Journal of the American Mosquito Control Association, 37(2):98–100, 2021

SCIENTIFIC NOTE

BILATERAL ANOMALY IN A MALE OF *EVANDROMYIA LENTI* (DIPTERA: PSYCHODIDAE) IN PERNAMBUCO, BRAZIL

DÉBORA ELIENAI DE OLIVEIRA MIRANDA,[1] JOANNA ALEXANDRE,[1] FERNANDO JOSÉ DA SILVA,[1] JOSÉ DILERMANDO ANDRADE FILHO,[2] FILIPE DANTAS-TORRES,[1] AND SINVAL PINTO BRANDÃO-FILHO[1]

ABSTRACT. More than 1,000 species of phlebotomine sand flies have been described in the literature, many of which are vectors of *Leishmania* spp. In addition to the morphological similarities between some species groups, the occurrence of anomalies within one species may lead to erroneous description of new taxa. This study describes a case of bilateral anomaly in the number of spines in the gonostyle in a male of *Evandromyia lenti*. In this anomalous male, a 5th spine is present between the upper external spine and the lower external spine. The occurrence of such an anomaly can potentially result in misidentifications in this important group of insects.

KEY WORDS Anomaly, *Lutzomyia lenti,* gonostyle, identification, taxonomy

Phlebotomine sand flies (Diptera: Psychodidae: Phlebotominae) are small insects of major medical and veterinary significance, due to the bloodfeeding habits of the females. More than 1,000 species of phlebotomine sand flies have been described to date, many of which are recognized vectors of *Leishmania* spp. (Maroli et al. 2013). In the Americas, there are around 530 species and >20 species have been listed as vectors of *Leishmania* spp. to humans in this region (Maroli et al. 2013, Brazil et al. 2015, Shimabukuro et al. 2017).

Phlebotomine sand fly identification is mostly based on morphological features. Many species belonging to certain genera, or species groups, are morphologically similar, which in turn makes their identification a complex task (Galati 2018). In addition, misidentifications may result in wrong conclusions about the geographical distribution and even the vector role of a given species (Andrade Filho et al. 2004, Cutolo et al. 2009). Morphological anomalies have been documented in phlebotomine sand flies around the world (Kassem et al. 1988, Marcondes 1999, Florin et al. 2010). For instance, anomalies can occur in the antennae, palpomeres, male terminalia, and female spermathecae. These anomalies may eventually lead to misidentifications, including the wrong description of a new species (e.g., Mangabeira 1942). Nonetheless, the biological impact of such anomalies (e.g., on sexual behavior and reproduction) remains poorly understood. Considering the eminent taxonomic implications of morphological anomalies in phlebotomine sand flies, we describe herein a case of morphological anomaly in a male of *Evandromyia lenti* (Mangabeira).

This anomalous male was found during one of a series of phlebotomine sand fly collections conducted in Salgadinho Village, Pesqueira municipality (8°16.47′S, 36°40.2′W; 709 m above sea level), Pernambuco State, northeastern Brazil. The climate of Pesqueira is semiarid, with an average annual rainfall of 53.3 mm and an average annual temperature of 23.8°C (data obtained from Instituto de Tecnologia de Pernambuco meteorological station: 82890). The vegetation is predominantly composed of semideciduous and deciduous forests.

Sand fly collections were performed under the framework of different research projects in January 2017, July 2018, and January 2019. During 2 consecutive nights of each month, Centers for Disease Control and Prevention light traps (2 per night) were installed from 5:00 p.m. to 6:00 a.m., positioned 1.5 m above the ground in the peridomicile (chicken coop) of a house. The anomalous male and additional phlebotomine sand flies collected were identified morphologically and classified according to Galati (2018). The specimens were prepared as microscopy slides, and examined by light microscopy. The following structures were observed: first 3 flagellomeres, palps, labrum–epipharynx, clypeus, sperm pump (= ejaculatory pump), aedeagal ducts (= genital filaments), epandrial lobes (= lateral lobes), parameres, gonocoxites, and gonostyles were also analyzed. The anomalous male described herein is deposited (accession number: 91.623) in the collection of sand flies (FIOCRUZ-COLFLEB) of René Rachou Institute (Fiocruz), Minas Gerais, Brazil. We also compared the anomalous male to a normal male from Lassance, Minas Gerais, the type locality of *E. lenti*, which is deposited in the FIOCRUZ-COLFLEB collection (accession number: 81.272).

[1] Department of Immunology, Aggeu Magalhães Institute, Oswaldo Cruz Foundation (Fiocruz), Avenida Professor Moraes Rego s/n, Cidade Universitária, Recife, 50740-465 Pernambuco, Brazil.

[2] René Rachou Institute, Oswaldo Cruz Foundation (Fiocruz), Avenida Augusto de Lima, 1715, Barro Preto, Belo Horizonte, 30190-009 Minas Gerais, Brazil.

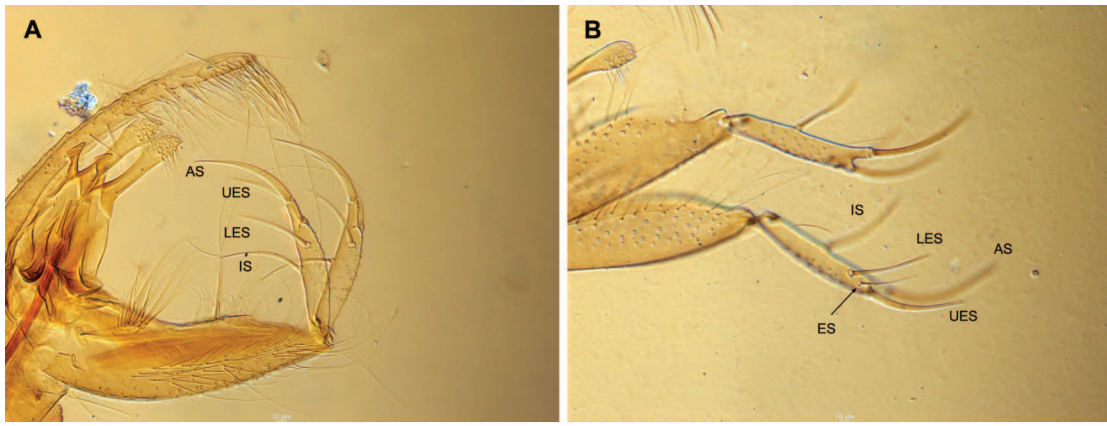

Fig. 1. Terminalia of *Evandromyia lenti* males. (A) Normal male from Lassance, Minas Gerais, Brazil; and (B) anomalous male, showing one of the extra spines (arrow) between the upper external spine and the lower external spine. Abbreviations: AS, apical spine; UES, upper external spine; LES, lower external spine; IS, internal spine; ES, extra spine.

A total of 388 phlebotomine sand flies belonging to 4 species (i.e., *Lutzomyia longipalpis* (Lutz and Neiva), *E. lenti*, *Nyssomyia intermedia* (Lutz and Neiva), and *Micropygomyia villelai* (Mangabeira)) were identified. The predominant species was *L. longipalpis* (85.8%; 232 males and 101 females), followed by *E. lenti* (9.8%; 11 males and 27 females), *N. intermedia* (2.6%; 8 males and 2 females), and *M. villelai* (1.8%; 6 males and 1 female).

Among the 388 phlebotomine sand flies identified, we found the anomalous male of *E. lenti*. A careful examination of the terminalia of this male revealed an extra spine in both gonostyles. The gonostyle of the *E. lenti* male bears 4 spines (apical spine, upper external spine, lower external spine, and internal spine) and a spiniform preapical seta (Mangabeira 1938, Galati 2018). On the anomalous male, a 5th spine is present between the upper external spine and the lower external spine (Fig. 1). All other morphological features allowed us to unequivocally assign this male to the species *E. lenti*.

Evandromyia lenti is a phlebotomine sand fly distributed in several Brazilian states, including Pernambuco (Dantas-Torres et al. 2010, Costa et al. 2013, Sales et al. 2019). This species has been found naturally infected by *Psychodiella chagasi* (Adler and Mayrink), a gregarine species also found in other sand fly species (Rocha et al. 2015). On the other hand, Brazil et al. (1997) carried out laboratory tests through experimental infections with *E. lenti* (from Minas Gerais State) with *Leishmania amazonensis*, but no evidence of infection was observed in 45 females of *E. lenti* dissected, strongly indicating that this phlebotomine sand fly is refractory to *Leishmania amazonensis* (Lainson and Shah) infection. Rêgo et al. (2015) found 2 *E. lenti* females (one collected in the peridomicile and other on a trail) positive for *Leishmania infantum* Nicolle DNA, suggesting that this phlebotomine sand fly could be involved in the transmission cycle of this parasite. However, experimental studies and more conclusive field data are needed to support this hypothesis.

Cases of anomalies have been described in other phlebotomine sand fly species in the Americas (Sherlock 1958, Marcondes 1999, Ximenes et al. 2002, Andrade Filho et al. 2004, Pinto et al. 2010, Sanguinette et al. 2013). From a broad perspective, the case of bilateral anomaly described herein is important for the taxonomy of American phlebotomine sand flies, since the occurrence of such anomalies can result in misidentifications in this important group of vectors.

Morphological anomalies in phlebotomine sand flies may be of genetic (embryonic/postembryonic) or environmental origin (Andrade Filho et al. 2004). The biological significance of such anomalies in phlebotomine sand flies is still unknown. However, from a taxonomic perspective, anomalous specimens may eventually be wrongly identified as a new species, as has happened in the past with *Flebotomus nordestinus* (Mangabeira) (Mangabeira 1942), which was later considered to be a junior synonym of *Sciopemyia sordellii* (Shannon and Del Ponte) (Young and Morales 1987). In this context, our report confirms that morphological anomalies may occur in *E. lenti*, which may help in preventing future misidentifications of this species.

REFERENCES CITED

Andrade Filho JD, Carvalho GML, Saraiva L, Falcão AL. 2004. Bilateral anomaly in the gonostyle of *Mycropygomyia schreiberi* (Martins, Falcão & Silva) (Diptera, Psychodidae). *Rev Bras Entomol* 48:583–585.

Brazil RP, Carneiro VL, Andrade Filho JD, Alves JCM, Falcão AL. 1997. Biologia de *Lutzomyia lenti* (Mangabeira) (Diptera: Psychodidae). *An Soc Entomol Bras* 26:191–193.

Brazil RP, Rodrigues AA, Filho JDA. 2015. Sand fly vectors of *Leishmania* in the Americas—a mini review. *Entomol Ornithol Herpetol* 4:1–4.

Costa PL, Dantas-Torres F, da Silva FJ, Guimarães VC, Gaudêncio K, Brandão-Filho SP. 2013. Ecology of *Lutzomyia longipalpis* in an area of visceral leishmaniasis transmission in north-eastern Brazil. *Acta Trop* 126:99–102.

Cutolo AA, von Zuben CJ, Galati EAB. 2009. Anomalia bilateral no gonóstilo de *Sciopemyia sordellii* (Shanon & Del Ponte) (Diptera, Psychodidae). *Rev Bras Parasitol Vet* 18:67–69.

Dantas-Torres F, de Andrade AJ, Tenório KE, Andrade Filho JD, Balbino VQ, Brandão-Filho SP. 2010. Phlebotomine sand flies (Diptera: Psychodidae: Phlebotominae) in the State of Pernambuco. *Rev Soc Bras Med Trop* 43:733–736.

Florin DA, Lawyer P, Rowton E, Schultz G, Wilkerson R, Davies SL, Lipnick R, Keep L. 2010. Morphological anomalies in two *Lutzomyia* (*Psathyromyia*) *shannoni* (Diptera: Psychodidae: Phlebotominae) specimens collected from Fort Rucker, Alabama, and Fort Campbell, Kentucky. *J Med Entomol* 47:952–995.

Galati EAB. 2018. Phlebotominae (Diptera, Psychodidae): classification, morphology and terminology of adults and identification of American taxa. In: Rangel EF, Shaw JJ, eds. *Brazilian sand flies*. Cham, Switzerland: Springer International Publishing. p 9–212.

Kassem HS, Sattar SA, Shehata MG, Sawaf BE. 1988. Abnormalities in the sand fly *Phlebotomus papatasi* Scopoli (Diptera: Psychodidae) of Egypt. *J Egypt Public Health Assoc* 63:209–213.

Mangabeira Filho O. 1938. Sobre duas novas especies de *Flebotomus* (Diptera: Psychodidae). *Mem Inst Oswaldo Cruz* 33:349–365.

Mangabeira Filho O. 1942. 12ª contribuição ao estudo do *Flebotomus* (Diptera: Psychodidae) *Flebotomus nordestinus* n. sp. *Mem Inst Oswaldo Cruz* 37:327–331.

Marcondes CB. 1999. Anomalies of *Lutzomyia intermedia* (Lutz & Neiva, 1912) (Diptera, Psychodidae, Phlebotominae). *Mem Inst Oswaldo Cruz* 94:365–366.

Maroli M, Feliciangeli MD, Bichaud L, Charrel RN, Gradoni L. 2013. Phlebotomine sandflies and the spreading of leishmaniases and other diseases of public health concern. *Med Vet Entomol* 27:123–147.

Pinto IS, Santos CB, Ferreira AL, Falqueto A. 2010. Variations in the gonostyle of *Nyssomyia intermedia* (Lutz & Neiva) (Diptera: Psychodidae). *Neotrop Entomol* 39:732–735.

Rêgo FD, Rugani JM, Shimabukuro PH, Tonelli GB, Quaresma PF, Gontijo CM. 2015. Molecular detection of *Leishmania* in phlebotomine sand flies (Diptera: Psychodidae) from a cutaneous leishmaniasis focus at Xakriabá Indigenous Reserve, Brazil. *PLoS One* 10:e0122038.

Rocha LS, Santos CB, Falqueto A, Brazil RP. 2015. Natural infection of *Evandromyia lenti* (Mangabeira) (Diptera: Psychodidae) by *Psychodiella chagasi* (Adler & Mayrink) (Apicomplexa: Lecudinidae). *J Vector Ecol* 40:419–421.

Sales KGS, Miranda DEO, Costa PL, da Silva FJ, Figueredo LA, Brandão-Filho SP, Dantas-Torres F. 2019. Home sweet home: sand flies find a refuge in remote indigenous villages in north-eastern Brazil, where leishmaniasis is endemic. *Parasit Vectors* 12:118.

Sanguinette CC, Faustino JX, Serra e Meira PC, Botelho HA, Carvalho GM, Gontijo CM, Filho JD. 2013. Anomalies in the sand fly *Lutzomyia longipalpis* (Diptera: Psychodidae) in Brazil. *J Am Mosq Control Assoc* 29:54–58.

Sherlock IA. 1958. Anomalias de *Phlebotomus longipalpis* Lutz & Neiva, 1912 (Diptera: Psychodidae). *Rev Bras Biol* 18:433–437.

Shimabukuro PHF, de Andrade AJ, Galati EAB. 2017. Checklist of American sand flies (Diptera, Psychodidae, Phlebotominae): genera, species, and their distribution. *ZooKeys* 8:67–106.

Ximenes MFFM, Castellón EG, Freitas RA. 2002. Morphological genitalic anomaly in *Lutzomyia longipalpis* sensu lato (Lutz & Neiva, 1912) collected in State of Rio Grande do Norte, Brazil. *Entomotropica* 17:183–184.

Young DG, Morales A. 1987. New species and records of phlebotomine sand flies from Colombia (Diptera: Psychodidae). *J Med Entomol* 24:651–665.