Association between *Ichthyocladius* (Diptera: Chironomidae) and armored catfishes in the Guareí River basin, São Paulo State, Brazil

VALTER M. AZEVEDO-SANTOS, ERIKA M. SHIMABUKURO, FERNANDO M. PELICICE & RAOUl HENRY

Abstract: *Ichthyocladius* is a genus of chironomid (Diptera, Insecta) whose immature forms live attached to the bodies of some species of freshwater fishes. Here we investigate the association between *Ichthyocladius* spp. and armored catfish in streams of the Guareí River basin, Paraná River system, Brazil. We provide the first record of *I. lilianae* associated with fish in the São Paulo State and the Paraná River basin. In addition, this is the first report of *Ichthyocladius* associated with the armored catfishes *Hypostomus iheringii* and *H. tietensis*. Lastly, we record two undescribed species of Chironomidae (i.e., *Ichthyocladius* sp. 1 and *Ichthyocladius* sp. 2) associated with fishes.

Key words: fish fins, insects, larvae, Loricariidae, pupae, Upper Paraná River basin.

INTRODUCTION

Different organisms can associate with fishes, either as parasites, commensals or mutualists. Freshwater fishes are hosts to many forms of life, including protozoa (e.g., Eiras et al. 2012, Martins et al. 2012), immature insects (e.g., Roque et al. 2004, Sydow et al. 2008), annelids (e.g., Ranzani-Paiva & Silva-Souza 2004), crustaceans (e.g., Neves & Tavares-Dias 2019) and other fishes (e.g., Zuanon & Sazima 2004, 2005). Among the insects, immature stages (i.e., larvae and pupae) of some Chironomidae (Diptera) may live attached to the bodies of aquatic animals (Tokeshi 1993, Roque et al. 2004, Sydow et al. 2008, Henriques-Oliveira & Nessimian 2009). Larva and pupae of the genus *Ichthyocladius* Fittkau, 1974, in particular, have been reported to live attached to fishes (e.g., Mendes et al. 2004, Roque et al. 2004).

Currently, *Ichthyocladius* is represented by three species, *Ichthyocladius neotropicus* Fittkau, 1974, the type species of the genus, *I. kronichticola* Mendes, Andersen & Sæther, 2004, and *I. lilianae* Mendes, Andersen & Sæther, 2004 (Mendes et al. 2004), as well as several additional species awaiting formal description. Individuals of *I. kronichticola* and *I. lilianae* have been reported to live in association with armored catfish (Mendes et al. 2004, Roque et al. 2004). *Ichthyocladius lilianae* has been recorded in waterbodies of three Brazilian States: Rio Grande do Sul (Sydow et al. 2008, Dala-Corte & Melo 2018), Minas Gerais (Mendes et al. 2004), and Rio de Janeiro (Henriques-Oliveira & Nessimian 2009).

In a recent study, Azevedo-Santos et al. (2020) investigated the fish fauna of the Guareí River basin (Upper Paraná River basin), São Paulo, Brazil. When examining the material collected, we noted the presence of species of *Ichthyocladius* living on armored catfishes. In this short paper, we investigate the association between these insects with fishes. In particular, we report for the first time the association of *I. lilianae* and two other undescribed species of Chironomidae with
armored catfish (Loricariidae) in the Paraná River basin, São Paulo State, Brazil.

MATERIALS AND METHODS

Fish specimens were collected in the Guareí River basin, located in the upper reaches of the Paranapanema River (Paraná River system), São Paulo, Brazil. Armored catfishes were sampled in two tributaries of the right bank of the Guareí River during June and August 2017. Fish sampling is described in Azevedo-Santos et al. (2020). Sampling sites were designated as R1 (-23.434932°S -48.388695°W) and R2 (-23.460077°S -48.414122°W) (Figure 1).

We searched for chironomids on the exterior surfaces of captured loricariids. All larvae and prepupae were removed and mounted on slides with Euparal®. Chironomidae species were identified according to Fittkau (1974) and Mendes et al. (2004) and by comparison with type specimens deposited at the Museu de Zoologia da Universidade de São Paulo (hereafter MZ), São Paulo, Brazil. Immature individuals (at least one for each species of Ichthyocladius) were deposited as vouchers at the MZ. Standard Length (SL) of all hosts was measured with a digital caliper (in millimeters - mm). Classification of the catfishes (see Table SI) followed Fricke et al. (2019a, b).

RESULTS

We examined nine armored catfishes, of which six served as hosts for Ichthyocladius. We recorded 13 larvae and one prepupa of the genus Ichthyocladius, including I. lilianae and two taxa identified at the morphospecies level (Ichthyocladius sp.1 and Ichthyocladius sp. 2). This is the first record of I. lilianae for the Paraná River basin and for the State of São Paulo. The catfish

Figure 1. Sampling reaches (R1 and R2) in the Guareí River system - map modified from Azevedo-Santos et al. (2020).
hosts were individuals of *Hypostomus iheringii* (Regan, 1908) and *H. tietensis* (Ihering, 1905) (28.6 - 95.2 mm of SL). We found larvae attached to different regions of the body — including the eyes and fins (Table I; and examples in Figure 2a-d). Virtually all fish with associated chironomids had *Ichthyocladius* in the adipose fin region.

**Table I.** Chironomids associated with armored catfishes from the Guareí River basin, São Paulo, Brazil. Classifications and voucher numbers can be found in Table SI.

| Reach | Hosts | Associated species (local in the fish) |
|-------|-------|---------------------------------------|
| R1    | One *Hypostomus iheringii* (Regan, 1908) | Three larvae of *Ichthyocladius lilianae* (right eye; left eye; lateral of the adipose fin) |
| R1    | One *Hypostomus tietensis* (Ihering, 1905) | One larva of *Ichthyocladius* sp.1 (right eye); and one larva of *I. lilianae* (lateral of the adipose fin) |
| R2    | One *Hypostomus iheringii* (Regan, 1908) | One larva of *I. lilianae* (lateral of the adipose fin) |
| R2    | One *Hypostomus iheringii* (Regan, 1908) | Two larvae of *I. lilianae* (pectoral fin; lateral of the adipose fin) |
| R2    | One *Hypostomus iheringii* (Regan, 1908) | One larva of *I. lilianae* (lateral of adipose fin) |
| R2    | One *Hypostomus iheringi* (Regan, 1908) | Two larvae of *Ichthyocladius* sp. 2 (both on the lateral of the adipose fin); two larvae of *I. lilianae* (near to the adipose fin and other between the adipose fin and caudal fin); and one prepupa of *I. lilianae* (near to the adipose fin basin). |

**DISCUSSION**

*Ichthyocladius lilianae* was described by Mendes et al. (2004) based on specimens collected in the São Francisco basin, Minas Gerais State. These authors reported its association with two loricariid fishes, *Harttia* sp. and *Hypostomus* cf. *H. iheringii* and *H. tietensis*. The larvae associated with *Hypostomus* species were mainly attached to the adipose fin region, but some were also found on the eyes and fins. The presence of *Ichthyocladius* in these armored catfishes suggests a parasitic relationship, as they are known to form galls on the host’s adipose fin. Further studies are needed to understand the ecological significance of these associations and the role of chironomids in the ecosystem of the Guareí River basin.
garmani (Regan, 1904). This species of chironomid has since been recorded in four Brazilian states and in different hydrographic basins, including São Francisco and Paraíba do Sul (Sydow et al. 2008, Mendes et al. 2004, Dala-Corte & Melo 2018, Henriques-Oliveira & Nessimian 2009; see details in Table II). Our work expanded its recorded range to include São Paulo State and the Paraná River basin.

We recorded two undescribed Chironomidae species (e.g., *Ichthyocladius* sp. 1 and *Ichthyocladius* sp. 2) associated with catfishes in the Guareí River system. *Ichthyocladius* sp. 1 is the only one without anal seta among the known species. The procercus is also absent in this species. *Ichthyocladius* sp. 2 differs from other species of *Ichthyocladius* by presenting the mentum with more than 24 median teeth and the premandible with pale teeth. In addition to these two, three additional species of the genus await formal description, being one from Ecuador (Fittkau 1974), other from Maraú River, and the last from Argentina (Mendes et al. 2004). Therefore, the diversity of Chironomidae associated with catfishes is certainly underestimated.

Only two previous studies have reported an association between *Ichthyocladius* and *Hypostomus* Lacepède, 1803. Mendes et al. (2004) mentioned the presence of *Ichthyocladius* on *H. cf. garmani* while Roque et al. (2004) reported the interaction with *Hypostomus* species. Our work is the first to report *Ichthyocladius* on *H. iheringii* and *H. tietensis*. All hosts had at least one chironomid in the region close to the adipose fin. In the studied streams, the hosts were captured in rocky streambeds with moderate water flow, or with some turbulence. It is likely that the adipose fin, because of its particular position, is less disturbed by water turbulence, providing greater opportunities for adherence by *Ichthyocladius*. Individuals were also found in the eye, sometimes within the orbit (e.g., Figure 2b), possibly because it provides a more stable environment.

We finalize by emphasizing the need for more studies on the taxonomy, biogeography and ecology of *Ichthyocladius* — including field

| Host (fish) | Hydrographic basins | Brazilian State | Reference |
|------------|---------------------|-----------------|-----------|
| *Ancistrus* cf. *multispinis* (Regan, 1912) | Maquinê River | Rio Grande do Sul | Sydow et al. (2008) |
| *Harttia* sp. | São Francisco River | Minas Gerais | Mendes et al. (2004) |
| *Hypostomus* cf. *garmani* (Regan, 1904) | São Francisco River | Minas Gerais | Mendes et al. (2004) |
| *Hypostomus* *iheringii* (Regan, 1908) | Guareí River | São Paulo | This work |
| *Hypostomus* *tietensis* (Ihering, 1905) | Guareí River | São Paulo | This work |
| *Pareiorhaphis* *hypselurus* (Pereira & Reis, 2002) | Maquinê River | Rio Grande do Sul | Sydow et al. (2008), Dala-Corte & Melo (2018) |
| *Pareiorhaphis* *nudula* (Reis & Pereira, 1999) | Maquinê River | Rio Grande do Sul | Sydow et al. (2008) |
| *Pareiorhina* *rudolphi* (Miranda Ribeiro, 1911) | Preto River | Rio de Janeiro | Henriques-Oliveira & Nessimian (2009) |
| *Trichomycterus mirissumba* Costa, 1992 | Preto River | Rio de Janeiro | Henriques-Oliveira & Nessimian (2009) |
surveys and laboratory investigations. These organisms and their ecological interactions are poorly understood, and their possible effects on fish ecology and behavior remain largely unknown.

Acknowledgments

We are grateful to Hamilton A. Rodrigues (UNESP), who helped with the fieldwork. Three reviewers provided important corrections and suggestions. We wish thanks to Claudio G. Froehlich and James A. Nienow for corrections on this manuscript. Authors were supported by: Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (scholarship to VMAS - FC-001); Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP - fellowship to EMS - process number 2018/11068-9), and Conselho Nacional de Desenvolvimento Científico e Tecnológico (grants to FMP).

REFERENCES

AZEVEDO-SANTOS VM, PELICICE FM & HENRY R. 2020. Knowing biodiversity: Fishes from the Guareí River basin, a tributary of the Jurumirim reservoir, Paranapanema River, Brazil. Biota Neotrop 20(3): e20201031. https://doi.org/10.1590/1676-0611-bn-2020-1031.

DALA-CORTE RB & MELO AS. 2018. Living on a catfish: nested occupation of ectosymbiont chironomids on host body. Can J Zool 96(7): 692-699. https://doi.org/10.1139/cjz-2017-0141.

EIRAS JC, TAKEMOTO RM, PAVANELLI GC & LUQUE JL. 2012. Checklist of Protozoan parasites of fishes from Brazil. Zootaxa 3221: 1-25. http://dx.doi.org/10.11646/zootaxa.3221.11.

FITTKAU EJ. 1974. Ichthyocladius n. gen., eine neotropische Gattung der Orthocladiinae (Chironomidae, Diptera) deren Larven epizoisch auf Welsen (Astroblepidae und Loricariidae) leben. Nordisk Hygienisk Tidskrift Supplementum 95: 91-106.

FRICKE R, ESCHMEYER WN & FONG JD 2019b. Species by Family/Subfamily. (http://researcharchive.calacademy.org/research/ichthyology/catalog/fishcatmain.asp). Electronic version accessed 08/01/2019.

HENRIQUES-OLIVEIRA AL & NESSIMIAN JL. 2009. Phoresy and commensalism of Chironomidae larvae (Insecta: Diptera) in the state of Rio de Janeiro, Brazil. Lundiana 10(1): 11-18.

MARTINS ML, MARCHIORI N, ROUMBEAKIS K & LAMI F. 2012. Trichodina nobilis Chen, 1963 and Trichodina reticulata Hirschmann et Partsch, 1955 from ornamental freshwater fishes in Brazil. Braz J Biol 72: 281-286. http://dx.doi.org/10.1590/S1519-69842012000200008.

MENDES HF, ANDERSEN T & SÆTHER OA. 2004. New species of Ichthyocladius Fittkau, a member of the Corynoneura-Group (Diptera: Chironomidae: Orthocladiinae), with a review of the genus. Stud Neotrop Fauna E 39: 15-35. https://doi.org/10.1080/01650520412331270936.

NEVES LR & TAVARES-DIAS M. 2019. Low levels of crustacean parasite infestation in fish species from the Matapi River in the state of Amapá, Brazil. Rev Bras Parasitol Vet 28: 493-498.

RANZANI-PAIVA MJT & SILVA-SOUZA AT. 2004. Co-infection of gills by different parasite groups in the mullet, Mugil platanus Günther, 1880 (Osteichthyes, Mugilidae): effects on relative condition factor. Braz J Biol 64: 677-682. http://dx.doi.org/10.1590/S1519-69842004000400016.

ROQUE FO, TRIVINHO-STRIXINO S, JANCSO M & FRAGOSO EN. 2004. Records of Chironomidae larvae living on other aquatic animals in Brazil. Biota Neotrop 2: 1-9. http://dx.doi.org/10.11646/zootaxa.3221.11.

SYDOW VG, VILELLA FS, HARTZ SM & RODRIGUES GG. 2008. Ichthyocladius (Diptera, Chironomidae) on loricariid fishes in Atlantic Forest streams: influence of host size and corporal region on larval attachment. Acta Limnol Bras 20: 333-337.

TOKESHI M. 1993. On the evolution of commensalism in the Chironomidae. Freshw Biol 29(3): 481-489.

ZUANON J & SAZIMA I. 2004. Vampire catfishes seek the aorta not the jugular: candirus of the genus Vandellia (Trichomycteridae) feed on major gill arteries of host fishes. Aqua 8: 31-36.

ZUANON J & SAZIMA I. 2005. Free meals on long-distance cruisers: the vampire fish rides giant catfishes in the Amazon. Biota Neotrop 5: 109-114.
SUPPLEMENTARY MATERIAL

Table SI. Classification of chironomids and armored catfishes (*sensu* Fricke et al. 2019a, b) from the Guareí River Basin, São Paulo, Brazil.

How to cite
AZEVEDO-SANTOS VM, SHIMABUKURO EM, PELICICE FM & HENRY R. 2022. Association between *Ichthyocladius* (Diptera: Chironomidae) and armored catfishes in the Guareí River basin, São Paulo State, Brazil. An Acad Bras Cienc 94: e20201829. DOI 10.1590/0001-3765202220201829.

Manuscript received on November 23, 2020; accepted for publication on March 8, 2021

VALTER M. AZEVEDO-SANTOS1 https://orcid.org/0000-0001-8986-6406

ERIKA M. SHIMABUKURO2 https://orcid.org/0000-0001-8377-1844

FERNANDO M. PELICICE3 https://orcid.org/0000-0002-9700-1194

RAOUL HENRY4 https://orcid.org/0000-0002-4000-2524

1Universidade Estadual Paulista “Júlio de Mesquita Filho”, Departamento de Zoologia, Rua Prof. Dr. Antonio Celso Wagner Zanin, 250, Distrito de Rubião Junior, 18618-970 Botucatu, SP, Brazil
2Universidade de São Paulo, Museu de Zoologia, Avenida Nazaré, 481, Ipiranga, 04263-000 São Paulo, SP, Brazil
3Universidade Federal do Tocantins, Núcleo de Estudos Ambientais, Rua 03, s/n, Jardim dos Ipês, 77500-000 Porto Nacional, TO, Brazil

Correspondence to: Valter M. Azevedo-Santos
E-mail: valtereologia@gmail.com

Author contributions
VMAS collected and identified fish species, conceptualized the manuscript, wrote the first draft, and revised the new versions. EMS identified insect species and collaborated with ideas and writing. FMP collaborated with ideas and writing. RH collaborated with ideas and writing.

[CC BY]