Differences in the male and female genitalia between *Iphiclides podalirius* and *Iphiclides feisthamelii*, further supporting species status for the latter (Lepidoptera: Papilionidae)

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Abstract. The taxonomy of *Iphiclides podalirius* vs *I. feisthamelii* is reviewed historically and new genitalic evidence is presented, further supporting the view that these two taxa are specifically distinct.

Samenvatting. Verschillen in de mannelijke en vrouwelijke genitalia tussen *Iphiclides podalirius* en *Iphiclides feisthamelii*, ondersteuning voor de soortstatus van de laatste (Lepidoptera: Papilionidae)

De geschiedenis van de taxonomie van *Iphiclides podalirius* versus die van *I. feisthamelii* wordt geschetst en nieuw ontdekte verschillen in de genitalia worden besproken, welke het standpunt verdedigen dat beide taxa specifiek verschillend zijn.

Résumé. Différences entre les genitalia mâles et femelles d'*Iphiclides podalirius* et *Iphiclides feisthamelii*, confirmant le statut spécifique du dernier (Lepidoptera: Papilionidae) L'histoire de la taxonomie d'*Iphiclides podalirius* et d'*I. feisthamelii* et des différences récemment découvertes entre les genitalia sont discutées, établissant ainsi le statut spécifique des deux taxa.

Key words: Lepidoptera – Papilionidae – *Iphiclides – I. podalirius – I. feisthamelii –* genitalia – taxonomy – Spain – Portugal – France – Croatia – Greece – Morocco – Algeria – Tunisia.

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Abbreviations: ZMA = Zöologisch Museum Amsterdam; NNML = Nationaal Natuurhistorisch Museum Naturalis (Leiden).

Introduction

The taxonomic position of *Iphiclides feisthamelii* (Duponchel, 1832), a resident of the Iberian Peninsula, the extreme SW of France and the Maghreb States of Morocco, Algeria and Tunisia, still remains unsettled and unclear. It is being given specific status by Verity (1905–1911, 1925 & 1947–1950), Manley & Allcard (1970), Eitschberger (1993), Tennent (1996), Whalley & Lewington (1996), Wohlfahrt (1998), Maravalhas (2003), Lafranchis (2004), and Tarrier & Delacre (2008), while other authors, or the same, but at different instances, such as Munroe (1961), Higgins & Riley (1970), Gómez-Bustillo & Fernándes-Rubio (1974–1979), Higgins (1975), Hancock (1983), Higgins & Hargreaves (1983), Hesselbarth, van Oorschot & Wagener (1995), De Prins & Iversen (1996), Tolman & Lewington (1997), Lafranchis (2000), tentatively Häuser *et al.* (2005) and Racheli & Cotton (2009), have considered it as being a subspecies of *Iphiclides podalirius* (Linnaeus, 1758), whose range, it is said, extends from the Mediterranean area (excluding the Iberian Peninsula and N Africa), through

Phegea 39 (1) (01.III.2011): 12

Central Europe, to SW Siberia, the Middle East and Central Asia to NW China. These differences of opinion stem primarily from the degree of importance given to wing differentiating characters between the two taxa. To complicate matters even further, recent reports by Wiemers (2003) and Wiemers & Fiedler (2007), interpreting the results of mitochondrial DNA sequencing, have suggested a closer taxonomic relationship between Greek *I. podalirius* and Spanish *I. feisthamelii* than between Spanish *I. feisthamelii* and N African *I. feisthamelii*, thus probably denoting conspecificity between the first two and heterospecificity between the latter two. This particular find is mentioned by Racheli & Cotton (2009), but even though the conspecificity of Spanish *I. feisthamelii* to *I. podalirius* is accepted by them, the heterospecificity of the former to its N African counterpart is not, pending additional evidence.

Morphological differences

The morphological differences between these two taxa, as described in detail by many of the aforementioned authors, are related to the ground colour of and colour distribution on the wings, the degree of wing transparency, the width of the black wing stripes and the shape and colour tone of the orange bar right above the blue-speckled black ocellus at the hindwing anal angle. This bar, probably being the most important differentiating character, in *I. podalirius* is light orange, of uneven overall width, and wide distad of the outer edge of the HW anal angle, whereas in *I. feisthamelii* it is dark orange, of almost even overall width, and narrow distad of the outer edge of the HW anal angle.

Egg differences

These have been described by Eitschberger (1993), and relate to the shape and size of the egg chorion.

Hitherto published information concerning the genitalia

Information relating to the comparison between the genitalia of these two taxa is given by Higgins (1975), who figures and describes only the male genitalia of *I. podalirius*, but states that they are similar to those of *I. feisthamelii*, as well as by Lafranchis (2000) and Tarrier & Delacre (2008), who, without the support of figures and descriptions, and without giving the source of their information, simply refer to genitalic differences being present in both male and female. An attempt to contact these authors, in order to obtain the source of their information, brought about the necessary results, as the first one of the three had the kindness to inform us personally that when he was preparing his book on the butterflies of France (Lafranchis 2000), Gerard Luquet had informed him that Patrice Leraut had found differences between *I. podalirius* and *I. feisthamelii* "in the genitalia and the egg case". This information was apparently never published and thus, we have decided to provide below our own finds on the matter.

Material examined:

I. podalirius: Croatia. $2\sqrt[3]{2}$ Žirje Otok Island, sea level, 19.vii.1988. Greece. $1\sqrt[3]{3}$ Stereá Ellás, Mt. Pernassós, 1200 m, 6.vi.1965; $3\sqrt[3]{3}$ Attikí, one of which Katsimídi, 17.v.1965, the other two Athína, Likavittós Hill, one of which 5.v.1963, the other 12.vi.1963; $1\sqrt[3]{3}$ Égina Island, 5-6.iii.1979; $2\sqrt[3]{3}$ and $2\xrightarrow{2}$ Spétses Island, the two males Ágios Mámas, one of which 13.vi.1965, the other 7.viii.1969, one of the females 7.viii.1979, the other 11.vii.1959; $2\sqrt[3]{3}$ and $4\xrightarrow{2}$ Páros Island, Voutákos, sea level, one of the males 19.v.1982, the other 5.v.1997, two of the females 19.v.1982, the other two 20.viii.2001; $1\sqrt[3]{3}$ Lésvos Island, Mólivos, 25.vi.1967; $1\sqrt[3]{3}$ and $2\xrightarrow{2}$ Sámos Island, the male Mt. Karvúni, 800–1000m, 12.v.2000, the two females Xiropótamos, ca. 200 m, 22.vi.2003; $2\sqrt[3]{3}$ and $1\xrightarrow{2}$ Pátmos Island, one of the males Léfkes, sea level, 8.vii.2004, the other 0–150 m, 24–28.v.2008, the female Kípi, 0–20m, 11.vii.2004; $1\sqrt[3]{3}$ Kíthira Island, Áno Livádi, ca. 200 m, 3.vi.1999; $1\sqrt[3]{3}$ Kríti Island, Iráklio District, near Rogdiá, 350–400 m, 22.vi.1995. Morocco. $2\sqrt[3]{3}$ Tangiers.

I. feisthamelii: Spain. 23° Jaen, Los Propios, 600 m, 16.viii.1984; 13° and 19° Huesca, the male Salinas, 750 m, 3.viii.1986, and the female La Fortunada, 650 m, 1.viii.1986; 13° Málaga, Torrox, 8.v.1980; 13° Madrid, Las Matas, 23.vi.1982; 19° Jaca, Bernues, 27.vii.1983; 19° Granada, Las Vigoras, 1250 m, 15.vii.1984. France. 33° and 19° Pyrénées-Orientales, two of the males and the female Conat, 15.iv.1988, and one male Canigou, Col de Jou, 4.vii.1980. Algeria. 43° and 49° , two of the males and two of the females Aflou, the other two males and two females Algiers. Tunisia. 13° Ain Draham, 400–800 m, 8.vi.1980. Morocco. 23° and 19° , 1 male Middle Atlas, 120 m, the other High Atlas, 2100 m, and the female Fez.

Comparison between the male genitalia of *I. podalirius* and *I. feisthamelii*, based on recent finds

The male genitalia of these two taxa appear to be close enough to one another to have persuaded Higgins (1975) that they are similar. This, however, is not the case, as we have detected a constant and rather impressive difference in the distal end of the aedeagus, which, when viewed either dorsally or ventrally, constantly appears much wider and massive in I. podalirius (Figs. 1-16) than it does in I. feithamelii (Figs. 17-31), being at least twice, to almost two and a half times larger than in the latter. This last character may even suggest mechanical incompatibility in the pairing between male I. podalirius and female I. *feithamelii*, though, admittedly, the same may not hold true in reverse. One interesting aspect about the aedeagus of both taxa is that its distal end (when viewed either dorsally or ventrally) is asymmetrical, its right side carrying a short, rather flat spine, and its left side possessing the aperture through which the vesica is everted during copulation. Also worth noting is the fact that when the aedeagus is viewed laterally it does not give any hint of the noted difference, appearing identical in both taxa, and thus explaining Higgins' statement that their genitalia are similar.

Comparison between the female genitalia of *I. podalirius* and *I. feisthamelii*

These, likewise, are quite similar to one another, but the ductus bursae, in either dorsal, or ventral aspect, appears wider in *I. podalirius* (Figs. 32–40), than it does in *I. feisthamelii* (Figs. 41–49). The greater width in the former taxon seems to be compatible with the wider distal end of its aedeagus.

Sympatry and syntopism in France

I. podalirius and I. feisthamelii are sympatric in that country, at least at its southwestern extremity. Verity (1905–1911 & 1947) reports that the latter has been recorded in the extreme SW of France, at Caux, Hérault; Higgins & Riley (1970), as well as Tolman & Lewington (1997), report it from the E Pyrenees, S France; Lafranchis (2000 & 2004) records it from the eastern Pyrenees, parts of Ariège and Aude, the south of Hérault, and Roussillon, S France, and also mentions the presence of erratic individuals from the High- and Atlantic-Pyrenees; finally Borie (2009, personal communication) has personally recorded it in the neighbourhood of Perpignan, E Pyrenees. With the exception of Verity (who based his conclusions on a set of ten specimens from Caux, seven of which he identified as I. feisthamelii, two as I. podalirius, form zanclaeides Verity, 1911, and one as intermediate, which he named form *notha* Verity, 1947 (thus implying both syntopism and occasional hybridization between these two taxa), none of the other above mentioned lepidopterists report syntopism of I. feisthamelii with I. podalirius, and Borie in particular told us that in Perpignan, which he had visited on several occasions, he had never found I. feisthamelii together with I. podalirius, even though the latter is a well established and rather common insect in all the rest of the country. The fact however remains that for taxa whose ranges come into close proximity to one another, and which inhabit the same biotopes and fly in the same period and at the same altitudes there is always a good chance for syntopism, and therefore this condition cannot be ruled out.

Sympatry and syntopism in the Iberian Peninsula and in Morocco

Verity (1947) states that: "... la *podalirius* del rimanente della zone Palearctica, a cominciare dall'Altai e fino al Portogallo, donde l'ho della Serra de Gerez, e a Tangeri, sovrapponendosi alla *feisthamelii*, ma in modo scarsissimo nel territorio di quest'ultima." The report about the presence of *I. podalirius* in the Serra de Gerez, Portugal, well within *I. feisthamelii* territory, is not supported by Maravalhas (2003), whose book on the butterflies of Portugal presents recent findings, often based on personal collecting. His colour illustration, however, of an *I. "feisthamelli*" on p. 191, shows a butterfly that instead looks disturbingly like a Spring brood *I. podalirius*, thus lending support to Verity's record. The record of *I. podalirius* from "Tangeri" (= Tangiers, Morocco) at first appeared to us as being highly improbable, but two specimens from the same locality, obtained from the NNML in The Netherlands, turned out to be *I. podalirius*, both on the basis of wing morphology as well as by genitalia (Figs. 15, 16). This means that at least in the near past this butterfly did indeed exist in Morocco, possibly through involuntary introduction.



Phegea **39** (1) (01.III.2011): 16

Figs. 1–16. Dorsal aspect of distal end of aedeagus of *Iphiclides podalirus*. 1–12. Greece. 1. Stereá Ellás, Mt. Parnassós, 1200 m, 6.vi.1965; 2, 3, 4. Attikí. 2. Katsimídi, 17.v.1965. 3, 4. Athína, Likavittós Hill. 3. 5.v.1963. 4. 12.vi.1963. 5. Égina Island, 5–6.iii.1979. 6, 7. Spétses Island, Ágios Mámas. 6. 13.vi.1965. 7. 7.viii.1969. 8. Lésvos Island, Mólivos, 25.vi.1967. 9. Sámos Island, Mt. Karvúni, 800–1000 m, 12.v.2000. 10, 11. Pátmos Island. 10. 0–150 m; 24–28.v.2008. 11. Léfkes, sea level, 8.vii.2004. 12. Kíthira Island, Áno Livádi, ca. 200 m, 3.vi.1999. 13, 14. Croatia, Žirje Otok Island, sea level, 19.vii.1918. 15, 16. Morocco, Tangiers.



Figs. 17–26. Dorsal aspect of distal end of aedeagus of *Iphiclides feisthamelii*. 17–21. Spain. 17, 18. Jaén, Los Propios, 600 m, 16.viii.1984. 19. Huesca, Salinas, 750 m, 3.viii.1986. 20. Málaga, Torrox, 8.v.1980. 21. Madrid. Las Matas, 23.vi.1982. 22–24. France, Pyrénées-Orientales. 22, 23. Conat, 15.iv.1988. 24. Canigou, Col de Jou, 4.vii.1980. 25–28. Algeria. 25–27. Aflou, v. 28. Algers, v. 29. Tunisia, Ain Draham, 400–800 m, 8.vi.1980. 30, 31. Morocco. 30. Middle Atlas, 1200 m. 31. High Atlas, 2100 m.



Figs. 32–40. Ventral aspect of ductus bursae of *Iphiclides podalirius* from Greece. 32, 33. Spétses Island. 32. 7.viii.1959. 33. 11.viii.1959. 34–37. Páros Island, Vutákos, sea level. 34. 19.v. 1982. 35. 19.viii.1982. 36, 37. 20.viii.2001. 38, 39. Sámos Island, Xiropótamos, ca. 200 m, 22.vi.2003. 40. Kríti Island, Iráklio District, near Rogdiá, 350–400 m, 22.vi.1995.



Figs. 41–49. Ventral aspect of ductus bursae of *Iphiclides feisthamelii*. 41–43. Spain. 41. Huesca, La Fortunada, 650 m, 1.viii.1986. 42. Jaca, Bernues, 27.vii.1983. 43. Granada, Las Vigoras, 1250 m, 15.vii.1984. 44. France, Pyrénées-Orientales, Conat, 15.iv.1989. 45–48. Algeria. 45, 46. Aflou, v. 47, 48. Algers. 49. Morocco, Fez.

Recent works on the butterflies of the Maghreb States, such as the ones by Tennent (1996) and by Tarrier & Delacre (2008), make no mention of any such record, and therefore we are assuming, pending new evidence, that probably the butterfly may have since become extinct from that area.

The mitochondrial DNA issue

We are not questioning the mtDNA sequencing results regarding *Iphiclides*, arrived at by Wiemers (2003), and Wiemers & Fiedler (2007), first of all because we are simply ignorant of molecular genetics as a whole. We think, however, that we are entitled to have doubts about the interpretation of these results. It does not make sense to us to believe that two morphologically and structurally different and geographically totally separated from one another butterflies, such as are *I. feisthamelii* from Spain and *I. podalirius* from Greece, should be regarded as being conspecific, while two morphologically and structurally totally similar and geographically in close proximity to one another butterflies, such as are *I. feisthamelii* from Spain and its counterpart from N. Africa, should be considered as most probably representing two separate species. Perhaps if this sequencing study was not based on the mitochondrial gene COI alone, but included the study of other genes as well, the sequencing results may have been totally different. Doubts regarding the credibility of the interpretations of mtDNA sequencing are also expressed by Zakharov, Lobo, Nowak and Hellmann (2009).

Voltinism of I. podalirius from Greece

In Greece this butterfly is often on the wing from as early as beginning March, to as late as end October, during which time fresh specimens are still to be found flying about. This suggests that under the right climatic conditions *I. podalirius* probably has as many as four broods per year.

Conclusions

The presence of constant differences between *I. podalirius* and *I. feisthamelii*, as expressed in their wing morphology, their male and female genitalia and the chorion of their eggs, coupled with sympatry, as reported from Portugal and Morocco, and syntopism, as recorded at least once in SW France, all are factors that suggest that these two butterflies should best be considered as representing two distinct species. The single reported hybrid (Verity 1947) does not negate our views, as there are quite a few cases of successful hybridization between firmly established separate species. (In fact the term "hybrid" by definition implies the offspring between two different species, for if the parents are conspecific then the offspring, being necessarily conspecific with the parents, cannot be termed hybrid). The interpretation of mtDNA results should be at present viewed with caution, in anticipation of a broader approach to DNA sequencing.

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