

## **Article**



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# Scrobipalpa chardonnayi Huemer and Özden, sp. nov.: a new presumably endemic species from Cyprus (Lepidoptera, Gelechiidae)

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#### **Abstract**

Scrobipalpa chardonnayi Huemer & Özden, **sp. nov.** is described from the limestone mountains of northern Cyprus and considered as a possible island endemism. The new species shows closer phylogenetic relationships to *S. vasconiella* (Rössler, 1877) and some related species, but differs phenotypically and in male and female genitalia, as well as through significant divergences in DNA barcode. Morphologically relevant diagnostic characters are compared and figured. Finally, *S. vasconiella* is reported from Kyrgyzstan for the first time.

Key words: DNA barcoding, endemism, Gnorimoschemini, Mediterranean, morphology, new record, North Cyprus

#### Introduction

Scrobipalpa is a highly diverse genus of the tribe Gnorimoschemini (Gelechiinae, Gelechiidae), particularly in the Palaearctic region, with currently over 300 described species in this region (Povolný 2002; Huemer & Karsholt 2020; Bidzilya 2021). Especially from Asia, numerous new species have been described recently (Bidzilya *et al.* 2022; Bidzilya & Li 2010; Li & Bidzilya 2019), and many taxa, particularly from Kyrgyzstan, remain taxonomically unclear (Huemer unpubl.). Even the European fauna appears not fully documented, despite extensive revision (Huemer & Karsholt 2010; Huemer *et al.* 2020), with ongoing discoveries of new species (Bidzilya 2023; Bidzilya & Budashkin 2011; Huemer 2021; Leraut 2020; Varenne & Nel 2013, 2017, 2018) or publications of as yet unresolved species (Huemer & Mutanen 2022).

The implementation of genetic methods (DNA barcoding) has notably contributed to a more objectively defined species delimitation in *Scrobipalpa*, which historically often relied on subtle and diagnostically insufficient morphological characters (Povolný 2002). Here, we report on a genetically and morphologically distinct new species of *Scrobipalpa*, discovered during comprehensive sampling of the moth fauna of Northern Cyprus.

#### Materials and methods

A total of 11 specimens of the new species have been examined morphologically and compared with related species, particularly the genetically closest *Scrobipalpa vasconiella* (Rössler, 1877) and with *S. brahmiella* (Heyden, 1862). Male genitalia preparation implemented the so-called unrolling technique as introduced for the preparation of complex male genitalia in Gelechiidae by Pitkin (1986).

Tissue samples (a single hind leg) from three specimens of *Scrobipalpa chardonnayi* **sp. nov.** were prepared according to prescribed standards to obtain DNA barcode sequences of a 658 bp segment of the mitochondrial COI gene (cytochrome c oxidase subunit 1). The material was 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). In addition, 48 private barcode sequences of *Scrobipalpa* species in the Barcode of Life Data Systems (BOLD; Ratnasingham & Hebert 2007, Ratnasingham 2018) belonging to 8 species with a minimum p-distance of <7% were used for analysis. All these species differ morphologically from the new taxon and, with the exception of *S. vasconiella*, were considered exclusively for genetic analysis. All barcodes range between 551 and 658 bp. Further details including complete voucher data and images can be accessed in the public dataset DS-SCROBCYP "Scrobipalpa sp.n.—Cyprus" dx.doi.org/10.5883/DS-SCROBCYP in the Barcode of Life Data Systems BOLD (Ratnasingham & Hebert 2007).

All sequences were assigned to Barcode Index Numbers (BIN), algorithm-based operational taxonomic units that provide an accurate proxy for the true species. BINs were automatically calculated for records in BOLD that comply with the DNA Barcode standard (Ratnasingham & Hebert 2013).

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. (https://boldsystems.org). A Neighbor-Joining tree was constructed using the Kimura two-parameter model in MEGA7 (Kumar *et al.* 2016).

Photographs of adults were taken with an Olympus OM-D Mark III camera and a 60 mm macro lens, genitalia photographs with a Zeiss Axiolab 5 microscope, mounted with an Olympus OM-D Mark III camera. 60 to 90 stacked photographs were edited using Helicon Focus 4.8 and Adobe Photoshop 6.0.

Abbreviations. CHNHM = Cyprus Herbarium and Natural History Museum, Nicosia, Cyprus.

TLMF = Tiroler Landesmuseum Ferdinandeum, Innsbruck, Austria.

#### Systematic treatment

### Scrobipalpa chardonnayi Huemer & Özden, sp. nov.

(Figs. 1, 3, 5, 7, 9, 11, 13)

**Holotype** ♀ "N CYPRUS, Kyrenia/Girne Yilgaz E 280 m, 11.9.2023 leg. Huemer 33°13'39"E, 35°19'21"N leg. Huemer TLMF 2024-003" (gen. slide P. Huemer GEL 1359 ♀—DNA Barcode ID TLMF Lep 39121); coll. TLMF.

Paratypes: 3♀, 1♂, same label as the holotype (gen. slide P. Huemer GEL 1360 ♂—DNA Barcode ID TLMF\_Lep\_39127, TLMF\_Lep\_39168); 3♀, 3♂, N Cyprus, Iskele, Kaplica, Kantara Castle, 550 m, 33°55'02"E, 35°24'12"N, 5.6.2024, leg. Huemer; colls. CHNHM, TLMF.

Diagnosis. Scrobipalpa chardonnayi Huemer & Özden, sp. nov. is a medium-sized greyish species with some orange-brown mottling and the characteristic three black spots of Gnorimoschemini on the forewing. It differs from the related S. vasconiella by the less extensive orange-brown mottling (Figs. 1–2). Male genitalia are distinguished by the longer and more slender valva without a truncate apex, the distinctly shorter sacculus, the smaller vincular process, and the pointed saccus. The female genitalia of S. chardonnayi Huemer & Özden, sp. nov. differ from those of S. vasconiella by several features: longer apophyses posteriores and apophyses anteriores, the laterally excavated anterior edge of the medial depression of segment VIII, and the longer, more slender, and less curved signum with smaller basal teeth. Additionally, S. chardonnayi Huemer & Özden, sp. nov. and S. vasconiella show a significant divergence in DNA barcodes of approximately 6%. Apart from the less prominent orange-brown suffusion, the new species differs from the very similar S. brahmiella by the more slender tegumen and valva, the pointed apex of the sacculus, the strongly excavated medial depression of female segment VIII, and the small teeth at the base of the signum, which are largely reduced or absent in S. brahmiella (see Huemer & Karsholt 2010; Sattler 1986). Other species at a similar genetic distance are quite different in genitalia characters (see below and Huemer & Karsholt 2010). Significant differences in genitalia morphology are also found in some externally similar species, such as the Central and Eastern Asian S. punctulata Li & Bidzilya, 2019, S. intima (Povolný, 2001), and S. tannuolella Bidzilya, Huemer & Šumpich, 2022.

**Description**. Adult (Fig. 1). Head grey-brown, face bone white to cream-coloured; antenna grey-brown, distinctly annulated with cream; second segment of labial palpus predominantly grey-brown on outer surface, inner and dorsal surface cream, segment 3 mixed grey-brown with cream medial and dark brown apical part; thorax and tegula mixed grey-brown, tegula sometimes with few orange-brown scales. Forewing length  $3 \pm 5.5$ 0 mm (n=4),  $4.5 \pm 5.2$  mm (n=7). Forewing upper side mixed light grey-brown from light grey and dark tipped scales, distinct black plical, discal and discocellular spots, further small black spots in sub-basal and subapical area, black markings

frequently edged with orange-brown, furthermore orange brown scales particularly along dorsum and on veins; termen with light, apically dark brown tipped scales; grey-brown fringes without fringe line. Hindwing grey-brown with concolorous fringes. Underside of forewing dark grey-brown with light apical part of costa.



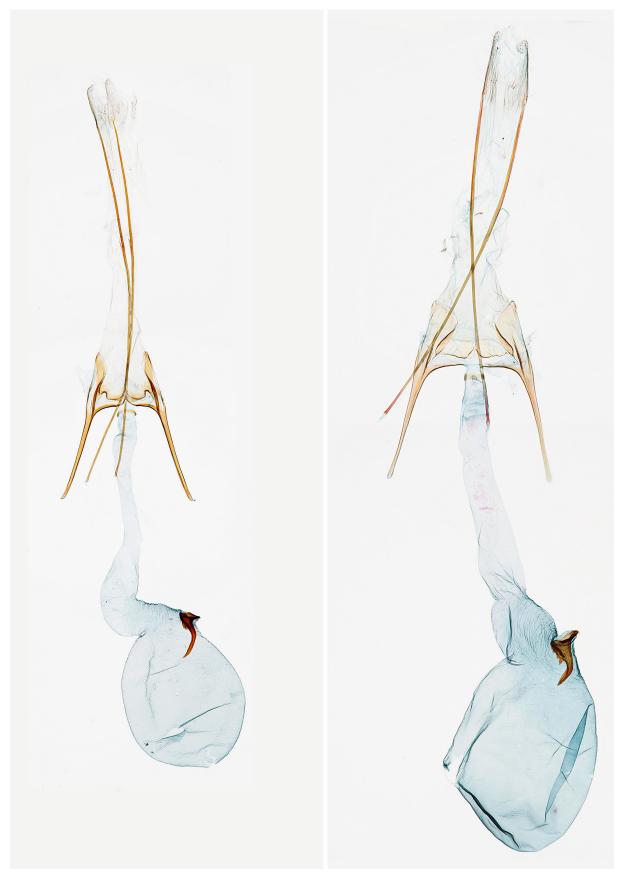
FIGURES 1–2. Adults of Scrobipalpa spp. 1, S. chardonnayi sp. nov., female, holotype. 2, S. vasconiella, male, Kyrgyzstan.



**FIGURES 3–4**. Male genitalia of *Scrobipalpa* spp. **3**, *S. chardonnayi* **sp. nov.**, paratype, gen. slide GEL 1360 P. Huemer. **4**, *S. vasconiella*, Kyrgyzstan, gen. slide GEL 1362 P. Huemer.



**FIGURES 5–8**. Male genitalia of *Scrobipalpa* spp., details of vinculum-valva (5–6) and uncus-tegumen (7–8). **5**, *S. chardonnayi* **sp. nov.**, paratype, gen. slide GEL 1360 P. Huemer. **6**, *S. vasconiella*, Kyrgyzstan, gen. slide GEL 1362 P. Huemer. **7**, *S. chardonnayi* **sp. nov.**, paratype, gen. slide GEL 1360 P. Huemer. **8**, *S. vasconiella*, Kyrgyzstan, gen. slide GEL 1362 P. Huemer.

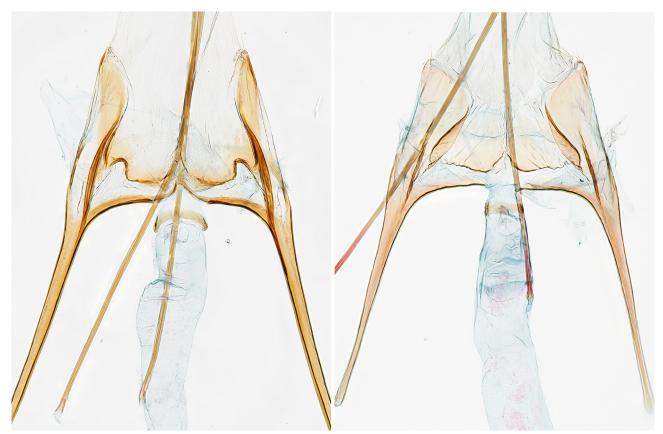


**FIGURES 9–10**. Female genitalia of *Scrobipalpa* spp. **9**, *S. chardonnayi* **sp. nov.**, holotype, gen. slide GEL 1359 P. Huemer. **10**, *S. vasconiella*, Kyrgyzstan, gen. slide GEL 1361 P. Huemer.

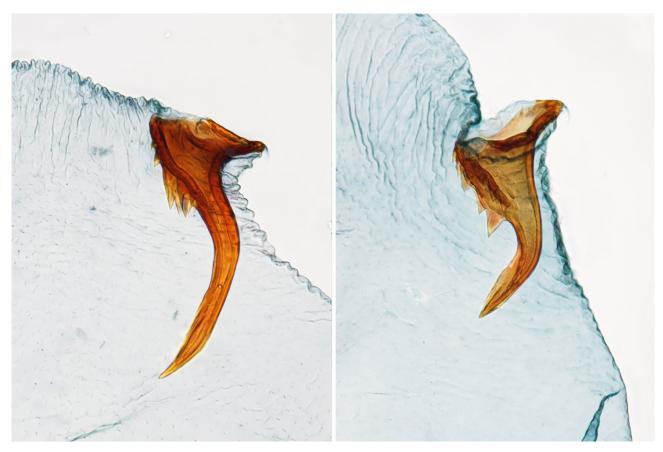
Variation. The extension of orange-brown mottling and black spots is quite variable, probably also depending on the age of specimens which increasingly loose their orange-brown.

Male genitalia (Figs. 3, 5, 7). Uncus moderately slender, longer than wide, apical corners rounded, posterior edge straight; gnathos-hook short, weakly curved; culcitula small; tegumen elongated, anteriorly broadly widened with sinusoid anterior edge, pedunculus sub-triangular; valva long, slightly exceeding apex of uncus, evenly slender, apex rounded; sacculus short, about one-fifth length of valva, nearly same width from base to apex, apically curved ventrad, with pointed apex; vincular process short, about half length of sacculus, tooth-like with outwardly turned and pointed apex, inner margin continuous with broad and deep V-shaped posteromedial emargination of vinculum; saccus shorter than vinculum, exceeding apex of pedunculus, basally moderately broad, distally evenly tapered to pointed apex; phallus moderately short, stout, ventrally with sclerotized ridge, apically with hooked sclerotization, coecum inflated.

Female genitalia (Figs. 9, 11, 13). Papillae anales sub-ovate, sparsely covered with short setae; apophysis posterioris nearly four times length of apophysis anterioris; segment VIII about as long as broad, smooth and without foamy sculpture; posterior margin of sternum VIII broadly emarginated, strongly sclerotized ventrolateral edges, anterior margin straight, with strongly sclerotized edge; subgenital plates distinctly broadened posteriorly and almost connected at 3/4 length, smoothly sclerotized, with several transverse folds at base of apophyses anteriores; ventromedial depression smooth, medially merged, strongly sclerotized anterior edge broadly rounded, medially with short triangular incision and few microtrichia, laterally with distinct excavation, sclerotized edge extending distinctly posteriad of anterior margin of segment VIII; apophysis anterioris straight, as long as segment VIII; colliculum narrow ring-shaped; ductus bursae anteriorly widened, with indistinct transition to sub-ovate corpus bursae; signum large, near entrance of ductus bursae, basal plate sub-oval, small, with several small teeth on inner side at base of hook, distal hook long and slender, weakly curved at about middle.



**FIGURES 11–12**. Female genitalia of *Scrobipalpa* spp., details of segment VIII. **11**, *S. chardonnayi* **sp. nov.**, holotype, gen. slide GEL 1359 P. Huemer. **12**, *S. vasconiella*, Kyrgyzstan, gen. slide GEL 1361 P. Huemer.



**FIGURES 13–14**. Female genitalia of *Scrobipalpa* spp., details of signum. **13**, *S. chardonnayi* **sp. nov.**, holotype, gen. slide GEL 1359 P. Huemer. **14**, *S. vasconiella*, Kyrgyzstan, gen. slide GEL 1361 P. Huemer.

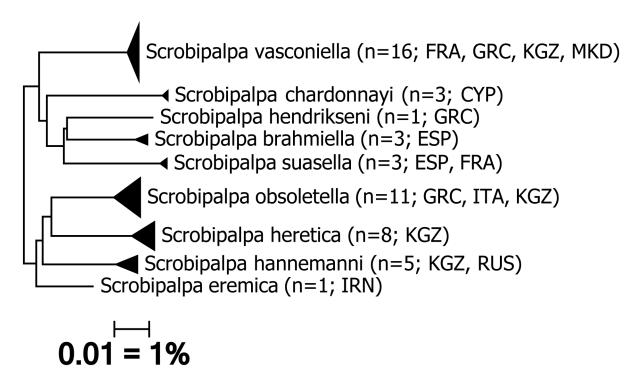
**Molecular data**. *Scrobipalpa chardonnayi* **sp. nov.** differs strongly in DNA barcodes from all related species of *Scrobipalpa*. The three successfully barcoded specimens cluster in an unique BIN: BOLD:AFN8036 with a maximum intraspecific divergence of 0.32% and a p-distance to the nearest neighbor, *S. vasconiella* (BIN BOLD: ABA3381), of 6.1%. In addition, seven congeneric species exhibit a minimum genetic distance to the new species, ranging from 6–7% (Fig. 15).

**Bionomy**. The early stages and the foodplant are unknown. The species was recorded in mid-September and early June, but the generation sequence is still unknown. The habitat is located on the northern slopes of an extensive mountain ridge (Besparmak/Pentadactylos), composed of carbonate rocks with Mediterranean maquis vegetation (Fig. 16).

**Distribution**. Cyprus (North), Kyrenia (Girne) and Iskele districts, only known from the northern exposed slopes of the Kyrenian mountain ridge (Besparmak/Pentadactylos) so far.

**Etymology**. The name is derived from the Chardonnay grape variety and recalls its widespread presence in the type region as well as the guesthouse named after it, which served as the central location for the surveys.

**Remarks.** Scrobipalpa vasconiella is widely distributed from the Mediterranean to the Middle East but absent in Cyprus (Huemer & Karsholt 2010). It was frequently mixed with S. brahmiella, and, i.e., genitalia figures of the female sex in Huemer & Karsholt (2010) in fact belong to the latter species, whereas Sattler (1986) gives a correct figure of female genitalia. Based on these errors, S. brahmiella was incorrectly reported from North Macedonia (Huemer et al. 2011). However, these records belong to S. vasconiella, which is here also recorded for the first time for Kyrgyzstan: Chüy Oblast, North Tian-Shan, Alexander Mountains near Bishkek, 1060 m, 74°32'2.7"E, 42°46'31.5"N, 22.6.2022, leg. P. Huemer.



**FIGURE 15.** Neighbor-Joining tree of selected species of *Scrobipalpa* (Kimura 2-parameter, built with MEGA7 (Kumar *et al.* 2016); Note: the scale bar only applies to internal branches between species. Width of triangles represent sample size, depth the genetic variation within the cluster. Source: DNA Barcode data from BOLD, DS-SCROBCYP "Scrobipalpa sp.n.—Cyprus" (Barcode of Life Database; Ratnasingham 2018).



FIGURE 16. Habitat of S. chardonnayi sp. nov. holotype.

#### **Discussion**

The lepidopteran fauna of Cyprus, the fourth largest Mediterranean island, includes approximately 900 species according to Fauna Europaea (Karsholt & Nieukerken 2013), but it seems to be far from fully documented (Barton 2015, 2018). This is especially true for the northern part of the island, which could not be effectively studied for several decades due to the complex political situation. Following initial comprehensive surveys, it is expected

that the inventory for Cyprus will significantly expand in this area and others. A final checklist and a subsequent biogeographical analysis are still pending, and current attempts must be considered incomplete (Arenberger 1994).

However, many species found in the Levant or the Near and Middle East are known to occur in Cyprus as their only locations within politically defined Europe. Some species are even considered endemic. The newly described *Scrobipalpa* might ultimately prove to be endemic to Cyprus. It has not been found in previous surveys in Lebanon or in the well-studied Gelechiidae fauna of Israel (Bidzilya *et al.* 2019; Povolný 2002). Since no additional findings have been reported from the southern and much better-studied part of Cyprus, it could be an endemic species of the limestone mountains (Kyrenia Mountain Range) in northern Cyprus, as these rock formations are absent in the south of the island. Alternatively, a wider distribution in Cyprus or even in the Levant remains a possibility, though this appears unlikely due to the more intensive sampling activities there.

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