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Taxonomy and systematics

## First record of the *Pseudodiamesa branickii* species-group (Diptera: Chironomidae: Diamesinae) from central Mexico

### *Primer registro del grupo Pseudodiamesa branickii* (Diptera: Chironomidae: Diamesinae) del centro de México

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#### Abstract

Larvae and pupae of the *Pseudodiamesa branickii* species-group (Makarchenko & Makarchenko, 1999) (Diptera: Chironomidae) were found in 2 high-altitude lakes (El Sol and La Luna, 4,200 m asl), in the crater of the Nevado de Toluca volcano, in central Mexico. This is the first record of the genus in Mexico, and its southernmost report from a location that represents a transitional zone between the Nearctic and Neotropical regions. New information on the ecology and biogeographical distribution of the species-group is given.

**Keywords:** Tropical high-altitude lakes; Benthos; Biodiversity

#### Resumen

Se identificaron ejemplares de larvas y pupas del grupo *Pseudodiamesa branickii* (Makarchenko y Makarchenko, 1999) (Diptera: Chironomidae) en 2 lagos de altura (El Sol y La Luna, 4,200 m snm), ubicados dentro del cráter del volcán Nevado de Toluca, en el centro de México. Éste es el primer reporte para México y la localización más austral del género, en una zona transicional entre las regiones Neártica y Neotropical. Se presenta nueva información sobre la ecología y la distribución biogeográfica de este grupo.

**Palabras clave:** Lagos tropicales de altura; Bentos; Biodiversidad

## Introduction

*Pseudodiamesa* Goetghebuer (1939) is a genus containing 12 species distributed in the Northern Hemisphere (Nearctic, Palearctic and Oriental regions). All of them are adapted to cold environments such as high-altitude or high-latitude lakes or found in the sedimentary (sub-fossil) record, correlating with past glacial phases (Ilyashuk et al., 2010). Based on the morphology of the adult male, Oliver (1959) divided this genus into 2 subgenera, *Pachydiamesa* and *Pseudodiamesa*. Later on, using different morphological characters of the larval head capsules Makarchenko and Makarchenko (1999) suggested applying a species-group nomenclature to the larvae of *Pseudodiamesa* dividing the genus into 2 species-groups that also have distinctive ecological preferences (Makarchenko & Makarchenko, 1999). The first is represented by 2 species, *P. branickii* (Nowicki) and *P. pertinax* (Garret), which are adapted to moderately cold conditions (littoral or non-glacial environments). While *P. branickii* has a wide geographical distribution in the Nearctic, Palearctic and Oriental regions *P. pertinax* is endemic to the Nearctic region (Ilyashuk et al., 2010). The second is the *P. nivosa* group which includes 10 species, their ecology is more difficult to define, but in general they are adapted to colder, peri-glacial conditions and have a preferential distribution in the Palearctic region (Fig. 1); however, in this group there is one species endemic to the Oriental region (*P. nepalensis*) and another one (*P. arctica*) to the Nearctic region (Ilyashuk et al., 2010; Manca et al., 1998). Therefore, for the Nearctic region only 3 species of this genus have been reported and 2 are endemic (Ilyashuk et al., 2010; Oliver, 1959) (Fig. 1): *P. arctica* (of the *P.*

*nivosa* group), and the 2 moderately-cold adapted species of the *P. branickii* group: *P. pertinax* and *P. branickii*.

In this paper we report the presence of chironomid larvae and pupae belonging to the *P. branickii* species-group in recent sediments of lakes El Sol and La Luna, both tropical high-altitude lakes from central Mexico. This is the first report of this species-group in Mexico and represents its southernmost record at a site located in a transitional area between the Nearctic and the Neotropical regions (Morrone, 2014, 2019).

## Materials and methods

The Nevado de Toluca (19°06'30" N, 99°45'30" W) is a stratovolcano located in the Trans-Mexican Volcanic Belt in central Mexico; with 4,680 m asl, it represents the fourth highest peak in the region. The crater of this volcano (4,200 m asl) contains 2 permanent lakes, El Sol and La Luna (Fig. 2). These are shallow (< 15 m) lakes with cold (< 10 °C), slightly acidic (pH < 7), clear waters of low alkalinity and nutrient levels (oligotrophic to ultraoligotrophic). The lakes lie above the local tree line (ca. 4,000 m asl) and have a cold climate with mean annual temperature of 3.8 °C and extreme temperatures on the range of -9 °C to 19 °C (SMN, 2019). Rainfall is concentrated from May to September, with average annual precipitation of 1,213 mm yr<sup>-1</sup>. Despite their high altitude, they do not freeze during winter, and their mixing regime is warm polymictic. Previous research reported the presence of chironomid larvae in the sediments of these lakes (Oseguera et al., 2016), *Pagastia* sp. and *Tanytarsus* sp. were reported for lake El Sol and *Pagastia* sp. was reported for lake La Luna.

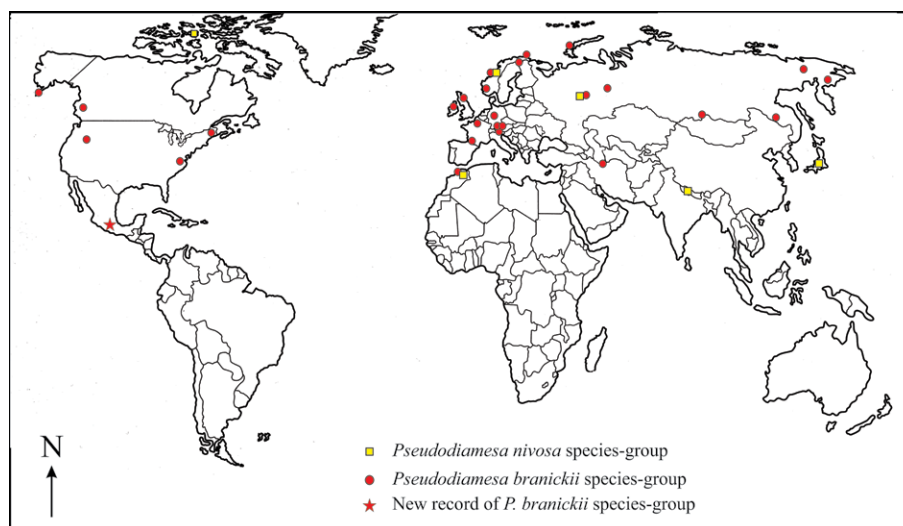


Figure 1. Map showing worldwide distribution of *P. branickii* (circles) and *P. nivosa* (squares) groups.

Sediment samples were collected from littoral and profundal zones in lakes El Sol and La Luna in November 2018 (Fig. 2). Profundal sediment was collected using an Eckman dredge, littoral sediment was collected with plastic jars, in both cases sediment was sieved using 100  $\mu$ m mesh and inspected for chironomid larva, pupae or exuviae which were hand-picked using tweezers. Surface sediment samples from the profundal zone were also collected in 250 ml plastic jars and preserved with alcohol for further examination in the laboratory. For laboratory rearing, third and fourth instar live larvae were deposited in individual, ventilated jars with some water and sediment and kept in the fridge at 4 °C, 3 of these individuals metamorphosed to pupae in the laboratory.

Chironomid larvae and pupae were slide-mounted using Hydromatrix®, identified and photographed under an optical microscope (Olympus BX50 - DP20 camera). For identification of larvae, morphological characteristics of the head capsules such as pecten epipharyngis, labral lamella, mentum and antenna were used (Ilyashuk et al., 2010; Makarchenko & Makarchenko, 1999). Identification of pupal exuviae was based on Serra-Tosio (1976). Specimens were deposited in the National Insect Collection at the Instituto de Biología, Universidad Nacional Autónoma de México (CNIN-IB-UNAM). Preserved chironomid larvae specimens collected previously from lakes El Sol and La Luna by Oseguera et al. (2016) were also re-inspected.

## Results

Six larvae and 3 pupae cultivated in the laboratory allowed the identification of *Pseudodiamesa branickii*

species-group (subfamily Diamesinae) in lake La Luna. Re-examination of the material collected previously from both lakes, El Sol and La Luna (Oseguera et al., 2016), allowed confirmation that the larvae reported as *Pagastia* sp. corresponds to the *P. branickii* species-group, which is documented for the first time in central Mexico.

Selected features of the head capsule morphology such as antennal ratio, pecten epipharyngis and mentum are key to separate the different species-groups (Makarchenko & Makarchenko, 1999). In *P. branickii* all the teeth in the mentum are similar in color and the antennal ratio is < 2.5 (Fig. 3). Regarding the labro-epipharyngeal region (Fig. 4), the pecten holds an odd number of median scales with a single median scale and 3 pairs of lateral scales, there is no labral lamella visible. These characteristics separate the larvae of the *P. branickii* species-group from the sister *P. nivosa* species-group and allow identification of the specimens from the Nevado de Toluca lakes as belonging to the *P. branickii* group (Ilyashuk et al., 2010). The larvae in the 2 sister species in this group (*P. branickii* and *P. pertinax*) appear to be identical, therefore the separation of both species based on larva morphology is impossible since the immature stages of *P. pertinax* are still unknown (Ilyashuk et al., 2010).

Identification was carried out using pupal exuviae of the reared specimens. In the VII and VIII abdominal segments of *P. branickii*, L the setae are non-branched and in the anal lobe the more external of the 3 anal macrosetae is fragile (often broken) and farthest from the other 2 (Fig. 5). In *P. nivosa*, L the setae of abdominal segments VII and VIII are branched and the anal macrosetae of the anal lobe are similar and equally distant between each other (Serra-Tosio, 1976).

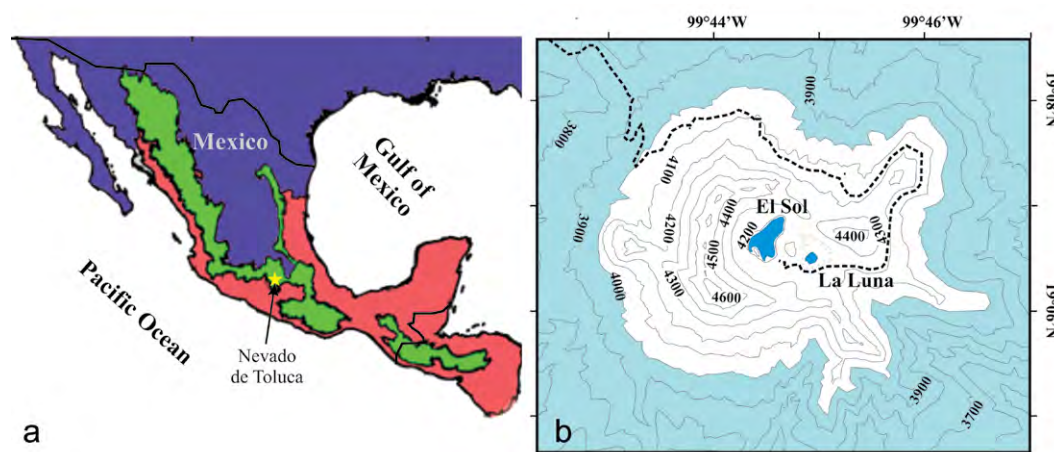


Figure 2. Location maps. a) Location of the Nevado de Toluca, in a transitional area (green) between the Nearctic (purple) and the Neotropical (pink) regions (according to Morrone, 2014, 2019); b) lakes La Luna and El Sol, at 4,200 m asl, in the crater of the Nevado de Toluca Volcano.

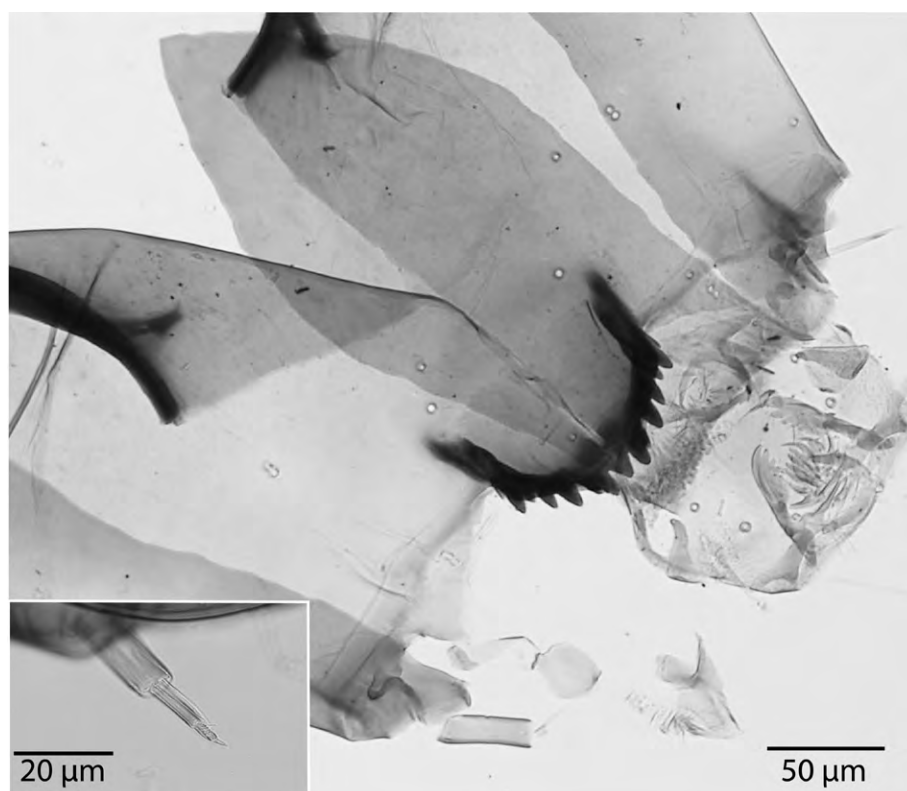


Figure 3. Larval head capsule showing mentum and labro-epipharyngeal complex. Details of the antenna are shown in the left-lower corner

## Discussion

*Pseudodiamesa* species are cold-stenothermic, with an ability to survive at below zero temperatures. This must be an evolutionary conserved guild in the subfamily Diamesinae, which in turn results as a strategy for survival since they spend most of their life as larvae while the pupal and adult stages last only a few days (Lencioni & Bernabò, 2017). The *P. branickii* group is characteristic of moderately cold conditions, in agreement with the climatic characteristics at the Nevado de Toluca lakes, which do not regularly freeze during winter. This species-group is more common in soft sediments of running waters and littoral zones of lakes (Boggero et al., 2006; Rossaro et al., 2006). First and second instars larvae of *P. branickii* are detritus feeders whereas third and fourth instars larvae are predators (Lencioni & Bernabò, 2017).

The *Pseudodiamesa branickii* species-group has a wide geographical distribution in the Northern Hemisphere, but thus far in the Nearctic region it has only been collected in a few localities, such as the Rocky Mountains in British Columbia and Alberta, Alaska and the northern part of the

East coast (Fig. 1, Appendix 1). *Pseudodiamesa branickii* group is part of the circumpolar fauna which suggests earlier land connections between Eurasia and North America. Its presence in central Mexico appears to be related with past climatic fluctuations and the existence of boreo-transitional conditions along the Mexican highlands, as a southward extension of the North American Rocky Mountains (Siciński, 1988). The highest mountains in central Mexico (including the Nevado de Toluca) show evidences of past glacier advances during intervals of colder climatic conditions during which this species-group could have extended its geographical range southwards, into the highlands of central Mexico (Vázquez-Selem & Heine, 2011). Therefore, this group may have remained as relict species by retreating to the favorable conditions on the highest peaks in the Mexican highlands during warmer periods, when mountain glaciers retreated. Indeed, the tolerance of the species to freezing temperatures suggests that populations isolated on different mountain ranges provide insights of historical changes probably associated to glacial periods (DeChaine & Martin, 2006; Knowles, 2000).

This new record of the *P. branickii* group found at the southern boundary of the Nearctic region, more specifically in the Mexican Transition Zone, extends the geographical distribution of the genus *Pseudodiamesa* (Morrone, 2019). In addition, our results provide information about the ecology and biogeographical distribution of the genus

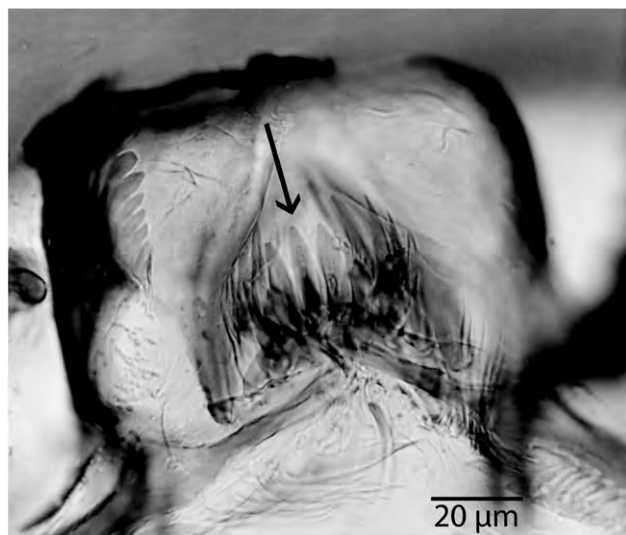


Figure 4. Details of the labro-epipharyngeal region. The arrow shows the single median scale.

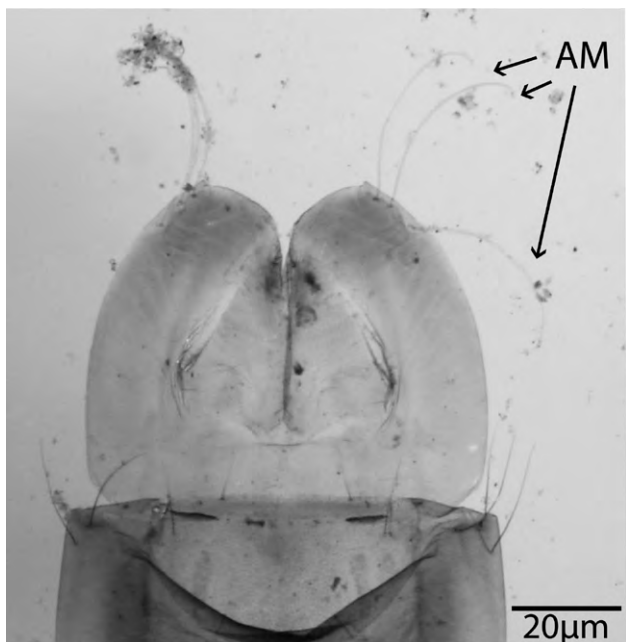


Figure 5. Abdominal tergite VII and anal lobe of pupae. AM: anal macrosetae.

and its potential isolation and further adaptation in a high mountain region in central Mexico. We also give insight on the Diamesinae and its relationships with temperature, which in turn, is a potential tool to evaluate the effect of global warming during the last century. More work is needed, especially dealing with taxonomical aspects of the adults, which in turn are key elements to species identification. On the other hand, molecular studies of different populations within the *P. branickii* species-group could be required to overcome the difficulties for the collection of adults and the taxonomic problems when dealing with immature stages of this group and may also provide valuable information on the history and biogeographical affinities of its population in central Mexico.

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