

Original article

First record of *Vacupernius packeri* (Allen) (Ephemeroptera: Leptohiphidae) from Colombia and South America with additional notes on egg morphology

Primer registro del efemeróptero *Vacupernius packeri* en Colombia y América del Sur y descripción de la morfología de sus huevos

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Abstract

In the present study, we recorded for the first time the mayfly species *Vacupernius packeri* (Allen, 1967) in Colombia and South America describing the egg of this species. The identification was based on nymphs, subimagos, and imagos from Tolima and Caldas, Colombia. The eggs are oval-shaped, with one polar cap, and chorionic plates that are a little overlapped near the uncapped pole but absent near the capped pole. The egg has similar characteristics to the *Traverhyphes* group eggs, such as chorionic plates slightly overlapped and absent on the chorion surrounding the polar cap. Future phylogenetic analyses would be important to clarify the relationships between *Vacupernius* and the *Traverhyphes* group.

Keywords: Pannota; *Traverhyphes* group; Distribution; Andean region; Eggs, Mayfly.

Resumen

Se registra por primera vez el efemeróptero *Vacupernius packeri* en Colombia y América del Sur y se describe la morfología de sus huevos. La especie se identificó a partir de ninfas, subimagos e imagos procedentes de los departamentos de Tolima y Caldas, Colombia. Los huevos tienen forma oval, una cápsula polar y placas coriónicas poco superpuestas cerca al polo sin tapa. Además, se observó que presentan características similares a las del grupo *Traverhyphes*: placas coriónicas poco traslapadas y ausentes en el corion que rodea la cápsula polar. Es importante aclarar las relaciones entre los grupos *Vacupernius* y *Traverhyphes* en futuros análisis filogenéticos.

Palabras claves: Pannota; Grupo *Traverhyphes*; Distribución; Región Andina; Huevos.

Introduction

The genus *Vacupernius* was proposed by Wiersema & McCafferty (2000) to include the following species: *V. packeri* (Allen), *V. paraguttatus* (Allen), and *V. rolstoni* (Allen), which were originally described by Allen (1967) in *Leptohiphidae* Eaton. According to the key proposed by Wiersema & McCafferty (2000), *Vacupernius* can be distinguished from *Leptohiphidae* and the other genera of *Leptohiphidae* by a combination of characters in nymphs: Middle and hind tibiae without medial longitudinal row of branched setae on the dorsal region; hind tarsi approximately half of the length of the respective tibiae, and spatulate setae in the transverse row at mid-length of fore femora. Adults can be

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differentiated by male genitalia with three-segmented forceps mounted on elevated columnar bases acutely pointed, penes almost completely fused with a unique morphology, and mid- and hind tarsi slightly less than one-half the length of the respective tibiae.

Among the three species of the genus, *V. packeri* has the widest distribution (Belize, Costa Rica, Guatemala, Honduras, México, and the USA) and has not been registered at more than 900 MASL while *V. paraguttatus* is reported in the United States and *V. rolstoni* in the Dominican Republic (**Wiersema & McCafferty, 2000**). It should be noted that **Emmerich (2004)** first recorded the genus in South America but could not identify the species because the captured individual was a subimago and the morphology of the genitals was not clearly visible. In that study, the author observed that the penis and the styliger plate of *Vacupernius* were like those of *Traverhypes*, *Allenhypes*, and *Yaurina*, but this has not been phylogenetically evaluated yet. To expand the knowledge on *Vacupernius*, in this study, we recorded for the first time a species in Colombia and South America identified based on adults and nymphs. Additionally, we described for the first time the egg of this species and the genus.

Materials and methods

The nymphs, subimagos, and adults of *V. packeri* were collected in the departments of Tolima and Caldas (Colombia). The nymphs were collected qualitatively with manual nets and adults with light traps between 6:00 p.m. and 8:30 p.m. Some adult specimens were manually captured in spider webs in riparian vegetation.

Mouthparts, legs (nymphs), and genitalia (imagos) were mounted and analyzed by stereomicroscope. We used taxonomic keys and original descriptions (**Allen, 1967; Wiersema & McCafferty, 2000**) for species identification while eggs were described using the terminology proposed by **Koss & Edmunds (1974)**. The collected material was stored in alcohol 96% and deposited at the *Colección Entomológica del Programa de Biología de la Universidad de Caldas - CEBUC*.

Pictures were taken using a Leica M205C stereomicroscope with an attached Leica MC-170HD camera. Some characters were examined through Scanning Electron Microscope (SEM) with a QUANTA 250 (11 kV) at low vacuum except those in **figure 1C-D**, which were washed with a neutral soap, ultrasonicated (for further cleaning), dehydrated, gold-coated, and then observed under high vacuum at the *Instituto de Investigaciones en Estratigrafía (Universidad de Caldas)*. The map of species distribution was done in a digital elevation model of the Neotropic with the software ESRI® ArcMap 10.0 using the records published in the literature (**Allen, 1967; Kilgore & Allen, 1973; Henry, 1986; Allen, 1978; Wiersema & McCafferty, 2000; Emmerich, 2004**) and the new records of this study.

Results and discussion

The nymphs and adults collected corresponded to the species *V. packeri* (Allen) (**Figure 1A**), which was originally registered for Belize, Costa Rica, Guatemala, Honduras, México, and the United States (**Allen, 1967; Kilgore & Allen, 1973; Henry, 1986; Allen, 1978; Wiersema & McCafferty, 2000**) and now it is documented for the first time in the northern part of South America. The characters used in the determination of the species were the shape of the forceps and penis of the imagos (**Figure 1B**). **Figure 1C-D** shows the transversal row of the spatulate setae in the middle region of the fore femur in nymphs.

Description of the eggs

The eggs were extracted from female mature nymphs (**Figure 2A**) and female subimago (**Figures 2 B-D**). The following features were observed: Yellowish coloration pattern, 135-152 µm length, 80-95 µm width (n=2), oval shape, one polar cap, chorionic plates slightly overlapped and absent near the polar capsule, and circular micropylar area surrounded by six chorionic plates located at the equatorial region of the egg.

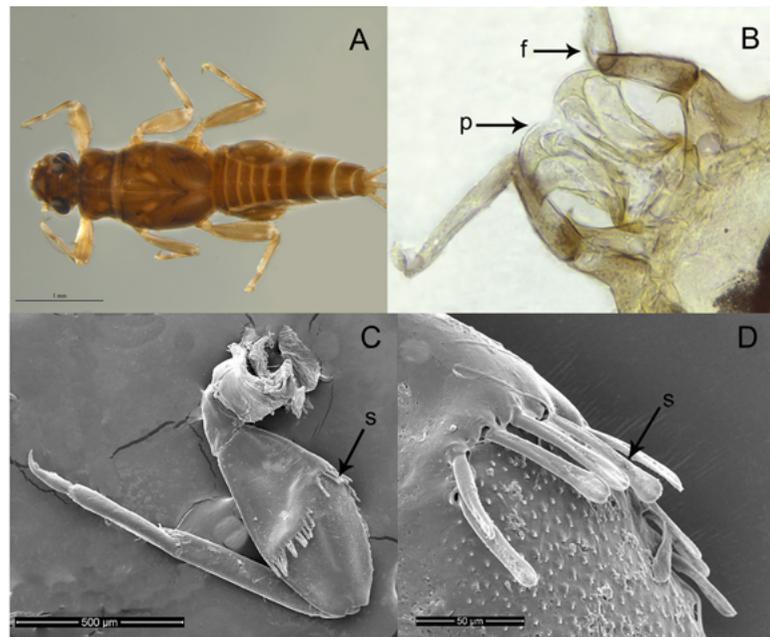


Figure 1. *Vacupernius packeri*: **A.** Nymph dorsal view. **B.** Detail of an adult male genitalia. **C.** Nymph fore leg. **D.** Detail of the spatulate setae in the fore femora. f: forceps. p: penis; s: setae

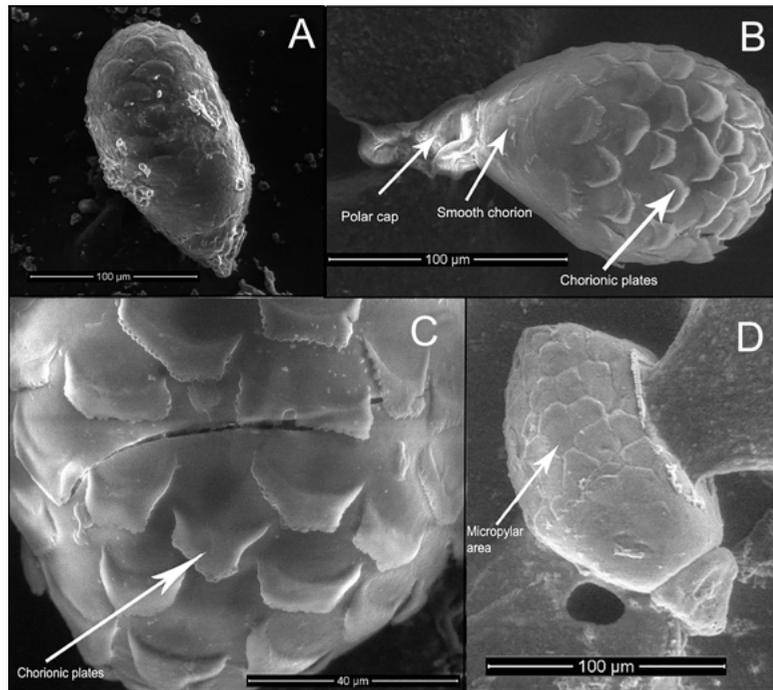


Figure 2. *Vacupernius packeri* eggs. **A.** Egg extracted from a nymph. **B-D.** Eggs extracted from subimago

Brittain (1982) reported that the spermatogenesis and oogenesis in Ephemeroptera are completed in the final nymphal instar and the eggs and sperm are physiologically mature in the subimago. Here we underline the importance of oology as a promising tool for associating Ephemeroptera nymphs and adults since the eggs from *V. packeri* nymphs,

subimagos, and imagos had the same form and similar chorionic ornamentation as proposed by **Domínguez & Cuezco (2002)** for other Ephemeroptera taxa. Concerning the contribution of the description of the eggs to the systematic relationship of *Vacupernius*, we observed that the species and the genus share similar oological characteristics to those of the *Traverhyphes* group (composed by *Allenhyphes*, *Lumahyphes*, *Traverhyphes*, and *Yaurina*), such as chorionic plates slightly overlapped and absent on the chorion surrounding of the polar cap (**Figure 3**). On the other hand, the study also allowed us to corroborate the observations of **Emmerich (2004)** regarding the affinities found in the shape of the penis and the styliger plate of *Vacupernius* and the *Traverhyphes* group.

Despite the affinities between *Vacupernius* and *Traverhyphes*, they can be differentiated by the styliger plate with posterolateral projections and slender penes with spines in *Traverhyphes* while in the *Vacupernius* genus the styliger plate has no projection and the penes are broad (**Domínguez, et al., 2006**). Future phylogenetic analyses would be important to clarify the relationships between *Vacupernius* and the *Traverhyphes* group and their possible synonymization.

Regarding the ecology of the genus, the information is preliminary. The nymphs sampled in this study were found in low altitudes in the tropical dry forest region. The Norcasia and Armero-Guayabal municipalities in Colombia (**Figure 4**) where the species was found are located in the Magdalena River median and low regions, an interesting biogeographical region similar to the Chocó region as suggested by **Hernández-Camacho, et al. (2007)**.

Examined material

26 nymphs, 1 male imago, and 1 female imago (in spider web), Colombia, Tolima, Universidad del Tolima farm, Santo Domingo Stream, Armero-Guayabal, 5° 0' 18.1"N, 74° 54' 24.7"W, 120 MASL, 18/iii/2017, Dias, Bacca & Marulanda cols.; 1 nymph, Colombia, Tolima, Universidad del Tolima farm, Santo Domingo Stream, Armero-Guayabal, 5° 0' 18.1"N, 74° 54' 24.7"W, 120 m, 18/x/2015, Dias & Marulanda cols.; 2 female subimagos (eggs extracted from one of these subimagos), Colombia, Tolima, Universidad del Tolima Farm, Armero-Guayabal 5° 0' 18.1"N, 74° 54' 24.7"W, 120 MASL, 22/iii/2017, Dias, Bacca & Marulanda. col.; 1 nymph Colombia, Tolima, Cajamarca, Coello Stream, 4° 16'

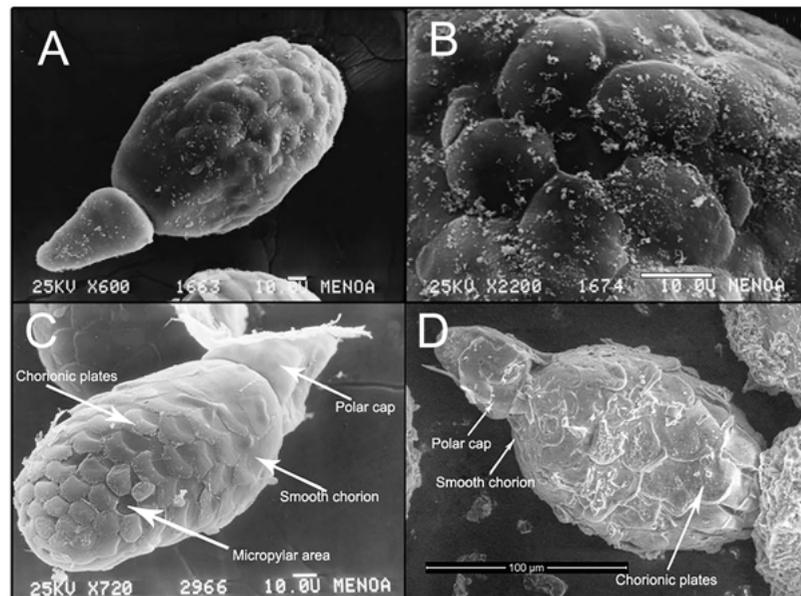


Figure 3. *Traverhyphes* eggs. **A, B.** *Traverhyphes (Mocoihyphes) edmundsi* (modified from **Molineri, 2004**). **C.** *Traverhyphes indicator* (modified from **Molineri, 2006**). **D.** *Traverhyphes* sp

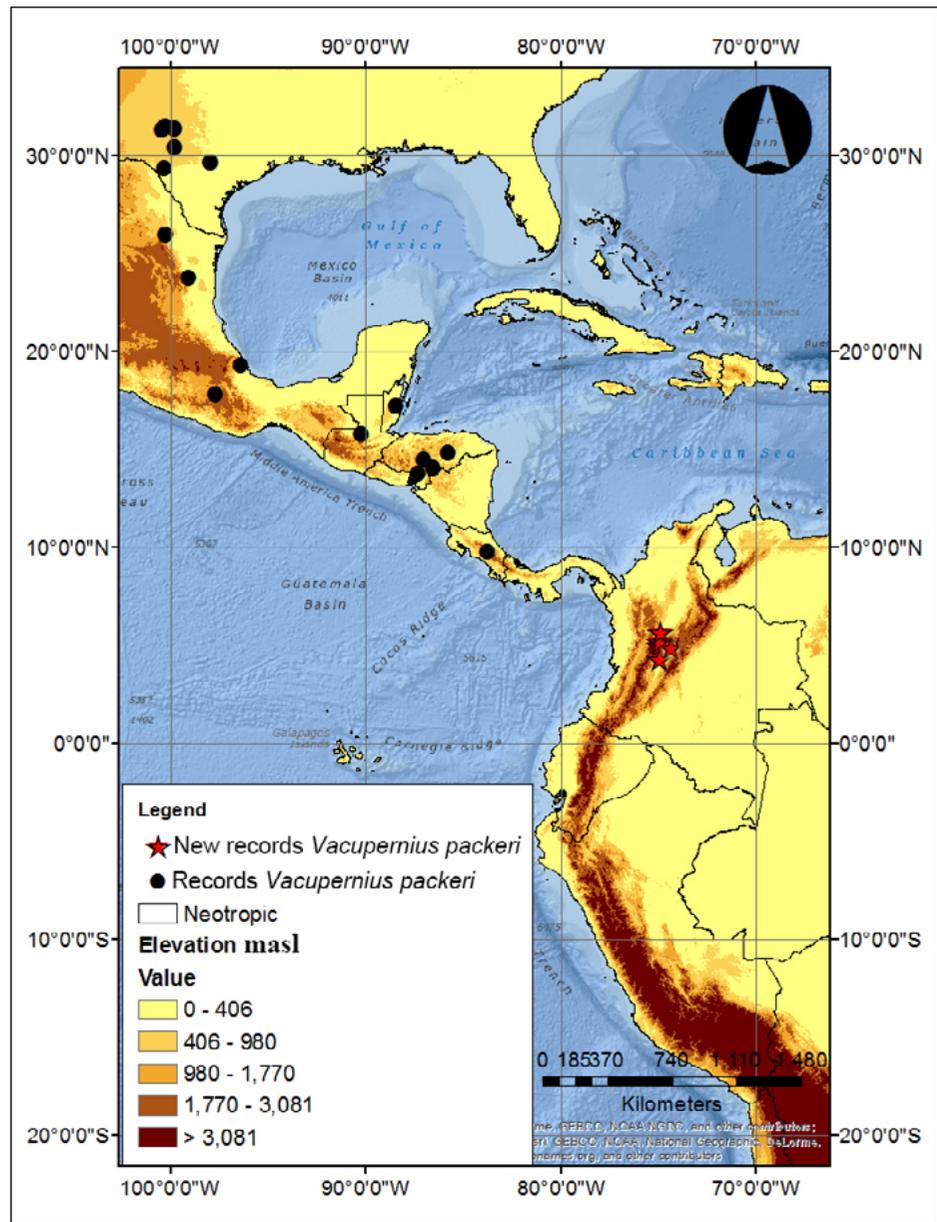


Figure 4. Occurrence of *Vacupernius packeri*

57.8°N, 74° 53' 50.1°W, 327 MASL, 1/iv/2013, Meza, Llano & Benavides cols.; 1 nymph, Colombia, Tolima, Lumbi Stream, Mariquita, 5° 9' 21.4°N, 74° 53' 58.3°W, 120 MASL, 28/v/2017, Dias, Bacca & Marulanda cols.; 3 mature nymphs (eggs were extracted from two of them; two eggs were measured), Colombia, Caldas, Norcasia, Reserva Natural de Río Manso, Río Manso Stream, 5° 39' 25.8°N, 74° 51' 44.6°W, 611 MASL, 21/xi/2014, Cardenas & Dias cols.

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Author contributions

TB wrote the manuscript and helped to collect the *Vacupernius packeri* specimens. JFM took the photographs, made the maps, and helped in the drafting of the manuscript. LGD helped in the drafting of the manuscript, the collection of all *Vacupernius packeri* specimens, the identification of the material collected, the photographs, the mounting, and genitalia photographs.

Conflict of interests

The authors declare that they have no conflicts of interest.

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