



A NEW GRASS-MINER MOTH, *ELACHISTA MARIAE* SP. N., FROM SLOVAKIA AND BULGARIA (LEPIDOPTERA, ELACHISTIDAE)

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Abstract: *Elachista mariae* sp. n. is described from moths taken in xerothermic localities in south-western Slovakia and south-western Bulgaria. The new species belongs to the genus *Elachista* Treitschke, 1833, subgenus *Apheloesetia* Stephens, 1834, and the *E. bedellella* species group according to KAILA (2019). It is similar to several species of this group in the colour and pattern elements of the forewing; however, it differs from them primarily in the structure of the male and female genitalia. The immature stages of the new species are unknown. Colour photographs of the adults and of the habitat are included with photographs and illustrations of the female and male genitalia.

Key words: Lepidoptera, Elachistidae, *Elachista mariae*, new species, Slovakia, Bulgaria

INTRODUCTION

Elachistinae Bruand, 1850 is a subfamily of the Elachistidae, a large and diverse family of small to very small grass-miner moths. KAILA (2019) in his annotated catalogue of the world species of Elachistinae lists 10 genera with 805 valid species. According to him, the number of described species of Elachistinae will undoubtedly continue to increase in the future, as large geographical areas of the world still remain virtually unexplored in respect of this family and subfamily. The type genus of the family is *Elachista*, described by TREITSCHKE in 1833. 221 species of this genus are known in Europe (Lepiforum 2024).

The second author, while surveying the fauna of the small moths of the Kováčovské kopce (the Blacksmiths hills, now the Burda Mountains) in south-western Slovakia, found six specimens of an unknown *Elachista* species on the forest-steppe slope in the early evening in late June 2010, which in appearance resembled *E. nolckeni* Šulcs, 1992. However, after examining the male genitalia of one specimen, he recognised that it was not one of the species known to him. Later, similar specimens came to him for determination, one male, caught by Ivan Richter near the village of Plášťovce, taken in 2000, and two specimens of each sex, found by the first author near the villages of Rybník and Kozárovce. In 2012, the second author sent four specimens (two males and two females) to Lauri Kaila

who is an expert on the Elachistidae family. Kaila sent these specimens for DNA analysis, which was successful and indicated that they were of a hitherto unknown species from the *Elachista bedellella* (Sircom, 1848) species group of the subgenus *Apheloseitia* Stephens, 1834. This is a large group, which in Kaila's catalogue (2019) had 47 species. Another species, *Elachista laurikailai* Varenne & Nel, 2020, has since been added to this group. The entire group is poorly researched, the described species are often very similar in appearance, as is the structure of the male genitalia.

In the following years, the first author collected more specimens of this species at three localities in Štiavnické vrchy (the Štiavnica Mountains). He prepared and examined in more detail the male and female genitalia, and the structure of the female genitalia of this species proved to be specific, different from those of known species and this was subsequently confirmed by Kaila. The specimens from Slovakia were supplemented by a male specimen originating from Bulgaria, which was caught by Jari-Pekka Kaitila in 2011 and about which we obtained detailed information from Kaila. It was the study of the structure of the male genitalia, partially supported by the combination of the original structure of the female genitalia and the results of the barcode analyses, that led the authors to consider this to be a new *Elachista* species, which is described below.

MATERIAL AND METHODS

Specimens of the new species were taken as adults, collected with a net or attracted to light traps. The genitalia were dissected following the usual procedure for small Lepidoptera and stored in small plastic vials filled with glycerol or embedded in Euparal in glass slides.

Photographs of adults were taken using a Canon Macro Lens EF 100mm 1:2.8 USM. Photographs of the genitalia were taken with a Meopta microscope using a 6x objective without an eyepiece, and with a Canon EOS 450 camera without an objective attached. All photographs were taken and edited, using a Zoner 12 software, by the first author. In addition, drawings of the genitalia were made by the second author using Indian ink and a coloured pencil on transparent drawing paper.

Molecular analysis. Tissue samples (dry legs) from the study specimens were successfully processed at the Canadian Centre for DNA Barcoding (CCDB; University of Guelph) using standard protocols. These sequences, along with details of the sequenced specimens, were uploaded to the Barcode of Life Data Systems (BOLD; RATNASINGHAM & HEBERT 2007) and to the project ELACA Elachistinae of the World (https://v4.boldsystems.org/index.php/MAS_Management_DataConsole?codes=ELACA) managed by Marko Mutanen. We compared the sequences initially with all records accessible to us and then selected a representative of each species and BINs (Barcode Index Numbers) of the *Elachista bedellella* species group for further analyses. A Maximum Likelihood

tree was constructed under the GTR model for nucleotide substitution, with node bootstrap support values based on 500 pseudoreplicates. The tree was rooted to *E. cretula* Kaila, 2011.

Abbreviations

ALMD	Aquazoo Löbbecke Museum Düsseldorf, Germany
MZUF	Museum Zoology University Florence, Italy
FK	František Kosorín
Gs, Gp	Genitalia slide(s), preparation(s)
LK	Lauri Kaila
ZT	Zdenko Tokár

TAXONOMY

Elachista mariae sp. n. (Figs 1–9)

Material. Holotype: 1♀, pinned, genitalia in a separate slide. Original labels: “Slovakia, Štiavnické vrchy, Kozárovce, 48°18'18" N, 18°31'40" E, 230 m, 8.VI.2008, leg. František Kosorín”, “HOLOTYPE *Elachista mariae* Kosorín & Tokár” (red label), “Genitalia slide FK/1827”, “DNA sample 22159 Lepid Phyl” (ELACA1696-12), coll. František Kosorín (to be deposited in the National Slovak Museum in Bratislava).

Paratypes: Slovakia, 1♀, 4♂, Štiavnické vrchy, Rybník, 310 m, 29.VI.2008, Gs ♀ FK/1826, 20.VI.2009, Gs ♂ FK/2240, 11.VI.2012, Gs ♂ FK/3074, 23.V.2014, Gs ♂ FK/3405, 19.VI.2021, Gs ♂ FK/4723; 1♂, Štiavnické vrchy, Tlmače, 200 m, 15.VI.2021, Gs FK/4721; 3♂, Štiavnické vrchy, Mochovce, 300 m, 26.V.2012, Gs FK/3210, FK/3211, FK/3212, all leg. & coll. F. Kosorín; 1♂, Krupinská pahorkatina, Plášťovce, 27.V.2000, leg. & coll. Ivan Richter, Gp ZT 6974, DNA sample 21352 Lepid Phyl (ELACA1289-12); 2♂, 4♀, Kováčovské kopce – Burda, 19.VI.2010, Gp ZT ♂ 11723, ♂ 14532, ♀ 14497, Gs FK/5081, DNA samples 22197, 22198 Lepid Phyl (ELACA1734-12, 1735-12), all leg. & coll. Zdenko Tokár. Bulgaria, 1♂, Kozhuh by Rupite, 41.458 N, 23.258 E, 300 m, 1.VI.2011, Gs LK/6356, DNA sample 25325 Lepid Phyl, leg. & coll. J.-P. Kaitila. All paratypes with red labels “PARATYPE *Elachista mariae* Kosorín & Tokár”.



Figs 1–2. *Elachista mariae* sp. n., adult female, paratype. 1. Upperside; 2. Underside.

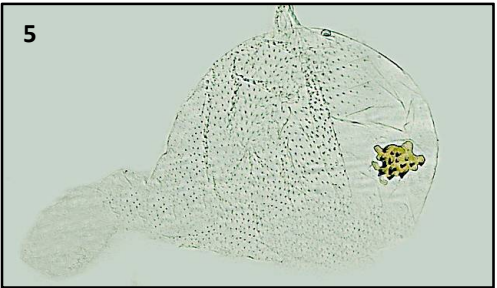
Description. Adult (Figs 1–3).

Wingspan 7.0–9.0 mm, head, thorax and tegula white. Labial palpus straight or slightly upcurved, length 2x diameter of eye, cream-coloured, outside of second segment predominantly grey-brown. Antenna white, dorsally ringed brown in females, but less obvious in males. Ground colour of forewing white, silky sheen with yellowish pattern and scattered dark brown dusting (in females pattern and dusting less distinctive). Cilia white to yellowish-white with dark brown ciliary line. Hindwings grey, cilia grey-white. Underside of both wings grey-brown with darker veins (Fig. 2).

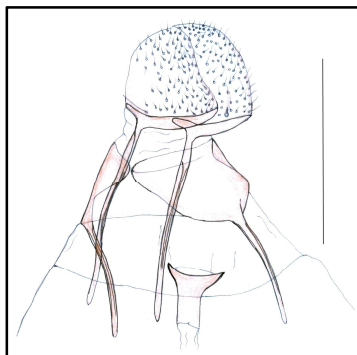


Fig. 3. *Elachista mariae* sp. n., adult male, paratype, Slovakia, Mochovce, 26.V.2012.

Female genitalia (Figs 4–6). Papillae anales broadly triangular, apophyses posteriores one third longer than apophyses anteriores, which are bent almost at right angles at the base in ventral view. Antrum wide at the base, strongly narrowed distally, colliculum cylindrical, moderately sclerotised (Fig. 6). Ductus



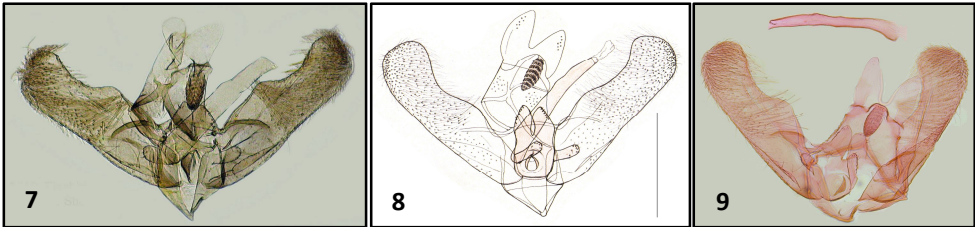
Figs 4–5. Female genitalia of *Elachista mariae* sp. n. 4. Holotype, Gs FK/1827; 5. Paratype, Gs FK/1826, details of bursa copulatrix, appendix bursae, and signum.



bursae long and narrow, almost same width throughout. Bursa copulatrix oval (Fig. 5), covered with minute spines, except for the part around the signum; laterally it passes into the oval, smaller appendix bursae. Signum prominent, irregularly shaped, dentate.

← **Fig. 6.** Female genitalia of *Elachista mariae* sp. n., paratype, Gp ZT 14497, scale bar 0.3 mm.

Male genitalia (Figs 7–9). Uncus lobes wide triangular-shaped, rounded at apex, a little longer than broad, separated by V-shaped incision with a few fine setae. Gnathos elongate oval, widest medially, approximately 3 times longer than wide, tapered towards pointed apex. Valva 3.7 times longer than wide at its widest; ventral margin straight; costa evenly convex before the middle; cucullus widening and rounded, slightly convex and somewhat bent towards costa. Juxta lobes triangular-shaped, similar in shape to uncus lobes, lateral margin with setae. Digitate process relatively small, almost parallel-sided, distally rounded and setose. Vinculum short, wide V-shaped. Phallus about $\frac{3}{4}$ length of valva, slightly bent at $\frac{2}{3}$ length and tapered towards apex, vesica without cornuti.



Figs 7–9. Male genitalia of *Elachista mariae* sp. n., paratypes. **7.** Gs FK/3212; **8.** Gp ZT 14532, scale bar 0,3 mm; **9.** Bulgaria, Kozhuh by Rupite, 1.VI.2011, Gs LK/6356, J.-P. Kaitila leg. & coll.

Diagnosis

Elachista mariae sp. n. is mostly similar externally and in the male genitalia to the following species in the *E. bedellella* species group: *E. ohridella* Parenti, 2001, *E. exigua* Parenti, 1978, *E. maculata* Parenti, 1978, *E. laurikailai*, and *E. nolckeni* Šulcs, 1992.

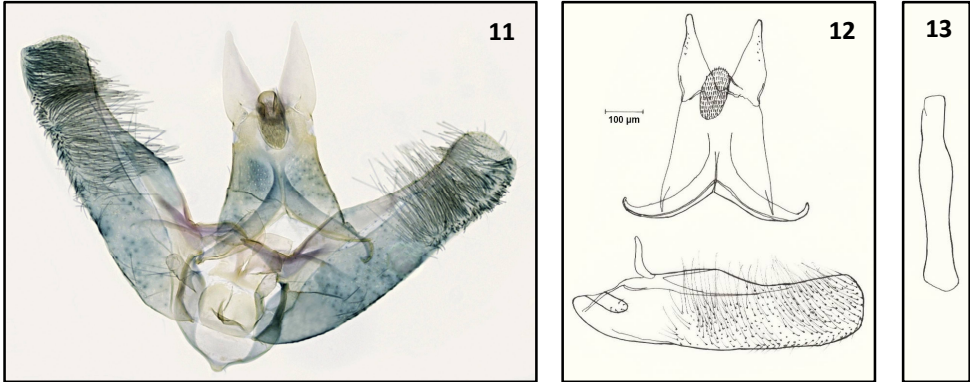
To clearly distinguish the new species from the given similar species, examination of the genitalia is necessary because the dusting and forewing pattern of specimens is often variable due to the easy loss of scales. Here, however, we can point out the differences of *Elachista mariae* from the most similar species *E. ohridella* (Fig. 10). The latter is more narrow-winged and the wingspan of the males in the type series is from 10 to 11.5 mm (measured by G. F. Turrisi, pers. comm.), while the wingspan of specimens of the new species are from 7 to 9 mm. In the other mentioned species, these differences in size are negligible or not so striking.



Fig. 10. *Elachista ohridella* Parenti, adult, holotype, Macedonia, Ohrid, Petrina Planina, 17–26.VI.1959, wingspan 11.5 mm.

In the male genitalia:

In *E. ohridella* (Figs 11–13), the uncus lobes are narrow, sharply pointed, almost straight, the costa is gradually and only slightly convex in the first third, and the cucullus is hardly expanded, whilst in *E. mariae* the uncus lobes are wide, triangular, rounded at the apex, distinctly convex, the costa is more significantly convex before the middle and the cucullus is wider and bent towards costa.



Figs 11–13. Male genitalia of *E. ohridella*. **11.** Holotype, Gs U. Parenti 9271, North Macedonia, Ohrid, Petrina Mt., 17.-26.VI.1959, J. Klimesch leg., coll. MZUF (phallus is missing); **12.** Parts of paratype, Gs U. Parenti 12821, Turkey, Malatya, Reşadiye – Sürgü, 1500 m, 15.-16.VI.1974, J. Gross leg, coll. ALMD; **13.** Phallus, paratype, Gs U. Parenti 8732, Bulgaria, Slavyanka (Ali Botush) Mts, 28.VI.1929, Kr. Tuleschkow, coll. MZUF.

E. laurikailai (Fig. 14) differs from the new species mainly by the narrower, triangular-shaped uncus lobes and the shape of the costa, which is convex medially, whilst in *E. mariae* the costa is convex before the middle.

The new species differs from *E. exigua* by the wide triangular-shaped uncus lobes, and the slightly convex and bent cucullus, whilst in the latter the uncus lobes are nearly parallel-sided and the cucullus is not convex or bent (Fig. 15).

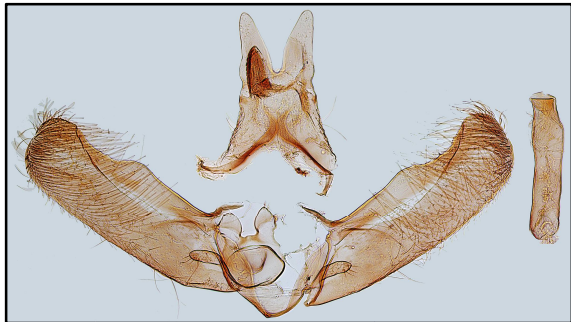
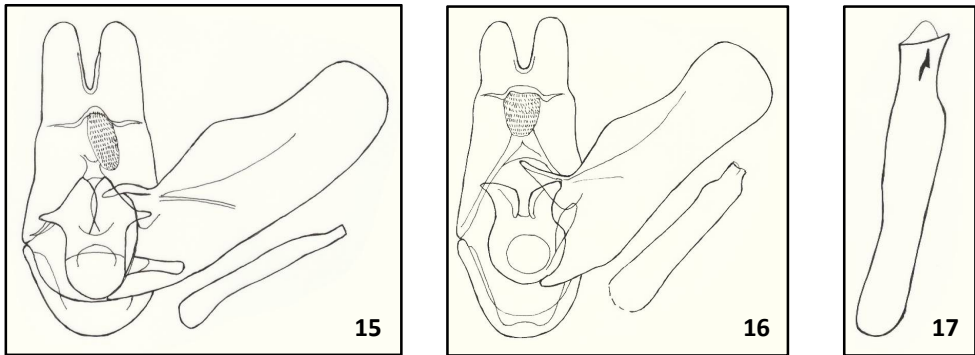


Fig. 14. Male genitalia of *E. laurikailai*, holotype, Gp J. Nel 4400, France, Alpes-Maritimes, La Brigue, 900 m, 21.VI.1995, J. Nel leg., coll. Th. Varenne.

E. maculata (Fig. 16) differs from the new species mainly in the shorter and rounded uncus lobes and gnathos.

From *E. nolckeni* the new species is readily separated by its phallus without cornuti, whilst in the latter the phallus contains a prominent cornutus (Fig. 17).



Figs 15–17. Male genitalia. **15.** *E. exigua*, according to PARENTI 1978; **16.** *E. maculata*, according to PARENTI 1978; **17.** *E. nolckeni*, phallus, Gp J. Liška, Bohemia, Český kras, Srbsko, 22.VI.1994, coll. ZT.

In the female genitalia:

The female genitalia of *E. mariae* (Figs 4–5) differ conspicuously from those of *E. exigua*, *E. laurikailai* and *E. nolckeni* mainly in the structure of the bursa copulatrix with an appendix bursae, while an appendix bursae is absent in these three species.

The female genitalia of *E. exigua*, *E. maculata*, and *E. ohridella* (see Discussion and notes) are unknown.

Because of this limited knowledge of the female genitalia of related species, it is not possible to use them for diagnosis.

Distribution

So far known from several localities in SW Slovakia and one locality in SW Bulgaria. All specimens were collected on forest-steppe slopes with areas of rocky steppe (Fig. 18).



Fig. 18. Locality of *Elachista mariae* sp. n. near Mochovce (Slovakia).

Biology

The immature stages and the host plants are unknown. Adults were collected at the end of May and in June. Some of them were disturbed from the low undergrowth before sunset (Burda Mts, Mochovce, Tlmače).

Etymology

The naming of the new species is dedicated to the wife of the first author for her life-long support.

Molecular data (Fig. 19)

The DNA barcoded specimens form a unique BIN: BOLD:ABX1341. Five specimens of the new species have been successfully sequenced, here only given with samples ID (MM21352, MM22159, MM22197, MM22198, MM25325). Details concerning collecting site and sequence quality are found in the type list (paragraph "Material"). Further data is accessible via the public dataset DS-ELAMAR New *Elachista* species (https://v4.boldsystems.org/index.php/MAS_Management_DataConsole?codes=DS-ELAMAR).

DISCUSSION AND NOTES

The structure and shape of the male genitalia of *E. mariae* sp. n. are typical for the *E. bedellella* species group, in which the costa of the valva is medially convex.

The structure of the female genitalia of *E. mariae* is unique and atypical for this group as far as is known: having a bursa copulatrix with minute spines, dentate signum, and an appendix bursae.

However, according to Kaila (pers. comm.), the problem with females of this group is that they are rarely collected compared to males and only barcodes can match females to males. There are quite a few species with significant differences in barcodes that are known only from males with minor differences in the genitalia, making certain identification impossible.

According to Kaila, the female genitalia of *E. ohridella* are also unknown and those drawn in PARENTI's description (2001) belong to another species from an unrelated group near *E. pollinariella* Zeller, 1839. The only female genitalia that match this drawing are known from the type series named as *E. ohridella*, as the genitalia slide of the second female is missing (G. F. Turrisi, pers. comm.). Parenti, very unhelpfully, does not describe either the male or female genitalia that he illustrates or compare them with a closely related species.

We also present the results of DNA analyses of the new species and species of the *Elachista bedellella* species group, whose genetic data are known so far. In the generated DNA barcoded tree (Fig. 19), there are large distances between *E. mariae* and other *Elachista* species. However, we recognize that, as with female

genitalia, molecular data are still incomplete as they are lacking from some known species of this group, for example from *E. ohridella* and *E. laurikailai*.

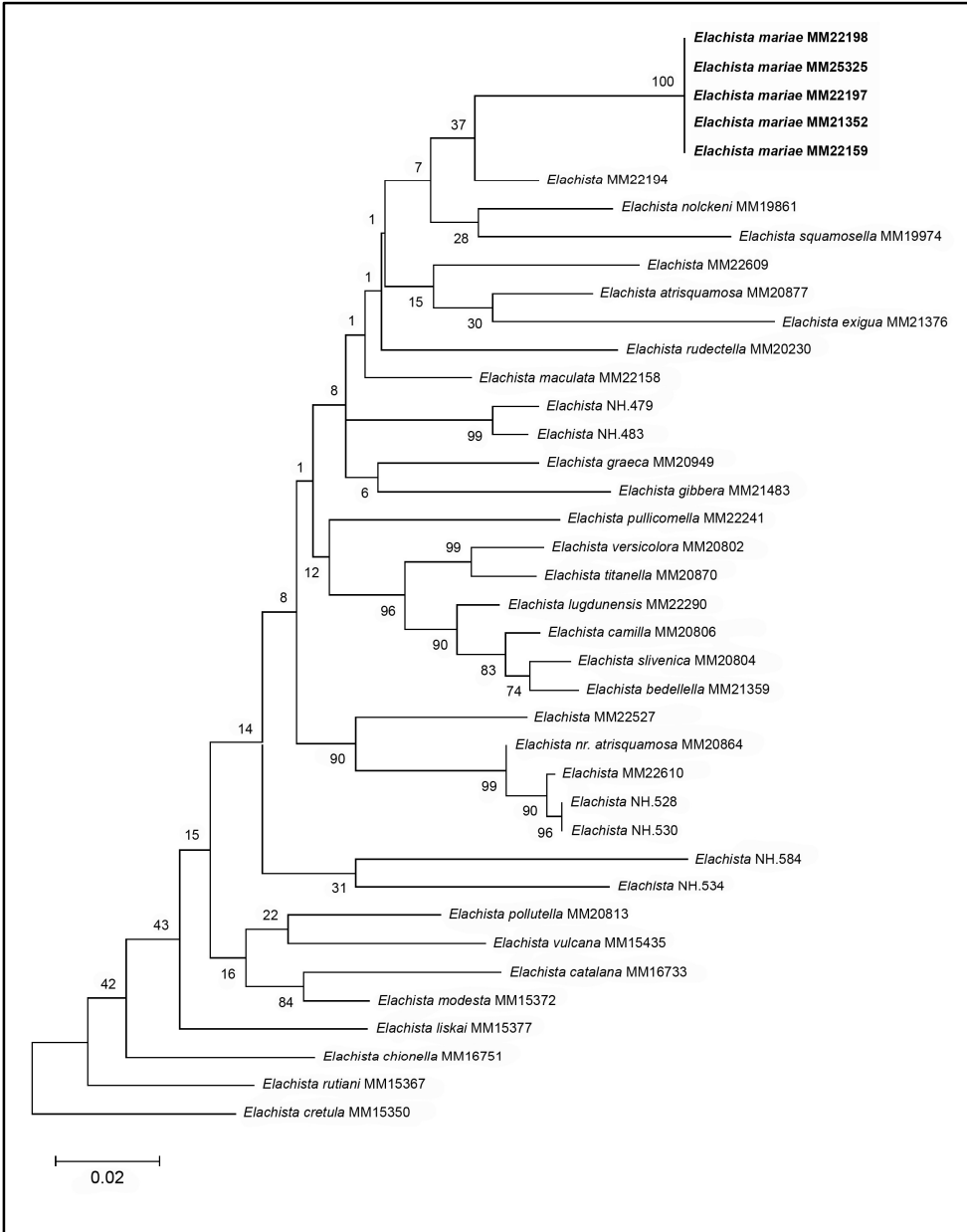


Fig. 19. A Neighbor-joining tree of *Elachista mariae* sp. n. (shown in bold type) and selected closely related species based on COI sequences (created by M. Mutanen). Scale bar represents 2 % K2P genetic divergence between sequences.

We collected males and females of *E. mariae* from the same localities in Slovakia and even some at the same time. This fact, combined with the study of the structure of the male genitalia and supported partly by the study of the original female genitalia and by the unique BIN, enable us to describe this as a new species.

We included a Bulgarian specimen in the Slovak type series due to its identical BIN and the similar structure of the male genitalia (Fig. 9), with the exception of the phallus, which is significantly narrower than in the Slovak specimens, which may be due to different ecological conditions. This could be the subject of further investigation.

We would like our contribution to stimulate a thorough revision of the *Elachista bedellella* species group, although due to the large number of species in this group, their similarity and the lack of material or insufficient knowledge of some of them, especially females, this represents a very difficult task for the future.

ACKNOWLEDGEMENTS

We express our sincere gratitude to Lauri Kaila (Finnish Museum of Natural History, Helsinki, Finland) for comprehensive help in the identification of the new species, providing the DNA barcoding of the specimens, the photograph and data of the Bulgarian specimen of the new species and for very constructive comments on the manuscript. Our thanks are also due to Marko Mutanen (University of Oulu, Finland) for his kind assistance in processing barcoding data into the Maximum Likelihood tree, to Ivan Richter (Prievidza, Slovakia), who generously provided his specimen for our study, to Thierry Varenne (Nice, France) for kindly allowing use of his photograph of the holotype of *Elachista laurikailai*, to Manuel König (Aquazoo Löbbecke Museum Düsseldorf, Germany), and to Giuseppe Fabrizio Turrisi (Museum of Natural History La Specola University of Florence, Italy) for valuable photographic assistance with the type material of *Elachista ohridella*. Finally, we are grateful to Robert J. Heckford (Plympton, UK), and Stella Beavan (Zeal Monachorum, UK) for checking and correcting the English, and helpful comments.

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