



The description of *Anacroneuria suerre* sp. nov. from Costa Rica (Plecoptera: Perlidae) and using nymphs in ecotoxicological studies

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Abstract

The lack of nymph-adult associations limits our knowledge of the ecology, history and sensitivity of aquatic insects. In this study, the new species, *Anacroneuria suerre* sp. nov. is described from Costa Rica based on the male, female, and nymphal stages. We briefly discuss the use of Plecoptera in ecotoxicological studies.

Key words: stoneflies, neotropics, nymph association, Central America, ecotoxicology

Introduction

With approximately 1,000 described species, the Perlidae Latreille, 1802 is the most diverse family of the order Plecoptera, and the only family that occurs in Costa Rica (Fochetti & De Figueroa, 2007). Of the 32-described species of *Anacroneuria* Klapálek, 1909 in Costa Rica, nymphs of only ten species have been previously associated with the adult (Table 1) (Stark, 1998; Gutiérrez-Fonseca & Springer, 2011; Gutiérrez-Fonseca & Springer, 2015; Fenoglio, 2007; Kondratieff & Armitage, 2019). This lack of nymph-adult associations limits our knowledge of the natural history, and ecological sensitivity at the species level.

Aquatic insects such as stoneflies play a key role in tropical and temperate freshwater ecosystems and have been used as indicators of anthropogenic disturbances in lentic and lotic environments (Zuñiga, 2010; Springer, 2010; Peterson *et al.*, 2001a). Their sensitivity to changes in temperature and dissolved oxygen has already been acknowledged (Genkai-Kato *et al.*, 2000; Haidekker & Hering, 2008). Furthermore, Rico & Van den Brink (2015) determined their high sensitivity and vulnerability to all insecticide classes. Among the anthropogenic stressors present in water bodies, pesticides used in agriculture are emerging as the main driver of biodiversity loss for many aquatic macroinvertebrate communities in rivers and streams (Beketov *et al.*, 2013).

Unfortunately, information gaps regarding the description of aquatic insect species, especially their aquatic stages, hinder the development of ecotoxicological tests at the species level. Furthermore, reported differences in sensitivity between organisms of the same family (even the same genus) in the tropics (e.g., Tomanova & Tedesco, 2007; Rubach *et al.*, 2010), justify the urgent need to associate environmental stressors to organisms at the lowest taxonomic level. In fact, in Central America, information about the ecotoxicological effects of any anthropogenic stressor in a native aquatic insect species is not available. In this paper, a new species of *Anacroneuria* from Costa Rica is described from both adult and nymph stages. We also highlight the importance of developing ecotoxicological studies with native aquatic insects in the tropics.

Materials and methods

Specimens collected. Stoneflies were collected between February and September 2018 in upper basin of Río Jiménez, Limón, Costa Rica, 10° 10' 24.38" N, 83° 45 '7.35" W (Figure 1).

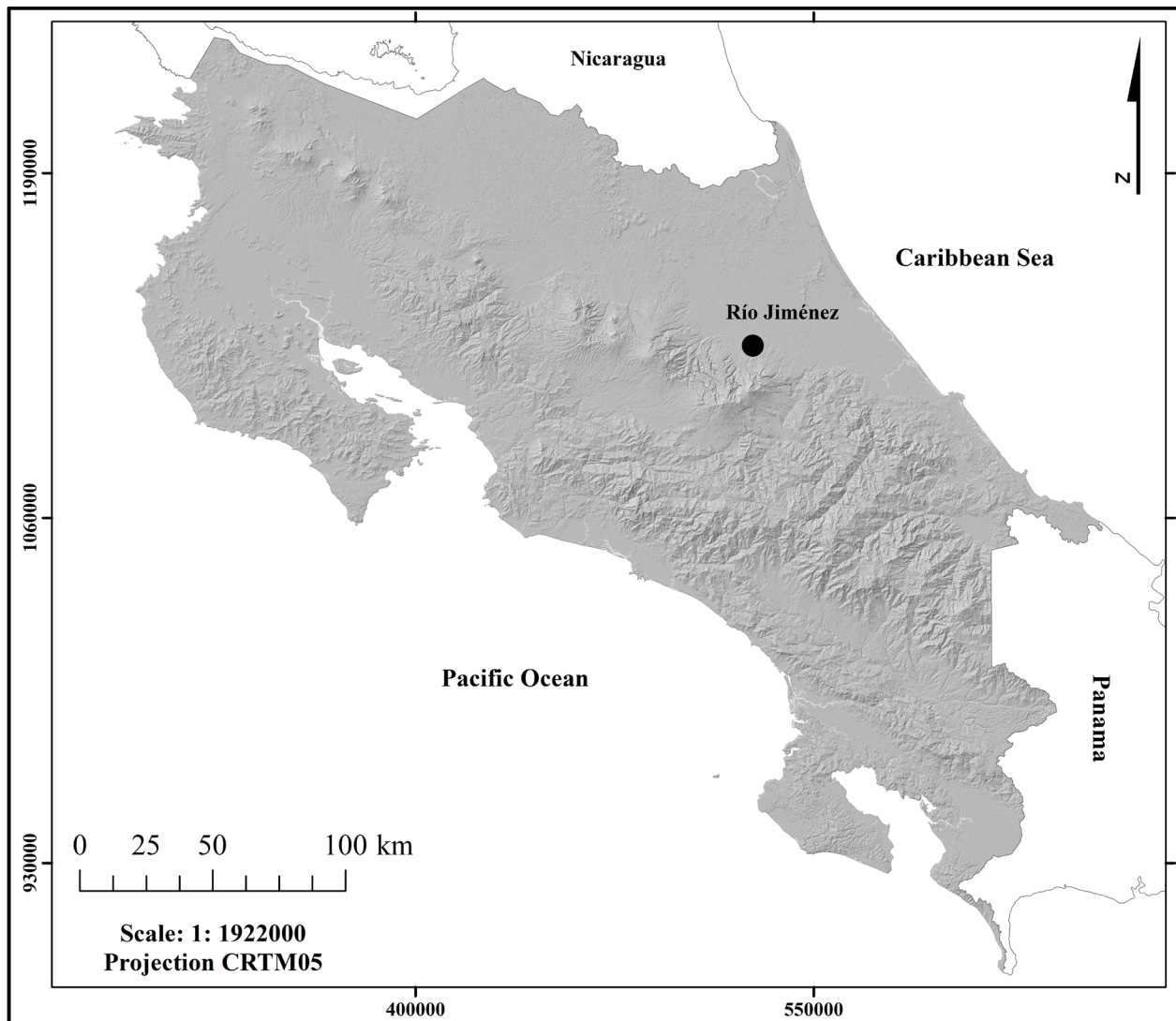


FIGURE 1. Geographic distribution of *Anacroneuria suerre* sp. n in Costa Rica.

Nymph description and associations. Nymphs were collected using D-kick nets, grouped in perforated plastic containers (maximum 4 nymphs per container) which were partially submerged during the sampling time (4-5 hours in total). Once the containers were completely removed from the river, they were placed in plastic bags with water from the site and if necessary, ice was added to maintain similar temperatures as measured in the field (i.e., 23°C).

Nymphs were transported to Laboratorio de Acuicultura Continental, Universidad Nacional, Costa Rica and placed into a rearing system adapted from Novaes *et al.* (2012), which consists of a continuous flow of water system with 27 separate cells; water temperature was controlled using coolers in the collector tank. Additional adult material was collected by using a light trap. Specimens studied are deposited in the following collections: United States National Museum of Natural History, Washington, D.C., U.S.A. (USNM), Laboratorio de Entomología de la Universidad Nacional (LEUNA), Heredia, Costa Rica; Colección de Historia Natural del Museo Nacional de Costa Rica (MNCR), Santo Domingo, Heredia; Museo de Zoología, Universidad de Costa Rica (MZUCR), San Pedro, Costa Rica.

TABLE 1. List of *Anacroneuria* species from Costa Rica with associated nymphs.

Species	Locality	Author
<i>A. benedettoi</i> Stark, 1998	CR, HO, PA	Gutiérrez-Fonseca & Springer, 2011
<i>A. divisa</i> Stark, 1998	CR, NI	Gutiérrez-Fonseca & Springer, 2011
<i>A. holzenthali</i> Stark, 1998	CR, HO, NI	Gutiérrez-Fonseca & Springer, 2011
<i>A. lineata</i> (Stark, 1998)	BE, CR, GU, HO, MX	Gutiérrez-Fonseca & Springer, 2011
<i>A. marca</i> Stark, 1998	CR	Gutiérrez-Fonseca & Springer, 2011
<i>A. maritza</i> Stark, 1998	CR	Stark, 1998
<i>A. perplexa</i> Stark, 1998	BE, CR, GU, HO, MX	Gutiérrez-Fonseca & Springer, 2011
<i>A. suerre</i> sp. n.	CR	described herein
<i>A. talamanca</i> Stark, 1998	CR, NI, PA	Fenoglio, 2007
<i>A. uatsi</i> Stark, 1998	CR, HO	Stark, 1998
<i>A. varilla</i> Stark, 1998	CR, PA	Gutiérrez-Fonseca & Springer, 2011

BE=Belize, CR=Costa Rica, GU=Guatemala, HO=Honduras, MX=Mexico, NI=Nicaragua, PA=Panama.

Results

Anacroneuria suerre **Bravo & Romero, sp nov.**

Material examined. Holotype ♂: Costa Rica, Limón, upper basin Rio Jimenez, 10° 10' 24.38" N, 83° 45 '7.35" W, 428 masl, 02 February-25 September 2018, F. Bravo & D. Romero ((USNM. **Paratypes**, same data as holotype: 1♂ MNCR, 1♂ MZUCR, 1♂ LEUNA.

Adult Habitus: General color brownish. Head with a dark brown spot from behind ocelli to M-line in a broad patch covering occiput, extending forward and lateral of callosities in a light brown tone; M-line discontinuous with well-defined limits; lappets brown and mesal field forward of M-line light brown; antennae brown (Fig. 2A). Pronotum with lateral dark bands; narrow pronotal stripe brown with a median pale line, bordered by light median region, irregular midlateral strips brown with scattered pale rugosities (Fig. 2A). Femora mostly pale distally brown, foreleg tibia and mesothoracic legs mainly yellow with some darkened areas, hind tibia yellowish brown with basal and apical sections darkened, tarsi dark.

Wing membrane brown, veins brown except C and Cs pale. Cerci yellowish with the last four segments dark.

Male. Forewing length 10-12 mm; antennae length 8-9 mm. Hammer thimble shaped, basal diameter slightly wider than at the apex. Aedeagal apex widened at the base, extending forward from broad sinuate shoulders, apically rounded and gradually narrowed forming a scoop; ventral membranous lobes well developed, formed basally by inward folds, aedeagal hooks slender projecting beyond the shoulders (Fig. 2D); dorsal keel poorly developed with two widely separated and apically divergent fringes (Fig. 2F), lateral view of dorsal keel concave (Fig. 2E).

Female. Forewing length 14-15 mm; cerci length 9-10 mm; antennae length 10-11 mm. Subgenital plate with four subequal lobes; external lobes much larger than inner, slightly wider than high; mesal notch deep, V-shaped. Transverse sclerite of sternum nine absent, posterior margin slightly sinuated, mesal sclerite T-shaped scarcely setose and with a marginal patch white and devoid of bristles (Fig. 2C).

Comments. The general habitus of the male of *A. suerre* is most similar to *A. chiriqui* Stark & Armitage, 2018 described from Panama, but the aedeagal apex of the new species has well-defined subapical membranous lobes, and tibiae and tarsi have yellow markings (Fig. 2A). In addition, the lateral lobes in *A. chiriqui* are longer, curved and extending laterally unlike the new species (Fig. 2F); the dorsal aedeagal keel in *A. suerre* is less conspicuous than in *A. chiriqui*, and the hammer is shorter, wider and less distinctly thimble shaped. Furthermore, the inner lobes of the subgenital plate of the female adult of *A. suerre* are more rounded, not acute, as in *A. chiriqui* (Fig. 2C). The new species also resembles the widespread Central American *A. lineata* (Navas, 1924), but it has a thimble shaped hammer and lateral view, the dorsal keel of the aedeagus is concave. Adults of *A. suerre* also differs from both *A. chiriqui* and *A. lineata* by having a medial pronotal brown stripe with a medial pale line in both teneral and fully mature specimens (Fig. 3).

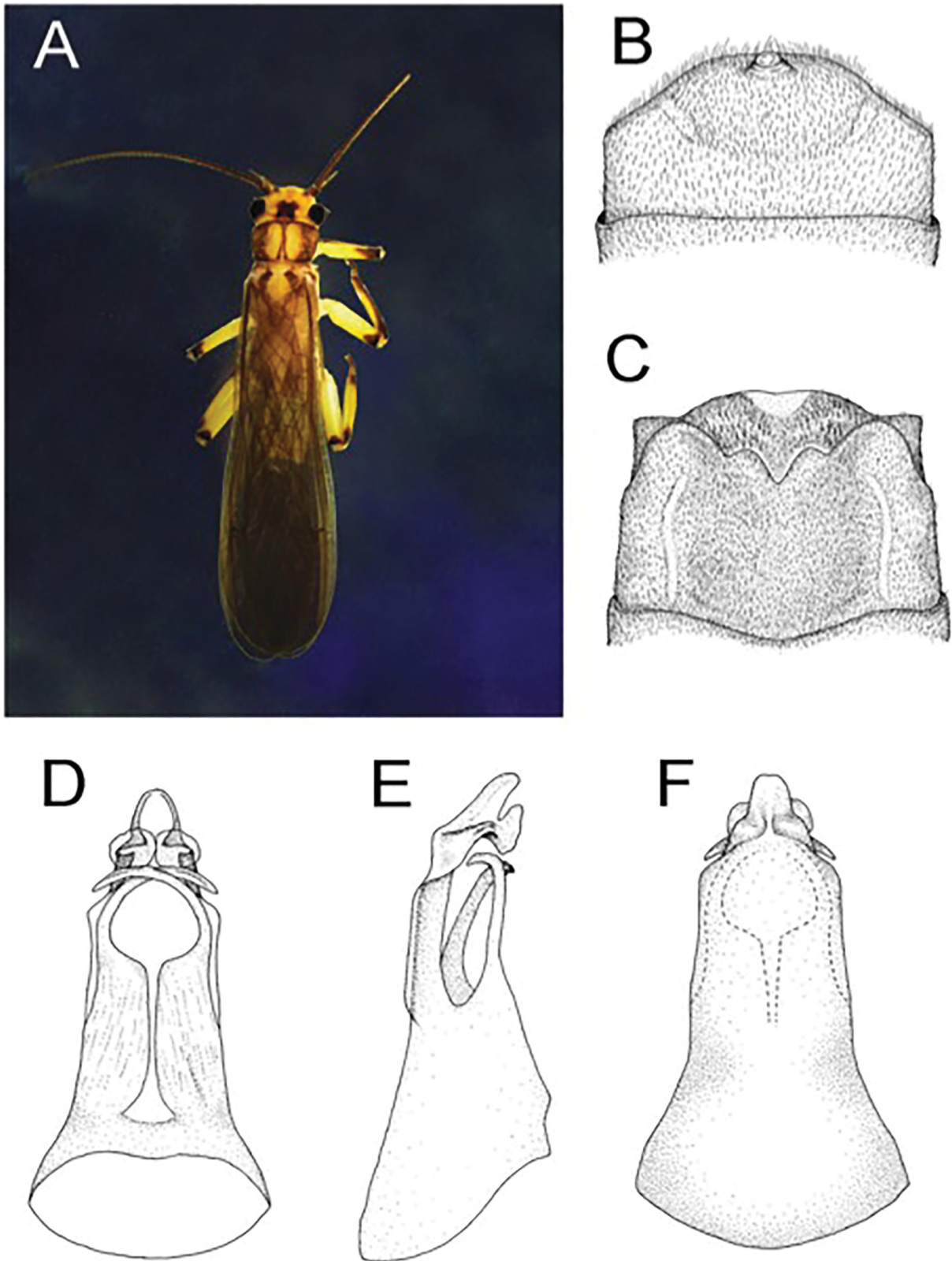


FIGURE 2. *Anacroneuria suerre* sp. n. male-habitus (A) hammer (B), female subgenital plate (C), aedeagus ventral (D), aedeagus lateral (E), aedeagus dorsal (F).



FIGURE 3. *Anacroneuria suerre* sp. n. pronotal stripe brown with median pale line. Teneral adult specimens (A, B), fully developed specimen (C).

Nymph. Body length: 9-10mm. General color brownish in dorsal view, yellow in ventral position. Head brown mainly from ocelli to frontoclypeal region, with lighter areas besides and behind ocelli; “M” line well defined (Fig. 4A). Eyes and ocelli dark. Irregular line of setae from dark spots of the base of head to the posterior dorsal region of the eyes. Labrum, labium, right maxillae, and right mandible as in Figs. 4B, 4C, 4D & 4E. Pronotum rectangular with rounded margins, median line pale, with a pale and well-defined pattern. Edge of pronotum with short and fine bristles. Rest of pronotum dark brown, covered by fine, short and dark hairs.

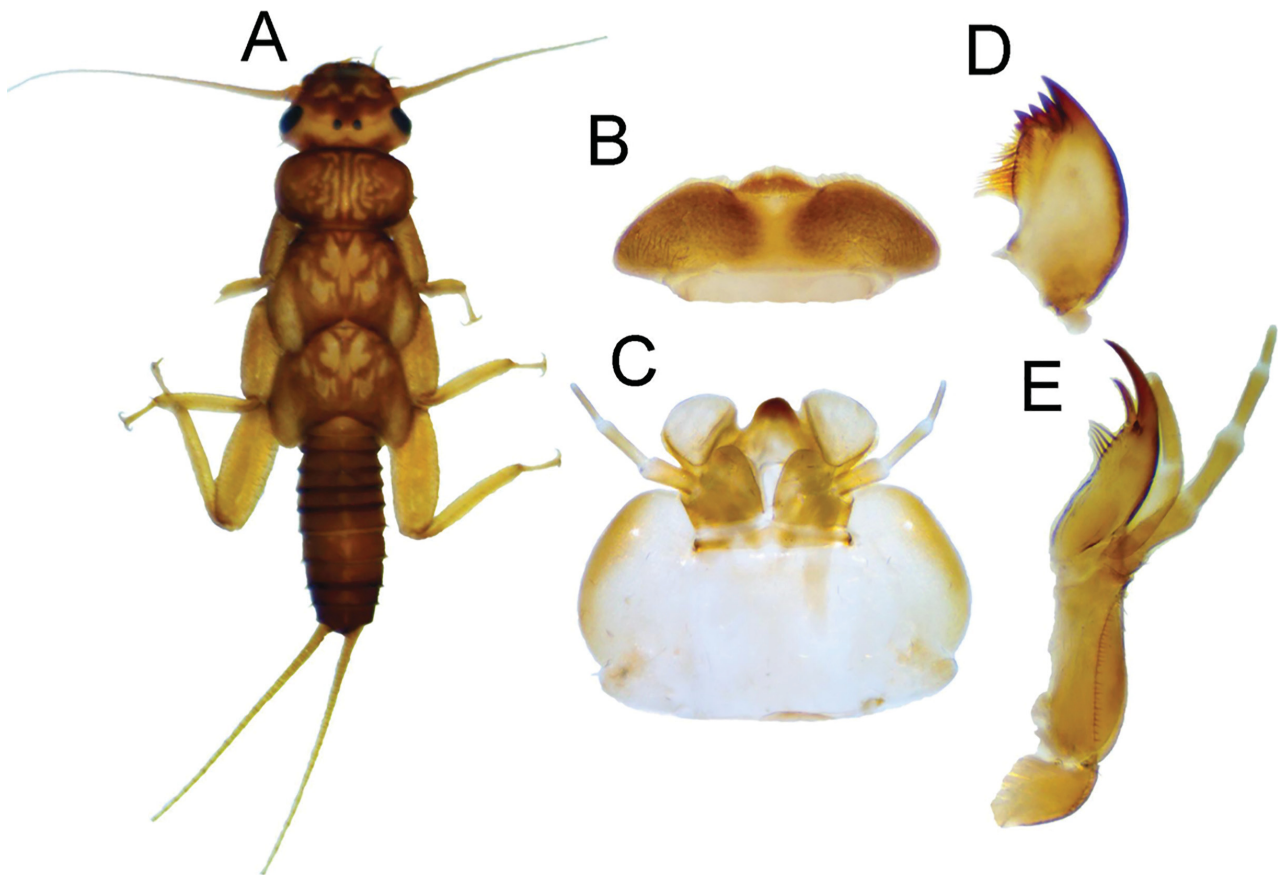


FIGURE 4. Nymph of *Anacroneuria suerre* sp. n.: Body-dorsal (A), labrum (B), labium (C), right mandible (D) & right maxilla (E).

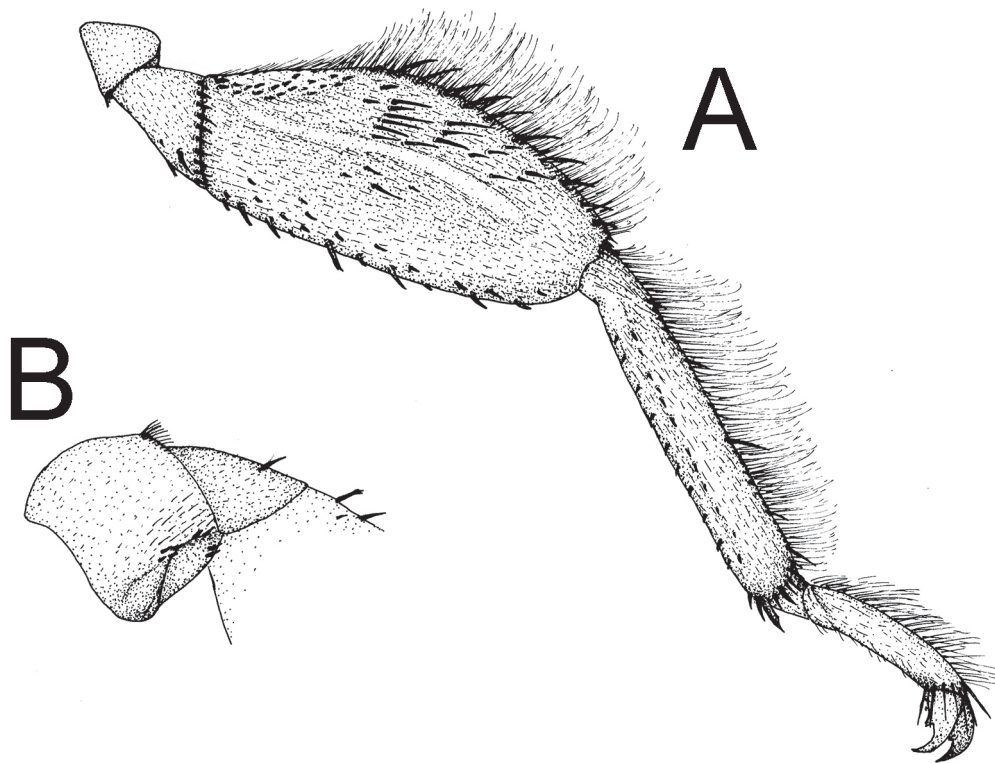


FIGURE 5. Nymph of *Anacroneuria suerre* sp. n.: Foreleg (A), ventral area of coxa (B).

Meso- and metanotum almost alike in color and brown pattern, forming a “v” when mature and pale in apical regions. Median pale spots creating a butterfly-like figure (Figure 4A). Rest of meso- and metanotum dark brown, covered by fine, short and dark hairs. Groups of short but width setae are distributed randomly in these structures. Forefemur with a line of 6-7 bristles, close to the front apical area (Fig. 5A). Median and hairless line that goes over $\frac{3}{4}$ of forefemur longitude. Most of forefemur covered by fine, short and dark hairs. Ventral area of coxa as in Fig. 5B.

Comments. The nymph of *A. suerre* appears most similar to the known nymphs of *A. lineata* and *A. varilla* Stark, 1998, but the “M” line is incomplete and pronotum more rectangular with rounded margins, and not as wide anteriorly.

Etymology. The species name refers to the stream where the specimen was collected, located in the upper basin of Río Jimenez, Limón, Costa Rica. It is used as a noun in apposition.

Discussion

Even though *Anacroneuria* is the predominant stonefly genus occurring in Central America, additional efforts are needed to elucidate not only the diversity of stoneflies in the region (Stark & Armitage, 2018; Gutierrez-Fonseca *et al.*, 2015) but determine nymph-adult associations. Of the 32 species of *Anacroneuria* known from Costa Rica, only 11 species have the nymph associated (Table 1). We recommend either rearing nymphs in the laboratory or using molecular tools (Molina *et al.*, 2017; Almeida *et al.*, 2018) to accurately identify nymphs to the species level.

There has been little use of Plecoptera as a model organism for ecotoxicological studies in the tropics, primarily due to difficulties in transporting and rearing individuals in the laboratory (Almeida *et al.*, 2018). However, in this study, 97% of nymphs survived after transportation and 48 hours of acclimatization. The use of local water and a rearing system with a continuous flow were crucial to this high success. In addition to nymph associations, biologi-

cal factors such as relative abundances of each taxon in study streams are important criteria for ecotoxicological studies (Peterson *et al.* 2001b). We collected 55–60 individual nymphs over a four-hour sampling period. Nevertheless, our success should be taken with caution, since relative densities of different species of *Anacroneuria* at different times of the year may vary and as Jackson and Sweeney (1995) have indicated, species of *Anacroneuria* may be multivoltine. It is worth mentioning that the use of native species in these investigations provides an invaluable representation regarding the ecological roles, functional feeding strategies, taxonomy, and physiology of the community and local aquatic ecosystems (Peterson *et al.*, 2001b), especially in the tropics.

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