and *Rhadinorhynchus* species already described

received a research productivity grant from the Conselho Nacional de Desenvolvimento Científico e Tecnológico CNPq (304687/2020-0).

References

- [1] Capra L.G., Bennemann S.T. 2009. Low feeding overlap between *Plagioscion squamosissimus* fishes introduced in tropical reservoir of South Brazil. *Acta Limnologica Brasiliensia* 21(3): 343–348.
- [2] Smales L.R. 2014. The genus *Rhadinorhynchus* (Acanthocephala: Rhadinorhynchidae) from marine fish in Australia with the description of four new species. *Acta Parasitologica* 59(4): 721–736. doi:10.2478/s11686-014-0305-4
- [3] Amin O.M., Rubtsova N.Y., Ha N.V. 2019. Description of three new species of *Rhadinorhynchus* Lühe, 1911 (Acanthocephala: Rhadinorhynchidae) from marine fish off the Pacific Coast of Vietnam. *Acta Parasitologica* 64(3): 528–543. doi:10.2478/s11686-019-00092-2
- [4] Thatcher V.E. 1980. *Rhadinorhynchus plagioscionis* n. sp. (Acanthocephala: Rhadinorhynchidae) da pescada (*Plagioscion Squamosissimus*) da Amazônia brasileira. *Acta Amazonica* 10(4): 835–839 ((in Portuguese with summary in English). doi:10.1590/1809-43921980104835
- [5] Lacerda A.C.F., Takemoto R.M., Tavares-Dias M., Poulin R., Pavanelli G.C. 2012. Comparative parasitism of the fish *Plagioscion squamosissimus* in native and invaded river basins. *Journal of Parasitology* 98(4): 713–717. doi:10.1645/GE-2882.1
- [6] Souza D.C., Eiras J.C., Adriano E.A., Corrêa L.L. 2020. Metazoan parasites of *Plagioscion squamosissimus* (Osteichthyes: Sciaenidae) of two rivers from the eastern Amazon (Brazil). *Annals of Parasitology* 66(2): 217–225. doi:10.17420/ap6602.257
- [7] Bennemann S.T., Capra L.G., Galves W., Shibatta O.A. 2006. Dinâmica trófica de *Plagioscion squamosissimus* (Perciformes, Sciaenidae) em trechos de influência da represa Capivara (rios Paranapanema e Tibagi) [Trophic dynamic of *Plagioscion squamosissimus* (Perciformes, Sciaenidae) in stretches under influence of the Capivara dam (Paranapanema and Tibagi rivers)]. *Iheringia. Série Zoologia* 96(1): 115–119 (in Portuguese with summary in English). doi:10.1590/S0073-47212006000100020
- [8] Eiras J.C., Takemoto R.M., Pavanelli G.C. 2006. Métodos de estudo e técnicas laboratoriais em parasitologia de peixes. 2nd ed. Maringá: Eduem (in Portuguese).
- [9] Amin O.M., Heckmann R.A. 2014. First description of *Pseudoacanthocephalus lutzi* from Peru using SEM. *Scientia Parasitologica* 15(1–4): 19–26.
- [10] Bray D.F., Bagu J., Koegler P. 1993. Comparison of hexamethyldisilazane (HMDS), Peldri II, and critical-point drying methods for scanning electron microscopy of biological specimens. *Microscopy Research and Technique* 26(6): 489–495. doi:10.1002/jemt.1070260603
- [11] Bush A.O., Lafferty K.D., Lotz J.M., Shostak A.W. 1997. Parasitology meets ecology on its own terms: Margolis et al. revisited. *The Journal of Parasitology* 83(4): 575–583. doi:10.2307/3284227
- [12] Reiczigel J., Marozzi M., Fábíán I., Rózsa L. 2019. Biostatistics for parasitologists – a primer to quantitative parasitology. *Trends in Parasitology* 35(4): 277–281. doi:10.1016/j.pt.2019.01.003
- [13] Stefani P., Rocha O. 2009. Diet composition of *Plagioscion squamosissimus* (Heckel, 1840), a fish introduced into the Tietê River system. *Brazilian Journal of Biology* 69(3): 805–812. doi:10.1590/S1519-69842009000400007
- [14] Ferreira Filho V.P., Guerra T.P., Lima M.C.S., Teixeira D.F., Costa R.R., Araújo I., Moura G.J. 2014. Padrões ecomorfológicos associados à dieta de *Plagioscion squamosissimus* (Perciformes, Scianidae) em reservatório permanente, no Nordeste do Brasil [Ecomorphological patterns with diet of *Plagioscion squamosissimus* (Perciformes, Scianidae) in permanent reservoir in northeastern Brazil]. *Iheringia. Série Zoologia* 104(2): 134–142 (in Portuguese with summary in English). doi:10.1590/1678-476620141042134142
- [15] Lourenço F.D.S., Morey G.A.M., Malta J.C.D.O. 2018. The development of *Neoechinorhynchus buttnerae* (Eoacanthocephala: Neoechinorhynchidae) in its intermediate host *Cypridopsis vidua* in Brazil. *Acta Parasitologica* 63(2): 354–359. doi:10.1515/ap-2018-0040

Received 21 August 2021

Accepted 03 October 2021