

On the status of *Bembecia zebo* Špatenka & Gorbunov, 1992; *Bembecia pamira* Špatenka, 1992; *Bembecia kreuzbergi* Špatenka & Bartsch, 2010 and *Bembecia martensi* Gorbunov, 1994 (Lepidoptera: Sesiidae)

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Abstract. Based on external morphology and DNA analysis, *Bembecia pamira* Špatenka, 1992 appears to be conspecific with *Bembecia zebo* and is considered here as a subjective junior synonym of the latter. The name priority is discussed since both taxa were described in the same year. Additionally, the DNA analysis demonstrates a similar profile to *Bembecia kreuzbergi* Špatenka & Bartsch, 2010 but this species shows clear differences in external morphology, especially in the female, and therefore its status as bona species appears justified. Also *Bembecia martensi* Gorbunov, 1994 is reviewed briefly. Illustrations of most of the discussed species are shown together with the genitalia preparations, when available.

Samenvatting. Gebaseerd op de externe morfologie en DNA analyses blijkt *Bembecia pamira* Špatenka, 1992 conspecifiek te zijn met *Bembecia zebo* Špatenka & Gorbunov, 1992 en wordt hier als een subjectief junior synoniem van *Bembecia zebo* beschouwd. De prioriteit in naamgeving wordt ook besproken vermits beide soorten hetzelfde jaar beschreven werden. De DNA analyse toonde ook een zeer gelijkend profiel voor *Bembecia kreuzbergi* Špatenka & Bartsch, 2010 maar deze soort vertoont duidelijke verschillen in externe morfologie, vooral bij het wijfje, en daarom wordt de status van deze soort als bona species behouden. Ook *Bembecia martensi* Gorbunov, 1994 wordt kort behandeld. Afbeeldingen van de meeste van de besproken soorten worden getoond, samen met de beschikbare genitaalstructuren.

Résumé. Selon une étude de la morphologie externe et des analyses ADN, *Bembecia pamira* Špatenka, 1992 semble être conspécifique avec *Bembecia zebo* Špatenka & Gorbunov, 1992 et est donc considérée ici comme synonyme subjectif plus récent de *Bembecia zebo*. La priorité des noms est discutée aussi parce que les deux espèces ont été décrites au cours de la même année. L'analyse ADN montrait aussi un profil semblable à celui de *Bembecia kreuzbergi* Špatenka & Bartsch, 2010 mais cette espèce montre des différences claires dans la morphologie externe, surtout chez la femelle, et le statut spécifique de ce taxon est donc retenu. *Bembecia martensi* Gorbunov, 1994 est discuté brièvement. La plupart des espèces discutées, ainsi que les genitalia disponibles, sont figurés.

Key words: Tajikistan – Pamir – Sesiidae – *Bembecia* – *zebo* – *pamira* – *kreuzbergi* – *martensi*.

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Abbreviations

MWM – Museum Witt München
CDB – collection of D. Bartsch
CTG – collection of T. & W. Garrevoet
COG – collection of O. Gorbunov
CAL – collection of A. Lingenhöle

Introduction

The genus *Bembecia* Hübner, 1819 has distribution hot spots in the Mediterranean area and Central Asia. In the past, many species were described from the latter area but recently several more new taxa were discovered (Bartsch & Špatenka 2010, Špatenka & Bartsch 2010, Stalling et al 2010, Garrevoet & Garrevoet 2011, Garrevoet & Lingenhöle 2011, Lingenhöle & Bartsch 2011, Stalling et al 2011). All species of the genus develop over one or two years in the roots or lower parts of the stems of various species of Fabaceae and have a reduced, non-functional proboscis.

After an expedition into the Pamir region (Gorno Badachstan) in Tajikistan (Central Asia), studying the collected material, especially within the genus *Bembecia*, the authors attention was drawn to *Bembecia pamira* Špatenka, 1992. This species showed a striking similarity in external morphology to *Bembecia zebo* Špatenka & Gorbunov, 1992. Therefore, a more thoroughgoing

investigation of external morphology, genitalia structure and DNA profile appeared necessary.

The DNA results, performed in Guelph, Canada as part of the BOLD project, together with morphological resemblance, caused the authors to include also *Bembecia kreuzbergi* Špatenka & Bartsch, 2010 and *Bembecia martensi* Gorbunov, 1994 in this research. Thus the aim of this paper is to clarify the taxonomic status of *B. pamira* and *B. zebo* and to provide short notes on that of *B. kreuzbergi* and *B. martensi*.

Material

Types:

- B. pamira* (Figs 13–14), 'ASIA centralis, Pamir 3550 m, Sarez ozero, Ljanger Fluss, 26.7.1985'; 'Holotypus ♀, K. Špatenka des. 1989' (MWM).
- B. zebo* (Figs 1–2), 'USSR-Tadjikistan, Turkestan-chain, Kumbel 3000-3200m, 39,34N 68,33E, 15.-21.7.1987, e.l., K. Špatenka lgt.', 'Holotypus ♂, K. Špatenka des. 1989' (MWM).
- B. zebo* (Figs 9–10), 'USSR-Tadjikistan, Turkestan-chain, Kumbel 3000-3200m, 39,34N 68,33E, 15.-21.7.1987, e.l., K. Špatenka lgt.', 'Paratypus ♀, K. Špatenka des. 1989' (MWM).
- B. kreuzbergi* (Figs 5–6), 'Holotypus ♂, SO Usbekistan, Babatag Gebirge, Zar Kamar, 38°06N, 68°10O, 1250m, 29.V.2004, K. Špatenka leg.' (MWM).
- B. kreuzbergi* (Figs 15–16), 'Paratypus ♀, SO Usbekistan, Babatag Gebirge, Zar Kamar, 38°06N, 68°10O, 1250m, 29.V.2004, K. Špatenka leg.' (MWM).

B. martensi (Figs 7–8), 'Holotypus ♀, Kirgizstan, Alashtau Mts., Ermendy, N 41°15', E 72°40', 1550 m, 25-26.V.1993, ex l., leg. O. Gorbunov.' (COG).

B. martensi (Figs 17–18), 'Paratypus ♂, Kirgizstan, Alashtau Mts., Ermendy, N 41°15', E 72°40', 1550 m, 25-26.V.1993, ex l., leg. O. Gorbunov.' (COG).

Additional material of *B. zebo*: all specimens from Tajikistan (CTG)

Region Sughd

Iskander Kul, 2350 m, N 39° 5' 18.8" E 68° 23' 55.0", 13.vii.2010, 11♂-9♀.

Iskander Kul, 2350 m, N 39° 5' 16.3" E 68° 24' 5.9", 01.viii.2010, 6♂-5♀.

Iskander Kul, 2400 m, N 39° 2' 49.5" E 68° 18' 24.1", 05.viii.2010, 1♂.

Anzob Pass (North side), 2200 m, N 39° 9' 6.6" E 68° 50' 46.5", 17.vii.2010, 2♂.

Anzob Pass (North side), 2200 m, N 39° 9' 5.7" E 68° 50' 46.8", 07.viii.2010, 1♂.

Shahrston Pass, 2850 m, N 39° 34' 20.9" E 68° 33' 15.2", 20.vii.2010, 1♂-1♂ dead pupa.

Childara, 1875 m, N 38° 51' 44.8" E 70° 20' 29.6", 19.vii.2011, 2♂.

Region of Republican Subordination

Rufigar, 2100 m, N 39° 07' 29.7" E 69° 25' 13.9", 21.vii.2009, 1♂.

Magov, 1500 m, N 38° 41' 18.1" E 69° 05' 55.6", 08.vii.2010, 1♂.

Sangvor, 2300 m, 50 km S of Jirgatal (Dzergatal), N 38° 44' 53.4" E 71° 14' 07.0", 15.vii.2011, Ex larva, 4♂-2♀.

Moths emerged between 27.vii.2011 and 06.viii.2011. (Figs 3–4 ♂ and 13–14 ♀).

Garm, 1950 m, N 39° 00' 30.8" E 70° 28' 07.9", 29.vii.2013, 1♂.

Anzob Pass (South side), 2450 m, N 39° 03' 59.7" E 68° 51' 00.0", 25.vii.2009, 5♂.

Anzob Pass (South side), 2400 m, N 39° 03' 52.3" E 68° 50' 56.4", 29.vii.2010, 5♂.

Anzob Pass (South side), 2575 m, N 39° 04' 13.6" E 68° 51' 01.1", 23.vii.2011, 1♂.

Anzob Pass (South side), 2850 m, N 39° 04' 30.0" E 68° 51' 02.4", 26.vii.2011, 4♂.

Gorno Badachstan (GBAO)

Saghirdasht Pass (North side), 2225 m, N 38° 41' 57.9" E 70° 43' 30.2", 11.vii.2013, 3♂.

Saghirdasht Pass (South side), 1850 m, N 38° 32' 51.7" E 70° 48' 01.6", 12.vii.2013, 1♂; 27.vii.2013, 1♂.

Methods

Bembecia pamira was described from a single, rather poorly preserved female and no additional specimens have been collected since. From this holotype, kept in the MWM, the external morphology was carefully compared using detailed photographs of both upper- and underside with illustrations of both male (holotype) and female (paratype) of *B. zebo* (also in MWM). Additional *B. zebo* specimens, of both sexes, captured or reared during recent expeditions in Tajikistan and kept in several collections (CTG, CDB and CAL) were also included in this comparison.

B. zebo was described from north-west Tajikistan, near the border with Uzbekistan, from bred material of both sexes from the host plant *Onobrychis echidna* Lipsky

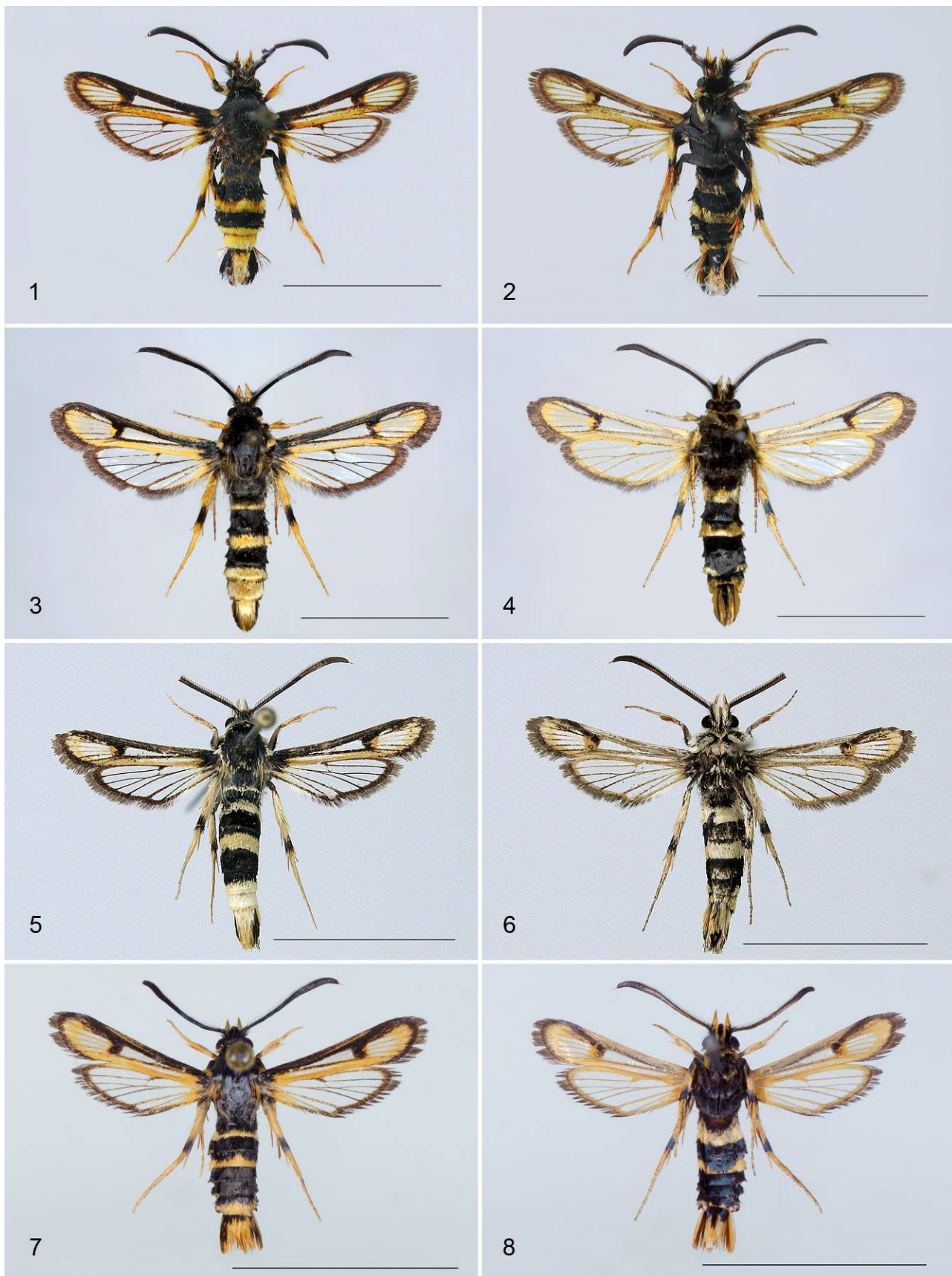
(Špatenka *et al.*, 1999). Since then, *B. zebo* has been captured, mainly with the use of pheromones, at different localities throughout Tajikistan. The best pheromone response was observed before noon using an old pheromone of unknown composition, for *Synanthedon myopaeformis* (Borkhausen, 1789) produced by the company BASF, Germany. Some individuals, including several females, were netted without the use of pheromones. Furthermore, at different localities, several specimens of both sexes were bred from *Hedysarum flavescens*, Regel & Schmalhausen, another species of Fabaceae. Infested plants had to be dug out from the hard and stony soil very carefully. The roots with larvae and pupae were kept in small containers and, once home, placed in terrariums containing a fine hydroculture granulate.

Preparations of genitalia of several specimens, including a dead pupa found in *Onobrychis echidna* at the type locality, were made using the standard techniques: maceration of the abdomen in 10% KOH, removal of the scales and cleaning in 70% ethanol. Genitalia of males were not stained, those of females were stained with Chlorazol Black, then embedded in Euparal on a cavity slide, males with opened valvae. DNA was extracted from a midleg of set specimens. DNA analysis ("Barcode" = 658 base pair sequences of COX1 mitochondrial DNA) of several Tajik *Bembecia* including the types of *B. zebo* and the type of *B. pamira* was carried out in Guelph (Canada). The PCR-primers used were LepF1 and LepR1; the distance model was Kimura 2 Parameter. For details see the "Barcode of Life Database" (BOLD) web pages (<http://www.barcodinglife.com/views/login.php>). The detailed data can be accessed with a login under the project "Global Sesiidae – Clearwing Moths of the World".

Results

When comparing the external morphology of the female holotype of *B. pamira* with a female paratype of *B. zebo*, supplemented with additional material of the latter species collected more recently, the striking similarity in external morphology of both taxa became obvious. Because the genitalia structure of the *B. pamira* type has not been examined, this auxiliary could not be included into the comparison but often the genitalia in this group of Central Asian *Bembecia* do not provide useful differential criteria anyway. Nevertheless, the genitalia of both sexes of *B. zebo* are depicted for completeness, but are adequately described in the original publication (Špatenka & Gorbunov, 1992).

The DNA analysis (BOLD, Guelph, Canada) (Fig. 28) confirmed the expectation that the taxa are conspecific and therefore *B. pamira* becomes a subjective junior synonym of *B. zebo*.



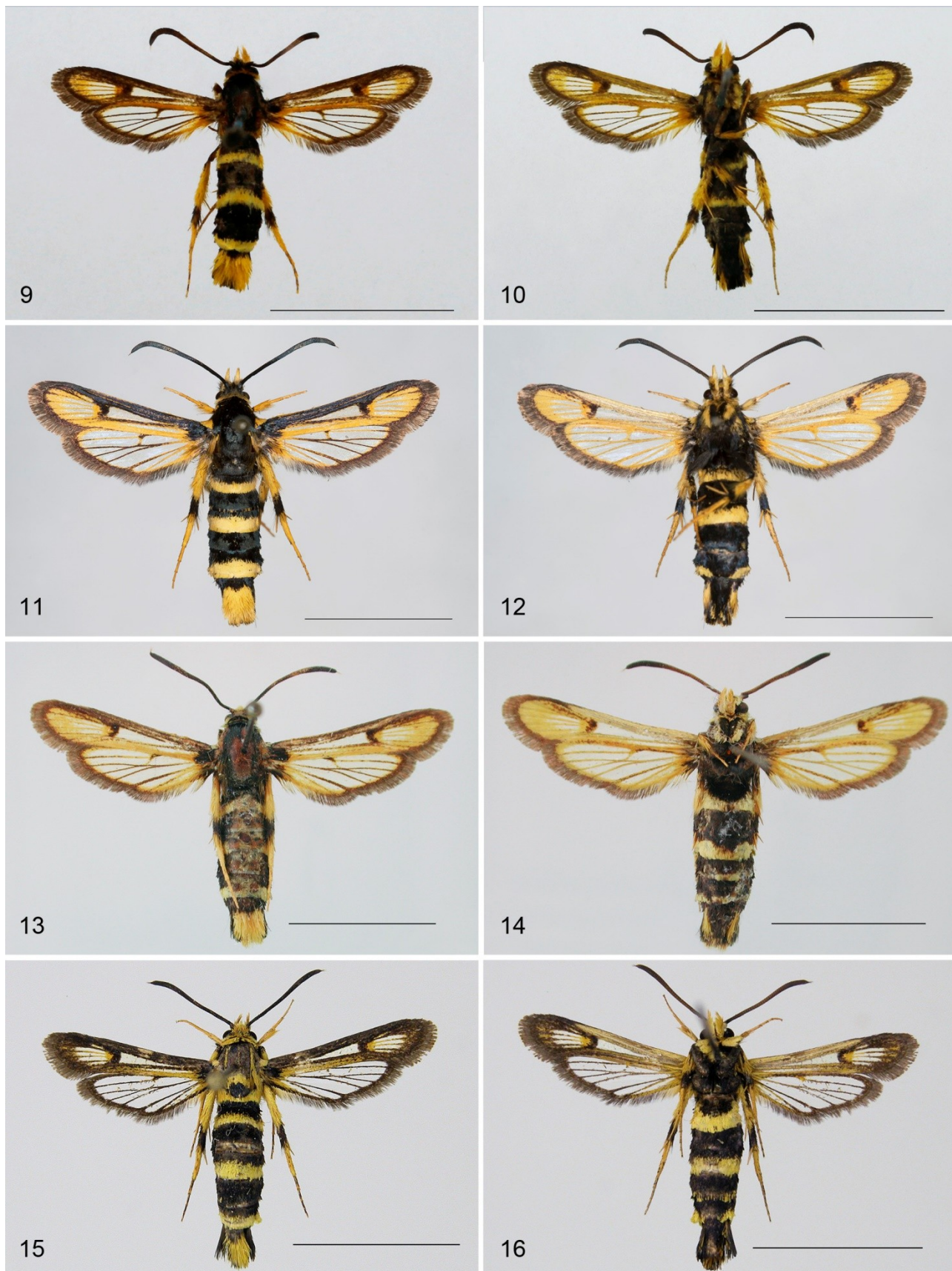
Figs 1–8. Males of *Bembecia* species, scale bars 10 mm.

1–2. *B. zebo* holotype, Tajikistan, Turkestan-chain, Kumbel Pass 3000-3200m, 15-21.VII.1987, e.l., leg. K. Špatenka (MWM).

3–4. *B. zebo*, Tajikistan, Region Republican Subordination, Sangvor, 50 km S of Dzergatol, N 38° 44' 53.4" E 71° 14' 07.0", 2300 m, 15.VII.2011, Ex larva, leg. T. Garrevoet (CTG).

5–6. *B. kreuzbergi* paratype, SE Uzbekistan, Babatag Range, Zar Kamar, N 38°06', E 68°10', 1250m, 29.V.2004, leg. K. Špatenka. (MWM).

7–8. *B. martensi* paratype, Kirgizstan, Alashtau Mts., Ermendy, N 41°15', E 72°40', 1550 m, 25-26.V.1993, ex. l., leg. O. Gorbunov. (COG).



Figs 9–16. Females of *Bembecia* species, scale bars 10 mm.

9–10. *B. zebo* paratype, Tajikistan, Turkestan-chain, Kumbel Pass 3000–3200m, 15–21.VII.1987, e.l., leg. K. Špatenka (MWM).

11–12. *B. zebo*, Tajikistan, Region Republican Subordination, Sangvor, 50 km S of Dzergatol, N 38° 44' 53.4" E 71° 14' 07.0", 2300 m, 15.VII.2011, ex larva, leg. T. Garrevoet (CTG).

13–14. *B. pamira* holotype, Tajikistan, Pamir, Sarez ozero, Ljangan River, 3550 m, 26.VII.1985, K. Špatenka des. 1989 (MWM).

15–16. *B. kreuzbergi* paratype, SE Uzbekistan, Babatag Range, Zar Kamar, N 38° 06', E 68° 10', 1250 m, 29.V.2004, leg. K. Špatenka. (MWM).



Figs 17–18. Females of *Bembecia* species, scale bars 10 mm.

17–18. *B. martensi* holotype, Kirgizstan, Alashtau Mts., Ermendy, N 41°15', E 72°40', 1550 m, 25–26.V.1993, ex l., leg. O. Gorbunov. (COG).

As both taxa were described in the same year, albeit in different journals, the problem of priority needed to be solved. *B. pamira* was described in the French journal *Alexanor* 17(7), 1992, giving the publication date and *B. zebo* was described in the Austrian journal *Entomofauna* 13(23), 1992 giving "Ansfelden 20. September 1992" as publication date.

Here the ICZN code is important: Art. 8.1.2 is not relevant here since both journals are published in a correct way but Art. 21.2. is important: "The date of publication specified in a work is to be adopted as correct in the absence of evidence to the contrary".

Although the latter journal arrived much later at several libraries – because the issues are sent to exchange libraries only once or twice a year to save postal charges – the publisher mentions he did send separata to some addressees even before the mentioned publication date. According to Art. 21.8.1, these are also valid as publication date, because the paper was published before the year 2000. So, at least 20 September 1992 can be considered as the valid publication date for *B. zebo*.

Because the title page of the specified *Alexanor* issue mentions "juillet-septembre 1992", Art. 21.3.1 stipulates the last day of the period mentioned has to be adopted as the publication date which in this case is 30 September 1992. Furthermore, regular subscribers to this journal indeed received the issue in the first half of October. Therefore, priority has to be given to the taxon name *B. zebo*.

In the DNA analysis, *B. kreuzbergi* was also included as this species showed a very similar pattern to that of *B. zebo*. The external morphological features, however, show obvious differences, especially in the female, which are clearly described in the original publication (Špatenka & Bartsch 2010). The colour of the metathorax is one of the most obvious ones: it is yellow in *B. kreuzbergi* and black in *B. zebo*. Therefore, notwithstanding the DNA results, the status of *B. kreuzbergi* is retained as bona species. For the integrity and comparison, illustrations of both sexes of *B. kreuzbergi* are given here (Figs. 5–6 and 15–16).

Based on both genital structure (not figured) and position in the DNA tree, *B. martensi* Gorbunov, 1994

also belongs to the same species group and has a similar general external appearance. However, it can be distinguished from *B. zebo* by the ground colour, which is yellow-orange instead of lemon yellow. Additionally, the markings on the dorsal side of the thorax are different, with a row of orange hair like scales on each side of the mesothorax and metathorax, which are absent in the other species. Furthermore, *B. martensi* has a small band on abdominal segment 1 which broadens significantly laterally, which is not the case in *B. kreuzbergi* and which is altogether absent in *B. zebo*.

Variability

The specimens of *B. zebo* which have been studied vary little in colour intensity. In older specimens the bright yellow colouration on both body and wings becomes more bleached pale yellow. On the other hand, differences in size are considerable and not directly related to the locality. However, dependence on different hostplants may be responsible, as the specimens from the type locality are in average somewhat smaller than the ones from other localities where the species develops in *Hedysarum flavescens*.

Genitalia

B. zebo belongs to the widespread Central Asian species group that lacks the medial crista of the gnathos and has a subdistal gap among the setae on the crista sacculi of the valva. The genitalia of both sexes are adequately described in the original description. In the male, the most important characteristics are the well-developed scopula androconialis, the absence of the crista medialis and the two groups of larger setae on the dorsal side of the crista sacculi. In the female, the slim, weakly sclerotised and funnel-shaped ostium bursae, together with the slender antrum and the reduced ductus bursae are the main characteristics.

As expected, since *B. kreuzbergi* belongs to the same species group, this species shows no significant differences in the male genitalia from those in *B. zebo*. The female genitalia of *B. kreuzbergi* have not been studied.



Figs 19–21. Genitalia of *Bembecia* males. Scale bars 1 mm. **19.** *B. zebo* (prep. TG2011-003) from a dead but fully developed pupa from the type locality, Tajikistan, Shahrison Pass, 2850 m, N 39° 34' 20.9" E 68° 33' 15.2", 20.vii.2010 (CTG). **20.** *B. zebo* (prep. TG2011-004), Tajikistan, Iskander Kul, 2350 m, N 39° 05' 16.3" E 68° 24' 5.9", 01.viii.2010 (CTG). **21.** *B. kreuzbergi* paratype (Bartsch, GU-prep. 2009-34), SE Uzbekistan, Babatag Range, Zar Kamar, N 38° 06', E 68° 10', 1250m, 29.v.2004, leg. K. Špatenka. (MWM).

For completeness, illustrations of the genitalia of the males of *B. zebo* (Fig. 19–20) and *B. kreuzbergi* (Fig. 21) are included, as well as the genitalia of a female of *B. zebo* (Fig. 22).

Bionomics

At the type locality *B. zebo* occurs on dry and stony soil with cushion forming flora including *Onobrychis* and *Astragalus* species and other typical high altitude plants such as *Eremerus* (Špatenka & Gorbunov, 1992 and authors' observation). Here *B. zebo* lives in *Onobrychis echidna* (Špatenka et al., 1999). In localities where it lives in *Hedysarum flavescens* (Fig. 27), its occurrence is almost entirely restricted to valleys with large or even

smaller riverbeds, erosion gullies and roadsides with sufficient humidity to sustain the growth of the hostplant.



Fig 22. Genitalia of *B. zebo* female (prep. TG2013-005). Scale bar 1 mm. Tajikistan, Iskander Kul, 2350 m, N 39° 5' 18.8" E 68° 23' 55.0", 2350 m, 13.VII.2010, leg. T., W. & J. Garrevoet (CTG).

At the type locality, the authors found only one dried dead pupa (genitalia investigated) in *Onobrychis echidna* and attracted only one male with pheromones, but the bionomics are well described in the original publication (Špatenka & Gorbunov, 1992). At other localities, where the species lives in *Hedysarum flavescens*, the larva apparently also lives two years in the root of the hostplant. Also here, prior to pupation, the larva constructs a short exit tube from the root to the soil surface, which is almost invisible. The pupal chamber is inside the upper part of the root. In culture, emergence of the imagines always took place in the morning which is most likely to be the case in nature also. The species appears emerge from mid-July to the second half of August, depending on altitude. Males are active from late morning to early afternoon. Females were captured in early afternoon flying near the host plant.

Distribution

B. zebo is known from the type locality in the Turkestan mountain range in Tajikistan and from several other locations in the western part of Tajikistan (Hissar-Alai) but also from localities in central Tajikistan including the Peter the First Range in the north-western Pamir and, as *B. pamira*, in the Pamir itself. Although it appears to be widespread, all currently known localities are in Tajikistan. However, it is considered likely to be present in neighbouring countries too.

Conclusion

Based on the results of the investigation of external morphology and DNA analysis of the *Bembecia* species discussed, *Bembecia pamira* becomes a subjective junior synonym of *Bembecia zebo*. Although *Bembecia kreuzbergi* and *Bembecia martensi* show strong resemblances in their DNA, and belong genitally in the same (sub)group, there are clear enough differences in external morphology to justify the validity of both taxa. In this respect, the morphology of the genitalia of these species, together with that of several other Central Asian species which

lack the medial crista of the gnathos and have a subdistal gap in the setae on the crista sacculi of the valva, does not fit nicely into the *Bembecia ichneumoniformis* ([Denis & Schiffermüller], 1775) subgroup (Pühringer & Kallies 2004). Raising a separate subgroup within the *ichneumoniformis* group should be considered.

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Figs. 23-27. In vivo pictures of *B. zebo*, biotope and hostplant. **23.** *B. zebo* ♂, ex larva, Sangvor, 50 km S of Jirgatol (Dzergatol), N 38° 44' 53.4" E 71° 14' 07.0", 2300 m, 15.VII.2011; moth emerged on 03.VIII.2011. **24.** *B. zebo* ♀, same data; moth emerged on 05.VIII.2011. **25.** *B. zebo* exuvium protruding from the stem base of *Hedysarum flavescens*, same data. **26.** Biotope of *B. zebo* near Anzob Pass (south side), 4500 m. **27.** *Hedysarum flavescens*, a hostplant of *B. zebo*. The insert shows the characteristic seeds.

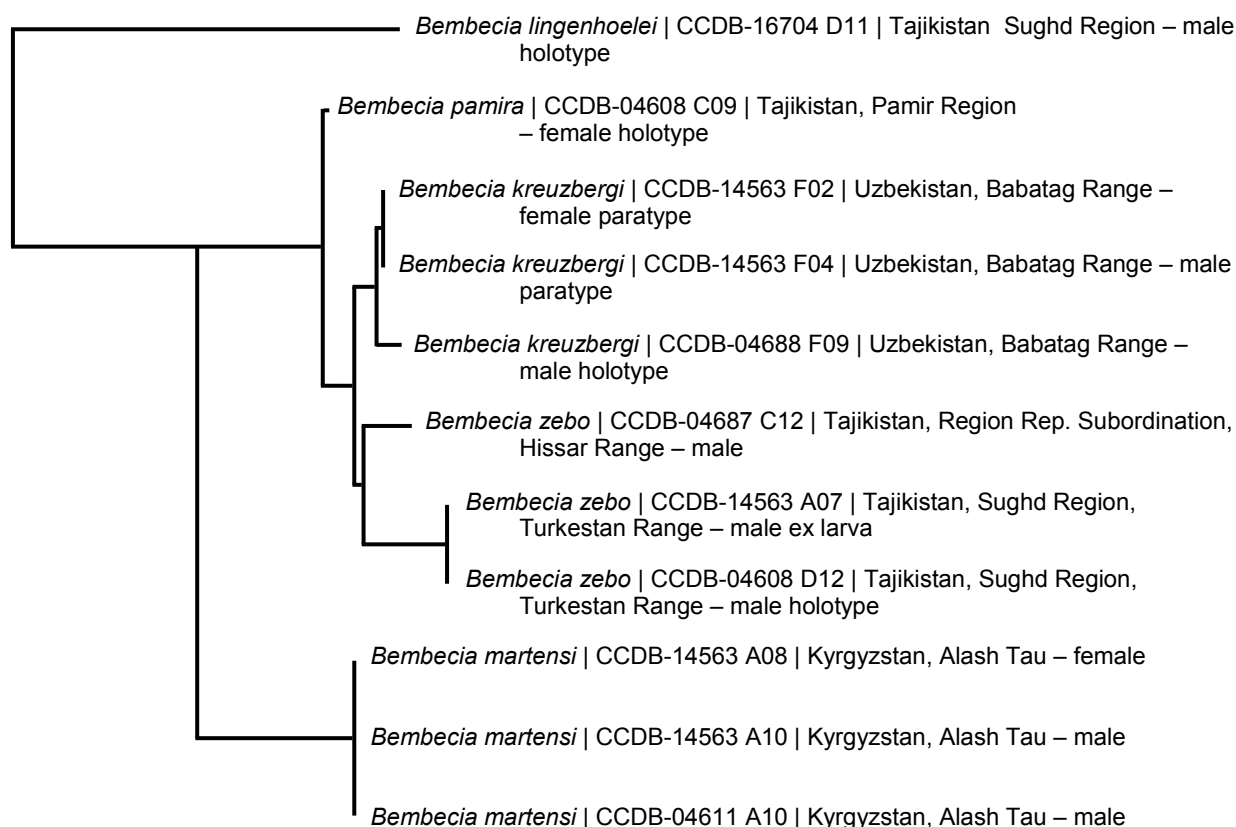


Fig. 28. Neighbour joining tree of DNA barcodes of *Bembecia* species, showing specimen registry numbers and localities of origin. *B. lingenhoelei* (Garrevoet & Garrevoet, 2011) is the outgroup.

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