A new endemic species of Baridinae (Coleoptera: Curculionidae) from the Mount Olympus on Cyprus

PETER E. STÜBEN^{1*}, ARIEL-LEIB-LEONID FRIEDMAN^{D2} & CHRISTOPH BRAUN^{D3}

¹CURCULIO-Institut, Hauweg 62, 41066 Mönchengladbach, Germany, https://peterstueben.com ²The Steinhardt Museum of Natural History, Tel Aviv University, Tel Aviv 69978, Israel ³Leibniz Institute for the Analysis of Biodiversity Change, Museum Koenig Bonn, Adenauerallee 127, 53113 Bonn, Germany

*Corresponding author: P.Stueben@t-online.de

ABSTRACT

A new baridine species, *Melanobaris troodi* Stüben, n. sp., associated with *Odontarrhena troodi* (Boiss.) Španiel, Al-Shehbaz, D.A. German & Marhold (Brassicaceae) is described from Mt Olympus ('Chionístra') of the Troodos massif in Cyprus. The new species has been distinguished morphologically and molecularly from its sister taxa *M. margaritae* Korotyaev & Friedman and *M. amanicola* (Pic) from the Middle Eastern mainland, using traditional and innovative (such as 3D-scanning) optical microscopy methods within the framework of integrative taxonomy. A Neighbour-joining tree and Bayesian tree for the mtCO1 gene are presented and a p-distance matrix is compiled for 12 related species of the genus *Melanobaris* Alonso-Zarazaga & Lyal, mainly from the Mediterranean Region. It is pointed out that a high endemism rate can be assumed for many ground-dwelling, flightless *Melanobaris* species in isolated mountain massifs and on islands. Morphological methods alone are certainly not helpful for recognition of these very similar, often cryptic, species. Molecular analysis is indispensable in such cases, above all in new descriptions.

KEYWORDS: Barcoding, integrative taxonomy, molecular analysis, morphology, neighbour-joining tree, new species, host plants, 3D scan, Cyprus, Israel, Turkey.

INTRODUCTION

Melanobaris Alonso-Zarazaga & Lyal, 1999 and *Aulacobaris* Desbrochers des Loges, 1892 are among the most speciose baridine genera in the Palaearctic Region. Some West Palaearctic species of *Melanobaris* and *Aulacobaris* have long been regarded as notorious cabbage pests, especially on the roots and stems. This has changed with the introduction of modern agricultural practices, pesticides and insecticides, and some of these species have actually become very rare (Curculio Team 2007; Prena *et al.* 2014). Recently, new flightless species of *Melanobaris* have been repeatedly reported from isolated mountain massifs (Korotyaev & Ismailova 2011; Korotyaev & Friedman 2011). The present case can even be seen as an illustration of a double geographical isolation manifested by an 'Inselberg' situated on an island. One such mountain is Mt Olympus, or 'Chionístra', in the centre of the densely forested Troodos Mountains, which is the highest (1952 m a.s.l.) peak on the island of Cyprus. In the complex geological setting of the island (McPhee &

DOI: 10.5281/zenodo.14552434; ISSN (online) 2224-6304

Received: 29 October 2024 / Revised: 29 November 2024 / Accepted: 3 December 2024 urn:lsid:zoobank.org:pub:2FDE2515-3482-4CF6-B4CE-1B31FE3EAA03

van Hinsbergen 2019), the uplift of the 90-million-year-old Troodos ophiolite started in the late Miocene and continued during the Plio-Pleistocene (Robertson 1977; Kinnaird *et al.* 2011; Morag *et al.* 2019). It is thus unsurprising that Mt Olympus is known to harbour unique flora and fauna. A total of 100 plant endemics are reported for the Troodos zone alone, including 45 local endemics (Viney 1994). A number of endemic weevils have already been reported from Chionístra (Stüben & Jacob 2023), including *Otiorhynchus crassicollis* Stierlin, 1861, *Psallidium chionistrae* Alziar, 2006 and *Strophomorphus exophthalmus* Pelletier, 1999. In this paper, we describe a new Baridinae species *Melanobaris troodi* and record it in association with the endemic Brassicaceae *Odontarrhena troodi* (Boiss.) Španiel, Al-Shehbaz, D.A. German & Marhold.

MATERIALS AND METHODS

The following institutional acronyms are used in this article: CURCI – Curculio Institute (Mönchengladbach, Germany); ZFMK – Zoological Research Museum Alexander Koenig (Bonn, Germany); SDEI – Senckenberg Deutsches Entomologisches Institut (Müncheberg, Germany).

The new species was first collected on the joint excursion of the Curculio Institute (Germany, Mönchengladbach) to Cyprus on Mt Olympus in April 2010 (Stüben *et al.* 2012), and on another three research trips by the first author; additional specimens were sifted from the detritus under the presumed host plant *Odontarrhena troodi* (Brassicaceae) in 2023 and 2024, and described and designated here as the holotype and paratypes.

All images in this article were taken by the first author using the focus stacking method. Various bridge cameras from the Panasonic Lumix series (FZ200 / FZ300 / FZ1000 II) and different attachment lenses (e.g. from Raynox) were used, which made it possible to generate extreme depth-of-field images—on which every bristle can be seen in detail—in a photo box (Puluz) with just a few (maximum 10–20) single shots. Further intermediate macro lenses were not necessary for the extremely miniaturised habitus images shown here.

The aedeagi and the female genitalia embedded in glycerine were photographed directly using a simple microscope with a standard Canon pocket camera (Stüben 2011).

The description of the new species is visually supplemented with 3D scanning by Christoph Braun (Fig. 8). The 3D reconstruction of the female paratype was done using a DISC3D Scanner (Ströbel *et al.* 2018; Fig. 1) and the associated control software provided by the manufacturer Small World Vision (https://smallworldvision.com). For the optical configuration, the pre-set "XS" was used. Scanning was done with the standard pose program of 360° and a step size of 10° for the azimuth and the elevation. With these settings, 89 individual images were taken for each stacked image and in total 396 stacked images from different perspectives were taken. The images were then used for 3D reconstruction of a digital model of the paratype in Agisoft Metashape Professional (Version 2.0.4) to create a PDF

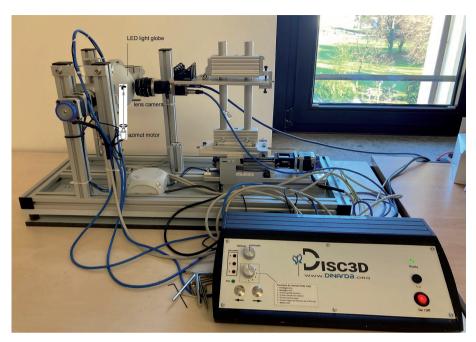


Fig. 1. Powerful 3D scanners such as the DISC3D in the Museum Koenig (Bonn, Germany) will play an increasing role in the description of new species in entomology in the future. We have now made a start on this in weevil research at the Curculio Institute.

file with an integrated 3D model and a scaled Wavefront file (.obj, Appendix 1), allowing for digital measurements of the model in millimetres.

In integrative taxonomy, however, every image-rich representation of a morphological description of a new species must also include a molecular classification, which should be more than just a supplement (in addition, the barcode of a new species must not be missing).

The sequencing of the CO1 mtDNA of the species reported herein was mostly done in the SDEI's molecular laboratory. Sequences of some previously barcoded *Melanobaris* and *Aulacobaris* species were contributed by the molecular laboratory of the ZFMK. Both barcoding endeavours were managed by the CURCI as part of the Molecular Weevil Identification Project, which has largely been run by the first author since 2008 (Schütte *et al.* 2013, 2023; Stüben *et al.* 2015). Specimens of all barcoded species listed here can be found in the reference collections of the ZFMK and the first author. The extracted DNA is stored at the ZFMK and SDEI facilities for further molecular studies. The sequences of the new species and most of the reference species used in this work can be found in Appendix 2, a few have already been deposited in GenBank.

Extraction of the DNA was performed on whole animals using the E.Z.N.A. Tissue DNA Kit (Omega Bio-tek Inc., Norcross, USA) and according to the manufacturer's

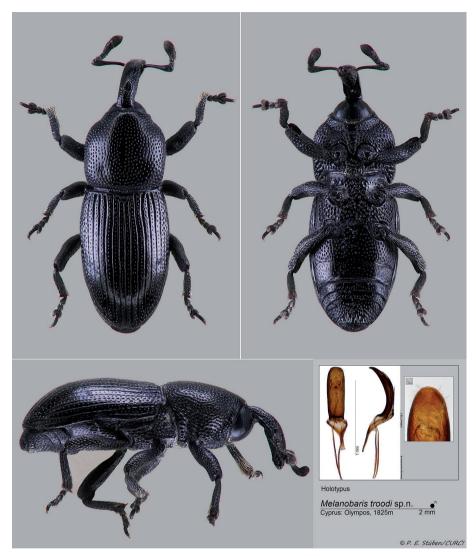


Fig. 2. Holotype of *Melanobaris troodi* n. sp., habitus (dorsal, lateral and ventral) and aedeagus (ventral and lateral).

instructions (Stüben & Kramp 2019). A section of the mtCO1 gene ('Folmer region', 658 bp) was amplified using the primers LCO1490-JJ and HCO2198-JJ (Astrin & Stüben 2008).

The holotype of *Melanobaris amanicola* (Pic, 1905) housed in the Museum National d'Histoire Naturelle, Paris, could not, most regrettably, be studied (Stüben 2023: 2).

TAXONOMY

Order Coleoptera Linnaeus, 1758 Family Curculionidae Latreille, 1802 Subfamily Baridinae Schoenherr, 1836 Genus *Melanobaris* Alonso-Zarazaga & Lyal, 1999 *Melanobaris troodi* Stüben, n. sp.

(Figs 2, 3, 6, 8, 9)

LSID: urn:lsid:zoobank.org:act:0C6D8F52-12A7-4C1C-8EF7-95DA76E0B0F4.

Etymology: The species name refers to the host plant *Odontarrhena troodi*, endemic of the Troodos Mountains on Cyprus.

Diagnosis: The new *Melanobaris* species is—in terms of its morphology—closely related to two other species from the southeast of the Mediterranean Region: *Baris amanicola* Pic, 1905 from the Turkish Amanus Mountains extending along the Mediterranean coast, a species that was transferred to the genus *Melanobaris* by Korotyaev and Friedman (2011), and *Melanobaris margaritae* Korotyaev & Friedman, 2011 from Israel, described in the same publication.

In fact, the new species *M. troodi* described herein shares the body outline with *M. amanicola*, especially the strongly constricted area immediately behind the anterior margin of the pronotum (Korotyaev & Friedman 2011: fig. 3). We acknowledge the excellent outline drawings of the aedeagus (Korotyaev & Friedman 2011: figs 5, 6, 8), according to which, there is not even an approximate resemblance to the species from Cyprus described here. This is obviously also true for the isolated punctate pits on the flanks of the pronotum (Korotyaev & Friedman 2011: fig. 1), which do not show any of the condensed puncture rows typical for *M. troodi*.

The habitus of the new species has much more in common with the endemic species *M. margaritae* from the Israeli part of the Hermon massif (south-western flank), which is known from an altitude of more than 1700 m a.s.l. However, the following differences in characteristics should be sufficient to distinguish the species morphologically (unfortunately, no molecular results are yet available for this species):

- The new species from Cyprus is—at the first glance—significantly larger, 3.5-4.0 mm (vs 2.9-3.5 mm);
- The pronotum is much more densely punctate, the deep punctures are more numerous (Fig. 1 vs Fig. 4);
- The deep punctures fused into rows on the sides of the pronotum is a remarkable feature (vs distinctly separate punctures in *M. margaritae*);
- Immediately behind the anterior margin of the pronotum the sides are rather suddenly strongly constricted; behind the constriction almost to the base subparallel (*vs* behind the front edge laterally significantly less constricted; the sides up to the base arcuate);
- Rostral dorsum noticeably humped and bulged at base; clearly visible in the



Fig. 3. Paratype of *Melanobaris troodi* n. sp., habitus (dorsal and lateral) and female genitalia (A, spermatheca; B, ovipositor; C, spiculum ventrale).

females of *M. troodi*, Fig. 3 (*vs* rostrum without basal bulging, evenly curved in lateral view, Fig. 4);

- Scutellum triangular (vs transverse and rectangular);
- Aedeagus with sides of median lobe slightly rounded (vs median lobe parallelsided);
- Cornu, but especially collum (duct lobe) (swollen in *M. troodi vs* narrower in *M. margaritae*) and ramus (gland lobe) (more humped, slightly slanted anteriorly in *M. troodi vs* less and evenly humped in *M. margaritae*) of the spermatheca differently shaped (Fig. 3 vs Fig. 5).

Another species—very similar to *M. margaritae* but also significantly larger—is the flightless *M. gulnarae* Korotyaev & Ismailova, 2011 with the host plant *Matthiola daghestanica* (Conti) N. Busch (Brassicaceae) from the northern Caucasus mountains of Inner Dagestan (Untsukulsky Distr.). However, this species has a matt upper side of the body, and a parallel-sided aedeagus with a strongly narrowed tip. In addition, the species is broader, especially the elytra $(1.15-1.19 \times \text{ as long as the}$ pronotum width!) in contrast to the slender habitus of *M. troodi*.

Description: The species is strikingly similar in many characteristics to *Melanobaris margaritae* Korotyaev & Friedman, 2011. In order to avoid repetition, the description focuses primarily on the differences of the new species.

Body length 3.5–4.0 mm (without rostrum); body, including rostrum and appendages, shining pitch-black.

Head: Rostrum, that of males $3.3\times$, that of females $4.1\times$ as long as wide between antennal insertions, almost parallel-sided, becoming abruptly somewhat narrower immediately near base; in lateral view strongly curved, hook-shaped, with hump-like obtuse bulge immediately near base (striking feature); more finely punctate dorsally, on sides of rostrum coarse punctures flow together into long and deep grooves; club-shaped scape of antennae about as long as funicle; 1^{st} antennomere (pedicel) ca. $1.2\times$ as long as wide, following six antennomeres clearly wider than long and becoming subsequently wider towards barely separated, short-oval club.

Pronotum: As long as wide, densely punctate, with puncture spacing mostly of diameter of a long-oval puncture; on flanks punctures often only separated from each other by narrow ridges and form long and deep grooves (flowing together) towards underside; pronotum abruptly constricted immediately behind anterior margin (particularly pronounced in males); sides subparallel or straight, slightly converging and only more strongly incurved immediately near base.

Scutellum: Triangular.

Elytra: Elongate, $1.5-1.6 \times$ as long as wide, widest at end of basal quarter, rounded to elongate oval towards tip, with very narrow, groove-like striae and many times wider intervals with single row of punctures each; 7th and 8th striae end well before base.

Legs: Short, profemora just reach posterior margin of eyes, metafemora end far in front of elytral apex (at level of 4th abdominal segment), densely punctate, each puncture with a fine, recumbent bristle.

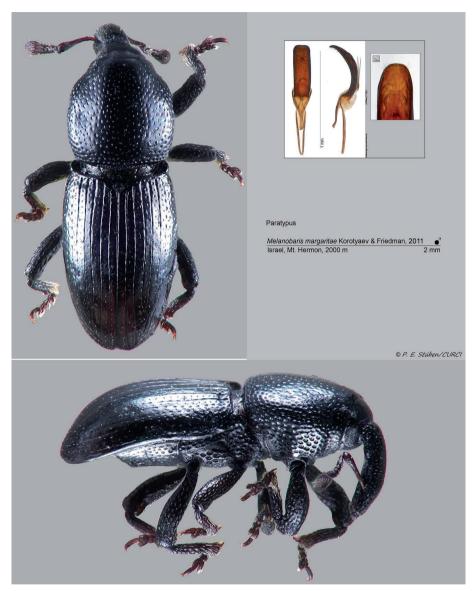


Fig. 4. Paratype of *Melanobaris margaritae*, habitus (dorsal and lateral) and aedeagus (ventral and lateral).

Underside: Pro- and mesothorax with dense, deep pit-like punctures, only separated from each other by narrow ridges. The 1st, long abdominal ventrite somewhat more densely (ca. half puncture diameter between punctures) and not quite as deeply punctate; following ventrites 2–4 with only very fine and sparse punctation, while

ventrite 5 densely punctate and covered with short bristles apically. Abdominal ventrites 3 and 4 combined about as long as ventrite 2.

Aedeagus: With long median lobe oval rounded towards tip and slightly rounded laterally, evenly bent dorso-ventrally.

Female genitalia: Spermatheca, ovipositor (genocoxites and styli) and spiculum ventrale as in Fig. 3A–C.

Holotype: ♂ "Cyprus, Olympos, Artemis Trail, 34°56'1"N 32°52'24"E, 1825 m, *Alyssum troodi* [*Odontarrhena troodi*], 11.11.2023, sifting, leg. Stüben (FO10)" (SDEI), DNA (mtCO1) collector no. 4019-PST (see CO1-sequence in Appendix 2).

Paratypes: Cyprus: 2° , same data as holotype, coll. Stüben (SDEI); 1° , same data as holotype, "13.11.2023 (FO15)", coll. Stüben; 2° , same data as holotype, "11.3.2024 (FO 18)", coll. Stüben; 1°_{\circ} , 2°_{\circ} , "CY – Lemesos, Troodos, Chionistre, 1900 m, 25.5.2023, under *Alyssum troodi & cypricum*, leg. Ch. Makris", coll. Makris, Stüben.

Distribution: Endemic to Cyprus.

Biology: This flightless and ground-dwelling species was sieved from under its supposedly host plant *Odontarrhena troodi* (Boiss.) (formerly *Alyssum troodi*) (Fig. 6). Ch. Makris (Limassol, Cyprus; pers. comm. 2023) also mentions *Odontarrhena cyprica* (Nyár.) (formerly *Alyssum cypricum*) as a probable host. The adults stay mainly near the root collar, so they cannot usually be knocked off the 10–15 cm high, perennial semi-shrub and probably, therefore, remained undetected for a long time. In the insectarium, the species was only occasionally found on the plant itself.

The occurrence of the new species seems to be restricted to the summit area of Mt Olympus at 1825 m a.s.l. However, as the crucifer can also be found at lower altitudes above 1300 m, the occurrence of *M. troodi* should also be checked there. The larvae and pupae of the new species remain unknown to this day.

The only allied to *M. troodi* species of which the circumstances of its collection are known is *M. margaritae* on Mt Hermon (Israel). *Melanobaris margaritae* is rarely collected and to date only eight specimens have been found, all on the top of Mt Hermon. All type specimens were collected at 2000 m a.s.l. The circumstances of the collecting of the older specimens in the 1970–1980s are unclear; however, they were most probably swept from the vegetation as both David G. Furth (Alticinae specialist) and the late dipterist Fini Kaplan were collecting nearly exclusively by sweeping, and it was unlikely they picked tiny weevils from the ground or dug them out of the soil. The two specimens found in 2010 were collected at the bottom of the Bol'an Valley (Fig. 7) with pitfall traps – the weevils probably stayed near the root collar, similarly to *M. troodi*. On 2 March 2018, two additional specimens were found by the second author at 1700 m a.s.l., most probably by sieving fallen leaves as sweeping was impossible in that season.

We assume that the host plant of *M. margaritae* from Mt Hermon is related to that of *M. troodi*, belonging to the genus *Odontarrhena* C.A. Mey. or to the closely related genus *Alyssum* L.

Thirteen species of *Alyssum* have been recorded recently from Israel and the adjacent countries, mainly from high altitudes, both in the Mediterranean zone



Fig. 5. Paratype of *Melanobaris margaritae*, habitus (dorsal, lateral and ventral) and female genitalia (A, spermatheca; B, ovipositor; C, spiculum ventrale).

and in the desert (e.g. Mt Ramon in the Negev), with four species occurring on Mt Hermon (Danin & Fragman-Sapir 2024). *Alyssum strigosum* Banks & Sol. is widely distributed throughout the country; the other three species are found only on Mt Hermon at high altitudes: the very rare *Alyssum baumgartnerianum* Bornm. in the alpine tragacanth belt on the west-facing wind-beaten slopes; scarce *Alyssum murale* Waldst. & Kit. in the alpine tragacanth belt; and a slightly more common *Alyssum szovitsianum* Fisch. & C.A. Mey. occurring throughout Mt Hermon and in the northern and north-eastern parts of the Golan Heights. These three *Alyssum*



Fig. 6. Summit region of Mt Olympus ('Chionístra') of the Troodos massif in Cyprus with the new baridine *Melanobaris troodi* and its supposedly host plant *Odontarrhena troodi*.

species are potential hosts of *M. margaritae*; in particular *A. murale*, which has been transferred from *Alyssum* to *Odontarrhena* (Španiel *et al.* 2015; Royal Botanic Gardens Kew 2024) and, therefore, is the closest candidate to the assumed host of *M. troodi* (Fig. 7).



Fig. 7. Mt Hermon (Israel) with last snowfields and the Bol'an Valley in May 2019; the yellow cushions with *Alyssum* are clearly visible between the rocks (photos by A.L.L. Friedman). Below: *Alyssum murale* Waldst. & Kit., one of the potential host plants of *M. margaritae* (photos by Ori Fragman-Sapir, from Danon & Fragman-Sapir 2024).

DISCUSSION

Nineteen species of the crucifer-associated Baridinae living on Brassicaceae and Resedaceae, including seven *Melanobaris* species and 12 *Aulacobaris* species, have already been molecularly classified by Prena *et al.* (2021) according to a different subsection of the mtCO1 gene (819 bp) from that used here. Five winged species of *Melanobaris* (*M. morio, M. carbonaria* (Boheman, 1836), *M. atramentaria* (Boheman, 1836), *M. laticollis* (Marsham, 1802) and *Melanobaris* sp. nr. *laticollis*) appear monophyletic in the Bayesian tree, even with low support values, while *M. erysimi* and *M. hochhuthi* clustered paraphyletically in a subtree of *Aulacobaris* (with either *A. lepidii* (Germar, 1823) or *A. ochsi* (A. Hoffmann, 1950)).



Fig. 8. 3D scan of the new species, a female of *Melanobaris troodi* (paratype) from Mt Olympus on Cyprus. Note: The 3D graphics can only be activated in the downloaded article. Then, click on the picture and allow 3D content if your PDF reader is asking you for permission (see info bar under the menu). Further information on the technical specifications of the scanner is provided in the Material and Methods section.

Twelve of a total of 30 *Melanobaris* species (15 of which are known from Europe) are barcoded here for the first time. The 'Folmer region', comprising standard base pairs in the CO1 gene and relatively well conserved, has been used for comparison for many decades and has a high variability to generate reliable results with a high differentiation potential at the species level (Folmer *et al.* 1994).

In addition to the already barcoded *Melanobaris atramentaria* (Czech Rep.), *M. laticollis* (Poland) and *M. carbonaria* (Slovakia) (Schütte *et al.* 2013, 2023; Stüben *et al.* 2015), and thanks to Jens Prena who kindly furnished us with further species from various locations for sequencing, we were able to include another eight *Melanobaris* species: *M. sinapis galliae* (Tempère, 1961) (France), *M. steppensis* (Roubal, 1935) (Slovakia), *M. cf. nigritarsis* (Boheman, 1844) (Romania), *M. dalmatina* (H. Brisout de Barneville, 1870) (Greece), *M. quadraticollis* (Boheman, 1836) (Italy), *M. elevata* (Reitter, 1899) (Spain), and *M. atronitens* (Chevrolat, 1861)

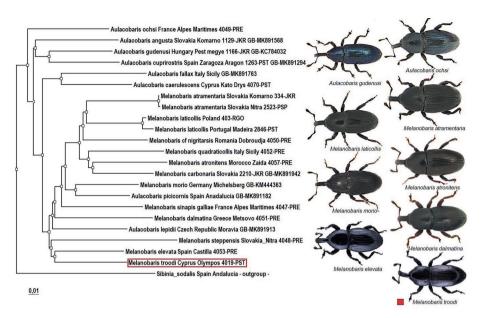


Fig. 9. Neighbour-joining tree for 12 *Melanobaris* species and 8 *Aulacobaris* species associated with Brassicaceae and Resedaceae.

(Morocco)—all determined by Jens Prena—and *M. troodi* n. sp. (Cyprus) (Appendix 2). With another species *Melanobaris morio* (Boheman, 1844) (in Hendrich *et al.* 2015), our effort results in an unbalanced mtCO1 neighbour-joining tree (which includes 12 *Melanobaris* species occurring throughout Europe and partly also in Northern Africa and the Near East) (Fig. 9), which does not take into account different mutation rates and back mutations, but which allows an initial assessment of phylogenetic relationships (especially at the species level). A Bayesian tree for 20 species of baridines associated with the Brassicaceae and Resedaceae with bootstrap support on branches is shown in Fig. 10.

At present, we do not anticipate a revision of the *Melanobaris* species, which include black-coloured taxa that are difficult to distinguish morphologically, especially in Asia, because the molecular database and the number of species available to us are still too small. As in the study by Prena *et al.* (2021), a few metallic-blue *Aulacobaris* species, *A. lepidii* (mainly on *Rorippa* species) and *A. picicornis* (monophages on *Reseda lutea* L.), cluster paraphyletically in the otherwise already quite 'monophyletic' *Melanobaris* tree. The new species *M. troodi* from Cyprus appears as a sister taxon of *M. elevata* from Siles (Spain), with the p-distance 13.83% (Table 1). The overall morphological appearance of the Iberian species, which differs from *M. troodi* in the laterally strongly convex pronotum and the broadly rounded elytra, suggests that we are dealing with a more distantly related species (Fig. 9), not only in geographical terms.

Table 1. P-distances of the mtCO1 gene of 12 *Melanobaris* species and 1 *Aulacobaris* species that were sequenced by us. The very high distance values of well over 13% among the *Melanobaris* species are striking, as we only know them from the more than 400 species of the Western Palaearctic Cryptorhynchinae (e.g. *Echinodera, Kyklioacalles*, Torneumatini), most of which are endemic and always flightless (see Schütte *et al.* 2023 for details).

	M. atramentaria	M. laticollis	M. sinapis galliae	M. steppensis	M. cf. nigritarsis	M. dalmatina	M. quadraticollis	M. elevata	M. atronitens	M. troodi	M. morio	M. carbonaria	A. ochsi
M. atramentaria		0.1338	0.1717	0.2345	0.1396	0.2046	0.1653	0.1918	0.1605		0.1644	0.1548	0.1938
M. laticollis	0.1338		0.1457	0.2101	0.1193	0.1854	0.1374	0.1816	0.1627	0.1800	0.1518	0.1358	0.1995
M. sinapis galliae	0.1717	0.1457		0.2157	0.1678	0.1782	0.1663	0.1692	0.1802	0.1713	0.1670	0.1775	0.1933
M. steppensis	0.2345	0.2101	0.2157		0.2146	0.2287	0.2522	0.1902	0.2408	0.2024	0.2101	0.2165	0.2133
M. cf. nigritarsis	0.1396	0.1193	0.1678	0.2146		0.1927	0.1426	0.1798	0.1485	0.1542	0.1459	0.1603	0.1806
M. dalmatina	0.2046	0.1854	0.1782	0.2287	0.1927		0.2084	0.1668	0.1876	0.1724	0.1947	0.1694	0.1928
M. quadraticollis	0.1653	0.1374	0.1663	0.2522	0.1426	0.2084		0.2055	0.1439	0.1945	0.1674	0.1451	0.2148
M. elevata	0.1918	0.1816	0.1692	0.1902	0.1798	0.1668	0.2055		0.1831	0.1383	0.1650	0.1623	0.1802
M. atronitens	0.1605	0.1627	0.1802	0.2408	0.1485	0.1876	0.1439	0.1831		0.1886	0.1849	0.1327	0.2123
M. troodi	0.1903	0.1800	0.1713	0.2024	0.1542	0.1724	0.1945	0.1383	0.1886		0.1492	0.1718	0.1765
M. morio	0.1644	0.1518	0.1670	0.2101	0.1459	0.1947	0.1674	0.1650	0.1849	0.1492		0.1623	0.1905
M. carbonaria	0.1548	0.1358	0.1775	0.2165	0.1603	0.1694	0.1451	0.1623	0.1327	0.1718	0.1623		0.2001
A. ochsi	0.1938	0.1995	0.1933	0.2133	0.1806	0.1928	0.2148	0.1802	0.2123	0.1765	0.1905	0.2001	

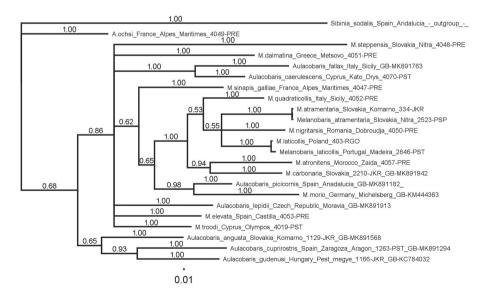


Fig. 10. Bayesian 50% majority rule consensus tree built from mitochondrial CO1 for 20 species of baridines associated with Brassicaceae and Resedaceae (Appendix 2), with bootstrap support on branches. The analysis was run for 20 million generations.

Unfortunately, the molecular characteristics of the morphologically considered (see the differential diagnosis above), closely related, also flightless (brachypterous) and endemic sister taxa from the neighbouring mainland of the Near East, *M. amanicola* and *M. magaritae*, are missing so far. This also applies to *M. gulnarae* described from the northern Caucasus mountains of Inner Dagestan. We suspect that these ground-dwelling species from very high altitudes in various mountain ranges are isolated endemics, which, with their strikingly slender habitus, probably fall into the species complex of *Melanobaris dalmatina*, whose type series possibly contains several species (J. Prena, pers. comm.). These very similar, sometimes even cryptic species can probably only be unambiguously identified through the molecular analysis.

And last but not least, the alleged proof of *M. dalmatina* in south-eastern Turkey (Diyarbakır province) and the hasty assumption that the species has recently spread on the Brassicaceae *Myagrum perfoliatum* L. (Özaslan & Bolu 2015) already appears highly questionable. Once again, it should be clearly emphasized that without knowledge of the type material and above all without a 'molecular corroboration', one should not embark on such thin ice of species identification, even in ecology.

ACKNOWLEDGEMENTS

Our greatest thanks go to Boris Korotyaev (Zoological Institute, Russian Academy of Sciences, St Petersburg) for the review of the manuscript and Jens Prena (Institute of Zoology, Chinese Academy of Sciences, Beijing), who provided us with *Melanobaris* species for the CO1 sequence analysis. As so often in the past, our special thanks go to Eva Kleibusch (SDEI) for the sequence analyses and to Benjamin Wipfler (ZFMK) for his help and technical information on the 3D scan. We would like to thank Andrè Schütte from the Curculio Institute for his help in preparing the Bayesian tree (Appendix 3). We cordially thank Ori Fragman-Sapir, the scientific director of the Jerusalem Botanical Gardens (The Hebrew University of Jerusalem, Israel) for granting us the permission to use his magnificent photographs of *Alyssum murale* on Mt Hermon. Finally, our thanks go to Adrian Fowles (Wales, UK) and Mike Mostovski (Tel Aviv University, Israel) for a critical review of the manuscript and for improving the language.

REFERENCES

- ASTRIN, J.J. & STÜBEN, P.E. 2008. Phylogeny in cryptic weevils: molecules, morphology and new genera of Western Palaearctic Cryptorhynchinae (Coleoptera: Curculionidae). *Invertebrate Systematics* 22: 503–522. https://doi.org/10.1071/IS07057
- CURCULIO TEAM. 2007. Digital-Weevil-Determination for Curculionoidea of West Palaearctic. Transalpina: *Baris / Limnobaris* (Baridinae: Baridini). *SNUDEBILLER, Studies on taxonomy, biology and ecology of Curculionoidea* **8** (97): 12–18. https://curci.de/institute/index.php?beitrag=97
- DANIN, A. & FRAGMAN-SAPIR, O. 2024. Flora of Israel and adjacent areas. Flora of Israel online. Analytical Flora. Alyssum.

https://flora.org.il/en/plants/systematics/alyssum (accessed 13.x.2024)

- FOLMER, O., BLACK, M., HOEH, W., LUTZ, R. & VRIJENHOEK, R. 1994. DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology* 3 (5): 294–299.
- HENDRICH, L., MORINIERE, J., HASZPRUNAR, G., HEBERT, P.D., HAUSMANN, A., KOHLER, F. & BALKE, M. 2015. A comprehensive DNA barcode database for Central European beetles with a

focus on Germany: adding more than 3500 identified species to BOLD. *Molecular Ecology Resources* **15** (4): 795–818.

https://doi.org/10.1111/1755-0998.12354

- KINNAIRD, T.C., ROBERTSON, A.H.F. & MORRIS, A. 2011. Timing of uplift of the Troodos Massif (Cyprus) constrained by sedimentary and magnetic polarity evidence. *Journal of the Geological Society* 168 (2): 457–470. https://doi.org/10.1144/0016-76492009-150
- KOROTYAEV, B.A. & FRIEDMAN, A.L.L. 2011. A new species of the weevil genus *Melanobaris* Alonso-Zarazaga et Lyal, 1999 (Coleoptera: Curculionidae: Baridinae) from Mt Hermon in Israel and commentaries on the composition of the genera *Melanobaris* and *Aulacobaris* Desbrochers, 1892. *Caucasian Entomological Bulletin* 7 (2): 169–172.
- KOROTYAEV, B.A. & GÜLTEKIN, L. 2003. Biology of two weevils, *Lixus ochraceus* Boheman and *Melanobaris gloriae* n. sp. (Insecta: Coleoptera: Curculionidae), associated with *Tchihatchewia isatidea* Boissier, a cruciferous plant endemic of Turkey. *Entomologische Abhandlungen Dresden* 61 (1): 93–99.

https://api.semanticscholar.org/CorpusID:56119335

- KOROTYAEV, B.A. & ISMAILOVA, M.SH. 2011. À new species of the weevil genus *Melanobaris* Alonso-Zarazaga et Lyal, 1999 (Coleoptera: Curculionidae: Baridinae) from Inner Daghestan. *Caucasian Entomological Bulletin* 7 (2): 173–175.
- MCPHEE, P.J. & VAN HINSBERGEN, D.J.J. 2019. Tectonic reconstruction of Cyprus reveals Late Miocene continental collision of Africa and Anatolia. *Gondwana Research* 68: 158–173. https://doi.org/10.1016/j.gr.2018.10.015
- MORAG, N., HAVIV, I. & KATZIR, Y. 2016. From ocean depths to mountain tops: Uplift of the Troodos ophiolite (Cyprus) constrained by low-temperature thermochronology and geomorphic analysis. *Tectonics* 35 (3): 622–637. https://doi.org/10.1002/2015TC004069
- MULCAHY, D.G., IBÁÑEZ, R., JARAMILLO, C.A., CRAWFORD, A.J., RAY, J.M., GOTTE, S.W., JACOBS, J.F., WYNN, A.H., GONZALEZ-PORTER, G.P., MCDIARMID, R.W., CROMBIE, R.I., ZUG, G.R. & DE QUEIROZ, K. 2022. DNA barcoding of the National Museum of Natural History reptile tissue holdings raises concerns about the use of natural history collections and the responsibilities of scientists in the molecular age. *PLoS ONE* **17** (3): e0264930. https://doi.org/10.1371/journal.pone.0264930
- ÖZASLAN, C. & BOLU, H. 2015. A new host *Myagrum perfoliatum* L. record for *Melanobaris dalmatina* (H. Brisout, 1870) (Col.: Curculionidae) from Turkey. *Scientific Papers. Series A. Agronomy* **58**: 266–268.

https://agronomyjournal.usamv.ro/pdf/2015/vol.LVIII/art48.pdf

- PRENA, J., COLONNELLI, E. & HESPENHEIDE, H.A. 2014. 3.7.9 Conoderinae Schoenherr, 1833. In: Leschen, R.A.B. & Beutel, R.G. (Eds), Handbook of Zoology: Coleoptera, Beetles Volume 3: Morphology and Systematics (Phytophaga). De Gruyter, Berlin/Boston, pp. 577–589.
- PRENA, J., LIU, N., REN, L., WANG, Z. & ZHANG, R. 2021. Three Eurasian malvid-associated Baridinae (Coleoptera, Curculionidae) with widely different vestiture: congeneric or not? *Zootaxa* 4948 (1): 42–50.
- https://doi.org/10.11646/zootaxa.4948.1.2 ROBERTSON, A.H.F. 1977. Tertiary uplift history of the Troodos massif, Cyprus. *GSA Bulletin* **88** (12):

1763–1772.

- https://doi.org/10.1130/0016-7606(1977)88<1763:TUHOTT>2.0.CO;2
- ROYAL BOTANIC GARDENS KEW. 2024. Odontarrhena C.A. Mey. Plants of the world online. https://powo.science.kew.org/taxon/urn:lsid:ipni.org:names:12855-1 (accessed 13.x.2024)
- SCHÜTTE, A., STÜBEN, P.E. & SPRICK, P. 2013. The Molecular Weevil Identification Project (Coleoptera: Curculionoidea), Part I. A contribution to Integrative Taxonomy and Phylogenetic Systematics. SNUDEBILLER: Studies on taxonomy, biology and ecology of Curculionoidea 14 (211): 1–77.

https://www.curci.de/?beitrag=211

SCHÜTTE, A., STÜBEN, P.E. & ASTRIN, J.J. 2023. Molecular Weevil Identification Project: A thoroughly curated barcode release of 1300 Western Palearctic weevil species (Coleoptera, Curculionoidea). *Biodiversity Data Journal* 11: e96438. https://doi.org/10.3897/BDJ.11.e96438

- ŠPANIEL, S., KEMPA, M., SALMERÓN-SÁNCHEZ, E., FUERTES-AGUILAR, J., MOTA, J.F., AL-SHEHBAZ, I.A., GERMAN, D.A., OLŠAVSKÁ, K., ŠINGLIAROVÁ, B., ZOZOMOVÁ-LIHOVÁ, J. & MARHOLD, K. 2015. AlyBase: database of names, chromosome numbers, and ploidy levels of Alysseae (Brassicaceae), with a new generic concept of the tribe. *Plant Systematics and Evolution* **301**: 2463–2491. https://doi.org/10.1007/s00606-015-1257-3
- STRÖBEL, B., SCHMELZLE, S., BLÜTHGEN, N. & HEETHOFF, H. 2018. An automated device for the digitization and 3D modelling of insects, combining extended-depth-of-field and all-side multi-view imaging. *ZooKevs* **759**: 1–27.
 - https://doi.org/10.3897/zookeys.759.24584
- STÜBEN, P.E., SCHÜTTE, A., BAYER, CH. & ASTRIN, J.J. 2015. The Molecular Weevil Identification Project (Coleoptera: Curculionoidea), Part II. Towards an Integrative Taxonomy. SNUDEBILLER: Studies on taxonomy, biology and ecology of Curculionoidea 16 (237): 1–294. https://www.curci.de/?beitrag=237
- STÜBEN, A. 2011. Schichtfotografie in der Entomologie. Weevil News 69: 1–13. https://www.curci.de/?beitrag=187
- STÜBEN, P.E., SPRICK, P., BEHNE, L., ALZIAR, G., COLONNELLI, E., GIUSTO, C., MESSUTAT, J. & TEODOR, L.A. 2012. The Curculionoidea (Coleoptera) of Cyprus. Results of a collecting journey on Cyprus by members of the CURCULIO Institute in April 2010. SNUDEBILLER, Studies on taxonomy, biology and ecology of Curculionoidea 13 (195): 80–137. https://www.curci.de/?beitrag=195
- STÜBEN, P.E. 2022. Madeiras Rüsselkäfer immer neue Überraschungen (Coleoptera: Curculionidae) (unter Mitarbeit von Rüdiger Jacob und Jiri Krátký). Weevil News 101: 1–20. https://curci.de/data/weevilnews/weevilnews 101.pdf
- STÜBEN, P.E. 2023. Schlüssel der westpaläarktischen Nanophyini (Coleoptera: Curculionoidea: Nanophyinae). Weevil News 111: 1–23.
- https://curci.de/data/weevilnews/weevilnews_111.pdf STÜBEN, P.E. & JACOB, R. 2023. Weevils of Cyprus - an image catalogue. Le Charançon. Catalogues & Keys No. 6. Curculio-Institute, Mönchengladbach, Germany.
 - https://cyprus.curci.de (accessed 13.x.2024)
- STÜBEN, P.E. & KRAMP, K. 2019. Neue Echinodera aus Griechenland Beitrag zur integrativen Taxonomie (Coleoptera: Curculionidae: Cryptorhynchinae). Contributions to Entomology 69 (2): 319–330.
 - https://doi.org/10.21248/contrib.entomol.69.2.319-330
- VINEY, D.E. 1994. An illustrated flora of North Cyprus, I–XVI. Koeltz Scientific Books, Königstein, Germany. xxix + 697 pp.

Appendix 1

3D scan of a female paratype of *Melanobaris troodi* n. sp. from Mt Olympus, Cyprus. More information on the technical specifications of the scanner can be found in the Materials and Methods section.

https://doi.org/10.5281/zenodo.14551900

Appendix 2

Overview of the exact location data and mtCO1 sequences (658bp) of the *Melanobaris* and *Aulacobaris* species associated with Brassicaceae and Resedaceae (see also the data already deposted in GenBank for some of the species in Fig. 9).

Note. In the opinion of the first author and others (Stüben 2022; Mulcahy *et al.* 2022), the barcode of new or first-time barcoded species should always be an integral part of the first description or re-description and should not be deposited separately elsewhere, often without illustrations or a morphological reference. This is the only way to ensure permanent availability and meaningful embedding in a descriptive context (integrative taxonomy). In addition, citation-free transfers from gene banks into sometimes arbitrarily composed selection potpourris of sequences should be avoided as far as possible. In practice, the PDF version also enables immediate access to the CO1 sequence listed below for further scientific studies.

>Melanobaris atramentaria: 334-JKR / Slovakia, Komárno, Virt env., 47°45'35.247"N 18°20'25.833"E, 112 m, Erysimum sp., 19.v.2011, sweeping, leg., det. J. Krátký, coll. ZFMK.

AACATTATATTTTATTTTGGAACTTGATCAGGCATAATAGGAACATCTTTGAGTATTAT TATTCGAGCAGAATTAGGAAATCCAGGTAACTTAATTGGAGACGACCAAATTTATAATA CAATTGTAACAGCCCATGCATTTATTATAATTTTTTTTTATAGTTATACCAATCATAATTGGAG GATTCGGAAATTGATTGGTACCTTTAATACTCGGAGCACCTGATATAGCTTTTCCTC GAATAAATAATAAAGATTTTGATTATTATAATTTACCCCAGGCATCTAACTGATAAGGTAGAAT TATTGATAAGGAGCTGGTACTGGGTGAACAGTGTATCCACCCTTATCAGCAAACATT GCCCATGAAGGAGCTTCTGTAGATTTAGCAATCTTCAGGATAAAGAAGAACAAT TATTCTAGGGCAATCAATTTTATTATTTTTTACTGATAAGAAAT AGATCAAATATCCCTATTCGTATGAGTTAGCAATCTTCAACATTAACACTCATCAGGAATAAAAT AGATCAAATATCCCTATTCGTATGAGCTGTTAAAATTAACACTCATCAGGAATAAAAAT TCTTTACCAGTACTTGCTGGAGCAATTACGATACTATTGACTGATCGAAATGTCAACA CATCTTTTTTGACCGGCAGGAGGAGCAATTACGATACTATTGACCGAACACTTATT >Melanobaris atramentaria: 2523-PSP / Slovakia, Nitra, SW Búč, 47°46'56"N 18°25'10"E,110 m, Syrenia cana, 18.v.2014, leg., det. P. Sprick, coll. ZFMK.

>Melanobaris laticollis: 403-RGO / Poland, ad. Janowiec, 51°32'22"N 21°53'25"E, 7.v.2011, leg., det. R. Gosik, coll. ZFMK.

>*Melanobaris laticollis*: 2846-PST / Portugal, Madeira, Sao Vincente: Rota da Cal, 32°47'52"N 17°01'46"W, 322 m, ruderal vegetation, 30.xi.2015, beating, leg., det. Stüben, coll. ZFMK.

>Melanobaris sinapis galliae: 4047-PRE / France, Col du Ferrier, Alpes-Maritimes, 43.714°N 6.864°E, 1040 m, 5.v.2018, leg., det. Prena, coll. Stüben.

>Melanobaris steppensis: 4048-PRE / Slovakia, Kolínanský vrch, Nitra, 48.349°N 18.192°E, 345 m, 8.ix.2022, leg., det. Prena, coll. Stüben.

>*Melanobaris* cf. *nigritarsis*: 4050-PRE / Romania, Măcin, Dobroudja, 45.246°N 28.123°E, 10 m, 20.v.2018, leg., det. Prena, coll. Stüben.

>*Melanobaris dalmatina*: 4051-PRE / Greece, Metsovo, West Makedonia, 39.770°N 21.183°E, 1160 m, 28.iv.2023, leg., det. Prena, coll. Stüben.

AACCTTATATTTTATTTTGGTGCCTGATCTGGAATAACCGGAACCTCCTTAAGAATCT TAATCCGAGCAGAATTAGGAAACCCGGGAAACCTTATTGGAGATGATCAAATCTATAA TACAATCGTTACTGCCCATGCATTTATTATGATTTTCTTCATAGTAATACCAATTATAATT GGGGGATTCGGAAATTGACTAGTTCCATTAATACTCGGTGCACCTGATATAGCCTT TCCCCGAATAAACAATATAAGATTCTGACTTTTACCCCCCTCTTTATTTTACTATAAGA GAAGAATTATTGATAAAGGAGCTGGAACAGGTTGAACAGTTTACCCCCCTTTATCAAC CAATATGCTCATGAAGGAACCTCTGTAGACCTAGCAATTTTAGCTTACATATAGCAG GAATTACCCAATCTTAGGGGCAATTAATTTATTTATTTACTATAAGAATCAAC CGGTATAAAAATAGACCAAATATCTTTATTTGATGAGCCGTAAAAAATTACCGCAATTCT TCTTCTTCTTCGTTACCAGAATATCTTATTTGATGAGCCGTAAAAATTACCGCAATTCT TCTTCTTCTTTCGTTACCAGTATTAGCAGGAGCCATCTCCATATTATTAACTGATCGAAAATT TAATACCTCTTTTTGATCCTGCAGGTGGGGGGGCGATCCAATTCTTTACAACATTTATTA

>Melanobaris quadraticollis: 4052-PRE / Italy, Gela, Sicily, 37.101°N 14.264°E, 32 m, 10.iv.2022, leg., det. Prena, coll. Stüben.

>Melanobaris elevata: 4053-PRE / Spain, Siles, Castilla, 38.377°N 2.518°W, 1410 m, 21.v.2019, leg., det. Prena, coll. Stüben.

AACCTTATATTTTATTTTGGAACTTGGTCAGGAATAGTAGGAACCTCATTAAGAATC CTAATTCGAGCAGAATTAGGGAATCCTGGAAATTTAATTGGTGATGATCAAATTTATAATA CAATTGTTACTGCACATGCATTTATTATAATTTTTTTCATAGTTATACCTATTATAATTGGAG GATTTGGTAATTGACTAGTACCACTAATATTAGGAGCCCCTGATATAGCCTTTCCTC GAATAAATAATATAAAGATTCTGACTACTTCCTCCTCTTCTTTTTTCATATAGCATATAAGAA GAATAATTGATAAAGGAGTAGGAACTGGGTGAACAGTCTACCCCCCTTTATCAACAAA TATTGCTCACGAAGGAGGCCCCAGTAGACCTTGCAATTTTAGTTACATATAGCATCTTCAG GAATAAATAGATCAAATACCTTAATTTTATTCAACAGTAGTAAAATAGCATTCTTCAG GAATAAATAGAACAATACCTTTATTTGTTTCAACAGTAAAATAGCATTCTTCAG GAATAAAAATAGAATCAAATACCTTTAATTTGTTTGAGCTGTAAAAATCACTGCGGATCCTC CTTCTTTTATCTTTACCAGTTCTAGCCGGAGCAATTACTATACCTATCACCACATATT CAATACTTCTTTTGATCCTGCAGGAGGAGGAGACCCCAATTTTAACCAACATTATT

>Melanobaris atronitens: 4057-PRE / Morocco, Zaida, 32.777°N 4.935°W, 1500 m, 17.iii.2018, leg., det. Prena, coll. Stüben.

>Aulacobaris ochsi: 4049-PRE / France, Col du Ferrier, Alpes-Maritimes, 43.714°N 6.864°E, 1040 m, 5.v.2018, leg., det. Prena, coll. Stüben.

CACATTATATTTTATTTTTGGAACATGAGCAGGAATAATTGGAACATCCATAAGAATTCTAAT TCGAGCAGAACTTAGAAATCCGGGAAATCTAATTGGAGATGATCAAATTTATAATAC TATCGTAACTGCTCATGCATTTATCATAATTTTCTTCATAGTAATACCTATTATAATTGGAG GATTTGGAAATTGATTGGTACCCTTAATACTAGGAGCTCCAGATATAGCCTTCCCAC GAATAAATAATAAAGATTTGACTTCTACCTCCGGCCTTATTTTTCTTACTAATAAGAA GAATTATTGATAAAGGAGCAGGAACTGGATGAACAGTCTATCCCCCTCTATCTTCAAA CATAGCCCATGAAGGAACCTCAATTGACTTAGCTATTTTAGATACATATAGCAGGAATT TCTTCAATTTTAGGAGCAATTAATTTTATTTCAACTGTTATTAATATACATTCTCAG GAATAAAACTAGATCAAATATCACTAATTTATTTCTGAGCTGGAAAATTACAATCTCAG GAATAAAACTAGATCAAATATCACTATTTATCTGAGCTGTGAAAATTACAGCAGCAATTCC CTTCTATTGTCCTTGCCTGTATTAGCAGGTGCAATTACTATATAACAGCCGAAATAT

>Aulacobaris caerulescens: 4070-PST / Cyprus, 1 km E Kato Drys, 34°51'01"N 33°18'57"E, 532 m, Brassicaceae, 3.iii.2024, leg., det. coll. Stüben.