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Short communication

Illustrated redescription of the male of Ixodes simplex Neumann, 1906

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ARTICLEINFO	A B S T R A C T
A R T I C L E I N F O Keywords: Bat-specialist ticks Identification Ixodes ariadnae I. vespertilionis Morphology	<i>Ixodes simplex</i> is a tick species of bats, mainly reported from Schreibers' Bent-winged bat, <i>Miniopterus schreibersi</i> , all over its host's range. While being a common parasite of cave-dwelling bat species, the initial description of males is poor (based on a sole damaged specimen), lacking distinctive features. This study describes the male of <i>I. simplex</i> also providing measurements for a number of characters, using a series 10 of specimens. Males of <i>I. simplex</i> show the following characteristics distinct from males of <i>I. ariadnae</i> and <i>I. vespertilionis</i> : idiosoma is shorter (2.6 mm vs. 3.4–4.7 mm); the palps of <i>I. simplex</i> males are laterally curved (bent) (while straight in <i>I. ariadnae</i> and gradually curving in <i>I. vespertilionis</i>), the genital opening is anterior in position between coxae II (whereas posterior to second intercoxal space in <i>I. vespertilionis</i>); coxae II are to some extent rectangular (vs. rounded in <i>I. ariadnae</i>); coxae IV have a prominent tuft of long hairs (missing in the other two species). Most importantly, the legs of <i>I. simplex</i> males are considerably shorter than in males of the other two species (2.7–3.3 mm vs. 7–10 mm). The description is completed with high-quality pictures highlighting all the mentioned features.

1. Introduction

The male of *Ixodes simplex* was described by J. C. Beaucournu (Beaucournu, 1962), but it is based on a single long-preserved and dried specimen. Even the author notes that the morphological characters described (and especially the biometrical measurements) are very basic (Beaucournu, 1962), and there is no drawing of the idiosoma. While *I. simplex* was later redescribed, the relevant sources either lack the description of the male (Yamaguti et al., 1971), or the male is poorly illustrated and its description is provided with few measurements, which are accessible only in Russian language (Filippova, 1977).

As the *Ixodes* species associated with bats are currently re-examined (initiated by the discovery of *I. ariadnae* in Europe (Hornok et al., 2014), there is a need for accurate and in-depth description of certain stages. We herein re-describe the male of *I. simplex*, based on material collected from nearby a colony of Schreibers' Bent-winged bat, *Miniopterus schreibersii*, the most commonly reported host for this tick species.

2. Materials and methods

2.1. Sample collection

Ticks were removed from the cave wall in a disused mine gallery, close to Băneasa, SE Romania (44.06776N; 27.64455E) hosting a large reproductive colony of Schreibers' Bent-winged bats. Ticks were collected individually or in copulating pairs on 22 May 2017 (altogether 157 ticks were collected, belonging to all development stages). After collection, the ticks were stored individually in vials containing 96% ethanol. Tick identification was performed in the lab, using morphological keys (Beaucournu, 1962, Yamaguti et al., 1971).

2.2. Sample analyses

Measurements of *I. simplex* males were performed with an Olympus BX61 microscope, using a DP72 digital camera equipped with the CellF software (Olympus Corporation, Tokyo, Japan). Ten intact males were selected for the measurements (from a total of 44 collected) and were measured 30 days after the collection. They were cleansed in water and placed individually on microscope slides, covered with cover slides,

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Fig. 1. Important morphological characters of male *Ixodes simplex*. Arrows point at structures referred to in the text according to encircled letters. I. Dorsal view. II. Ventral view showing gnathosoma and coxae. III. Enlarged dorsal view of the gnathosoma.

without fixation. For clarification we used one drop of Lactofenol, placed directly to the samples. Pictures of the same males were made with a VHX-5000 (Keyence Co., Osaka, Japan) digital microscope, and were used here for the figures.

3. Results

The sizes in the descriptions below are provided in millimetres, including the range and the means (in parentheses) of evaluated specimens.

3.1. Ixodes simplex Neumann, 1906, male

Length of the idiosoma (from the half point between scapular apices

to the posterior margin) 2.30–2.87 (2.56), breadth 1.45–1.77 (1.58), ratio of idiosomal length/breadth 1.49–1.90 (1.62). Conscutum elongated, in shape ovoid (to some extent hexagonal) with slightly converging postero-lateral edges (Fig. 1: I.a), broadest at or anterior to its mid-length. Length of scutum 2.11–2.67 (2.35), breadth 1.27–1.59 (1.47), ratio length/breadth 1.46–1.72 (1.6). Conscutum with scattered punctuations, and anteriorly with shallow cervical grooves (Fig. 1: I.b), rounded scapulae (Fig. 1: I.c). Idiosoma has sparse hair covering, more pronounced posteriorly (Fig. 1: I.d) and in perianal region. Genital aperture broad (> 0.1) and slightly curved backwards, in an anterior position between 2nd coxae (Fig. 1: II.a). Spiracular plates rounded (0.21–0.25 in diameter), with excentric opening. Anal groove posteriorly convergent.

Length of gnathosoma (from palpal apices to posterior margin of basis capituli) 0.53-0.60 (0.56), width of basis capituli dorsally 0.32-0.34 (0.33). Ratio of gnathosomal length to basis capituli width 0.67-0.87 (0.74). Basis capituli dorsally broadest anteriorly, at the palpal base, converging backwards, with broad U-shaped sclerotized posterior edge elevated behind two fields of scattered pores (Fig. 1: III.a). Basis capituli ventrally elongated, converging backwards, lightly colored posteriorly to a semi-circular edge (Fig. 1: II.b). Palps short, length 0.22-0.26 (0.24), breadth 0.11-0.13 (0.12), ratio length/ breadth 1.86-2.2 (2.0). Separation of palpal segments II-III indistinct. Segments II-III broad and laterally curved (bent) in an obtuse angle, anteriorly with nearly parallel edges (Fig. 1: III.b). Palpal hairs anteriorly short (0.02-0.03), laterally long (0.05-0.08) (Fig. 1: III.c). Hypostome shorter than palps, anteriorly bifid (bilobed), its length 0.13-0.15 (0.14), breadth 0.11-0.12 (0.117), ratio length/breadth 1.19-1.21 (1.20). Teeth underdeveloped (ill-defined).

Legs short (legs I–III: mean length 2.7–3, leg IV 3.3). Coxae lack spines or spurs. Coxa I nearly triangular, coxa II rounded rectangular, and coxae III–IV medially semi-circular, with few hairs, except coxa IV with a row of 10–13 hairs in a tuft (Fig. 1: II.c). Tarsus I. 0.51–0.57 (0.54). Haller's organ shorter than maximum diameter of tarsus I (Fig. 1: II.d).

3.2. Differential diagnosis from the males of other bat-associated tick species

Males of *I. simplex* show the following distinctive characteristics from males of *I. vespertilionis* (as shown and described in (Hornok et al., 2016a): idiosoma is shorter (2.6 mm vs. 4.7 mm); the genital opening is anterior in position between coxae II (whereas posterior to second intercoxal space in *I. vespertilionis*); the palps of *I. simplex* males are laterally curved (bent) in an obtuse angle (while gradually curving in *I. vespertilionis*); coxae IV have a prominent tuft of long hairs (missing in *I. vespertilionis*). Most importantly, the legs of *I. simplex* males are considerably shorter than those of *I. vespertilionis* males (2.7–3.3 mm vs. 8–10 mm).

Males of *I. simplex* show the following distinctive characteristics from males of *I. ariadnae* (Hornok et al., 2016a) idiosoma is shorter (2.6 mm vs. 3.4 mm); the palps of *I. simplex* males are laterally curved (bent) (while straight in *I. ariadnae*); coxae II are to some extent rectangular (vs. rounded in *I. ariadnae*); coxae IV have a tuft of long hairs (unlike in *I. ariadnae*); and the legs of *I. simplex* males are much shorter than those of *I. ariadnae* males (2.7–3.3 mm vs. 7–8 mm). We found no significant differences between the measurements reported by Beaucournu, (1962) and those measured by this study (Table 1.).

4. Discussion

A Pan-European survey among tick researchers (Estrada-Peña et al., 2017) revealed high degrees of misidentification rates for tick species non-familiar for the experts. This phenomenon was especially evident for development stages rarely seen by tick researchers. The authors also highlighted the need for good quality and accessible references for less

Table 1

Morphological measurements of male *Ixodes simplex* from Beaucournu (1962) and the present study. All measurements are in mm, showing the range and the mean for the 10 individuals measured.

	Beaucournu (1962) (n = 1)	This study (n = 10)
Idiosoma length	2.35	2.30-2.87 (2.56)
Idiosoma breadth	1.4	1.45-1.77 (1.58)
Gnathosoma length	0.56	0.53-0.60 (0.56)
Basis capituli width	0.38	0.32-0.34 (0.33)
Palp length	0.21	0.22-0.26 (0.24)
Palp breadth	0.13	0.11-0.13 (0.12)
Diameter of spiracular plate	0.25	0.21-0.25 (0.24)
Length of tarsus I	0.54	0.51–0.57 (0.54).

familiar tick species (Estrada-Peña et al., 2017). With the pictures and the detailed description of morphological characters this study provides primarily a much-needed re-description, which make the future identification of *I. simplex* males a straightforward task, completing this way the series of descriptions of bat-related tick species (Hornok et al., 2016a, 2015). While bat specialist ticks may seem less interesting from a zoonotic point of view, a series of pathogens molecularly identified in *I. simplex* in Hungary and Romania recently (Hornok et al., 2016b) may confer a particular importance to this species.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at https://doi.org/10.1016/j.ttbdis.2018.05.011.

References

- Beaucournu, J.C., 1962. Description du mâle d'Ixodes simplex simplex NEUMANN, 1906, parasite de Chiroptères. Bull. Soc. Zool. Fr. 87, 127–131.
- Estrada-Peña, A., D'Amico, G., Palomar, A.M., Dupraz, M., Fonville, M., Heylen, D., Habela, M.A., Hornok, S., Lempereur, L., Madder, M., Núncio, M.S., Otranto, D., Pfaffle, M., Plantard, O., Santos-Silva, M.M., Sprong, H., Vatansever, Z., Vial, L., Mihalca, A.D., 2017. A comparative test of ixodid tick identification by a network of European researchers. Ticks Tick Borne Dis. 8, 540–546. http://dx.doi.org/10.1016/ i.ttbdis.2017.03.001.

Filippova, N.A., 1977. Ixodid Ticks (Ixodinae). Fauna of the USSR, vol. IV/4.

- Hornok, S., Kontschán, J., Kováts, D., Kovács, R., Angyal, D., Görföl, T., Polacsek, Z., Kalmár, Z., Mihalca, A.D., 2014. Bat ticks revisited: *Ixodes ariadnae* sp. nov. and allopatric genotypes of *I. vespertilionis* in caves of Hungary. Parasite Vectors 7, 202. http://dx.doi.org/10.1186/1756-3305-7-202.
- Hornok, S., Kontschán, J., Estrada-Peña, A., de Mera, I.G.F., Tomanović, S., de la Fuente, J., 2015. Contributions to the morphology and phylogeny of the newly discovered bat tick species, *Ixodes ariadnae* in comparison with *I. vespertilionis* and *I. simplex*. Parasite Vectors 8, 47. http://dx.doi.org/10.1186/s13071-015-0665-0.
- Hornok, S., Kováts, D., Angyal, D., Dányi, L., Kovács, R., Kontschán, J., 2016a. Description of the male and the larva of *Ixodes ariadnae* Hornok, 2014. Ticks Tick Borne Dis. 7, 1252–1255. http://dx.doi.org/10.1016/j.ttbdis.2016.07.005.
- Hornok, S., Szőke, K., Kováts, D., Estók, P., Görföl, T., Boldogh, S.A., Takács, N., Kontschán, J., Földvári, G., Barti, L., Corduneanu, A., Sándor, A.D., 2016b. DNA of piroplasms of ruminants and dogs in ixodid bat ticks. PLoS One 11, e0167735. http:// dx.doi.org/10.1371/journal.pone.0167735.
- Yamaguti, N., Tipton, V.J., Keegan, H.L., Toshioka, S., 1971. Ticks of Japan, Korea, and the Ryukyu islands. Brigham Young Univ. Sci. Bull. Biol. Ser. 15, 1–226.