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First taxonomic revision of the *Ptilocephala albida*-species group in Europe (Lepidoptera: Psychidae, Oiketicinae)

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Abstract

The European part of the *Ptilocephala albida*-species group is revised by morphological and molecular characteristics. This supports the existence of six taxonomic separated species in Europe, of which one is described here as new: *Ptilocephala rutjani* Arnscheid & Weidlich, sp. n. *Ptilocephala lorquiniella* (Bruand, 1853) "stat. rev"., is again treated as a separate species. In addition, this paper gives an overview on the current status of the revision of the genus *Ptilocephala* Rambur, 1858 with special emphasis to DNA barcoding.
KEY WORDS: Lepidoptera, Psychidae, Oiketicinae, cryptic diversity, DNA barcode, morphology, new species, *P. lorquiniella*, *P. rutjani*, Balkans, Bulgaria, Greece, North-Macedonia, Ukraine.

Taxonomische Revision der *Ptilocephala albida*-Artengruppe in Europa (Lepidoptera: Psychidae, Oiketicinae)

Zusammenfassung

Der europäische Teil der *Ptilocephala albida*-Artengruppe wird anhand morphologischer und molekularer Merkmale revidiert. Als Ergebnis wird die Existenz von sechs taxonomisch deutlich differenzierten Arten in Europa festgestellt, von denen eine hier als *Ptilocephala rutjani* Arnscheid & Weidlich, sp. n. als neu beschrieben wird. *Ptilocephala lorquiniella* (Bruand, 1853) "stat. rev"., wird wieder in den Artrang erhoben. Zusätzlich wird in dieser Arbeit ein Überblick über den aktuellen Stand der Revision der Gattung *Ptilocephala* Rambur, 1858 unter besonderer Berücksichtigung der molekularen Daten gegeben.
SCHLÜSSELWÖRTER: Lepidoptera, Psychidae, Oiketicinae, kryptische Diversität, DNA Barcode, Morphologie, neue Art, *P. lorquiniella*, *P. rutjani*, Balkan, Bulgarien, Griechenland, Nord-Mazedonien, Ukraine.

Revisión taxonómica del grupo de las especies de *Ptilocephala albida* en Europe (Lepidoptera: Psychidae, Oiketicinae)

Resumen

Se revisa, por características morfológicas y moleculares, la parte europea del grupo de especies de *Ptilocephala albida*. Esto respalda la existencia de seis especies taxonómica separadas en Europa, de las cuales una se describe aquí como nueva: *Ptilocephala rutjani* Arnscheid & Weidlich, sp. n., *Ptilocephala lorquiniella* (Bruand, 1853) "stat. rev"., otra vez, es tratada como una especie distinta. Además, este trabajo da una vision general sobre el estado actual de la revisión del género *Ptilocephala* Rambur, 1858 con el énfasis especial al ADN código de barras genético.
PALABRAS CLAVE: Lepidoptera, Psychidae, Oiketicinae, diversidad críptica, AND Código de barras, morfología, nueva especie, *P. lorquiniella*, *P. rutjani*, Balcanes, Bulgaria, Grecia, Macedonia del Norte, Ucrania.

Introduction

The genus *Ptilocephala* Rambur, 1858 includes a total of 23 described species in the western Palaearctic (SOBCZYK, 2011; ARNSCHEID & WEIDLICH, 2017). The *Ptilocephala* species are mainly distributed on the Iberian Peninsula, in the Pyrenees, the Alps and on the Apennine Peninsula. Less species are known from Central Europe, the Balkans and Anatolia. Two species have been found in North Africa, and one single species is known from the Altai Mountains in Russia. Moreover, several currently undescribed species are also existent from various localities.

Ptilocephala was partially revised for the first time by BOURGOGNE (1967). Based on his studies, we introduce here the *Ptilocephala albida*-species group. This is a cluster of six European species that have a high probability of being closely related and that exist completely allopatric. The species *P. albida* (Esper, 1786), *P. lorquiniella* (Bruand, 1853) stat. rev., *P. rutjani* sp. n., *P. piae* Hättenschwiler, 1996, *P. wockei* (Standfuss, 1882) and *P. malvinella* (Millière, 1858) are grouped together because of their morphological similarities. This species group is characterised by showing 7 veins from dc on forewing, ciliated antennal pecten and spines on the membrane between the VII and VIII segment ventrally. Moreover, also phenological and molecular similarities exist. This paper deals with the European species *P. albida*, *P. lorquiniella* and *P. rutjani*. The revision of the North African species of the *P. albida*-species group is part of an ongoing study (ARNSCHEID et al., in prep.).

Material and methods

General

Netting the adults and visual searching for the larval cases in the wild are the valuable methods of gaining samples. Flying males are to be found on dry and rocky slopes, heaths, steppe and other xerothermic open landscapes. They are active morning until midday hours, and are, therefore, never attracted to light. Their caterpillars live polyphagous on moss and different herbs.

Altogether, more than 500 dried male specimen, mostly with their larval cases, from the whole distribution area were examined during these study.

DNA barcoding

For the DNA analyses, tissue samples were sent in a lysis plate to the Canadian Centre for DNA barcoding, in Ontario, Canada, especially within the framework of our private project, POESE (Psychidae of East- and South Europe). DNA extraction, amplification, and sequencing of the barcode region of the mitochondrial cytochrome oxidase I (COI) gene (658 base pairs at the 5' terminus) were carried out following protocols by DEWAARD *et al*. (2008). The taxonomic and collection data, voucher image, COI sequence and/or GenBank accession numbers are available for all specimens in BOLD (Barcode of Life database, http://www.boldsystems.org). The neighbor-joining tree is based on the Kimura 2 parameter (K2P) model of nucleotide substitution (KIMURA, 1980) as recommended in the barcoding protocol (RATNASINGHAM & HEBERT, 2007, 2013) and was obtained using MEGA 7 (KUMAR *et al*., 2016).

During the author's project, samples of 19 species of *Ptilocephala* were barcoded. This is more than 85 % of the hitherto described species. Within the framework of this project, we found several references to cryptic species. Also results to the intraspecific relationship within the genus, as well as to other Psychidae genera, were obtained. Their complete presentation is part of an ongoing study.

Images

Most images of the adults were taken with an Olympus E1 digital camera with a 35-50 mm macro

lens. A series of 12 single shots were stacked with COMBINE ZP using Soft Stack and post-processed with PHOTOSCAPE V.37 (www.photoscape.org/) and PAINT.NET v4.2.13 (www.getpaint.net). Several other images were taken with digital cameras Nikon 5500 and 7000 with Nikkor lens 105 mm. Images of male genitalia (procedure as described in ARNSCHEID & WEIDLICH, 2017) were taken with an Olympus OMD EM10 Mark II digital camera using an Olympus stereomicroscope with photo adapter and stacked with COMBINE ZP using Soft Stack; sharpened and denoised with NEAT IMAGE V8 and post-processed with PHOTOSCAPE V.37.

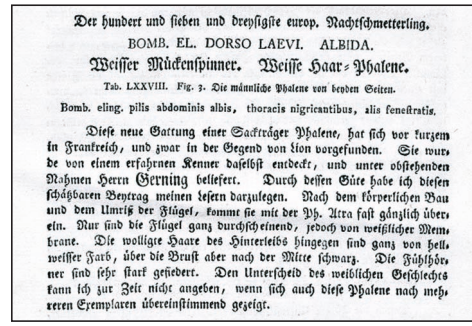
Abbreviations

NHM	The Natural History Museum, London
BOLD	Barcode of Life database
CER	Coll. Evgeniy V. Rutjan, Kiev
CMW	Coll. Michael Weidlich, Neißemünde
CWA	Coll. Wilfried R. Arnscheid, Bochum
dc	discal cell
e. l.	ex larva
e. o.	ex ovum
e. p.	ex pupa
LMW	Landesmuseum Wiesbaden, Germany
MNHN	Musée National d'Histoire Naturelle, Paris
MWM	Museum Witt, München
SMNK	Staatliches Museum für Naturkunde Karlsruhe, Germany
ZSM	Zoologische Staatssammlung München, Germany
FI	Forewing-Index (Maximal length wing-base to apex divided through maximal wing width).

Fig. 1.– The syntypes of *P. albida* (Esper, 1786) in the Gerning collection (LMW): **a.** The original box of the Gerning collection with the two syntypes. **b.** The lectotype. **c.** The drawing of the type specimen in the book of ESPER (1786).

Historical hints

The first taxon still presently including in the genus *Ptilocephala* was first described by Johann Jacob Esper (1742-1810) in 1786 from Lyon in France. He originally named it *Phalaen[a] Bom[yx] albida*. Hence, it is one of the very early described species ever still today classified as a Psychidae. Esper himself was a scientist at the University of Erlangen in South Germany and author of a series of tremendous scientific books about minerals, birds, plants, mollusks and insects. He described many insect species as new for science. Esper's Lepidoptera collection is preserved in the ZSM. Parts of the species described by Esper were loaned to him by Johann Christian Gerning (1745-1802) who had one of the biggest private Lepidoptera collections of the entire Europe at that time. With reference to Esper's description (fig. 2), type material in the Gerning collection is only valid if Esper affirms that he received the respective material from Gerning ("they were communicated to him"). With very few exceptions, Esper later returned all these specimens back to Gerning. Also *P. albida* has been described from material housed in the Gerning collection. After 250 years the two syntypes of *P. albida* are still in good condition (fig. 1, a-b). One specimen has been designated as lectotype by HACKER (1999).



Der hundert und sieben und dreyßigste europ. Nachtschmetterling.

BOMB. EL. DORSO LAEVI. ALBIDA.

Weisser Mückenspinner. Weisse Haar-Phalene.

Tab. LXXVIII. Fig. 3. Die männliche Phalene von beyden Seiten.

Bomb. eling. pilis abdominis albis, thoracis nigricantibus, alis fenestratis.

Diese neue Gattung einer Sackträger Phalene, hat sich vor kurzem in Frankreich, und zwar in der Gegend von Lion vorgefunden. Sie wurde von einem erfahrnen Kenner daselbst entdeckt, und unter obstehenden Nahmen Herrn Gerning beliefert. Durch dessen Güte habe ich diesen schätzbaren Beytrag meinen Lesern darzulegen. Nach dem körperlichen Bau und dem Umriß der Flügel, komme sie mit der Ph. Atra fast gänzlich überein. Nur sind die Flügel ganz durchscheinend, jedoch von weißlicher Membrane. Die wolligte Haare des Hinterleibs hingegen sind ganz von hellweisser Farb, über die Brust aber nach der Mitte schwarz. Die Fühlthörner sind sehr stark gefiedert. Den Unterscheid des weiblichen Geschlechts kann ich zur Zeit nicht angeben, wenn sich auch diese Phalene nach mehreren Exemplaren übereinstimmend gezeigt.

Fig. 2.– Facsimile of the original description of *P. albida* by ESPER (1786).

Taxonomic part

Ptilocephala Rambur, 1866
Cat. Syst. Lep. Andalousie, (2): 307
Type species*: Psyche agrostidis* Schrank, 1802, by subsequent designation by SOBCZYK (2011).
Hyalina Rambur, 1866 *nec* Schumacher, 1817
Cat. Syst. Lep. Andalousie, (2): 310
Type species: *Bombyx albida* Esper, 1786, by subsequent designation by TUTT (1900).
Hyalina was established to denote an unnecessary subgenus of *Ptilocephala* Rambur, 1866, and is a junior homonym of *Hyalina* Schumacher, 1817 (Mollusca). Already treated as a junior subjective synonym of *Ptilocephala* by STRAND (1913) and replaced with *Deuterohyalina* Dalla Torre, 1913 (SOBCZYK, 2011).

Ptilocephala albida (Esper, 1786), fig. 3, a, b.
Bombyx albida Esper, 1786.
Type locality: Lyon, FRANCE. Lectotype ♂, designated by HACKER (1999) (LMW).
= *Phalaena (Bombyx) semiluctifera* Villiers, 1789. *Car. Linn. Ent.*, **2**: 159
= *Bombyx hyalina* Fabricius, 1793. *Ent. Syst*., **3**(1): 472
= *Tinea vitrella* Hübner, [1813]. *Sammlung. Eur. Schmett. Tin*.: pl. 55, fig. 272
= *Psyche albella* Meigen, 1832. *Syst. Beschr. Schmett. Eur*., **3**: 12, nr. 9, pl. 88 fig. 15
= *Psyche albivitrella* Bruand, 1850. *Mem. Soc. Emil. Doubs*, (3) **1**: 28
= *Psyche millierella* Boisduval, 1852. *Bull. Soc. Ent. Fr*., (2) **10**: 22
= *Psyche tabanivicinella* Bruand, 1853. *Mem. Soc. Emil. Doubs*, (2) **3**: 43, nr. 23, pl. 1 fig. 23

Diagnosis: Wingspan 14-16 mm, FI = 1.8. Costal margin slightly convex, apex roundish, termen oblique and convex. Whitish grey, wings hyaline with fine short blackish hair-like scales. The whitish grey hyaline base-color and the density of scalation is somewhat variable also the intense of the blackish color. Head, thorax and abdomen are also characterised with long blackish hair. Fringe scales are short, and whitish grey. Forewings with 7 veins emanating from dc, r2 and r3 always free originating from dc. Antenna bipectinate, long ciliated, whitish scaled on the rear side, with 24-28 segments. The spines on the membrane between segments VII and VIII ventrally. Female apterous. Male genitalia: Variable, trapezoidal in general appearance, Tegumen and vinculum melted. Tegumen weakly indented and showing two small hocks distally, slightly folded laterally. Valva short and broad, not reaching the distal end of the tegumen, cucullus roundish, densely covered with very short hair-like setae distally, clasper of sacculus short, slightly curved, slender or broader, sclerotized, with short spines distally. Anellus roundish distally, laterally covered with short curved thorns. Vinculum broad, trapezoid, distinctly indented laterally, saccus very long and stretched, slightly shorter than genitalia

length, phallus thick, weakly curved, slightly longer than length of both tegumen and vinculum (fig. 8 a-b).

Larval cases: length 14-15 mm, width 5-11 mm, tubular with short silky tube at rear end, irregularly covered with moss (fig. 4, a, b).

Distribution: *P. albida* is distributed from South France northwards to the Saarland, the south-westernmost part of Germany (SOBCZYK & WERNO, 2011). In Switzerland it has a very restricted population near Geneva (HÄTTENSCHWILER, 1997). In Italy it is found very locally in the Ligurian Alps and in the region Emilia-Romagna on the northern Apennine Peninsula (BERTACCINI, 2009).

***Ptilocephala rutjani* Arnscheid & Weidlich, sp. n.** (fig. 3, f-h)
http://zoobank.org/5EE78BC2-7499-4FCD-8C8D-D11E93BA5D1D

Holotype ♂, UKRAINE, Kherson prov., Tschernomorsky res., Ivano-Rybaltshansky, 12-17-IV-2001, leg. E. Rutjan. The holotype is deposited in SMNK.

Paratypes 102 ♂♂, same data as holotype (CER, CMW, CWA, SMNK); 1 ♂ with larval case Greece, Makedonia, Oros Vermio, near Kato Vermio W, 1.050 m, e. p. 04-IV-2011 (CMW); 1 ♂, with larval case, same data, e. l. 26-V-2011, leg. M. Weidlich (CMW). Larval cases: (on pins): 7 ♂♂ and 12 ♀♀, same data as holotype (CER). 1 larval case Makedonia, Oros Vermio, Umg. Kato Vermio W, 24-IV-2011, 1.050 m leg. M. Weidlich (CMW). North-Macedonia: 2 larval cases, near Ochrid, 1 km N, 700 m, 18-IV-2011, leg. M. Weidlich (CMW). Bulgaria: 1 larval case with ♂ exuvia, 1 ♀ larval case, near Stara Zagora W, Stare selo 2 km S, 400 m, 23-IV-2011 (CMW); 1 larval case near Stara Zagora W, Stare selo 2 km S, 400 m, e. o. 28-VIII-2011 (CMW); 6 larval cases, near Stara Zagora W, Stare selo 2 km S, 400 m, e. o. IV- 2012, leg. M. Weidlich (CMW, fig. 5 a-d).

Diagnosis: Wingspan 16-18 mm, FI = 2.0. Costal margin straight, apex pointed, termen oblique slightly convex in some specimen. Whitish grey, more blackish grey towards costa and outer margin. Wings hyaline with fine short hair. Head, thorax and abdomen short blackish hairy, also sometimes at the abdomen. Fringe scales short, whitish grey. Forewings with 7 veins emanating from dc. Veins r2 and r3 mostly from one point rising, rarely divided. Antenna bipectinate, unscaled, with 24-30 segments. The ciliae are slightly longer than in the other species of the group. The spines on the membrane between segments VII and VIII only ventrally. Female apterous. Male genitalia: Ovally in general appearance, tegumen and vinculum ovally melted. Tegumen weakly indented distally and slightly folded laterally. Valvae short, not reaching the distal end of the tegumen, slender, slightly enlarged distally, cucullus roundish, covered with short hair-like setae distally, clasper of sacculus short, curved inwardly, slender, sclerotized, with short spines distally. Anellus slightly pointed, laterally covered with short curved spines. Vinculum ovally, not indented laterally, saccus very long and stretched, slightly shorter than genitalia length, phallus thick, weakly curved, slightly longer than length of tegumen and vinculum (fig. 8 e-f). Larval case: Variable, length 14-15 mm, width 5-11 mm. Male larval cases from Ukraine are more tubular and bear a short silken tube at the rear-end, whereas female larval cases of the same origin are more bulging in general. The coating consists of debris of lichens. The larval cases of the Balkan populations are quite similar to *P. albida* and irregularly covered with moss and twigs (fig. 4, e-i).

Discussion: Apart from the genetic distance, it differs morphologically mainly by the higher FI: 2.0, which is 1.8 for each of the other species of the *P. albida*-species group. In *P. rutjani* sp. n. the veins m2 and m3 of the forewings always originate from one point whereas they are always separated in *P. albida* and originate separately or from one point in *P. lorquiniella* stat. rev. The white hairs on the abdomen are only rudimentary in *P. rutjani* sp. n. and are sometimes missing completely.

Distribution: Beside the type locality found in Ukraine, it has been recorded in Greece, North-Macedonia and Bulgaria.

Biology: *P. rutjani* sp. n. is bound to xerothermic habitats such as steppes and rocky slopes (figs 5 a-c, 6 a, b, d). It flies in bright sunshine from mid-April to early June from sea level to about 1000 m. The caterpillars live polyphagous on various herbaceous plants, grass and moss. They spin to pupate on dry branches or on the ground, more rarely on rocks or boulders (figs 5 d-f, 6 c).

Etymology: We dedicate this new species to Evgeniy Vladimirovich Rutjan, Kiev, who collected the first specimen. As a Psychidae specialist he has intensively studied the family Psychidae of the Ukraine and published various papers.

Ptilocephala lorquiniella (Bruand, 1853), **stat. rev.** (fig. 3 c-e)
Psyche lorquiniella Bruand, 1853. *Mem. Soc. Emil. Doubs*, (2) **3**: 46, nr. 25, pl. 1 fig. 25
= *Psyche plumosella* Bruand, 1853. *Mem. Soc. Emil. Doubs*, (2) **3**: 48, nr. 26
= *Psyche plumosella* Rambur, [1866]. *Cat. Syst. Lep. Andalousie*, (2): 311, pl. 3, fig. 3
= *Psyche albidella* Rambur, [1866]. *Cat. Syst. Lep. Andalousie*, (2): pl. 3 figs 4, 4a
Type locality: Andalousie [Andalucía], SPAIN.

There is no information on the syntypes in the original description and their deposition remains unknown.

Diagnosis: Wingspan 16-19 mm, FI = 1.78. Costal margin slightly convex, apex roundish, termen oblique and convex. Wings hyaline with fine short hair. Base-color from whitish grey to white, light grey or blackish grey beyond dc to outer margin. Populations are variable in base-color and the density of scalation. Head, thorax covered with long blackish hair combined with stits of longer whitish hair. The abdomen is distally covered with long white hair, distinctly protruding the distal end of abdomen. Fringe scales short, whitish grey. Forewings with 7 veins emanating from dc, r2 and r3 free originating or one point rising. Antenna bipectinate, whitish scaled on the rear side, with 24-30 segments. The spines on the membrane between segments VII and VIII ventrally. Male genitalia: Trapezoidal in general appearance, tegumen and vinculum melted. Tegumen weakly indented and showing two small hocks distally, slightly folded laterally. Valvae short, not reaching the distal end of the tegumen, broader, sometimes distinctly enlarged distally, cucullus roundish, densely covered with very short hair-like setae distally, clasper of sacculus short, straight, slender, sclerotized, with short spines distally. Anellus roundish distally, laterally covered with short curved thorns. Vinculum broad, trapezoid, distinctly indented laterally, saccus very long and stretched, slightly shorter than genitalia length, phallus thick, weakly curved, slightly longer than length of both tegumen and vinculum (fig. 8 c-d). Larval case: Variable, length 13-15 mm, width 5-11 mm, tubular, somewhat bulging in female. Male larval cases with short silken tube at rear-end, the coating differs between different populations. Mostly irregularly covered with dry grass stems and twigs, often with silken stitches (fig. 4 c, d).

Discussion: The type locality of *P. lorquiniella* stat. rev. is not stated. But there is no doubt, that Bruand received the type specimen from Pierre Joseph Michel Lorquin (1797-1873), who collected them in Andalusia. The number and recent deposition of the syntypes of *P. lorquiniella* stat. rev. is unknown (SOBCZYK, 2011). REBEL (1910) treated it as a "var." (= ssp.) of *P. albida*. He refers to the different shape of the larval cases and the slightly darker base-color of the males as the main differences. LERAUT (1980, 1997) interpreted *P. lorquiniella* stat. rev. as a junior subjective synonym of *P. albida* but it seems questionable whether he studied comprehensive material from the whole distribution area. Looking closer to a larger series several slight morphologic differences between the two taxa become visible. Beside the different FI the shape of the forewings is mostly different. The termen is slightly convex and the apex more roundish in *P. albida* whereas in *P. lorquiniella* stat. rev. the termen is mostly straight and the apex slightly pointed. However, the external morphologic characteristics are quite variable. The male genitalia also show differences. In general appearance it is more trapezoid in *P. lorquiniella* stat. rev. and more ovoid in *P. albida*. The vinculum is distinctly indented laterally in *P. lorquiniella* stat. rev. and not or only weakly indented in *P. albida*. Altogether the male genitalia of all *Ptilocephala* species are quite variable in shape and size. Much more striking is the DNA barcode. Both taxa differ more than 4 %, on the other hand the intraspecific divergence between remotely populations of *P. lorquiniella* is less than 1 %. We consider that *P. lorquiniella* stat. rev. is a separate allopatric species and not conspecific with *P. albida*.

Distribution: *P. lorquiniella* stat. rev. is probably distributed throughout the Iberian Peninsula (Portugal, Spain) north-eastwards to the Pyrenees. The distribution in the higher altitudes of the Pyrenees is still unclear. It also occurs in north-western Spain in the region of Galicia and it is striking that these populations are genetically more related to the populations of Andalusia than these to their next neighboring populations of Portugal. East of the Pyrenees it is most likely replaced by *P. albida*.

Molecular diagnostics

PATTERNS OF DNA BARCODE DIVERGENCES OF *PTILOCEPHALA*-SPECIES

Within the project POESE in BOLD, altogether 19 of the 23 currently described Palaearctic species of *Ptilocephala* were compared for their DNA barcodes. Altogether within the framework of BOLD, 76 samples have sequences. They are summarized in 24 Barcode Identification Numbers (BINs) (RATNASINGHAM & HEBERT, 2013) derivated from 16 countries. For this study we compared public data from the following BINs: BOLD:ABV9485 (GWOTD349-12, POESE284-19, PSYCH119-12), BOLD:ABW5202 (GWOTD350-12), BOLD:ABW7233 (PHLAH340-12, PHLAH341-12) and BOLD:ADY4384 (POESE015-15, POESE016-15). The extensive data in BOLD allowed e. g. a detailed evaluation of the phylogenetic relationship within the *P. albida*-species group and within the genus *Ptilocephala* itself. The DNA divergences among the this part of the *P. albida*-species group are shown in matrix 1 and fig. 9. The nearest neighbor of *P. rutjani* sp. n. is *P. albida* with a K2P minimum divergence of 7.6-7.8 %. The next closest species is *P. lorquiniella* stat. rev. with a K2P minimum divergence of 7.78-8.0 %.

Matrix 1 we have color-coded the numerical values according to certain criteria: Light blue: differences of all *P. lorquinella* stat. rev. against *P. albida* (inter-specific); purple: differences of all *P. rutjani* sp. n. against *P. albida* (inter-specific); red: differences of all *P. lorquinella* stat. rev. against each other (intra-specific); green: differences of all *P. rutjani* sp. n. against all *P. lorquinella* stat. rev. (inter-specific); brown: *P. rutjani* sp. n. against *P. rutjani* sp. n. (intra-specific).

Matrix 1.– DNA barcode K2P divergences of *Ptilocephala albida*-species group.

	Sequence	Species	Origin	1	2	3	4	5	6	7
1	GWOTD350-12	*P. albida*	Italy, Liguria							
2	GWOTD349-12	*P. lorquiniella*	Spain, Galicia	0,047						
3	POESE016-15	*P. lorquiniella*	Portugal, Manteigas	0,045	0,004					
4	POESE015-15	*P. lorquiniella*	Portugal, Manteigas	0,045	0,004	0,000				
5	POESE284-19	*P. lorquiniella*	Spain, Andalucía	0,041	0,013	0,009	0,009			
6	PSYCH119-12	*P. lorquiniella*	Spain, Andalucía	0,043	0,016	0,011	0,011	0,002		
7	PHLAH341-12	*P. rutjani*	Greece, Macedonia	0,078	0,080	0,076	0,076	0,076	0,078	
8	PHLAH340-12	*P. rutjani*	Ukraine, Kherson	0,080	0,083	0,078	0,078	0,078	0,080	0,002

Both the inter-specific and intra-specific data show the clear barcode divergences of the *P. albida* species group (fig. 13). For the distance summary graphs (figs 11-14) we used exclusively samples from the project POESE which are included in the following BINs:
BOLD:ADY8384, BOLD:ACY1399, BOLD:AED2768, BOLD:ABW4876, BOLD:ACX9498, BOLD:ACM6573, BOLD:ACY1802, BOLD:ACX9533, BOLD:ACY1693, BOLD:ACX9660, BOLD:ABV3659, BOLD:ABV4674, BOLD:ADD7072, BOLD:ADY0378, BOLD:ADY0330, BOLD:ADY4491, BOLD:ADV9485, BOLD:ACS1987, BOLD:AED2768 and BOLD:ACM6573.

The divergence for all sequences of *Ptilocephala* derivate from the project POESE, compared on the species level show a high divergence of 9 or 10 % at the intra-specific level of several sequenced species which is probably clear evidence for the existence of cryptic species within the genus (fig. 1) Matrix 2. Within the genus *Ptilocephala*, an inter-specific divergence of up to 17 or 18 % was detected (fig. 12). Comparing this result with other species clusters of several genera of the subfamily Oiketicinae, these clear differentiation is not surprisingly. For example, within the genus *Oiketicoides*, divergences of up to 17% are found even between morphologically barely distinguishable taxa such as *Oiketicoides lutea* and *O. tedaldii*. In fact the DNA barcode divergences of several species groups within the Psychidae are much better studied due a larger numbers of samples are available.

Matrix 2.– The average value und standard deviation for the value groups mentioned.

Species	Average value	Standard deviation	
P. lorquiniella vs. P. rutjani	0,078	0,002	
P. albida vs. P. rutjani	0,079	0,001	
P. albida vs. P. lorquiniella	0,044	0,002	
P. lorquiniella vs. P. lorquiniella	0,008	0,005	intra-specific
P. rutjani vs. P. rutjani	0,002		intra-specific

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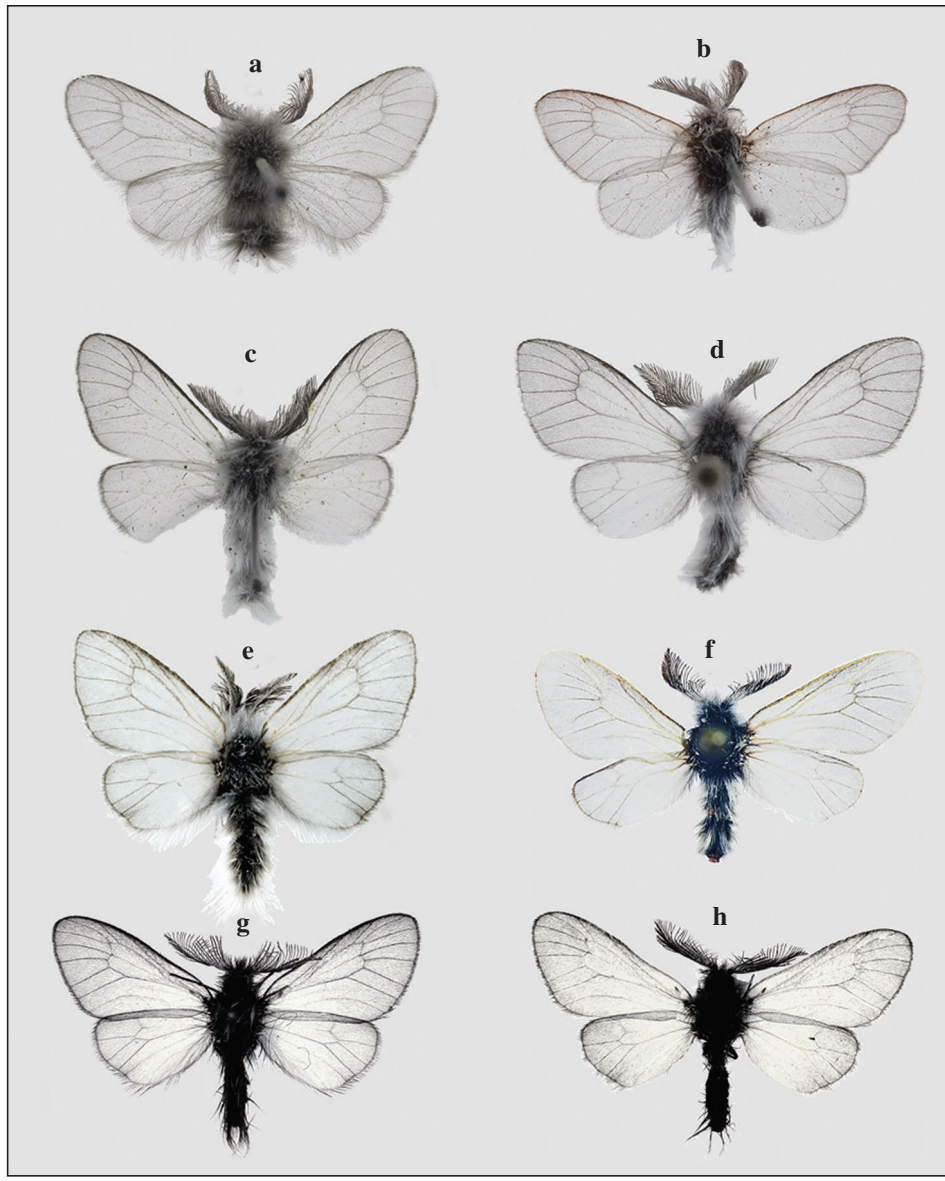


Fig. 3.– Male specimen of the *Ptilocephala albida*-species group: **a.** *P. albida*, "Frankreich", leg. C. Gatnar (MWM); **b.** *P. albida*, "Südfrankreich" ex. Coll. Daniel (MWM); **c.** *P. lorquiniella* stat. rev. Andalusia, Chiclana, 11-V-1914, groß V. Bartha (MWM); **d.** *P. lorquiniella*, Spain, Málaga, e. l. 1-V-1972, leg. Eitschberger (MWM); **e.** *P. lorquiniella* stat. rev. Portugal, Manteigas, Serra da Estrela, e. l. 10-V-2010 (CWA); **f.** *P. rutjani* Arnscheid & Weidlich, sp. n. paratype, Greece, Makedonia, Oros Vermio, near Kato Vermio W, 1050 m, e. p. 04-V-2011 (CMW); **g.** *P. rutjani* sp. n. holotype, Ukraine, Kherson prov., Tschernomorsky, 12-17-IV-2001, leg. E. Rutjan (SMNK); **h.** ditto, paratype, same data (CWA).

Fig. 4.– Male and female larval cases of the *Ptilocephala albida*-species group: **a.** ♂ larval case, *P. albida*, "Südfrankreich" ex. Coll. Daniel; **b.** ♀ larval case, *P. albida*, Spain, Huesca, Benasque, VI-2012 (CWA); **c.** ♂ larval case, *P. lorquiniella* stat. rev. Portugal, Manteigas, Serra da Estrela, V-2010 (CWA); **d.** ♂ larval case, *P. lorquiniella* stat. rev. Spain, Málaga, 8-V-1972, leg. Eitschberger (MWM); **e.** *P. rutjani* Arnscheid & Weidlich, sp. n. ♂ larval case, Greece, Makedonia, Oros Vermio, near Kato Vermio W, 1.050 m, e. p. 04-V-2011 (CMW); **f.** *P. rutjani* sp. n. ♀ larval case, North-Macedonia, near Ochrid, 1 km N, 700 m, 18-IV-2011, leg. M. Weidlich (CMW); **g.** *P. rutjani* sp. n. ♀ larval case, Bulgaria, near Stara Zagora W, Stare selo 2 km S, 400 m, 23-IV-2011 (CMW); **h.** *P. rutjani* sp. n. ♂ larval case, Ukraine, Kherson prov., Tschernomorsky, 12-17-IV-2001, leg. E. Rutjan (CER); **i.** *P. rutjani* sp. n. ♀ larval case, ditto (CER).

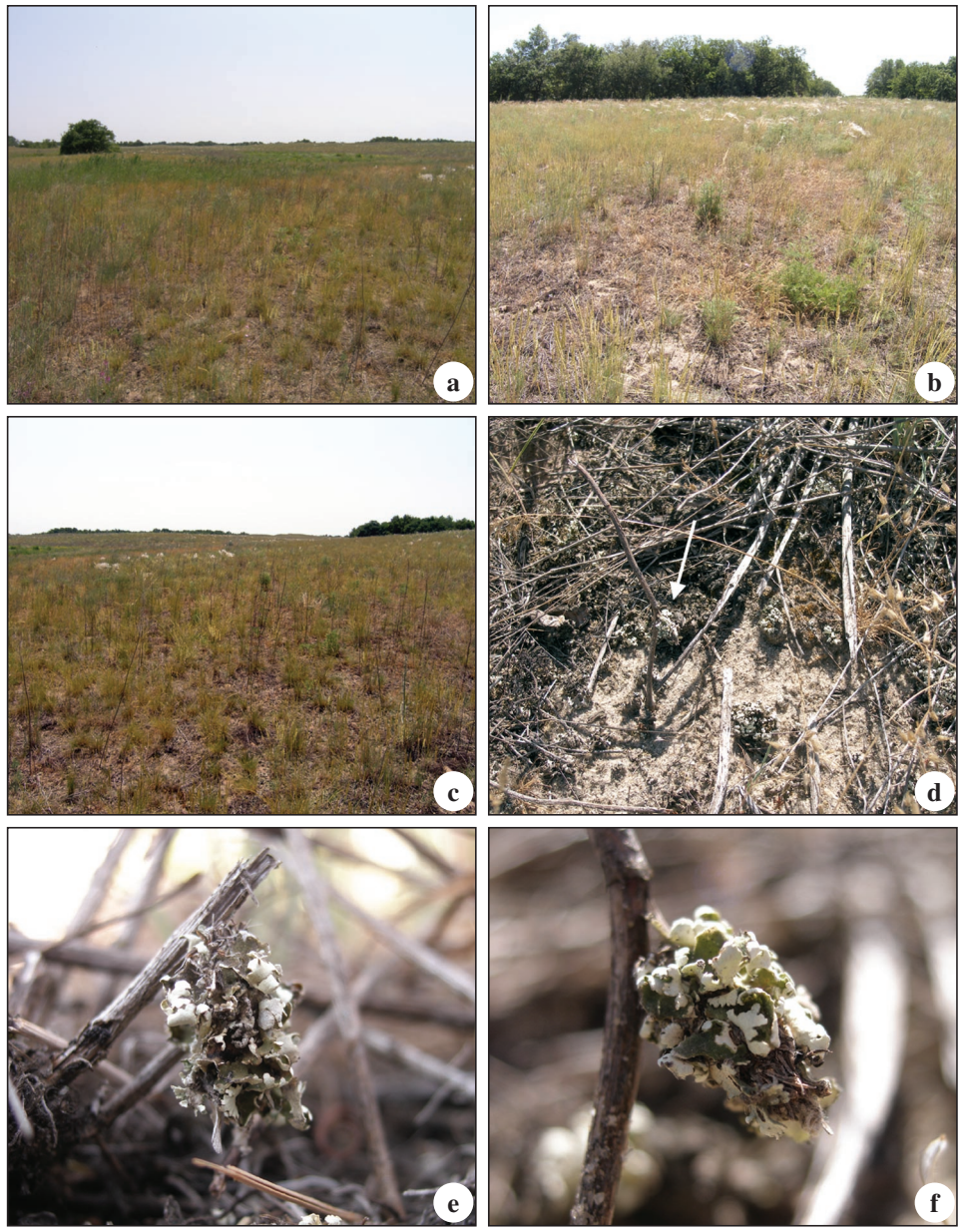


Fig. 5.– Habitat of *P. rutjani* Arnscheid & Weidlich, sp. n. in Ukraine: **a-c.** The habitat at the type locality in Ukraine in the Black Sea Biosphere Reserve (Ukrainian) which is located at the littoral zone of the northern Black Sea coast in the region of Kherson. The reserve is part of the National Academy of Sciences of Ukraine. **d., e., f.** Female larval cases, spun at twigs for pupation. Photos: E.V. Rutjan, 5-VI-2011.

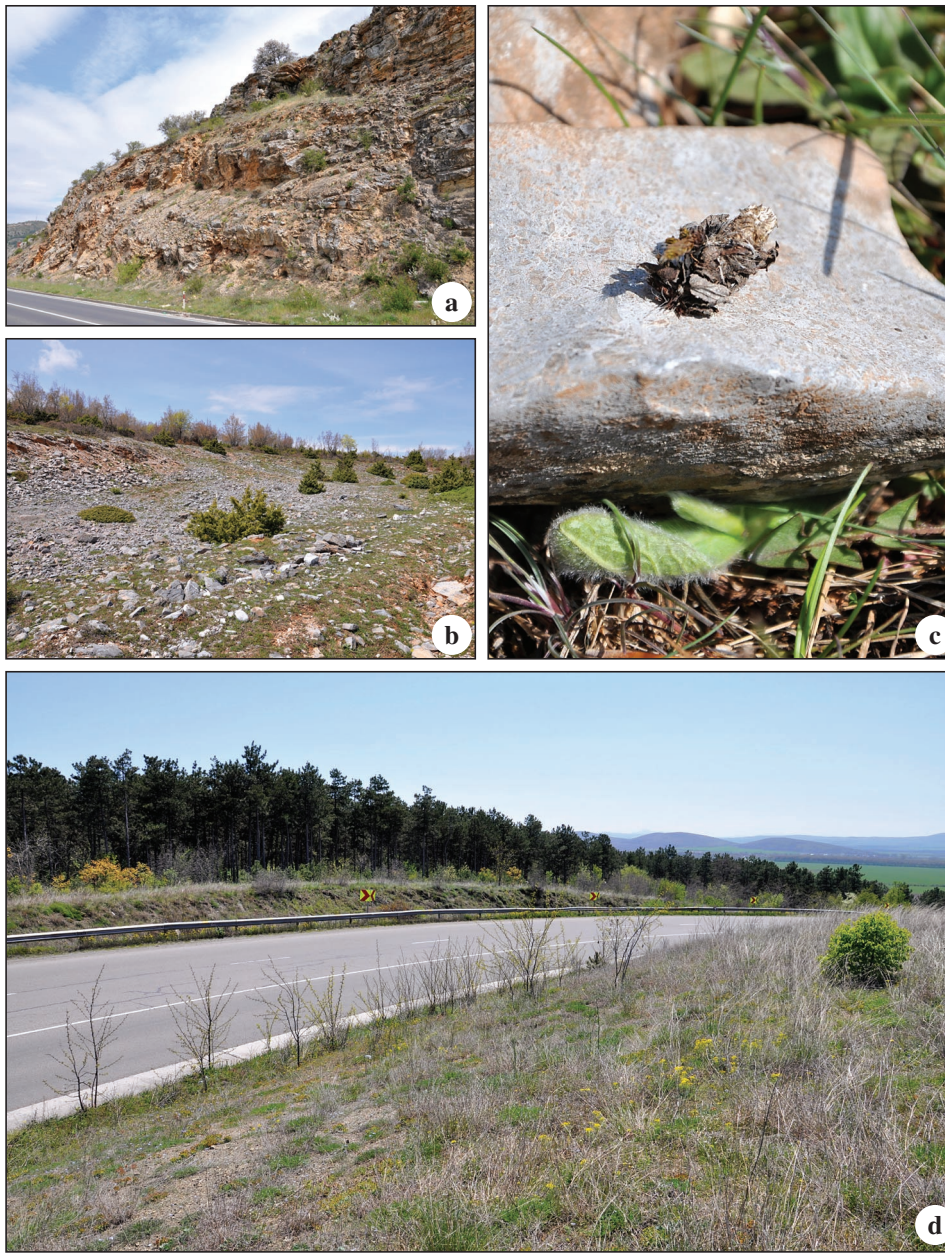


Fig. 6.– Habitat of *P. rutjani* Arnscheid & Weidlich, sp. n. at the Balkans: **a.** North-Macedonia, near Ochrid, 1 km N, 700 m, 18-IV-2011. **b.** Greece, Makedonia, Oros Vermio, near Kato Vermio W, 1050 m, 20-IV-2011. **c.** Caterpillar, same data. **d.** Bulgaria, Stara Zagora W, near Stare Selo 2 km S, 400 m, 23-IV-2011. Photos: M. Weidlich.

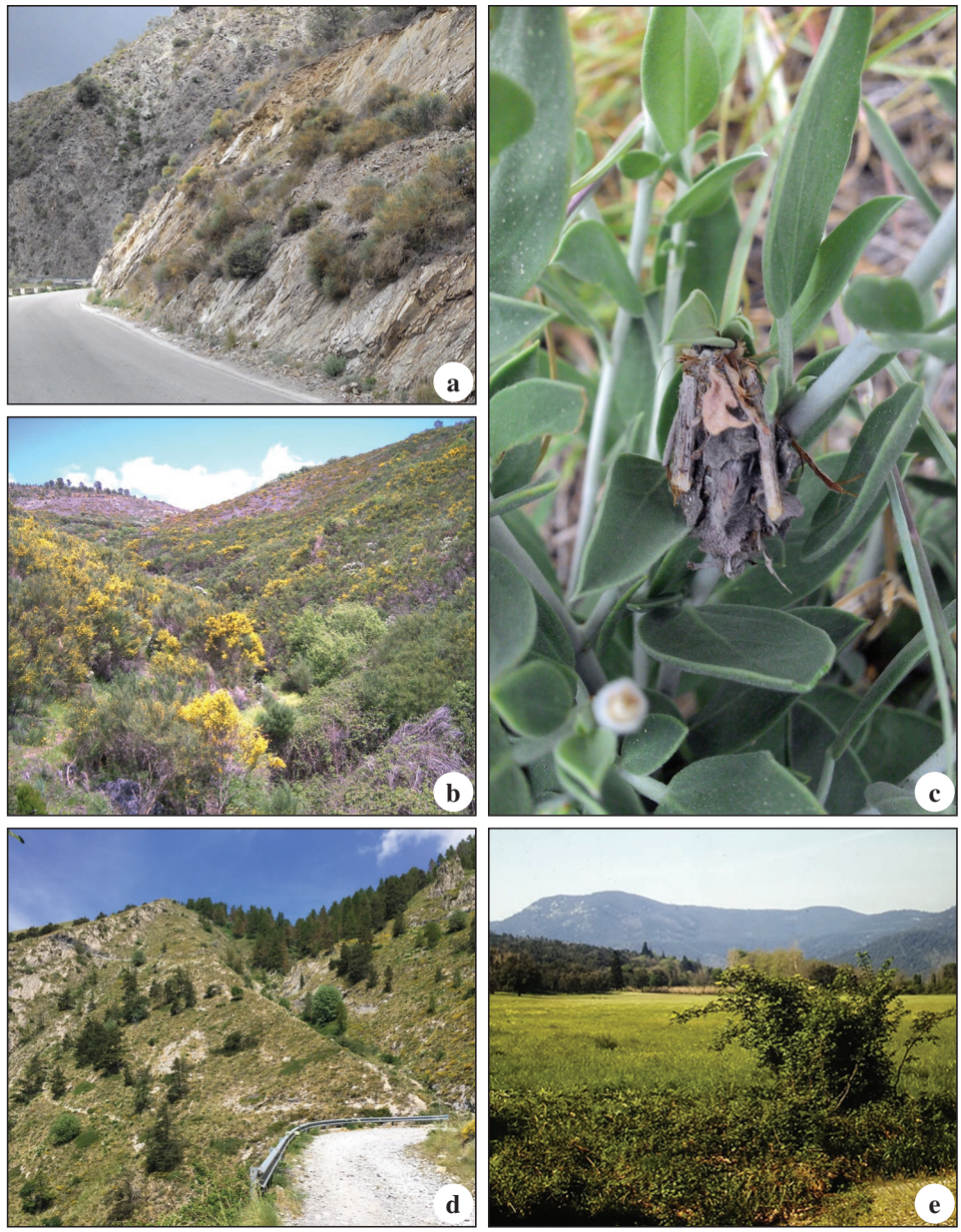


Fig. 7.– Habitat of *P. albida* and *P. lorquiniella* stat. rev. in South Europe. **a.** *P. lorquiniella,* stat. rev. Spain, Andalusia, near Algarrobo, 15-III-2015; **b.** *P. lorquiniella,* stat. rev. Portugal, Sameiro 1 km W, near Manteigas, 5-V-2010, **c.** *P. lorquiniella* stat. rev., larva with larval case, Spain, Andalusia, Algarrobo, 15-III-2015. Habitat of *P. albida*: **d.** *P. albida*, Italy, Liguria, Colle Melosa, 1650 m, 23-VI-2017; **e.** *P. albida*, South France, Massif des Maures, near Colobrières, 10-IV-1980. Photos: W. R. Arnscheid.

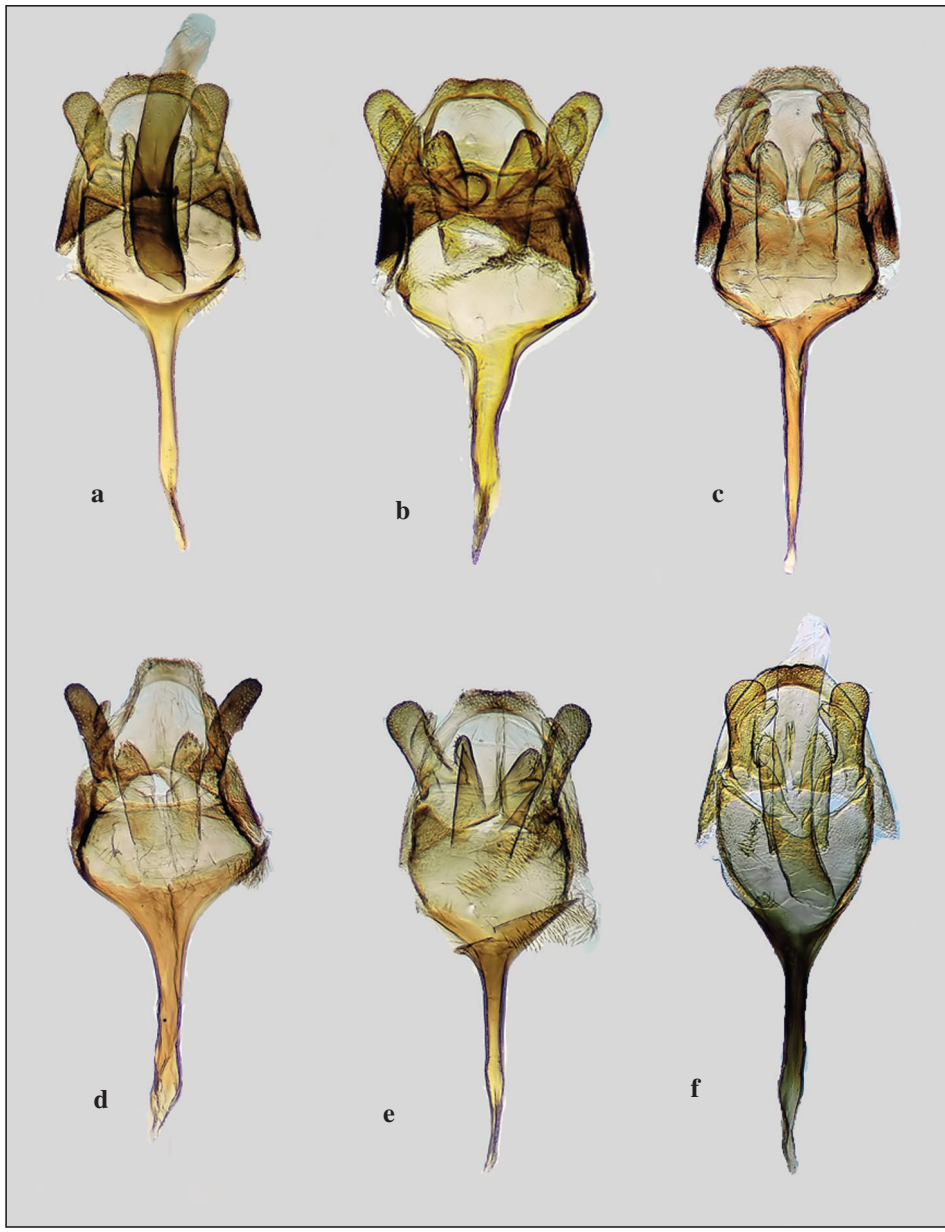
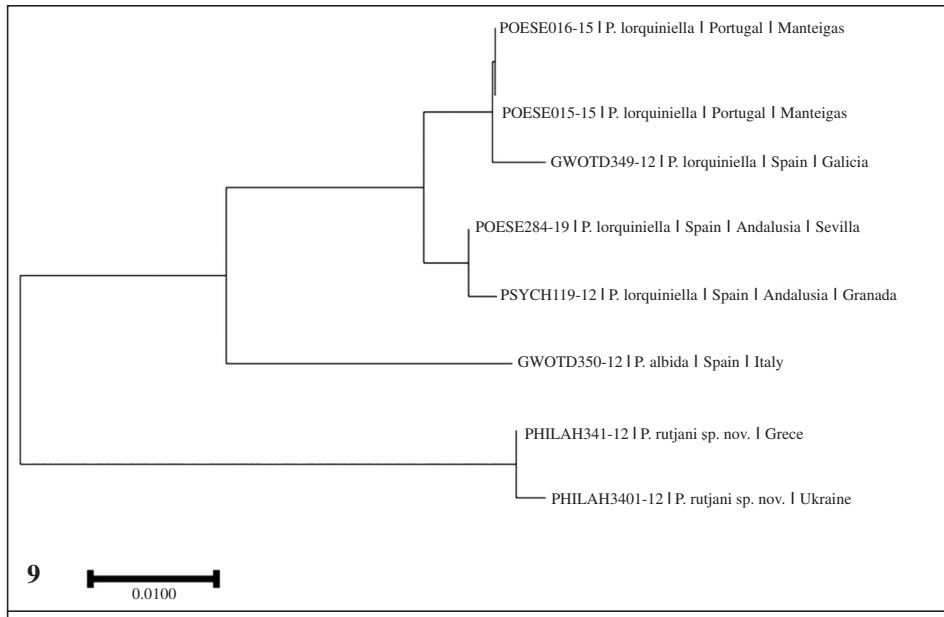
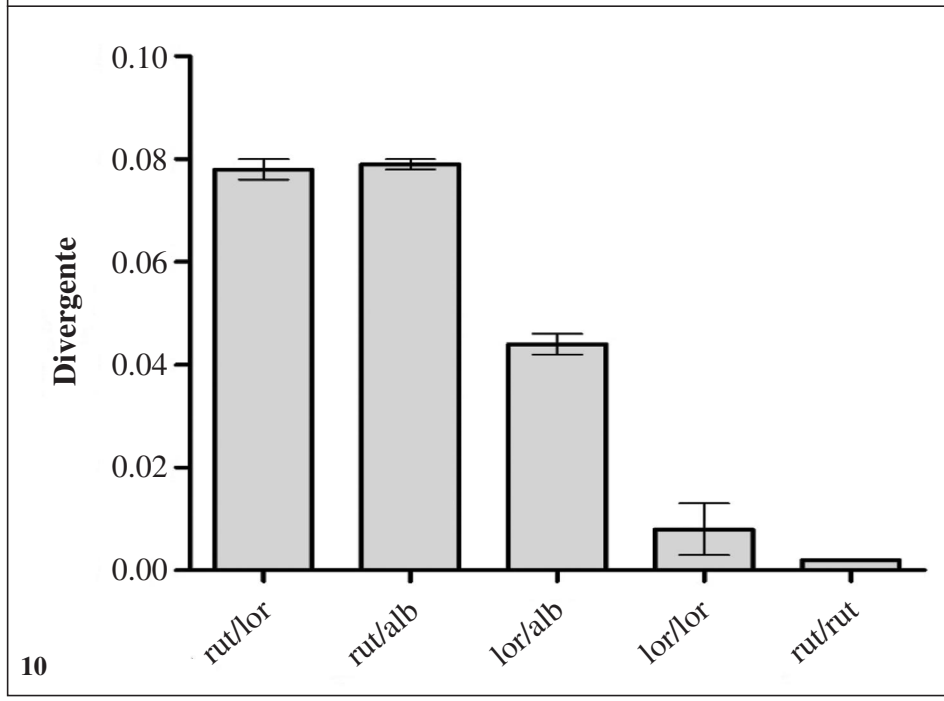
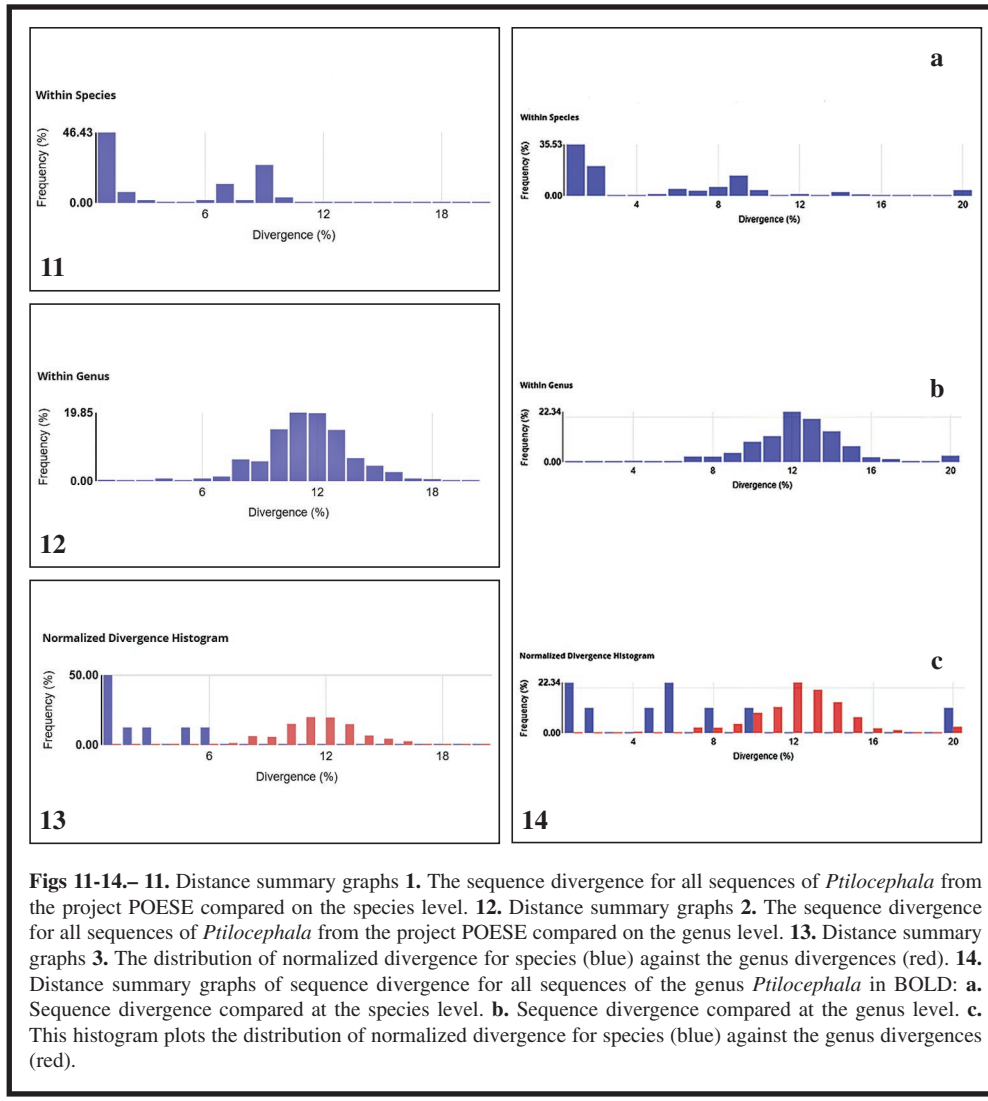


Fig. 8.– Male genitalia of species of the *P. albida*-species group: **a.** *P. albida,* genital slide 4009, France, Dpt. Lot. **b.** *P. albida*, genital slide 4105 (phallus removed), South France, Cannes. **c.** *P. lorquiniella* stat. rev., genital slide 4107 (phallus removed), Spain, Andalusia, Granada. **d.** *P. lorquiniella* stat. rev.*,* genital slide 4106 (phallus removed), Portugal, Singeverga. **e.** *P. rutjani* Arnscheid & Weidlich, sp. n.*,* genital slide 4109 (phallus removed), Ukraine, Kherson prov. **f.** *P. rutjani* sp. n., genital slide 4007, Ukraine, Kherson prov.





Figs 9-10.– 9. Neighbor-joining tree of a part of the European species of the *P. albida*-species group. **10.** The average value und standard deviation for the value groups as mentioned in matrix 2.



Figs 11-14.– 11. Distance summary graphs **1.** The sequence divergence for all sequences of *Ptilocephala* from the project POESE compared on the species level. **12.** Distance summary graphs **2.** The sequence divergence for all sequences of *Ptilocephala* from the project POESE compared on the genus level. **13.** Distance summary graphs **3.** The distribution of normalized divergence for species (blue) against the genus divergences (red). **14.** Distance summary graphs of sequence divergence for all sequences of the genus *Ptilocephala* in BOLD: **a.** Sequence divergence compared at the species level. **b.** Sequence divergence compared at the genus level. **c.** This histogram plots the distribution of normalized divergence for species (blue) against the genus divergences (red).