

Morphological studies of two *Denisiella* species (Collembola, Symphypleona) with an updated identification key to *Denisiella* species based on males

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Received 3 March 2016 | Accepted 16 September 2016

Published online at www.soil-organisms.de 1 December 2016 | Printed version 15 December 2016

Abstract

For the first time, Scanning Electron Microscopy (SEM) techniques were used to study the morphology of the genus *Denisiella*. Two species were examined in detail: *Denisiella bretfeldi* Schulz & van Harten, 2013 and *D. serroseta* (Börner, 1908). An updated key for identifying *Denisiella* species on the basis of morphological characters of their males is presented including all presently known species world-wide, for which males have been reported.

Keywords *Denisiella* | SEM-photos | key males

1. Introduction

According to Betsch (1980), the genus *Denisiella* Folsom & Mills, 1938 belongs to the family Sminthurididae, which so far holds nearly 150 described species and 10 genera (Palacios-Vargas 2007; Bellinger et al. 1996-2016). The genus is characterized by the absence of tibiotarsal organ III and a distinct modification in the male clasp organ: the antenna exhibiting characteristic trichobothrial elements and spines on antennal segment III and modified setae on antennal segment II (Massoud & Betsch 1972). Further characteristics of *Denisiella* are the presence of serrated spines on tibiotarsi I – III in males and females as well as of setae and a single spine on coxa III. Some *Denisiella* species show bladder-shaped organs on the basis of tibiotarsus I in males (Ospina & Palacios-Vargas 2009). As a rule, the mucro holds two lamellae, of which the inner is serrated and the outer smooth (Ospina & Palacios-Vargas 2009). The males of some species are armed with a nasal organ consisting of two spine-like setae and one big alveolus with four strong setae with fine ciliations (Palacios-Vargas 2007).

Worldwide there are so far ten described species (Table 1). The present study aims at 1) giving additional morphological characters of two *Denisiella* species, which are at present difficult to distinguish and 2) developing an update of the Ospina & Palacios-Vargas identification key to the species of *Denisiella*, based only on males.

2. Materials and methods

All studied specimens were collected in the United Arab Emirates: *D. bretfeldi* origins from Wadi Madaq where specimens were sampled in March 2005 by hand or using water-filled traps, *D. serroseta* from Wadi Wurayah, in December 2006, exclusively via water-filled traps (for details see Schulz & van Harten, 2013). Light microscopy investigation was performed using z-stack imaging (Leica DM 5500 B, Leica Microsystems GmbH, Wetzlar, Germany, 1000x magnification, oil immersion). SEM investigations were carried out with an analytical Scanning Electron Microscope JSM-6510LV (JEOL Germany).

Table 1. Species of *Denisiella* Folsom & Mills, 1938 together with the number of ommatidia they hold, and the countries/regions where they are presently known to occur.

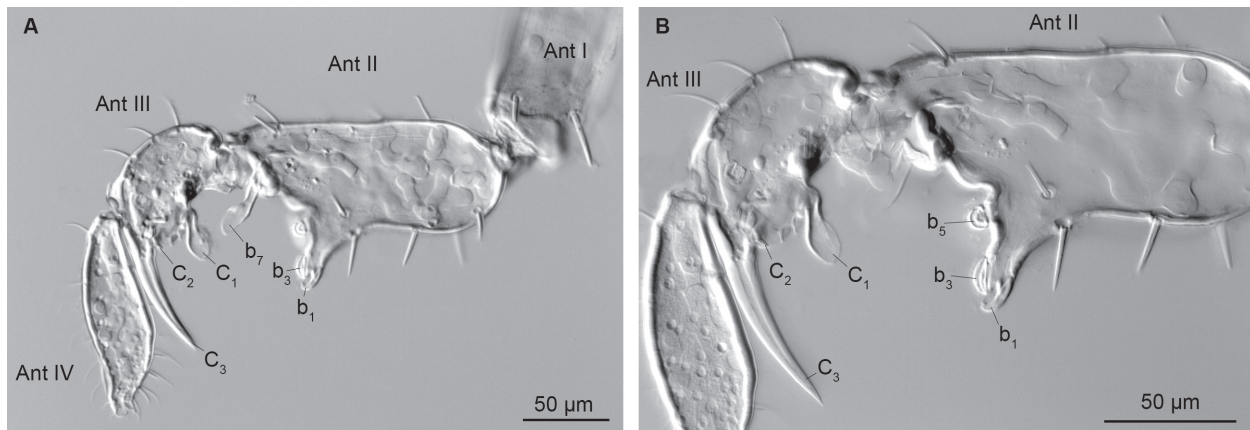
Species	Number of ommatidia	Occurrence
<i>bretfeldi</i> Schulz & van Harten, 2013	6	United Arab Emirates
<i>colombiana</i> Ospina & Palacios-Vargas, 2009	6	Colombia
<i>diomedesi</i> Palacios-Vargas, 2007	6	Panama
<i>lithophila</i> Snider, 1988	6	USA, Georgia
<i>maesorum</i> Palacios-Vargas, 1995	6	Nicaragua
<i>nayarita</i> Palacios-Vargas & Bernava, 1999	6	Mexico
<i>ramosa</i> (Folsom, 1932)	8	Hawaii
<i>seurati</i> (Denis, 1925)	6	Polynesia
<i>serroseta</i> (Börner, 1908)	6	South Africa, United Arab Emirates
<i>sexpinata</i> (Denis, 1931)	8	USA, Mexico, Nicaragua, Costa Rica

3. Results

Light microscopy (Figure 1) and SEM photos (Figures 2 and 3) of the two *Denisiella* species show, on the one hand, all morphological characters important in species differentiation and also the typical characters of the genus. For the determination of males the following morphological characters are of high taxonomical value: presence or absence of (four) bladder-like organs, their shape, (Fig. 2A) and presence or absence of the nasal organ (Fig. 2B). Other characters concern the shape of the mucro (Figs 2C, D) and its lamellae, the organization of the clasping organ with spines and trichobothrial

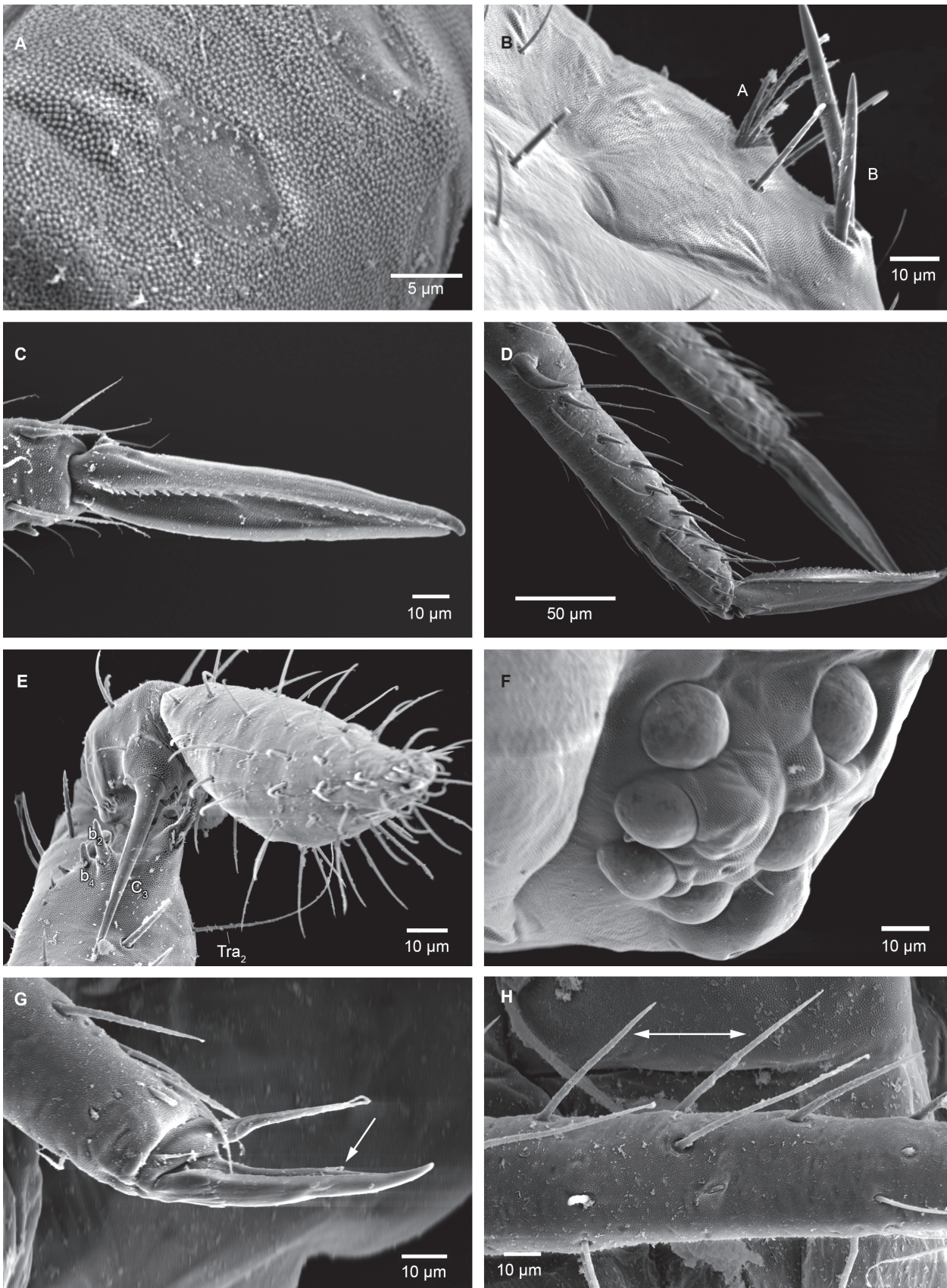
elements (Fig. 2E), the number of ommatidia (Fig. 2F), the presence or absence of an inner tooth on claw III (Fig. 2G) and the shape of the setae on tibiotarsus III (Fig. 2H). Characters of lower taxonomic value are the fourth antennal segment, outer mouth parts (chaetotaxy of the labrum), mandibles and the surface structure of eversible vesicles of the ventral tube (Figs 3C–F).

There are two recent keys to the world's *Denisiella* species: Bellinger et al. 1996–2016 and Ospina & Palacios-Vargas 2009. The identification key published by Ospina & Palacios-Vargas 2009 was used to develop an updated key to *Denisiella* species which is based solely on morphological characters of male specimens.



▲ **Figure 1 (A, B):** Light microscopy z-stack images (1000x, oil immersion, DIC) of the clasping organ of *Denisiella bretfeldi*. Important elements are designated: Antennal segment III with club-shaped element C1, spine-shaped seta C2 and strong spine C3, Ant II with modified club-shaped elements b_1 , b_3 , b_5 and b_7 .

► **Figure 2:** Scanning electron microscopy details of *Denisiella bretfeldi* and *Denisiella serroseta*. (A) *D. bretfeldi* male: Three of the four oval bladder-shaped organs, which are nearly twice as long as broad and not elevated. (B) *D. bretfeldi* male: Lateral view of nasal organ (A – three of the four strong setae visible in one big alveolus, B – 2 spine-like setae of different size). (C) *D. bretfeldi* female: Mucro (inner side serrated, outer smooth). (D) *D. serroseta* female: Dens with spine-like setae in a row and the typical narrow mucro. (E) *D. bretfeldi* male: Details of clasping organ (Antennal segment III with strong spine C_3 , Ant II with spines b_2 and b_4 and trichobothrial element Tra.). (F) *D. serroseta* female: 6 + 6 ommatidia of equal size. (G) *D. bretfeldi* female: Claw III with inner tooth (arrow). (H) *D. bretfeldi* female: Tibiotarsus III with rough (not serrated) setae.



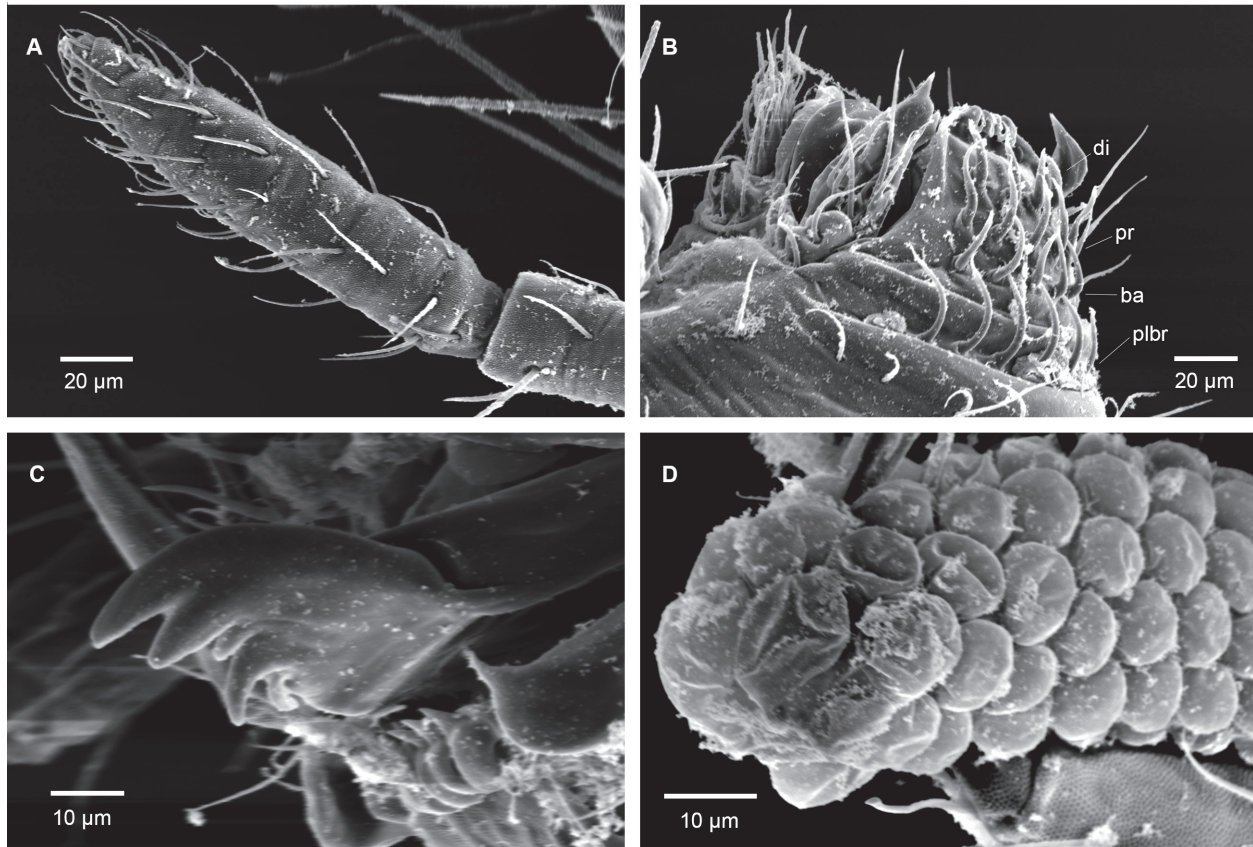


Figure 3: Scanning electron microscopy details of female *Denisiella bretfeldi*. (A) Ant IV with slightly thickened setae. (B) Labrum with 6 praelabral (plbr), 5 basal (ba), 5 proximal (pr) and 4 distal (di) setae. (C) Mandible with 4 teeth. (D) Terminal structure of an eversible vesicle of the ventral tube.

Updated key of *Denisiella* males

Male specimens having clasping organ on antennae (Figs 1A, B), thus being easily discernible from females. Three species of *Denisiella* (*D. nayarita*, *D. seurati* and *D. sexpinnata*) are not included since their males were not found so far.

- | | | |
|---|---|----------------------|
| 1 | Tibiotarsus I with bladder-shaped organs (Fig. 2A) | 2 |
| - | Tibiotarsus I without bladder-shaped organs | 6 |
| 2 | Bladder-shaped organs elongate (nearly four times as long as broad) | <i>D. ramosa</i> |
| - | Bladder-shaped organs oval (nearly twice as long as broad, (Fig. 2A) or roundish | 3 |
| 3 | Bladder-shaped organs small-oval & highly elevated; Nasal organ present (Fig. 2B); Tibiotarsus III with four serrated setae, claw III with inner tooth (similar to <i>D. bretfeldi</i> , Fig. 2G) | <i>D. diomedesi</i> |
| - | Bladder-like organs small-oval (slightly elevated) or large-oval; Tibiotarsal setae and nasal organ variable; Claw III with or without inner tooth | 4 |
| 4 | Bladder-like organs small-oval (slightly elevated); Nasal organ absent; Tibiotarsus III with four serrated setae, claw III without inner tooth | <i>D. maesorum</i> |
| - | Bladder-like organs large-oval (Fig. 2A), not highly elevated | 5 |
| 5 | Nasal organ present; Tibiotarsus III with not-serrated setae (Fig. 2H) | <i>D. bretfeldi</i> |
| - | Nasal organ absent; Tibiotarsus III with four strong serrated setae | <i>D. colombiana</i> |
| 6 | Tibiotarsus III with 3 strong serrated setae, claw III with inner tooth (similar to <i>D. bretfeldi</i> , Fig. 2G) | <i>D. lithophila</i> |
| - | Tibiotarsus III with not-serrated setae (Fig. 2H), claw III without inner tooth | <i>D. serroseta</i> |

4. Discussion

One of the characters difficult to discern in *Denisiella* species is the number of ommatidia. Describing *D. ramosa*, Folsom (1932) stated for females: 'Eyes at least twelve (fig. 113), possibly sixteen'. Subsequently Christiansen & Bellinger (1992) noted for the same species that it has '6 + 6 large clear eyes; 2 + 2 small inner eyes present at least as corneas, (Plate 138G)'. The same was reported by Denis (1931) for *D. sexpinnata*: '8 cornéules par côté, dont deux (C et D) mal formées'. The genus *Denisiella* belongs to the family Sminthurididae. According to the diagnosis of Sminthuridida proposed by Bretfeld (1999) 'Each eye-patch usually has 6 normal ommatidia and 2 smaller ones (C, D), the latter may be completely reduced'. After the present SEM investigation of two species of *Denisiella* it can be concluded that the genus *Denisiella* is a characteristic representative of Sminthurididae. Both species under study show 6 + 6 normal-sized ommatidia (Table 1, Fig. 2F). The two smaller ommatidia are reduced except for *D. ramosa* and *D. sexpinnata* showing 8 + 8 ommatidia, of which 2 + 2 are very small and difficult to see. All other important characters for separation of *Denisiella* species e. g., the four bladder-shaped organs, the nasal organ and the serrated setae of tibiotarsus III, are easier to identify, even in older collections, in alcohol as well as on permanent slides.

5. Acknowledgements

I would like to express many thanks to Jacqueline Gitschmann for the excellent revising of the SEM photos and to Diana Goernert for preparing the digital photos of the clasping organ.

6. References

- Bellinger, P. F., K. A. Christiansen, & F. Janssens (1996-2016): Checklist of the Collembola of the World [http://www.collembola.org].
- Betsch, J.-M. (1980): Éléments pour une monographie des Collemboles Symphypleonés (Hexapodes, Aptérygotes). – Mémoires du Muséum National d'Histoire Naturelle, Nouvelle Série A, Zoologie **116**: 1–227.
- Bretfeld, G. (1999): Synopses on Palaearctic Collembola. Symphypleona. – Abhandlungen und Berichte des Naturkundemuseum Görlitz **71**: 1–318.
- Christiansen, K. & P. Bellinger (1992): Insects of Hawaii. Collembola. – University of Hawaii Press, Honolulu **15**: 1–445.
- Denis, J. R. (1931): Contributo alla conoscenza del "Microgenton" di Costa Rica. – Bolletino del Laboratorio di Zoologia Generale e Agraria della R. Scuola Superiore d'Agricoltura in Portici **25**: 60–170.
- Folsom, J. W. (1932): Hawaain Collembola. – Proceedings of the Hawaiian Entomological Society **8**: 51–80.
- Massoud, Z. & J.-M. Betsch (1972): Étude sur les Insectes Collemboles II. – Les caractères sexuels secondaires des antennes des Symphypléones. – Revue d'Ecologie et de Biologie du Sol **IX**, 1: 55–97.
- Ospina, M. & J. G. Palacios-Vargas (2009): A new *Denisiella* Folsom and Mills, 1938 (Collembola: Sminthurididae) from Colombia. – Zootaxa **2168**: 63–68.
- Palacios-Vargas, J. G. (2007): A new species of *Denisiella* (Collembola: Sminthurididae) from Panama and new records for *D. sexpinnata* (Denis, 1931). – Zootaxa **1637**: 63–68.
- Schulz, H.-J. & A. van Harten (2013): Subclass Collembola, order Symphypleona. – Arthropod Fauna of the UAE **5**: 13–21.

