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A new species *Anthrenus bakaloudisi* sp. nov. (Coleoptera, Dermestidae, Megatominae) from Macedonia, Greece and comparison with *Anthrenus pfefferi* Kalík, 1954, *Anthrenus delicatus* Kiesenwetter, 1851, and *Anthrenus warchalowskii* Kadej, Háva & Kalík, 2007

Graham J. Holloway, George Thanasoulias, Andreas Herrmann

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A new species *Anthrenus bakaloudisi* sp. nov. is described from Greece. Images of external and internal features are illustrated. The aedeagus of *A. bakaloudisi* has slim parameres so is compared with species that also have slim parameres from middle and eastern Mediterranean and just into Asia: *A. pfefferi*, *A. delicatus*, and *A. warchalowskii*. The new species differs internally from *A. pfefferi* in subtle ways, but it is easy to distinguish between the species using external colour pattern. *Anthrenus delicatus* differs from *A. bakaloudisi* internally and ways of distinguishing between the two species externally are described. *Anthrenus warchalowskii* is only known from Iran and differs from *A. bakaloudisi* internally. No information could be found on the likely true range of *A. bakaloudisi*, so the species is currently only known from Macedonia, Greece in the region of Thessaloniki.

Key words: Carpet beetle, Anthrenini, aedeagus, sternite IX, dissection, identification

Graham J. Holloway and George Thanasoulias, Cole Museum of Zoology, School of Biological Sciences, HLS Building, The University of Reading, Whiteknights, Reading, Berkshire, GG6 6EX, UK; e-mail: g.j.holloway@reading.ac.uk;
ORCID: https://orcid.org/0000-0003-0495-0313

Andreas Herrmann, Bremervörder Strasse 123, 21682 Stade, Germany. e-mail: herrmann@coleopterologie.de;
ORCID: https://orcid.org/0000-0001-5700-1125

INTRODUCTION

The family Dermestidae Latreille, 1804 contains over 1800 species (Háva 2023), of which over 280 species belong to the genus *Anthrenus* Geoffroy, 1762 (Háva 2023). The genus is broken into 10 subgenera, but research suggests that the taxonomy of the genus has yet to be resolved since many of the subgenera are polytypic (Kadej 2018).

The one subgenus that Kadej (2018) concludes is monotypic is *Anthrenus* s. str. with over 70 species. Over one third of the species within *Anthrenus* s. str. belong to the Palaearctic *A. pimpinellae* (Fabricius, 1775) complex (Háva 2023). Nearly all species within the complex are typified by possession of a sub-basal elytral band consisting of white or cream colour scales (Holloway & Cañada Luna 2022; Herrmann 2023) which has made species recognition a difficult process. Beal (1998) found that aedeagal structure varied among several Nearctic species that also have white subbasal elytral bands. Kadej et al. (2007) extended Beal's (1998) work and applied dissection to the Palaearctic *A. pimpinellae* complex species and found three new species. Kadej & Háva (2011) found three more species and Holloway (2019, 2020, 2021) added a further three species bringing the total number of species in the complex to 23. Unquestionably, there remains more species in the complex still to discover and other taxonomic issues to resolve (Holloway et al. 2020).

Dissection has become a fundamental technique in the study of the *A. pimpinellae* complex. Some species resemble each other externally, for example *A. pimpinellae* and *A. amandae* Holloway, 2019 which drove some authors to declare they were synonymous despite difference at the genital level (Háva & Herrmann 2019, 2020). *Anthrenus amandae* has now been widely accepted as a valid species, but others, such as *A. chikatunovi* Holloway, 2020 and *A. isabellinus* Küster, 1848, continue to be confused (Háva 2023). Confusion occurs because the genitalia sometimes vary among species in subtle ways. Another issue is that several species have colour variants, such as *A. delicatus delicatus* Kiesenwetter, 1851 and *A. d. armstrongi* Mroczkowski, 1952 (see Herrmann 2023), or colour pattern plasticity such as *A. isabellinus* (Holloway et al. 2022).

Kadej et al. (2007) illustrated 17 *A. pimpinellae* complex species. Most of them differed obviously from each other, but the aedeagi of two species, *A. delicatus* and *A. pfefferi* Kalík, 1954, appeared to be very similar. The original aim of this study was to describe *A. delicatus* and *A. pfefferi* in detail to establish whether *A. pfefferi* is just a variant of *A. delicatus*. In the pursuit of this aim, a new species, *Anthrenus bakaloudisi*, was discovered and described here.

MATERIALS AND METHODS

Specimens of *A. delicatus* var. *delicatus* (hereafter referred to as *A. delicatus*) were collected from Greece in 2019 and 2021 and supplemented with specimens from Andreas Herrmann's collection (AHEC) and the Natural History Museum, London (NHML). *Anthrenus pfefferi* specimens were obtained from AHEC and NHML. All specimens were macerated in a solution of 2% acetic acid for five days to soften and where required to enable removal from staging prior to dissection. Dissection was carried out under a Brunel BMSL zoom stereo LED microscope and involved detaching the abdomen from the rest of the insect using two entomological pins. The soft tergites were then peeled away from the harder ventrites to expose the genitalia. For males, the aedeagus was detached from the ring sclerite, and then sternite IX was detached from the ring sclerite and the aedeagus. Females were similarly dissected to confirm sex, but no further examination of female genitalia was carried out. Images of male and female habitus, both upper and under sides, were captured at ×20 magnification using a Canon EOS 2000D camera mounted on the BMSL microscope. Images of aedeagi and (male) sternite IX were captured at ×200 magnification for measurement using a Canon EOS 1300D camera mounted on a Brunel monocular SP28

microscope. After dissection, all body parts were mounted on card. The antennae were teased out (where possible) and images were taken at ×200 magnification through the SP28 microscope. All images were fed through Helicon Focus Pro version 8.0 focus-stacking software. All measurements were made using DsCap.Ink software version 3.90.

Measurements taken:

- Body length (BL): distance from anterior margin of pronotum to the apex of the elytra.
- Body width (BW): maximum distance across the elytra
- Antennal club length (AL): length of the last three antennomeres
- Antennal club width (AW): maximum width across the terminal antennomere
- Paramere length (PL): distance from the anterior end of the parameres to the apex of the parameres
- Sternite IX length (SL): distance from the tip of one anterior horn to the tip of the posterior lobe

The data for the distribution maps (Shorthouse 2010) were derived from field collection sites and data on specimens from AHEC. Statistical analysis (General Linear Modelling) was carried out using Minitab version 21.2.

RESULTS

***Anthrenus* (*Anthrenus*) *bakaloudisi* sp. nov.**
(Figs 1, 2)

Specimens examined. Holotype (male) NHML. Thessaloniki, Macedonia, Greece (40.656030, 22.978385), 8th May 2019, D.E. Bakaloudis leg.
Paratypes, (two female) Thessaloniki Greece (40.656030, 22.978385), 8th May 2019, D.E. Bakaloudis leg, (one female paratype NHML), (one male, four females) Serres, Greece (41.095242, 23.543851), 5th May 2021 G. Thanasoulias leg (six paratypes in authors' collections).

Holotype habitus (Fig. 1A) (BL = 2.654 mm, BW = 1.850 mm), antenna (Figure 1E) (AL = 171 µm, AW = 140 µm).

Description holotype, external characteristics. *Anthrenus* (*Anthrenus*) *bakaloudisi* (Fig. 1A) has a single dark amber ocellus in the middle of the head just below the level of the top of the eyes (an ocellus is a feature of all species within Dermestidae, except for *Dermestes*) and an emarginated inner side of the eye, typical of the subgenus *Anthrenus*. The body integument is very dark, black or dark brown. The dorsal surface of the habitus is covered in white, pale brown and black (or very dark brown) scales. The scales are parallel sided evenly round at the posterior end. The anterior end of each scale is hidden as the scales lie over each other like tiles. Most of the white scales form a sub-basal elytral fascia which is broad at the outer elytral margin, becoming even broader just inside each margin, and then becoming progressively narrower as it sweeps up towards the small, dark, triangular scutellum. More patches of white scales on each elytron are sub-marginal just below the white fascia, on the margin halfway between the sub-marginal spot and the elytral apex, sub-apical, and next to the elytral suture loosely joined to the white fascia. The white fascia is surrounded by very dark scales. Most of the scales along the base of each elytron are pale brown. The elytral margins below the fascia carry pale brown scales down to the marginal white patch, with a larger patch of pale brown scales from the marginal white patch spreading across half of each elytron towards the suture. From the white marginal patch, the margins are covered in dark scales to the elytral apex where most pale brown scales are found which extend as a narrow line up

the elytral suture. On the elytral disc below the fascia three loose lines of pale brown scales are found orientated anterior posterior. All pale brown (and white) scales below the fascia are set in a background of very dark scales.

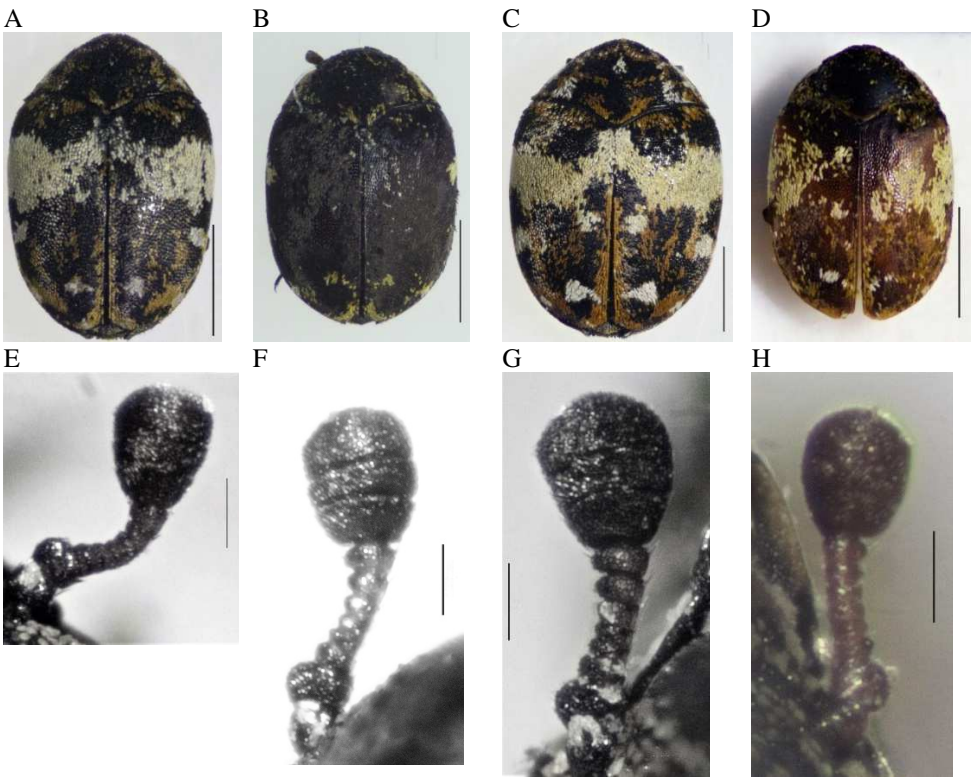

Fig. 1. Habitus and antenna, A and E: *Anthrenus bakaloudisi* **sp. nov.**, B and F: *Anthrenus pfefferi*, C and G: *Anthrenus delicatus*, D and H: *Anthrenus warchalowskii*, respectively. Scale bars: habiti = 1 mm, antennae = 100 µm

The pronotum also carries dark, pale brown and white scales. The pronotal disc is covered in dark scales, the pale brown and white scales concentrated largely at the margins. The few white scales are found towards the outer corners of the pronotum and surrounded completely by pale brown scales. More pale brown scales are found along the anterior margin of the pronotum forming circular patterns around patches of dark scales.

The head is mostly covered in dark scales with most of the pale brown scales concentrated along the inner edges of the eye around the emargination. The legs are brown. The femora are covered in scales on the anterior face, the tibiae and tarsi are largely devoid of scales.

The sternites are covered in white scales. The outer margins of sternites II-V carry spots of black scales. The spots on sternite II are large, larger than those of sternite III, which in turn are larger than those on

sternite IV. The whole of the sternite margins are covered by these black scales apart from a narrow patch of white scales on the posterior end of each sternite margin. Sternite V also carries marginal spots of black scales but also a single apical spot forming a semi-elliptical shape. Sternite I does not have any marginal spots of black scales, but just has a small number of submarginal black scales.

The antenna (Fig. 1E) is dark red consisting of 11-antennomeres which end in a large three-segmented club. The club is vase-shaped with a flatly rounded apical antennomere.

Internal characteristics. The aedeagus (Fig. 2A) is longer (PL = 385 µm) than broad. The parameres are very narrow, parallel-sided, and diverge from each other for most of their length before bowing in towards each other. The terminal part of each paramere is paler than the rest of the paramere suggesting that it consists of less heavily sclerotinized material. The inner margin and surface of the paramere carries long, straight setae. The median lobe (ML = 391 µm) has a broad base but narrows continually for the length of the median lobe so that the terminal ¼ of the median lobe is very narrow. The bulb-shaped tip of the median lobe falls short of the paramere tips. Sternite IX (Fig. 2E) is longer than the parameres (SL = 423 µm). The tip of the posterior lobe is evenly rounded and devoid of setae. Setae are present from the corners of the posterior lobe, where they are long, and continue down the margins of the posterior lobe gradually becoming shorter to the neck where they terminate. Beyond the neck the margins diverge before forming two anterior pointing horns.

Differential diagnosis (Figs. 1, 2). *Anthrenus bakaloudisi* should be compared with the other species with slim parameres that occur around or close to the Mediterranean, namely: *A. pfefferi*, *A. delicatus* and *A. warchalowskii* Kadej, Háva & Kalík. They are considered sequentially here.

Anthrenus bakaloudisi can be distinguished easily from *A. pfefferi* (Fig.1B) using external characters. *Anthrenus pfefferi* is considered a member of the *A. pimpinellae* complex but shows only the merest remnant of the white fascia at the outer margins on each elytron. There are scatterings of pale brown and white scales along the outer margins of the pronotum and the apices of the elytra. There is usually an isolated patch of pale scales around the scutellum. Otherwise, *A. pfefferi* is covered in black scales. The antennal club (Fig. 1F) is cubic whereas the antennal club of *A. bakaloudisi* is vase shaped.

Anthrenus pfefferi parameres (Fig. 2B) are thin and bowed like *A. bakaloudisi* but they are slightly broader before the tips and carry denser, shorter setae along the inner margin and surface than *A. bakaloudisi*. *Anthrenus pfefferi* parameres also have spiny setae on the outer margins of the paramere which *A. bakaloudisi* does not have. The median lobe is similar to *A. bakaloudisi* but shows no sign of a bulb-shaped tip. The sternite IX of *A. pfefferi* (Fig. 2F) and *A. bakaloudisi* are very similar.

Differentiation of *A. bakaloudisi* from Greek *A. delicatus* var. *delicatus* (Fig. 1C) using external features is possible but more difficult than separation from *A. pfefferi*. *Anthrenus delicatus* usually has three bold and almost circular spots of scales on each elytron, sub-marginal, sub-apical, and subsutural. These spots consist of bright white scales that contrast with the creamy colour of the scales in the fascia. Equivalent spots of scales are present in *A. bakaloudisi*, but they are poorly developed. The submarginal spot is close to and almost joined with the fascia, the sub-apical spot is often

chevron shaped, and the sub-sutural spot is very weak, often no more than scales loosely attached to the fascia. In addition, these spots of scales in *A. bakaloudisi* are the same colour as the scales in the fascia. The patches if white scales on the pronotum are well developed and clear in *A. delicatus* but usually barely visible in *A. bakaloudisi*. Finally, the outer edge of the white fascia is broader in *A. bakaloudisi* than in *A. delicatus*. The antennal club in both *A. bakaloudisi*and *A. delicatus*is vase-shaped so differentiation using the shape of the club is not possible.

The parameres of *A. delicatus* (Fig. 2C) are slim like *A. bakaloudisi* but differ from *A. bakaloudisi* in having straight, spikey setae that extend further down the inner margin of the paramere. The median lobe of *A. delicatus* narrows quickly from a moderately broad base and is thereafter slim, slimmer for most of its length than *A. bakaloudisi*. *Anthrenus delicatus* sternite IX (Fig. 2G) is very different having a pointed rather than a rounded tip to the posterior lobe.

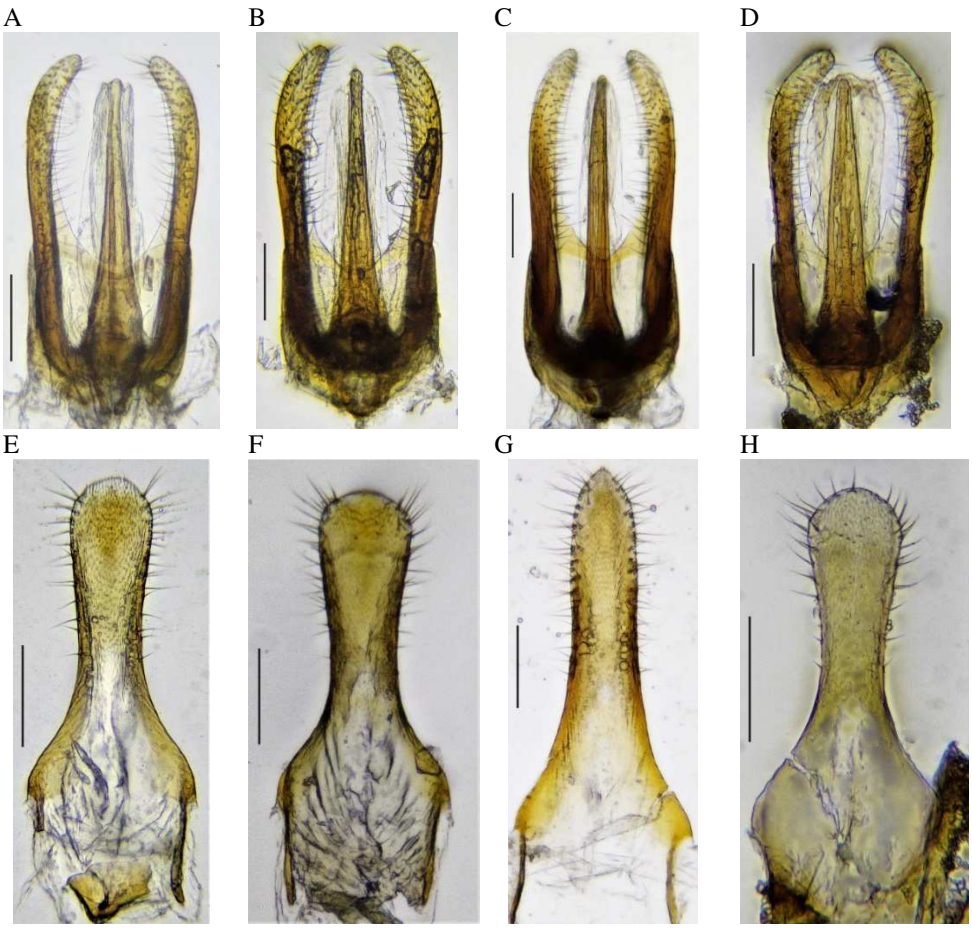

Fig. 2. Aedeagus and sternite IX, A and E: *Anthrenus bakaloudisi* **sp. nov.**, B and F: *Anthrenus pfefferi*, C and G: *Anthrenus delicatus*, D and H: *Anthrenus warchalowskii*, respectively. Scale bars = 100 µm

The white elytral fascia in *A. warchalowskii* (Fig. 1D) is broad across its entire width, much broader than *A. bakaloudisi*. A good image of *A. warchalowskii* habitus can be found in Kadej et al. 2007. The antennal club of *A. warchalowskii* is cubic (Fig. 1H) rather than vase shaped as in *A. bakaloudisi*.

The parameres of *A. warchalowskii* (Fig. 2D) are slim like *A. bakaloudisi* but the median lobe of *A. warchalowskii* is broader than *A. bakaloudisi*. The margins of the median lobe in *A. warchalowskii* are straight and gradually converge to end in a broad, rounded tip. The sternite IX of *A. warchalowskii* (Fig. 2H) has a broader tip to the posterior lobe, a more accentuated neck, and setae on the tip of the posterior lobe. Sternite IX of *A. bakaloudisi* has longer marginal setae that continue to the neck region whereas those of *A. warchalowskii* are short around the neck.

Abundance and distribution. In the field, *A. bakaloudisi* is most likely to be confused with *A. delicatus*. Eighty nine *A. delicatus* all collected from within 40 km of Thessaloniki, Greece were examined, eight of which were *A. bakaloudisi*. *Anthrenus delicatus* were also examined from Croatia, Albania, Cyprus, Turkey, Syria, Jordan, Iran, Israel, Azerbaijan, Turkmenistan. No *A. bakaloudisi* were found beyond Greece. The sites of collection of *A. bakaloudisi* are shown in Figure 3.

Fig. 3. Locations of collections of *Anthrenus bakaloudisi* from Greece.

Morphometrics. There is no difference in BL among *A. bakaloudisi*, *A. pfefferi* and a random sample of Greek *A. delicatus* ($F_{2,29}$ = 2.07, ns [not significant]), but there is a significant difference in BL between the sexes ($F_{1,29}$ = 9.49, p<0.01). *Anthrenus bakaloudisi* mean male BL = 2.528 ± 0.163 [standard deviation] mm, mean female BL = 3.077 ± 0.292 mm; *A. pfefferi* mean male BL = 2.677 ± 0.0223, mean female BL = 2.857 ± 0.405 mm; *A. delicatus* mean male BL = 2.883 ± 0.163 mm, mean female BL = 3.032 ± 0.22 mm. The single *A. warchalowskii* male BL = 2.485 mm.

There was a significant difference in BW/BL among *A. bakaloudisi*, *A. pfefferi* and *A. delicatus* ($F_{2,29}$ = 11.79, p<0.001) but no difference in BW/BL between the sexes ($F_{1,29}$ = 1.32, ns). *Anthrenus bakaloudisi*

mean BW/BL = 0.7 ± 0.009, *A. pfefferi* mean BW/BL = 0.696 ± 0.011, *A. delicatus* mean BW/BL = 0.718 ± 0.011. *Anthrenus delicatus* has a significantly broader body profile ($p<0.05$) than *A. bakaloudisi* and *A. pfefferi* (the latter two species are not significantly different).

Etymology. *Anthrenus bakaloudisi* is named after Professor Dimitris Bakaloudis, Aristotle University of Thessaloniki, who collected the first specimen.

DISCUSSION

The study started to establish whether *A. pfefferi* is a synonym of *A. delicatus*. It is not; this study has demonstrated that they differ at a variety of levels, not just the obvious external differences. Some unusual specimens were noted that initially were thought to be a variant of *A. pfefferi*, but several differences were found that differentiated the specimens from *A. pfefferi* and *A. delicatus* indicating that they belonged to a new species; *Anthrenus bakaloudisi*. The study contributes to an increasing list of studies showing the importance of dissection as a basic technique in the study of *Anthrenus* species. Work has been done to establish how to differentiate among some *Anthrenus* species using colour patterns (Holloway and Cañada Luna 2022), but only after dissection studies to definitively confirm identification. For most species, this work has not been carried out, but where it has many confusion species have been revealed, such as *A. amandae* and *A. pimpinellae* (Holloway and Bakaloudis, 2020), *A. festivus* Erichson, 1848 and *A. mumbaiensis* Holloway, 2023 (Holloway 2023). There are more species that are difficult to differentiate from each other externally which renders redundant many components of faunistic studies where specimens have not been dissected to confirm identification. In the current study, dissection was carried out first that highlighted *A. bakaloudisi*. After that efforts were made to locate colour pattern features that could be used to differentiate *A. bakaloudisi* from *A. delicatus*. Applying these results enabled the discovery of more *A. bakaloudisi* in uncarded examples of *A. delicatus* so the features described here appear to be robust. *A. bakaloudisi* was only considered here against other species with slim parameres (*A. delicatus*, *A. pfefferi*, and *A. warchalowskii*). *Anthrenus isabellinus* is also a common species in Greece that could be confused with *A. bakaloudisi*, but how to recognize *A. isabellinus* under field conditions has been covered elsewhere (Holloway and Cañada Luna 2022). There is also a difference between *A. bakaloudisi* and *A. delicatus* in body profile with *A. delicatus* broader than *A bakaloudisi*. Specimens with a BW/BL > 0.72 are likely to be *A. delicatus* (from Greece), a feature evident through comparison of Figure 1A with Figure 1C.

Examination of *A. delicatus* in the current study failed to find any specimens of *A. bakaloudisi* from anywhere beyond Greece. *Anthrenus bakaloudisi* was only found in samples from Thessaloniki and Serres, but it is likely that the species is found across a wider range. In the region where it was found in the current study, it does not appear to be very scarce. *Anthrenus delicatus* is a very common species in Greece. For every 10-12 specimens of *A. delicatus* found, one specimen of *A. bakaloudisi* was found. *Anthrenus pfefferi* and *A. warchalowskii* were examined as potential confusion species but subsequently dismissed because *A. pfefferi* can be distinguished easily courtesy of its colour pattern and *A. warchalowskii* is only known from Iran. There was no evidence that *A. bakaloudisi* is found so far east.

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