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The female of *Megacraspedus peslieri* Huemer & Karsholt, 2018 (Lepidoptera, Gelechiidae), a new case of brachyptery in alpine Lepidoptera

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Abstract

Megacraspedus peslieri was described from the Pyrenees in 2018 and subsequently also discovered at a few localities in the northern Cottian Alps (Italy). The hitherto unknown female was collected in these localities after a targeted search. As with some other representatives of the genus, the female is flightless and shows a strikingly strong reduction of the wings (brachyptery). Whereas the forewings are significantly shortened and narrowed, the hindwings are strongly reduced. Both the external morphology and the genitalia of the female are described and illustrated in detail, together with previously unpublished photographs of live adults of both sexes. The identification of specimens from the Alps as well as of the male and female were determined by means of a DNA barcode comparison with the holotype.

Key Words

Alps, brachyptery, DNA barcoding, female, *Megacraspedus peslieri*, wing reduction

Introduction

Wing reduction (brachyptery) or the complete loss of wings (aptery) are rare phenomena in Lepidoptera (Sattler 1991) and only reported for 35 families (Heppner 1991), with few recent additions i.e. for Notodontidae (Sattler and Wojtusiak 1999), Nymphalidae (Viloria et al. 2003) or Ethmiidae (Shovkoon 2008). Aptery is rarely a largely consistent group-specific trait, e.g. in the Psychidae and Heterogynidae (de Freina 2011; Arnscheid and Weidlich 2017). However, in most taxonomic groups with brachyptery there are different gradations of wing reduction, mainly of the hindwings and in the female, and the associated inability to fly (Huemer and Sattler 1989; Sattler 1991). Such tendencies are known in particular from climatically unfavourable areas such as subantarctic islands and mountain regions, but also from arid habitats, and in late autumn or winter-active species (Heppner 1991; Sattler 1991).

The gelechiid genus *Megacraspedus* is an example of widespread wing reduction, with the females of the majority of the 89 described species probably being brachypterous (Huemer and Karsholt 2018). Only in few closely related taxa such as the *M. fallax* species group do females seem to be normally winged (Huemer and Karsholt 2018; Huemer and Tokár 2021). Though more than half of the species are known exclusively from males, more or less pronounced reduction of wings, particularly hindwings, is widespread in species with described females, i.e. about 20 species. Here a hitherto unknown case of brachyptery is reported for *M. peslieri*, a species placed in a species group of its own.

Descriptive terminology follows Huemer and Karsholt (2018).

Material and methods

Sixteen males and two female specimens of *Megacraspedus peslieri* from France (holotype) and Italy have been examined. The material is preserved in the research collection of the Tiroler Landesmuseum Ferdinandeum (Hall, Austria) and kärnten.museum (Klagenfurt, Austria). The specimens were pinned and either spread or set. Initial species identification of males was based firstly on phenotypic characteristics (wing markings, colour, size), using the holotype of *M. peslieri* in the Tiroler Landesmuseum Ferdinandeum as a reference. This was confirmed by dissections and by DNA barcoding. Females were assigned to the species due to simultaneous occurrence with males and by the DNA barcode.

Tissue samples from a single hind leg of eight specimens were prepared according to prescribed standards to obtain the DNA barcode sequences of a 658 base-pair long segment of the mitochondrial COI gene (cytochrome c oxidase subunit 1). The tissue samples were successfully processed at the Canadian Centre for DNA Barcoding (CCDB, Biodiversity Institute of Ontario, University of Guelph) using the standard high-throughput protocol described in deWaard et al. (2008). The only available barcode sequence of the nearest neighbor *Megacraspedus ibericus* Huemer & Karsholt, 2018 in BOLD was added to the analysis, whereas several already published sequences of *Megacraspedus* were not (Huemer and Karsholt 2018).

All sequences were submitted to GenBank and details including complete voucher data and images can be accessed in the public dataset "*Megacraspedus peslieri* [DS-MEGAPESL]" dx.doi.org/10.5883/DS-MEGAPESL in the Barcode of Life Data Systems BOLD (Ratnasingham and Hebert 2007). Degrees of intra- and interspecific variation of DNA barcode fragments were calculated using the Kimura two-parameter model on the platform of BOLD systems v. 4.0. (http://www.boldsystems.org). A neighbor-joining tree was constructed under the Kimura two-parameter model in MEGA7 (Kumar et al. 2007). The photographs of live adults were taken with an Olympus OM-D Mark III camera and an Olympus 60 mm *f*/2.8 ED macro lens. The photographs of spread adults were taken with a Zeiss Stemi 508 KMAT stereo microscope, genitalia photographs with a Zeiss Axiolab 5 microscope, both adapted to an Olympus OM-D Mark III camera. Stacked photographs were edited using Helicon Focus 4.8 and Adobe Photoshop 6.0.

Results

Molecular analysis

Sequencing resulted in full length DNA barcodes of 658 bp for eight specimens of *M. peslieri*. The intraspecific p-distance was low with 0.32% on average and a maximum distance of 0.48%, mainly due to the slight deviation of the sequence of the holotype (Fig. 1), whereas

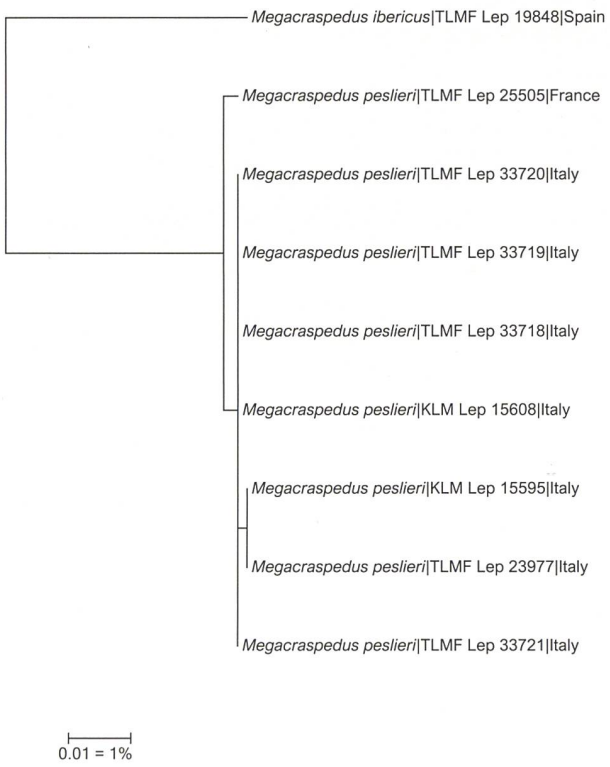

Figure 1. COI neighbor-noining tree of *Megacraspedus peslieri* and the nearest neighbor *M. ibericus*. Note: TLMF Lep 25505 represents the holotype.

the distance to the nearest neighbor *M. ibericus* was 7.37%. All specimens of *M. peslieri* clustered together and were assigned to the unique BIN:BOLD:ADM8362 (n = 9). Female specimens (specimen identifier TLMF_Lep_33721 and TLMF_Lep_3719) fully corresponded with males of the same cluster, thus strongly supporting the conspecifity of the males and females.

Taxonomic part

Megacraspedus peslieri Huemer & Karsholt, 2018

Material examined. 24♂, 2♀: Italy, prov. Torino, PN Orsiera - Rocciavré, Fenestrelle, Forte Serre Marie, 1830 m a.s.l., 45°02'57"N, 07°03'03"E, 21.8.2022, leg. Huemer (DNA Barcodes TLMF Lep 33718-33721; gen. slide P. Huemer GEL 1351♀; 1♂: Italy, prov. Torino, PN Orsiera - Rocciavré, Villaretto, Gran Faetto, Colletto, 1445 m a.s.l., 45°00'28"N, 07°08'28"E, 21.9.2019, leg. Huemer (all coll. Tiroler Landesmuseum Ferdinandeum, Innsbruck, Austria).

Description (Figs 2–6). For a detailed description of the male including the male genitalia see Huemer and Karsholt (2018). Males (Figs 2, 4) are more variable in size than originally described, with a forewing length ranging from 7.2–9.0 mm; furthermore, the cream-coloured dorsum is clearly separated from the remaining and predominantly brownish mottled part of the forewing with mainly cream-coloured veins in fresh samples, and the third segment of the labial palpus is entirely cream-white.

Figure 2. *Megacraspedus peslieri*, male in natural resting position (Italy, Alpi Cozie).

Figure 3. *Megacraspedus peslieri*, female in natural resting position (Italy, Alpi Cozie).

Female (Figs 3, 5–6). Segment 2 of labial palpus with long scale brush, dark brown on outer and lower surface, cream-white mottled with brown on inner surface, cream-white on upper surface; segment 3 cream-white. Antennal scape without pecten; flagellum cream-white, annulated with light brown. Head and thorax cream-white with some light brown mottling, particularly on tegula. Forewing length 3.8–4.4 mm. Forewing distinctly reduced, shorter than abdomen, with strongly convex dorsal margin, cream-white ground colour intensely mottled with

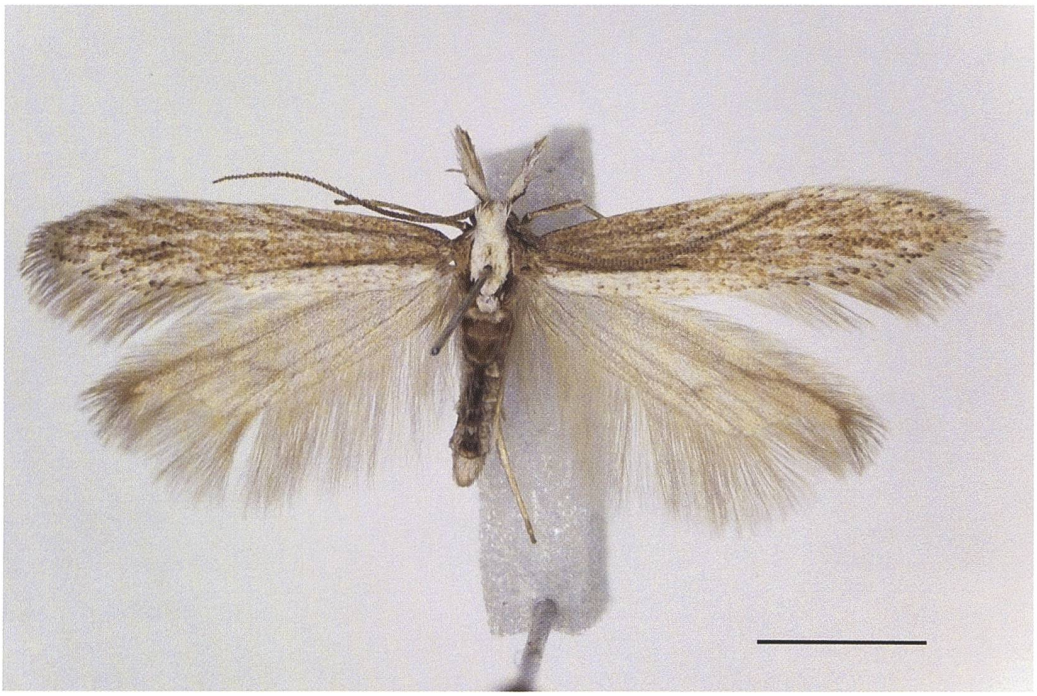

Figure 4. *Megacraspedus peslieri*, set male specimen (scale bar: 3 mm).

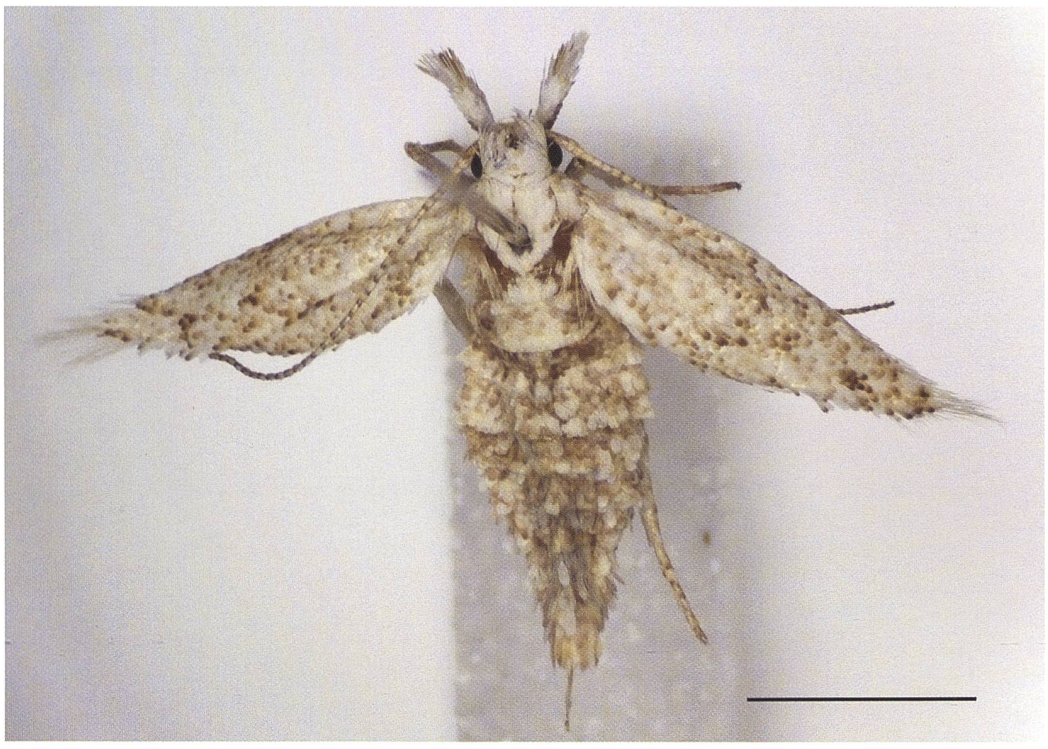

Figure 5. *Megacraspedus peslieri*, set female specimen (scale bar: 2 mm).

light brown; with few darker brown spots in middle and at apex; fringes reduced to group of very long, bristle-like hairs around apex. Hindwing reduced to minute sub-oval flap, with narrow long scales near apex, frenulum with one to two well-developed bristles.

The male and female are easily distinguished by the largely reduced wings of the female with an indistinct wing pattern compared to the male (Figs 2–5).

Female genitalia (Figs 7–8). Papilla analis large, weakly sclerotized, apically evenly rounded, slightly longer than segment VIII, lateral part with anteriorly widened sclerotized area; apophysis posterioris rod-like, short, about 1.65 mm long, apex slightly widened, rounded, anteriorly membranous intersegmental zone; segment VIII about 0.6 mm long, smooth, laterally sclerotized, medially membranous with microsculpture in anterior and posterior

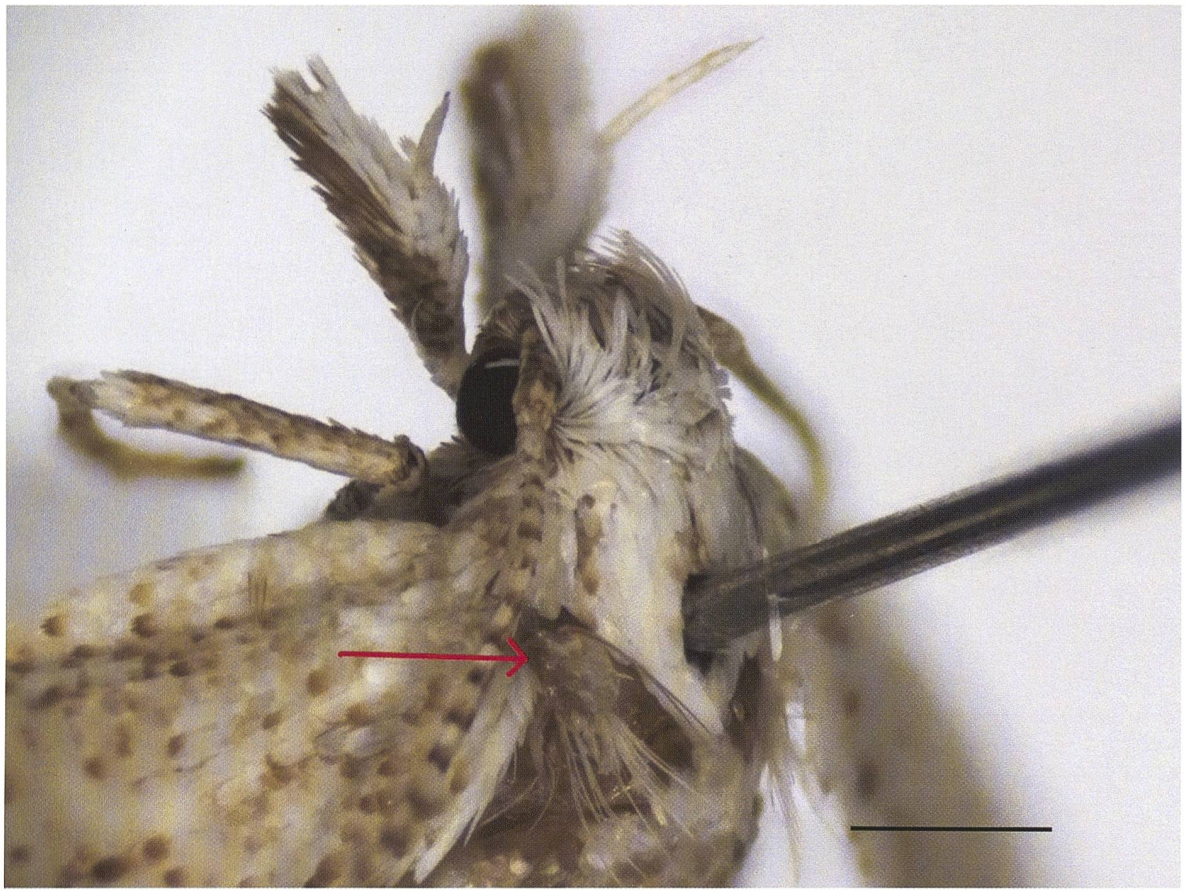

Figure 6. *Megacraspedus peslieri*, details of female hindwing (marked with red arrow) (scale bar: 0.5 mm).

parts; subgenital plate without specialized sclerotizations, anterior edge with short sinusoid projection delimiting ostium bursae; apophysis anterioris rod-like, about as long as segment VIII; colliculum about 1/2 length of apophysis anterioris, wrinkled, with small sclerotization anteriorly; ductus bursae slender, about 1.5 mm long; corpus bursae clearly delimited, about as long as ductus bursae, slender; signum moderately small, laterally oblong spiny plate, with about two dozen small to strong spines.

Remarks. The female genitalia support the unique position of *M. peslieri* and clearly differ from all other species groups particularly by the simple structure of the subgenital plate without specialized sclerotizations in combination with the peculiar signum. Furthermore, the distally rounded papilla analis combined with a short apophysis posterioris is rarely observed in other species.

Biology. The species is on the wing late in the season, from late August to the last third of September and active even at low temperatures of ca 6 °C. Both males and females of *M. peslieri* were observed sitting on grass stems or on other herbaceous plants and detected by illumination with a headlamp in the first two to three hours of the night. Simultaneously, males were attracted to UV light in large numbers.

Distribution. Only known from few localities in the Pyrenees (France, Spain) and the Cottian Alps (Italy) (Huemer and Wieser 2020).

Habitat (Fig. 9). The habitat in the Cottian Alps is predominated by xeromontanous grassland intermixed with rock formations on siliceous soil, at montane elevations from approximately 1400 to 1800 m a.s.l. In the Pyrenees the species was collected at lower altitudes from ca 250 to 900 m a.s.l.

Discussion

Brachyptery is a relatively widespread morphological adaptation in alpine Lepidoptera, which is interpreted as an avoidance strategy against strong winds and the increased risk of wind drift. However, it is limited exclusively to the female sex, while in other regions such as subantarctic island faunas, for example, both sexes can be flightless in extreme cases (Sattler 1991). In a few genera restricted to the alpine and subnival altitudinal zones of the Alps and other European high mountains, brachyptery is a consistent phenomenon, including in particular the genera *Sattleria* (Gelechiidae) (Pitkin and Sattler 1991; Huemer and Hebert 2011; Huemer and Timossi 2014; Timossi and Ruzzier 2020; Timossi and Huemer 2022) and *Sphaleroptera* (Tortricidae) (Whitebread 2006; Timossi and Ruzzier 2023). In other genera such as *Kessleria* (Yponomeutidae) (Huemer and Mutanen 2015; Huemer and Tarmann 1992), *Oxypteryx* (Gelechiidae)

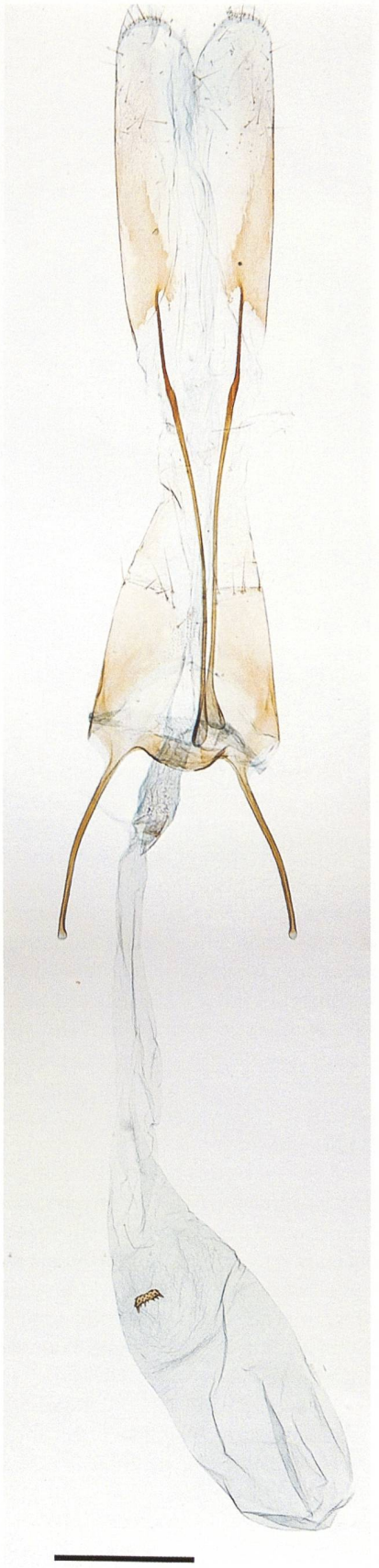

Figure 7. *Megacraspedus peslieri*, female genitalia (scale bar: 0.5 mm).

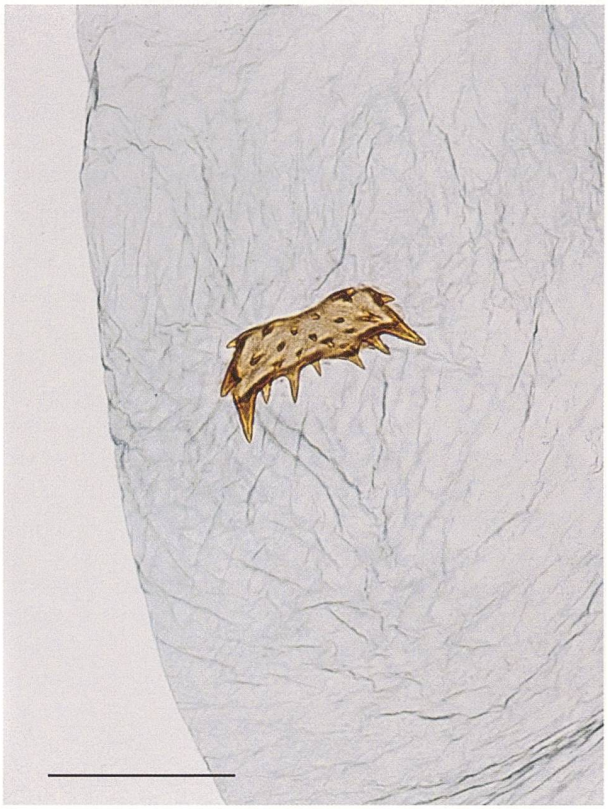

Figure 8. *Megacraspedus peslieri*, female corpus bursae with signum enlarged. (scale bar: 0.1 mm).

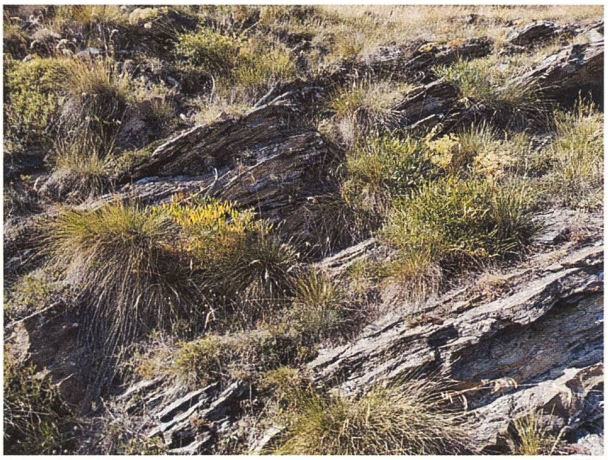

Figure 9. Habitat of *Megacraspedus peslieri* in the Cottian Alps, above Fenestrelle.

(Huemer et al. 2013), *Elophos* (Geometridae) (Müller et al. 2019) and *Agrotis* (Noctuidae) (Ronkay and Huemer 2018) there are different stages of brachyptery, from nearly fully winged to strongly pronounced wing reduction in high altitude taxa. Furthermore, brachyptery can also be restricted to few species within a genus, for example *Elachista brachypterella* (Elachistidae) (Klimesch 1990). In addition, the females of many fully winged species in mountain regions are generally not very active flyers.

The inability to fly, mostly manifested by wing reduction, is also found in some xeromontane groups of Lepidoptera

in the Alps. A particularly striking example is the genus *Megacraspedus*. Representatives of this very diverse, palaearctic genus are found in many montane regions, with the Alps being highly important for species diversity. Despite extensive revisionary work by Huemer and Karsholt (2018), new species have been discovered or recognized in this region in the recent past, not least thanks to the increasing implementation of molecular methods (Huemer et al. 2020a, 2020b; Timossi and Huemer 2021). Though female brachyptery is widespread in *Megacraspedus*, the female remains unknown for about half of the species (Huemer and Karsholt 2018), despite some recent advances (Nel and Varenne 2019; Huemer and Tokár 2021). However, the inability to fly does not necessarily reflect an adaptation strategy to certain habitat conditions, but rather seems to be related to morphologically defined species groups (Huemer and Karsholt 2018). For example, in the Pannonic region of eastern Austria *M. podolicus* (Toll, 1942), a member of the *Megacraspedus fallax* species group, which is fully capable of flying can be found cohabiting with the strongly brachypterous species of the *Megacraspedus dolosellus* and *M. binotella* species groups. However, in the absence of any closely related species to the highly isolated *Megacraspedus peslieri* it has not been possible to predict the ability of the females to fly. Since the newly detected female strongly differs from the male by the much smaller size, shorter and relatively broader and pointed forewings, the largely reduced hindwings, and in the forewing colour and pattern, the question of conspecifity of the sexes arose, but it finally could be proved by a DNA barcode analysis.

The currently known and extremely disjunct distribution pattern (Pyrenees, southwestern Alps) of a species with such a pronounced brachyptery raises attention to further yet unresolved questions. Due to the very small and possibly irrelevant differences in the DNA barcodes between these populations, a (formerly) continuous distribution must be assumed, at least in post-glacial periods. Alternatively, however, the species might be much more widespread and simply overlooked in many places. However, this scenario seems unlikely due to the intensive collecting activities in southern France over a long period.

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