



## A new black fly species from Brazil, closely related to *Simulium guianense* Wise (Diptera, Simuliidae), revealed by morphology and DNA barcoding

NEUSA HAMADA<sup>1</sup>, MATEUS PEPINELLI<sup>2</sup>, ALINE MATTOS-GLÓRIA<sup>1,3</sup>  
& SERGIO LUIZ BESSA LUZ<sup>4</sup>

<sup>1</sup>Coordenação de Pesquisas em Entomologia, Caixa Postal 478, 69011-970, Manaus, AM, Brazil. E-mail: nhamada@inpa.gov.br

<sup>2</sup>Laboratório de Entomologia Aquática, Departamento de Hidrobiologia, Universidade Federal de São Carlos, Caixa Postal 676, 13565-905, São Carlos, SP, Brazil. E-mail: mateuspepi@yahoo.com.br

<sup>3</sup>Divisão de Curso em Genética, Conservação e Biologia Evolutiva, Instituto Nacional de Pesquisas da Amazônia, Caixa Postal 478, 69011-970, Manaus, AM, Brazil. E-mail: mattosaline@hotmail.com

<sup>4</sup>Centro de Pesquisa Leônidas e Maria Deane - Biodiversidade em Saúde, Fundação Oswaldo Cruz, Rua Teresina n. 476, Adrianópolis, 69.057-070, Manaus, AM, Brazil. E-mail: sergioluz@amazonia.fiocruz.br

### Abstract

The male, female, pupa and larva of *Simulium litobranchium* **n. sp.** are described and illustrated. This new species has 12 gill filaments, as do *S. duodenicornium* Pepinelli, Hamada & Trivinho-Strixino, *S. guianense* Wise, *S. hirtipupa* Lutz, *S. perplexum* Shelley, Maia-Herzog, Lunas Dias & Couch and *S. scutistriatum* Lutz, but it can be distinguished from these other species by a combination of characters observed at the pupal and adult stages. DNA barcoding showed that *Simulium litobranchium* **n. sp.** exhibits more than 4% nucleotide divergence in cytochrome oxidase I from three other closely related species in the Neotropical subgenus *S. (Thyrsopelma)*. The new species was collected in the Brazilian states of Goiás and Minas Gerais and inhabits rivers 30–40 m in width in the Paraná River hydrographic basin. Females were not observed engaging in anthropophilic behavior.

**Key words:** aquatic insects, taxonomy, COI, genetic distance, sibling species

### Introduction

Shelley *et al.* (2002) described morphological variation in larvae of *S. guianense* Wise collected in the state of Goiás, denominating them as atypical. They reported that the only reliable character to distinguish them from the typical *S. guianense* is the presence, in the larva, of a pair of raised tubercles on the dorsolateral surface of abdominal segments 1 to 6. They also reported differences in wing vein setation of both genders in comparison with the typical form of this species. At the pupal stage they did not observe any difference.

*Simulium guianense* is the main vector of *Onchocerca volvulus* (Leuckart) in locations in the Brazilian and Venezuelan Amazonian where onchocerciasis is confined, and this simuliid is widely distributed in Brazil. It is therefore very important to establish the morphological boundaries of this species.

Molecular methods have been used to understand relationships between populations or between closely related species. Among the available molecular tools, sequencing of gene fragments allows direct evaluation of DNA polymorphism, providing additional data to make inferences about relationships between specimens, populations, and species. Different segments of mitochondrial and nuclear DNA evolve at different rates. Regions with rapid evolution are adequate for studying closely related taxa, while more conservative regions are appropriate for comparing more divergent taxa (Avice 1994). Molecular methods of analysis have permitted many innovations in insect systematics (Roderick 1996; Caterino *et al.* 2000).