

Contribution to the knowledge of two endemic Corsican butterflies, *Polyommatus coridon nufrellensis* and *Plebejus bellieri* (Lepidoptera: Lycaenidae) in relation to *Hippocrepis conradiae*: first evidence as hostplant, discovery of a new locality and update on distribution, biology and conservation

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Abstract. During an entomological excursion from 14th till 21st July 2012 in Corsica, France, a new locality for the Corsican Blue *Polyommatus (Lysandra) coridon nufrellensis* (Schurian, 1977) has been discovered by the authors in the Fango valley. Since its discovery in 1975 this *Polyommatus* was only known from the vicinity of the a Muvrella Mountain and from the nearby Spasimata valley after its rediscovery in 2001. The new location, with a different habitat, is situated at a lower altitude (below 1000 m) and about 8 km south from the known distribution places. In this article, the authors present for the first time evidence by its behaviour (oviposition of female, egg on plants) and on the two known localities (Spasimata valley, Fango valley) for *Hippocrepis conradiae* Gamisans & Hugot, 2011, being a hostplant for caterpillars of *P. coridon nufrellensis*. Furthermore, DNA-analysis results of all eggs, found on the same plant, suggests *H. conradiae* to be also a hostplant for *Plebejus bellieri* Oberthür, 1910. Therefore, the presumed unilateral endemic plant-butterfly relationship is more complex and needs to be updated. Next to this, the discovery of a second biotope situated in the upper Fango valley raises hope that the poorly known distribution of the elusive *P. coridon nufrellensis* may be wider than expected. Finally, the new distribution area, habitat, ecology and conservation status are discussed. In particular, the lower altitude of the new locality increases the potential zone for further research.

Samenvatting. Bijdrage tot de kennis van twee Corisicaanse endemische vlinders, *Polyommatus coridon nufrellensis* en *Plebejus bellieri* (Lepidoptera: Lycaenidae) in relatie tot *Hippocrepis conradiae*: eerste bewijs als waardplant, de ontdekking van een nieuwe vindplaats en actualisering van de verspreiding, biologie en het behoud ervan
Gedurende een entomologische excursie van 14 tot 21 juli 2012 in Corsica, Frankrijk, werd door de auteurs een nieuwe biotoop voor het Corsicaans Bleek Blauwtje *Polyommatus (Lysandra) coridon nufrellensis* (Schurian, 1977) ontdekt in de Fango vallei. Sedert de ontdekking in 1975 was gekend dat deze ondersoort enkel voorkwam rond de berg a Muvrella en na de herontdekking ervan in 2001 ook in de hoger gelegen gedeeltes van de Spasimata vallei. De nieuwe vindplaats met een andere habitat situeert zich echter lager (net onder 1000 m) en zowat 8 km ten zuiden van de, tot op heden, enige gekende verspreidingsplaatsen. In dit artikel leveren de auteurs voor de eerste keer het eenduidige bewijs (vrouwje in ovipositie, ei op plant), en op meerdere plaatsen (Spasimata vallei, Fango vallei) dat *Hippocrepis conradiae* Gamisans & Hugot, 2011 een voedselplant is voor de rupsen van deze endemische vlinder. Verder suggereren DNA-analyseresultaten van alle eitjes, gevonden op dezelfde plant, dat *H. conradiae* ook een waardplant is voor *Plebejus bellieri* Oberthür, 1910. Bijgevolg is de veronderstelde eenzijdige endemische plant-vlinder relatie complexer en moet deze worden herzien. Ten slotte wordt het nieuwe verspreidingsgebied, biotoop, ecologie en natuurbehoud besproken. De ontdekking van een tweede biotoop in de Fango vallei doet de hoop rijzen dat het slecht gekende verspreidingsgebied van deze bijzondere *P. coridon nufrellensis* groter kan zijn dan verwacht. Vooral de lagere hoogte van de nieuwe plaats biedt nieuwe kansen voor verder onderzoek.

Résumé. Contribution à la connaissance de deux papillons endémiques de Corse, *Polyommatus coridon nufrellensis* et *Plebejus bellieri* (Lepidoptera : Lycaenidae) en relation avec *Hippocrepis conradiae*: première preuve que cet *Hippocrepis* est bien la plante hôte, découverte d'une nouvelle localité et éléments nouveaux concernant distribution, biologie et conservation
Au cours d'une excursion entomologique du 14 au 21 Juillet 2012 en Corse (France) une nouvelle localité fréquentée par *Polyommatus (Lysandra) coridon nufrellensis* (Schurian, 1977) a été découverte dans la vallée du Fango par les auteurs. Depuis sa découverte en 1975, ce *Polyommatus* n'était connu que des environs de la montagne, a Muvrella, et de la haute vallée de la Spasimata, après sa redécouverte en 2001. La nouvelle localité, correspondant à un habitat différent, se situe en moyenne altitude (au-dessous de 1000 m) et à environ 8 km au sud des localités déjà connues. Dans cet article, les auteurs confirment *Hippocrepis conradiae* Gamisans & Hugot, 2011 comme plante hôte des chenilles de *P. coridon nufrellensis* en apportant les premières preuves d'observations in situ (oviposition, œufs sur les plantes), réalisées dans les deux localités connues (la vallée de la Spasimata et la vallée du Fango). De plus, les résultats des analyses ADN d'autres œufs trouvés sur la même plante, suggèrent que *H. conradiae* est aussi une plante hôte de *Plebejus bellieri* Oberthür 1910. Par conséquent, la relation présumée unilatérale d'une plante et d'un papillon endémiques est plus complexe et doit être étudiée. La découverte d'un deuxième biotope, situé dans la vallée du Haut Fango, laisse espérer que la distribution encore peu connue du rare *P. coridon nufrellensis* soit plus étendue qu'il n'était soupçonné auparavant. Finalement, la nouvelle distribution, les biotopes, l'écologie et son statut de conservation sont discutés. En particulier, la plus basse altitude de la nouvelle localité augmente la zone potentielle de recherche.

Zusammenfassung. Beitrag zur Kenntnis zweier auf Korsika endemischen Schmetterlingen, *Polyommatus coridon nufrellensis* und *Plebejus bellieri* (Lepidoptera: Lycaenidae) in Bezug auf *Hippocrepis conradiae*: erste Beweise als Wirtspflanze, Entdeckung einer neuen Lokalität und Aktualisierung der Verbreitung, Biologie und Naturschutz
Während einer entomologischen Exkursion zwischen dem 14. und dem 21. Juli 2012 auf Korsika, Frankreich, wurde von den Autoren ein neuer Standort für die auf Korsika endemische Bläulingunterart, *Polyommatus (Lysandra) coridon nufrellensis* (Schurian, 1977) (Lepidoptera: Lycaenidae), im Fango Tal entdeckt. Seit der Entdeckung im Jahr 1975 war bekannt, dass diese Unterart nur um den Berg Muvrella und in den oberen Teilen des Spasimata Tals, nach der Wiederentdeckung im Jahr 2001, vorkommt. Der neue Standort, mit einem anderen Lebensraum, befindet sich in niedriger Höhe (knapp unter 1000 m) und liegt

ca. 8 km südlich von den bis dahin einzigen Verbreitungsstandorten. In diesem Artikel bringen die Autoren zum ersten Mal die eindeutigen Beweise (Weibchen in Eiablage, Ei auf Futterpflanze), und dies sowohl an mehreren Standorten (Spasimata Tal, Fango Tal), dass *Hippocrepis conradiae* Gamisans & Hugot, 2011 eine Futterpflanze für die Raupen dieser endemischen Schmetterlingsart ist. Darüber hinaus belegen DNA-Analysen von übrigen Eiern auf derselben Pflanze *H. conradiae*, dass sie auch eine Futterpflanze für *Plebejus bellieri* Oberthür 1910 ist. Daher ist die vermutete einseitige endemische Pflanze-Schmetterlings-Beziehung komplexer und muss aktualisiert werden. Schließlich werden neue Verbreitung, Lebensraum, Ökologie und Schutz-Status diskutiert. Die Entdeckung eines zweiten Biotops im Fango Tal weckt die Hoffnung, dass die wenig bekannte Verteilung dieser besonderen *P. coridon nufrellensis* Unterart weiter ist als erwartet. Besonders die neue Stelle in tieferen Lagen bietet neue Möglichkeiten für die weitere Forschung.

Keywords: Corsica – endemism – Lycaenidae — *Polyommatus (Lysandra) coridon nufrellensis* — *Plebejus bellieri* – hostplant – Fabaceae – *Hippocrepis conradiae* – *Hippocrepis comosa* – oviposition – co-evolution – ecology – distribution area – habitat conservation.

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In the main text abbreviated as LP and EZ.

1. Introduction

Polyommatus coridon nufrellensis (Schurian, 1977) is one of the rarest European Blues, with a very local and limited distribution area in the higher mountains of Corsica, France. Because of its small and restricted distribution area, it took several decades until the entomological community accepted its occurrence and description. K. Schurian (1977) received the first specimen collected by Dr. R. Mayr, and described the new subspecies from “Mt. Nuvrella”, wrongly interpreted from a letter written by the latter (Tolman 1997). Dr. R. Mayr discovered the first specimen west of Mount a Muvrella between 23 and 27 July 1975 at an altitude of 1900–2200 m (Schurian *et al.* 2006).

For the next couple of decades no more specimens were found despite several attempts to locate populations, which made the first description by Schurian controversial for the non-believers (Kleinebuhle 1999). It took 26 years, until August of 2001, before it was rediscovered by M. Guidi, an Italian entomologist, who captured a female of this subspecies in the Asco valley at 1450 m. Two years later, this observation was confirmed by M. Gascoigne-Pees, who found it common between an altitude of 1200–1300 m in the Spasimata valley (Schurian *et al.* 2006). This rediscovery was confirmed by other entomologists, and described by G. Sala, who added extra knowledge about its biology (Sala *et al.* 2005). Surprisingly, the distribution area is at a much lower altitude (between 1200 and 1450 m) than that given in the initial description. Subsequent attempts by various entomologists added little knowledge, and the biology of this Blue became shrouded in mystery (Cuvelier 2003, 2008; Schurian *et al.* 2006, 2011; Diringer 2009). Indeed, this rare subspecies had never been observed in copulation nor had oviposition by the females been seen. Also, in spite of intensive searching in 2005, 2006, 2007 and 2008, the hostplant remained unknown (Diringer 2009; Schurian *et al.* 2006, 2011).

Inspired by these mysteries, in the summer of 2011 the first author (LP) decided to make an entomological excursion to Corsica to search for *P. coridon nufrellensis* and to try to find out more on its biology. During this trip, LP found 8 males and 6 females in the Spasimata valley

between 18th and 24th of July. Most of them were feeding intensely on the yellow flowers of *Doronicum corsicum* (Loiseleur-Deslongchamps) Poiret, confirming previous observations describing this plant as a major nectar source for this subspecies (Schurian *et al.* 2006), but no copulations or females in oviposition were seen. However, on 18 July, a worn female of *nufrellensis* seen in oviposition behaviour around a bunch of *Genista salzmanii* var. *lobelioides* Gamisans in the Spasimata valley, around 1200 m became a subject for discussion with other lepidopterists, a.o. S. Cuvelier, who visited the island in 2008 (Cuvelier 2008) and observed in the same valley some females showing oviposition behaviour on a flowering *Allium* sp. (pers. comm.). After exchanging ideas, LP decided to return to the island, probably better earlier in the season, searching for additional information on all the possible endemic hostplants in the Haut-Asco mountain range around Mt. Cinto. With these ideas in mind, in December 2011, LP received the surprising news of an article describing a new *Hippocrepis* species for Corsica, different from *H. comosa* Linnaeus, discovered by Gamisans and colleagues in 2010: *Hippocrepis conradiae* Gamisans & Hugot, 2011 (Gamisans *et al.* 2011). In that paper, its distribution is described from the Haut-Asco mountain range, occurring only in small populations (max. 50 m²) in and around the Spasimata and the Haut-Fango valleys between 1200 and 1865 m altitude: “*H. conradiae* croît dans des anfractuosités de falaises de rhyolites siliceuses, et au pied de ces falaises, entre 1200 et 1865 m d’altitude (...) Bunifatu, vallée de la Spasimata, partie S, crête rocheuse près de Bocca di Pitinaghia, sur une falaise rhyolitique, en versant WNW, 1610 m, (...) Bunifatu, vallée de la Spasimata., sommet de Meta di Filu, en versant S, falaise rhyolitique, 1865 m, (...) Haut Fangu, au-dessus de Saltare, Haute Cavicchia, Val de Ghiarghja Minuta, dans des pierriers de rhyolites au bord du torrent, 1200–1250 m” (Gamisans *et al.* 2011). Interestingly, the search for *Hippocrepis* in Corsica by botanists in the 1980’s led to the discovery of *H. comosa* from the Spasimata valley, only some years after *nufrellensis*’ description. This discovery, by Mrs. Conrad, was confirmed by J. Gamisans in 1985 (Schurian *et al.* 2006). In turn, but later, the rediscovery of *P. coridon nufrellensis* in 2001 by M. Gascoigne-Pees was facilitated

by his knowledge of the distribution information of *H. comosa* in Corsica, which is the usual foodplant of nominotypical *Polyommatus coridon coridon* (Poda, 1761) on the mainland, provided by J. Gamisans (Schurian *et al.* 2006).

In the same article, however, and after first describing *H. conradiae*, the two authors, together with third author D. Jutzeler suppose *H. conradiae* to be the foodplant for *P. coridon nufrellensis*. Unfortunately, this hypothesis is not supported by any field observation or breeding experiment (Gamisans *et al.* 2011, Dodinval 2012). Further, *H. comosa* had already been described from the same valley (Conrad 1980) and proposed as a foodplant for *nufrellensis* (Schurian *et al.* 2006, 2011, Diringer 2009, Descimon *et al.* 2010, Tshikolovets 2011). However, our understanding is that all the references to *H. comosa* in Corsica are based on the single observation by Mrs. Conrad in the Spasimata valley (Conrad 1980, Gamisans *et al.* 2010). More detailed information by Mrs. Conrad about the distribution of *H. comosa*, revealed this plant was growing “in the lower parts, stations abyssales, around 1000 m in the Spasimata valley” but only “in small quantities” and in granite soil, which is unusual for this species of Fabaceae (Schurian *et al.* 2006, Jeanmonod & Gamisans 2007). In this respect, it is noteworthy that in the description of *H. conradiae*, its altitudinal range is not mentioned in the observation by Mrs. Conrad (Gamisans *et al.* 2011) although this observation had been confirmed by Gamisans before (Schurian *et al.* 2006). Therefore, it seems that the distribution of *Hippocrepis* in Corsica is not fully understood and it remains possible that both *H. conradiae* and *H. comosa* might occur in the same area near the habitat of *P. coridon nufrellensis*.

With all questions unanswered, EZ and LP decided to make a new entomological trip to the Haut-Asco mountain range of Corsica, between the French and Belgian National Days.

2. Material and methods

2.1 Field trip on Corsica

Butterflies collected in Spasimata valley and Fango valley, using a standard butterfly net, were retained for identification. Some females from Spasimata valley were taken alive to obtain eggs as a reference for further determination.

2.2 Identification of eggs, caterpillars, imago

During our excursion, adults and eggs were found (see 3.1.2 for detailed description). Because Lycaenidae eggs are small and difficult to identify accurately in the field, they were taken home for further investigation. Determination was initially made by using focus stacking pictures. In the meantime some eggs had hatched, and we applied a DNA analysis method on both eggs and caterpillars as a control to obtain an unambiguous determination. DNA analysis was also used on tissue from an imago. A detailed description of the techniques used:

a. Focus stacking pictures of the eggs

A focus stacking program (Zerene Stacker vs. 1.04 by Zerene Systems LLC.) was applied to a series of sharply focused photos, taken using a Nikon D90 camera with a modified objective (Nikon Plan 10/0.30 - 160/0.17) placed on an automatic stacking rail (Stackshot Macrorail by Cognysis). The number of photos taken per focus stacking picture was determined by the program controlling the automatic rail using following settings: begin position, end position, distance between photos (generally 10 μ m). Based on observations described by Schurian *et al.* (2006), we used pictures of unknown eggs (Egg 1 and Egg 2) taken in both dorsal and lateral views and in close up of the micropylar region to compare with reference eggs in the same positions.

b. DNA analysis method

Following samples were used: 1) emerged caterpillar (L1 stage) and empty egg shell from “Egg 2” Fango valley, 2) emerged caterpillar (L1 stage) from “Egg 3” Fango valley, 3) a leg from a specimen collected in Fango valley (see 3.1.2 for detailed description). Egg or empty egg shell from Spasimata valley, laid by females taken at home, were used as a control. DNA extraction and amplification were performed in the Butterfly Diversity and Evolution Lab at Institut de Biologia Evolutiva, Barcelona, Spain. Total genomic DNA was extracted using Chelex 100 resin, 100–200 mesh, sodium form (Biorad), under the following protocol: a larval body part (for samples 12-Q572 and 12-Q573) or one leg (sample 12-Q574) was removed and immersed into 100 μ l of Chelex 10%; 5 μ l of Proteinase K (20 mg/ml) were added. The samples were incubated overnight at 55°C and then at 100°C for 15 minutes. Samples were then centrifuged for 10 s at 3000 rpm. A 658-bp fragment at the 5' end of the mitochondrial gene COI was amplified by polymerase chain reaction (PCR) using the primers LepF1 (5' TTCAACCAATCATAAAGATATTGG-3') and LepR1 (5' TAAACTTCTGGATGTCCAAAAATCA-3') (Hebert *et al.* 2004). Double-stranded DNA was amplified in 25- μ l volume reactions containing: 14.4 μ l autoclaved Milli-Q water, 5 μ l 5x buffer, 2 μ l 25 mM MgCl₂, 0.5 μ l 10 mM dNTPs, 0.5 μ l of each primer (10 μ M), 0.1 μ l Taq DNA Polymerase (Promega, 5U/ μ l) and 2 μ l of extracted DNA. The typical thermal cycling profile began with denaturation at 92°C for 60 s, followed by five cycles of 92°C for 15 s, 48°C for 45 s and 62°C for 150 s, and then by 35 cycles of 92°C for 15 s, 52°C for 45 s and 62°C for 150 s and a final extension at 62°C for 420 s. PCR products were purified and sequenced by Macrogen Inc. Sequences were edited and aligned using GENEIOUS PRO 4.7.5 created by Biomatters (www.geneious.com).

Species level identifications were achieved by using the Blast Local Alignment Search Tool (BLAST) of the National Center for Biotechnology Information (NCBI) (www.ncbi.nlm.nih.gov/BLAST/Blast.cgi?CMD=Web&PAGE_TYPE=BlastHome). On the 6th of December 2012, nucleotide blasts were run for each obtained COI sequence, by using the entire nucleotide collection (nr/nt) and optimizing the process for highly similar sequences (megablast).

3. Results

3.1 Field observations

3.1.1. Spasimata valley

Between 14th and 21st July 2012, the authors explored different localities in the Haut-Asco mountain range. On 16th of July the weather was ideal to make a first climb into the Spasimata valley. After a few hours walking, partly along the GR20 route, the most famous hiking path in Corsica, LP saw a female *P. coridon nufrellensis* nectaring on *D. corsicum* at an altitude of 1230 m, near the same spot where LP observed his first specimen in 2011. Remarkably, this female was already a little worn, and a male in the same locality even more. According to previously published data (Cuvelier 2008), this seemed to be indicative that 2012 was an early season for *nufrellensis*. In the higher parts of the valley, ca. 1300 m, a second spot was visited. Although somewhat disturbed by sunbathing tourists in and around the valley, both sexes were again seen nectaring on *D. corsicum*, confirming previous observations (Schurian *et al.* 2006, 2011, Gamisans *et al.* 2011). At this altitude, *P. coridon nufrellensis* was clearly more numerous (a total of approximately 25 adults were observed in about two hours) and females were seen flying to and coming from higher portions of the valley, a behavioural observation corresponding with descriptions in literature (Schurian *et al.* 2006). Given the description of *H. conradiae* and accurate information of its occurrence in the Spasimata valley (Gamisans *et al.* 2011), together with a detailed map of localities (kindly provided by J. Gamisans) we moved higher in the valley to discover new data.

At ca. 1450 m (Fig. 1a), close to the type locality (TL) of *H. conradiae* (Gamisans *et al.* 2011), LP was surprised to observe some pink flowering plants of *Erica terminalis* Salisbury (Plate 1.a & b), covered with bluish butterflies. These proved to be numerous *P. coridon nufrellensis* and *Plebejus bellieri* (Oberthür, 1910), with a few other Lycaenidae including *Lycaena phlaeas* (Linnaeus, 1761) and one male of *Lampides boeticus* (Linnaeus, 1767). Redirecting our attention to *nufrellensis*, we observed both sexes flying in and around this place. Shortly afterwards, EZ found two small tufts of *H. conradiae* on the cliffs near the type TL. Despite intensive searching, we failed to observe copulation or oviposition.

The next morning, July 17th, climbing again along the same path between 8:30 a.m. and 9:30 a.m. we passed the places where we had observed butterflies nectaring the day before, but at this time of the day we saw none, probably because the sun was not yet shining directly into the valley. Around 10 a.m., with the sun reaching the lower parts of the valley, the first *P. coridon nufrellensis* appeared. Arriving at the locality of *H. conradiae*, visited the day before, we found it still in shade, so we went to the lower place at ca. 1450 m which was sunlit and again with many lycaenids feeding on *E. terminalis*. Here, we

studied the flight and feeding behaviour of *nufrellensis* for the next hours.

1) Flowering plants are abundant here and of many species, mostly *D. corsicum* and *E. terminalis*, but also some smaller, rather similar plants, later identified from illustrations as being of at least two different species, *Armeria leucocephala* Koch and *Allium schoenoprasum* Linnaeus (S. Cuvelier, pers. comm., based on information from J. Gamisans);

2) *D. corsicum* with its typical yellow flowers only grows on the wettest places, around stationary pools of water and small streamlets in the valley;

3) *E. terminalis* shrubs grow in different spots on the slopes and cliffs, some 10 to 20 m above *D. corsicum*. They are larger, with a typical pink inflorescence (Plate 1.a & b), and attracted even more butterflies;

4) *Armeria leucocephala* and *Allium schoenoprasum* (Plate 1.c & d): are both smaller, with a single white-pinkish inflorescence which was attractive to both sexes for nectaring. However, both species were less conspicuous, sometimes growing between bushes of bigger plants such as *G. salzmanii*. Despite the fact that most of the plants had already dried out and the flowers were somewhat wilted, they still attracted a lot of *nufrellensis*. Although no actual oviposition was seen, some females showed “pseudo-oviposition-like” behaviour after feeding on these flowers, an observation made previously in the Spasimata valley (S. Cuvelier, pers. comm.);

5) Numbers of *nufrellensis* were higher than in the localities we visited the day before. About 70 specimens recorded in all and females appeared to outnumber males.

Bearing in mind these observations, we continued exploring to an altitude of 1485 m at the TL of *H. conradiae*. After EZ found the first small patches of this plant on very steep cliffs, LP noticed that it was growing much more abundantly a few meters higher up. Searching for a good way of reaching the more accessible plants, we screened the whole area in a radius of about 200 m. Despite our efforts, we did not find another location for this rare endemic *Hippocrepis*. In the early afternoon, LP began the short but dangerous climb to reach the exact TL of *H. conradiae*. Having reached it (Plate 1d), LP made following observations:

1) *H. conradiae* was abundant here, but most of the plants had already dried out or at least a lot of the leaves had lost their turgidity. Only those growing in the shade under higher cliffs and protected from the midday sun were still in good condition;

2) *A. schoenoprasum* was growing between *H. conradiae* in good numbers and in the same condition as observed a little lower down, at 1450 m, and still attracted a lot of *P. coridon nufrellensis* for nectaring. Some butterflies were feeding on almost completely dried flowers, probably due to the absence of alternatives at the TL;



Plate 1. Spasimata valley, 1450 m, around the TL of *Hippocrepis conradiae*. 16–17.vii.2012. a.–b. *Polyommatus coridon nufrellensis* ♀ feeding on *Erica terminalis*; c. *P. coridon nufrellensis* ♂ feeding on *Allium schoenoprasum*; d. Biotope of *P. coridon nufrellensis* with nectaring and hostplants growing together: *H. conradiae* is abundant here (TL), with some *A. schoenoprasum* (red arrow indicates inflorescence); e. Detail of *H. conradiae* leaves: recent feeding traces of a caterpillar (red arrow), probably of *P. coridon nufrellensis*; f.–g. Oviposition by female *P. coridon nufrellensis* in Spasimata valley, on a tuft of *H. conradiae*; h. Egg deposited by a female of *P. coridon nufrellensis* on a leaf of *H. conradiae* (red arrow). All photos: L. Parmentier.

3) After feeding at these flowers, some females clearly were attracted to the *Hippocrepis* tufts and showed oviposition behaviour, searching actively and when disturbed, flying away but often quickly returning to the same spot. A little lower down, females showed similar behaviour around identical flowers, but less intensive;

4) Exploring *H. conradiae* plants, LP observed some whitish feeding traces among the leaves. Their colour, shape and condition suggested they were from caterpillars of *nufrellensis*, but this could not be confirmed (Plate 1.e).

After about two hours waiting in the midday sun, LP noticed a female *nufrellensis* crawling around and somewhat into a *H. conradiae* plant, but actual oviposition was not seen. However, a second female was subsequently photographed egg laying on this plant (Plate 1.f & g) and shortly afterwards the egg was found on a leaf (Plate 1.h). Other females showed more attraction to the plants that had not dried out, but none

were seen in the proximity of fresh plants under the cliff in the shade. Oviposition was more likely when a female crawled more into the centre of the *Hippocrepis* tuft (seen twice). Thus the behaviour around a *G. salzmannii* bunch (observation LP, 2011) and around the flowers of *Armeria leucocephala* and *Allium schoenoprasum* (observations LP and EZ, 2012) can be regarded as “pseudo-ovipositioning-behaviour”. A small quantity of *H. conradiae*, some with feeding traces, was collected for further investigation at home and some females were kept for eggs.

Following the publication of *H. conradiae* and its distribution in Corsica (Gamisans *et al.* 2011) and with detailed information on its distribution in the Haut-Asco region, we focused our attention for the second part of our stay on the Fango valley. Apart from the lowest parts of this valley, tourist visits are negligible compared to those in the Spasimata valley. The Fango valley is not on the GR20-route, and we saw that it would be necessary to take camping equipment for our stay.



Fig. 1. Comparison between explored biotopes of Spasimata and Fango valleys. a. Spasimata valley (~1500 m) 16–17.vii.2012. Known biotope for *P. coridon nufrellensis* with foodplant *H. conradiae* and nectaring plants *D. corsicum*, *E. terminalis*, *Armeria leucocephala* and *Allium schoenoprasum*; b. Fango valley (just below 1000 m) 19.vii.2012. New locality for *P. coridon nufrellensis*. *H. conradiae* growing between the stones on the river side. Note differences in habitat in the two valleys where *H. conradiae* grows. Photos: a.: L. Parmentier; b.: E. Zinszner.

3.1.2. Fango valley

On 19th July, we started our second excursion early in the morning from the town of Barghiana. The road above Barghiana is forbidden to vehicles and the long climb has to be done on foot to reach the point indicated as ‘base camp’ (Fig. 2). Many endemic butterfly species were observed in the meanwhile, including *Lasiommata paramegaera*, *Argynnis paphia f. immaculata*, *Hipparchia aristaeus*, *H. neomiris*, *Coenonympha corinna* and *P. bellieri*. At an altitude of ca. 510 m, on the borders of the Fango valley (Plate 2.a), LP noticed some inflorescences of two *D. corsicum* plants, as far as we know, the first record at this altitude (Plate 1.b) (Gamisans *et al.* 2004, Paolini *et al.* 2007). In any case, these plants were growing at an altitude considerably lower than the lowest described altitudes from the Spasimata valley. Because this plant is known as a good nectar source for *P. coridon nufrellensis* (Diringer 2009, Schurian *et al.*

2006, pers. observation), this was an encouraging first observation, but a quick look around, revealed no *nufrellensis* butterfly.

After noon, we continued our exploration in the direction of the upper Fango valley. At a certain point, around 800 m altitude, the upper Fango valley divides but with the available information about *H. conradiae* distribution higher up, we intuitively took the southeasterly fork (Fig. 2). From this point on, progress is slow and very difficult because of dense Corsican “maquis” vegetation on the steep rocky sides of the valley. The best strategy is probably to climb up along the riverside but even so, it took a few hours to get only about 200 m higher. Meanwhile, other sites for *D. corsicum* were located along the riverside, always around the pools. Suddenly, when EZ was taking a picture of the biotope (Fig. 1.b), LP recognized a *Hippocrepis* tuft between the stones beside the river (Plate 2.c). After critical

examination of features and size of the leaves and seed structure, the authors decided it had to be *H. conradiae* and not *H. comosa*, a conclusion confirmed when field samples were compared with data in literature. Surprisingly, this site was at a mere 1000 m altitude (IGN 2004) and in a forested habitat (Fig. 2.a). However, a careful search around this place and some 50 m higher and lower, did not allow to discover another tuft. When looking at this plant, LP noticed again some typical whitish holes in the leaves, most probably feeding traces of a Lycaenidae caterpillar (Plate 2.d). Looking more closely into the tuft, EZ found a Lycaenidae egg on a small leaf (Plate 2.e) which we hoped was an egg of *nufrellensis*. Within half an hour, two more eggs were found deeper in the tuft. Two of the three eggs were found on dry leaves in the centre close to the roots, the third on a petiole, less dried out, situated more on the outer part of the tuft.

By this time it was getting late, and further search would have been futile, especially for adults of *P. coridon nufrellensis*, so we documented the biotope and decided to return. This higher part of the Fango valley has a tall and dense vegetation, but the river bed had almost dried out apart from some of the larger pools which were still filled with water. In these we noticed some dead Lepidoptera, mostly moths, and LP remarked jokingly it would be a last chance to find *nufrellensis* floating on the water surface. Only a few minutes later, on the way down, LP found a dead male *nufrellensis* floating in one of these pools (Plate 2.f). It was quite worn, but still showed enough features for a positive identification: the typical blue colour and submarginal ocelli of *P. coridon nufrellensis* (Plate 2.g & h). This was the first proof that this endemic subspecies occurs elsewhere than in the a Muvrella area. The site, at around 970 m, is only a few metres below the *H. conradiae* tuft the authors had just discovered. Further search in the vicinity yielded no more adults in the pools nor living adults feeding at *D. corsicum* flowers growing in the same area.

With a cloudless sky the next day, the authors climbed back to the divide. They decided to explore the north-east fork of the Fango valley towards the high peaks in the direction of the Spasimata valley (Fig. 2), with the idea of looking for other localities for *nufrellensis* between the South-East part of the Fango and the upper Spasimata valley. This climb even seemed tougher than that on the previous day but was slightly facilitated at some point when traces of an ancient path were found and used to reach a higher level. A full afternoon's work spent on climbing and searching this difficult terrain failed to reveal any sign of *nufrellensis* or the hostplant associated with it. Just a few *D. corsicum* plants were found in the area, in typical places but more enclosed at this altitude, and amongst dense vegetation consisting of trees and bushes along the riverside.

3.2 Identification of eggs found on *H. conradiae* in Fango valley

The three eggs found on 19 July were taken home for further investigation and proper determination, using the techniques described above.

a. Egg 1

This was found on a leaf petiole. Unfortunately, it had hatched before it could be properly examined, leaving an empty egg shell and a hatched caterpillar, which soon died. A picture of the empty egg shell was used for comparison with previously identified *nufrellensis* eggs obtained from the females taken in the Spasimata valley, and the caterpillar was used for DNA analysis as a positive control.

Results of a comparison of this egg shell with a series of pictures of eggs of nominotypical *coridon* and subsp. *nufrellensis* (Spasimata) eggs are shown on Plate 3 (from left to right): a. *Polyommatus coridon coridon* (origin: Inor, France); b. *P. coridon nufrellensis* from female collected in Spasimata valley; c. *P. coridon nufrellensis* egg shell, from same female; d. Egg 1 egg shell from Fango valley. Differences in egg chorion structure and the micropylar region of nominotypical *coridon* and *nufrellensis* are clearly visible. So we were able to differentiate between nominal *coridon* and *nufrellensis* eggs. The empty egg shell matched *nufrellensis*, but unfortunately the caterpillar had emerged via the micropyle, so impeding unambiguous identification. Nonetheless, we were confident that this egg had been laid by a *nufrellensis* female.

DNA analysis was then applied as a control on Egg 1. (RVcoll.12-Q572) and the dead caterpillar (RVcoll.12-Q574). Both samples RVcoll.12-Q572 and RVcoll.12-Q574 had identical COI sequences and the BLAST revealed them as most similar to several *P. (Lysandra) coridon* (four or five nucleotide differences), and also to a few specimens of *P. (Lysandra) bellargus* Rottentburg (five nucleotide differences). This is not surprising, as *P. coridon* and *P. bellargus* are suspected of introgression (e.g. Dincă *et al.* 2011). However, since *P. bellargus* is unknown on Corsica (Kudrna *et al.* 2011), our specimens can only belong to *P. coridon nufrellensis*, the only *Polyommatus* subgenus *Lysandra* taxon present on the island.

These results are consistent with the observations in the Spasimata valley, where a female was seen egg-laying on *H. conradiae* – further evidence that *H. conradiae* is the hostplant of *P. coridon nufrellensis*.

b. Egg 2 and Egg 3

These two eggs were found towards the centre of the plant on totally dried out leaves (brown coloured) close to the soil. When the time came for detailed study, Egg 2 was still unhatched, and focus stacking pictures were used satisfactorily for a confident identification. By then, however, Egg 3 had already hatched, and the same identification steps were taken as used in 3.2.a.

Pictures of Egg 2 and Egg 3 are shown on plate 4: a. Egg 2 (lateral, frontal view and detailed view of micropylar region), b. Egg 3 (only lateral view, because of hatched caterpillar), c. For comparison, we used SEM pictures of *P. bellieri*, published by Jutzeler *et al.* (2003). From our focus stacking pictures, we draw the following conclusions:

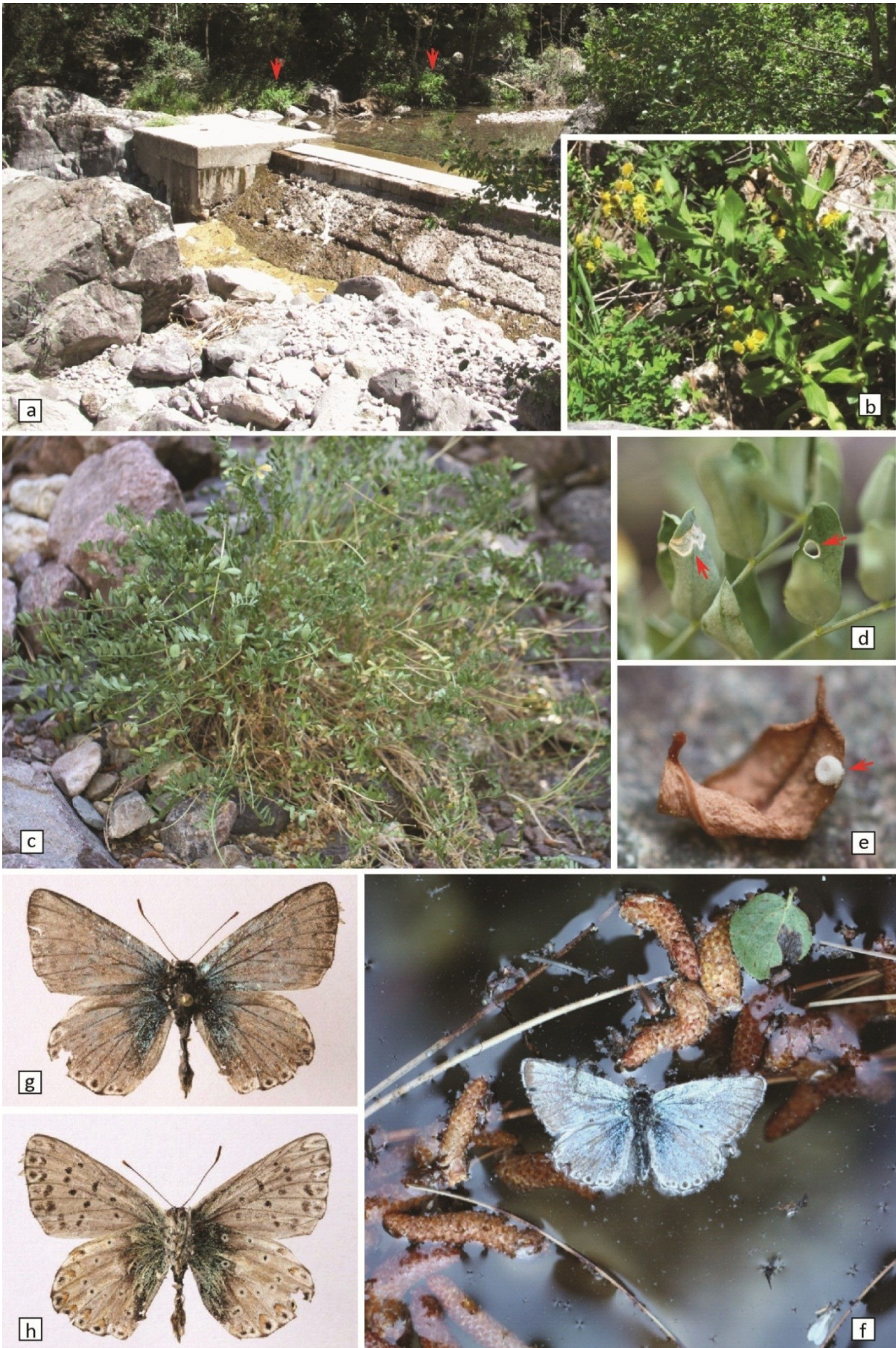


Plate. 2. Fango Valley. 19.vii.2012. New locality for *Polyommatus coridon nufrellensis* and *Hippocrepis conradiae*. a. View from base camp (ca. 510 m). First observation of yellow flowers of *Doronicum corsicum* in this valley (red arrows); b. Detailed picture of *D. corsicum*; c. "Abyssal" tuft of *H. conradiae* growing in the Fango valley below 1000 m; d. Feeding traces of a caterpillar on *H. conradiae* leaves; e. Some eggs were found on dead leaves and petioles on this plant (arrow indicates an egg of *Plebejus bellieri*); f. Dead *P. coridon nufrellensis* ♂ in a small pool in the river bed. g.-h. Upper and underside of set *P. coridon nufrellensis* ♂. All photos: L. Parmentier.

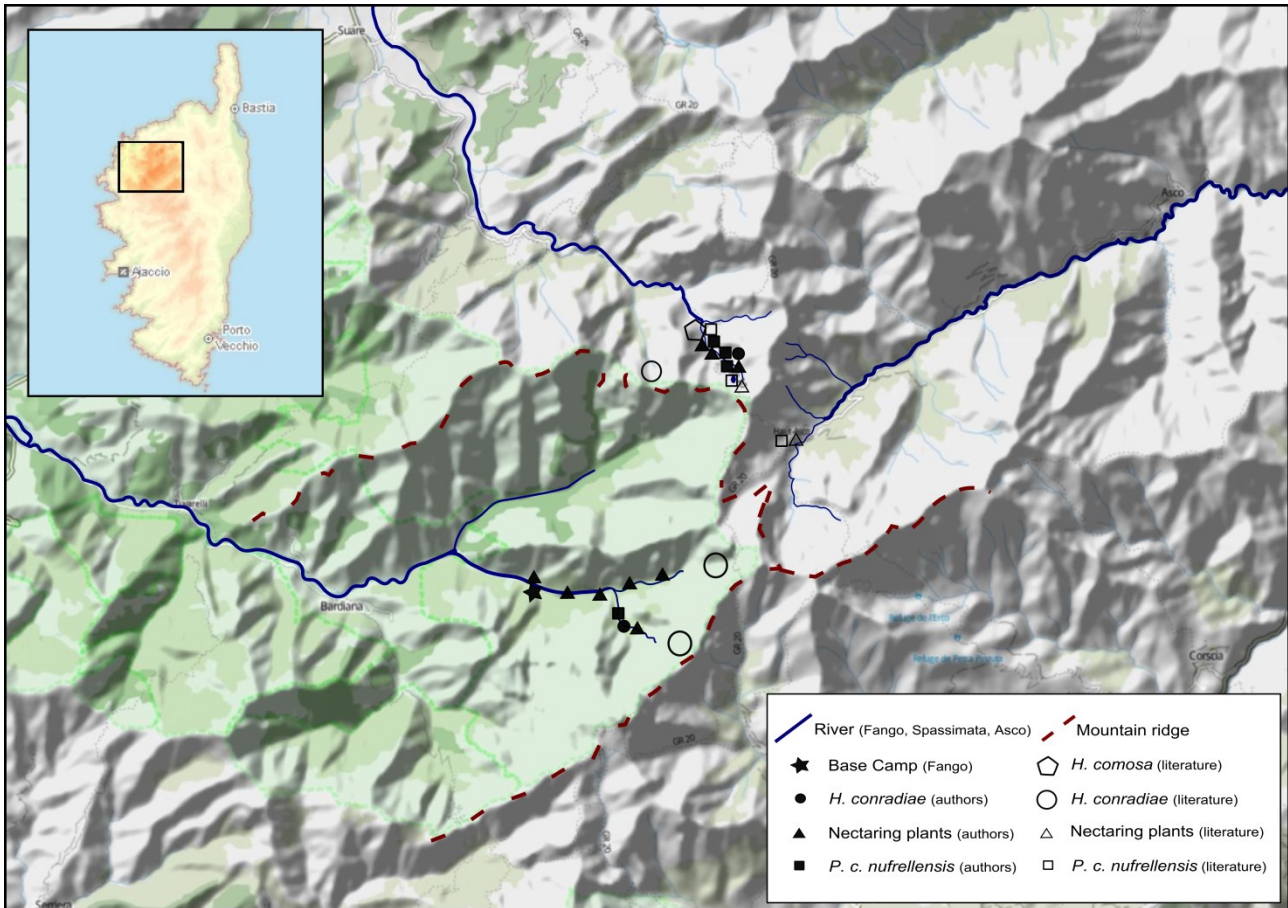


Fig 2. Map of Corsica, Haut-Asco region around Spasimata, Asco and Fango valley. Indicated on the map: big rivers (blue line) and mountain ridges (dotted red line), base camp in Fango valley; Indicated from literature or by authors' observations: *Polyommatus coridon nufrellensis* (eggs and/or adults), foodplants (*Hippocrepis conradiae* and/or *Hippocrepis comosa*) and nectaring plants (*Doronicum corsicum* and other species). Map by L. Parmentier.

1. Lateral view: Egg 2 and Egg 3 are identical; we conclude that both eggs belong to the same taxon;
2. The outer egg structures on the frontal pictures as well as the distance between egg chorion structures in the lateral view on pictures of Egg 2 correspond with the SEM-photos shown in c; we conclude Egg 2 and Egg 3 are those of *P. bellieri*.

As a control, the same DNA-identification method, described under 2.2.b and 3.2.a on the hatched caterpillar from Egg 3 (RVcoll.12-Q573) was applied. Sample RVcoll.12-Q573 was most similar to that of several specimens of *Plebejus argyrognomon* (Bergsträsser, 1779) (only one nucleotide difference), and also to several *Plebejus idas* (Linnaeus, 1761) and *Plebejus christophi* (Staudinger, 1874) (two nucleotide differences). These three species have been shown to have similar DNA barcodes (Lukhtanov *et al.* 2009, Dincă *et al.* 2011). However, none of them are found on Corsica, and therefore our samples are assigned to *P. bellieri*, a taxon with controversial status, but closely related to *P. idas* (Kudrna *et al.* 2011). Comparisons with unpublished COI sequences by Vila & Dincă confirmed the BLAST results.

Visual identification with focus stacking pictures and by DNA analysis both gave the same result, and we conclude that both these eggs were laid by a *P. bellieri*

female on *H. conradiae*, the first evidence that this is a hostplant of this Blue butterfly.

4. Discussion

4.1. The distribution of *Hippocrepis* in Corsica

Until now, various articles and field guides (e.g. Tolman & Lewington 1997, Lafranchis 2000, Tshikolovets 2011) cite *H. comosa* as the only hostplant of *P. coridon nufrellensis*, following the original report made by Mrs. Conrad of the presence of this plant in non-calcareous sediments (Conrad 1980). Subsequent attempts to relocate *H. comosa* on Corsica have been unsuccessful, but its presence, particularly in the "stations abyssales" (around 1000 m) cannot be excluded, as these were not included in the given altitudinal distribution area for *H. conradiae* (1200 to 1850 m) (Gamisans *et al.* 2011). However, the present authors made the unambiguous discovery of an "abyssal" *H. conradiae* (compare illustrations on Plate 1.e and Plate 2.d) just below 1000 m in the Fango valley. The altitude and biotope of our observation is consistent with that made by Conrad in the eighties, and is strongly suggestive that the old "abyssal *H. comosa*" in the Spasimata is in fact an "abyssal *H. conradiae*". Therefore, we conclude:

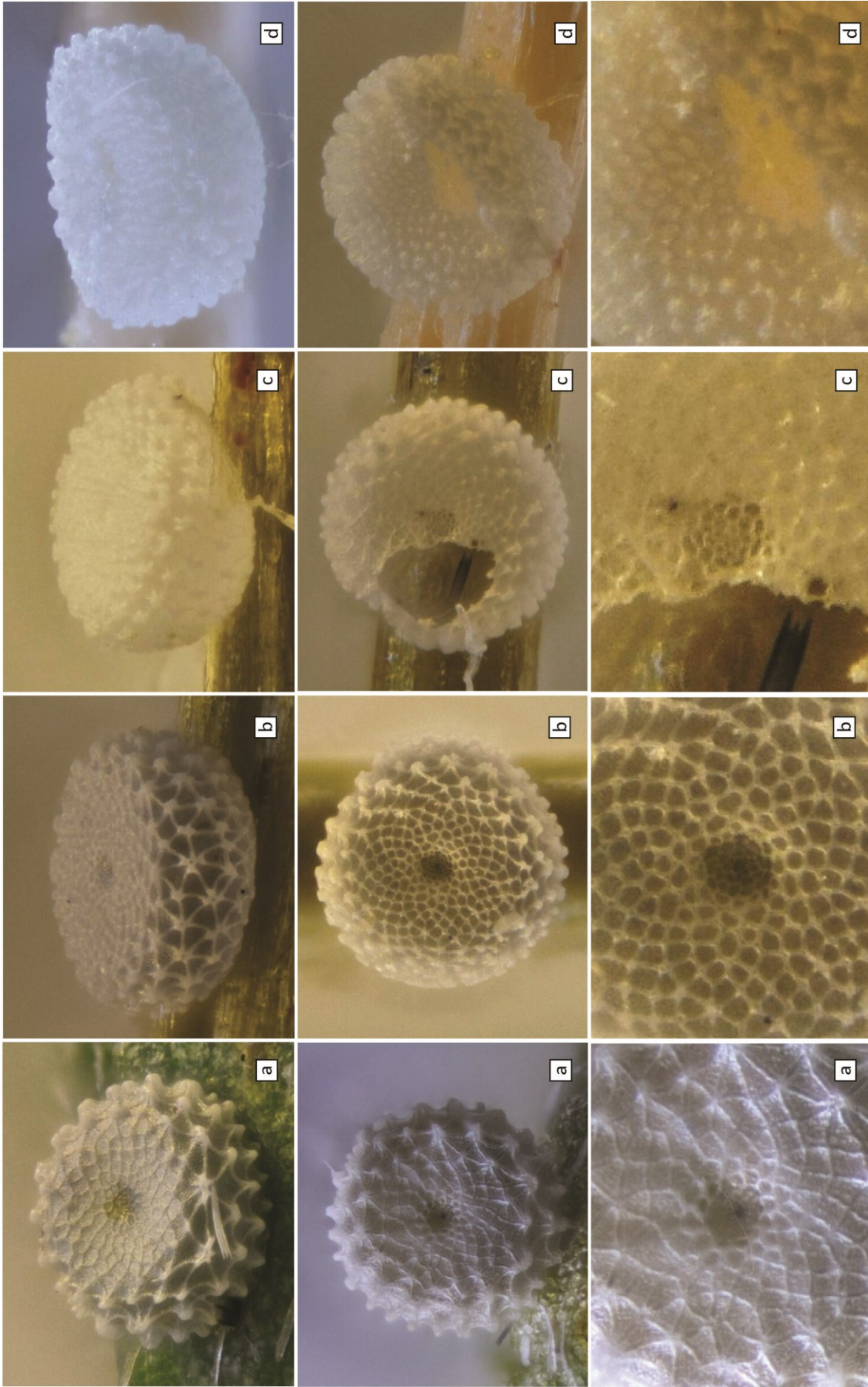


Plate 3. Focus stacking pictures of *P. coridon* ssp. eggs: a comparison between *P. coridon coridon* and *P. coridon nuffrellensis* (a. vs. b.), between un-hatched and hatched eggs (b. versus c.) and between *Spasimata* and Fango valley (unknown "Egg 1"); Origin: a. France, env. Inor, 18.vii.2012; b./c. France, Corsica, Spasimata valley, 16-17.vii.2012; d. France, Corsica, Fango valley, 19.vii.2012; Eggs: leg & coll. L. Pamentier (a., b., c.) and E. Zinszner (d); All focus stacking pictures: S. Cuvelier, processed by L. Parmentier.

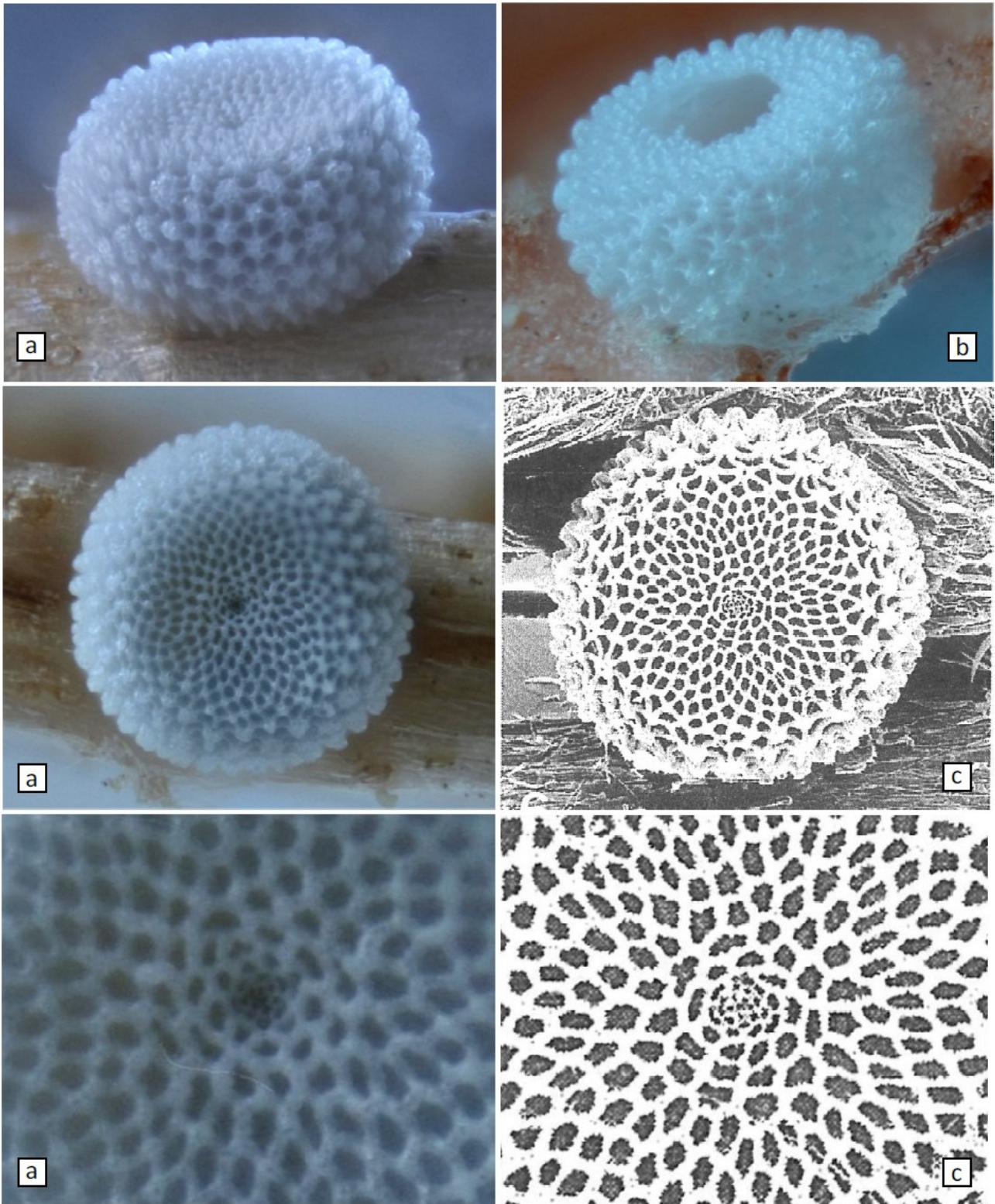
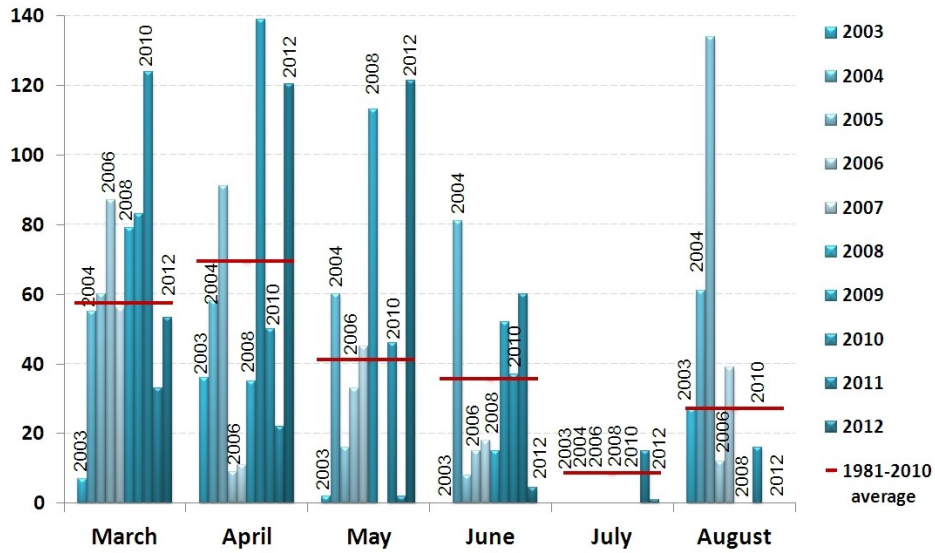


Plate 4. Focus stacking and SEM pictures of *Plebejus bellieri* eggs: a. comparison between outer egg chorion structures from a known *P bellieri* egg (c.) and unknown "Egg 2"(a.) and "Egg 3" (b.) found on a *H. conradiae* plant in upper Fango valley; origins: a./b. France, Corsica, Fango valley, 19.vii.2012; c. SEM pictures by U. Jauch (Jutzeler et al. 2003); Leg. & coll: L. Parmentier (a.) and E. Zinszner (b.); All focus stacking pictures: S. Cuvelier, processed by L. Parmentier.

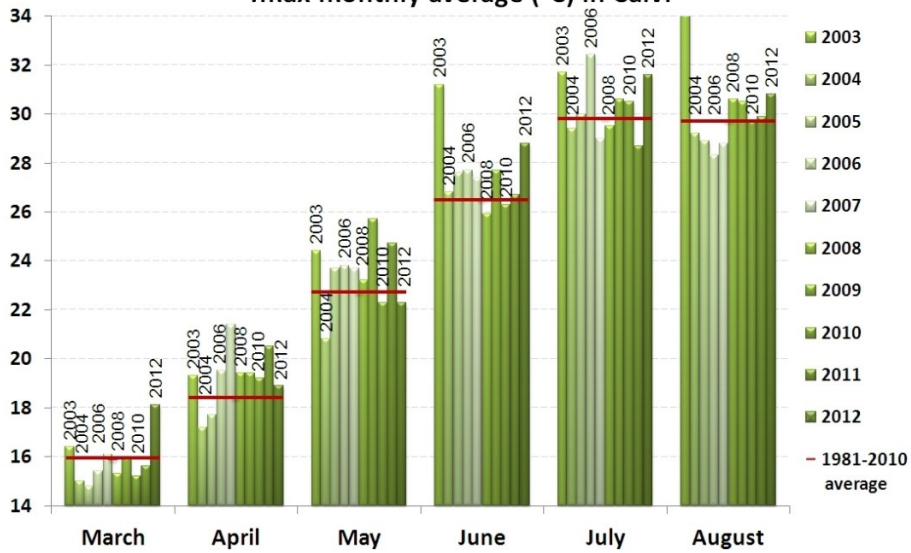
a) All Corsican references to *H. comosa* as the foodplant of *nufrellensis* given in the literature almost certainly refer to *H. conradiae* and this should be borne in mind in future articles and field guides.

b) The altitudinal distribution of *H. conradiae* is broader than hitherto described and should be extended in the Mt. Cinto area to between 995 m and 1865 m (Fig. 2).

Monthly rainfall (mm) in Calvi



Tmax monthly average (°C) in Calvi



hours of Sun in Month in Calvi

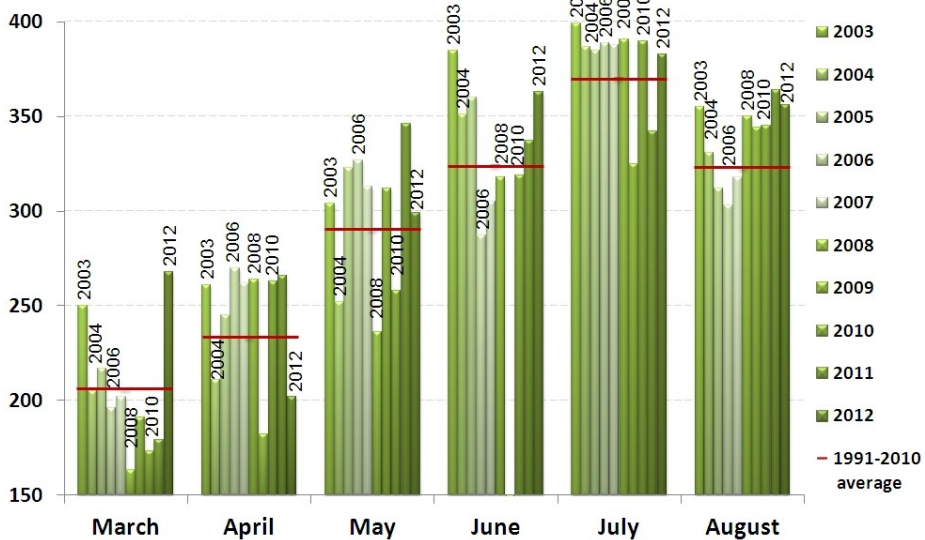


Plate 5. Average weather statistics for Calvi, period 2003 – 2012. 1. Monthly rainfall (mm); 2. Average monthly maximum temperature; 3. Average monthly hours of sunshine. Data: Météo-France, summarized by E. Zinsner.

c) *H. conradiae* grows in a wider range of habitats than previously thought: between stones in open, drier and higher habitat as well as in wetter lower “abyssal” localities in a forested habitat.

d) *H. conradiae* is likely to occur over a greater altitudinal range in the Spasiamata valley as well, especially in inaccessible places.

4.2. Flight period of *nufrellensis* and variation in abundance in different years

The optimal flight period indicated in literature is given as late July to early August (Tolman & Lewington 1997, Lafranchis 2000, Tshikolovets 2011). However, in 2012, *nufrellensis* was already well out by 16th July in the Spasimata valley. About 70% of the males were already worn, and many females were past their best, and it was already quite late to expect to see copulations in mid July. We sought a possible correlation between the variation in abundance of the butterfly and weather conditions. For this purpose, we compared the weather data over several years for the region, using data for Calvi from Météo-France (<http://climat.meteofrance.com>). Even though the mountainous microclimate on Monte Cinto can differ from that at the seaside in Calvi, the monthly climate bulletin by Météo-France shows that observed weather data trends for Calvi are valid to use for the Monte Cinto area, as shown on graphs in Plate 5.

First, we discuss the variation in abundance of *nufrellensis*. In 2006, K. Schurian and later H. Descimon (Diringer 2009) located just a single specimen, while in other years, e.g. the summers of 2008 (Cuvelier, pers. comm.) and 2012 (LP and EZ) several dozens of adults were seen in the same biotope. In the spring of 2008 and 2012 rainfall, was abundant and far above average in April and May (Plate 5.1). This may have resulted in a good growth of *H. conradiae*, which in turn would have allowed the caterpillars to flourish, and by the end of the spring in those years, to successful pupation. This reasoning is supported by an inverse observation: UK populations of *P. bellargus*, the larvae of which feed on *H. comosa*, suffered as a result of an extreme drought in 1976 (Thomas 1983). Many of the more isolated populations have not recovered from this event (Harper *et al.* 2003, 2008). Thus there is evidence that drought causing *Hippocrepis* to wilt can be a major threat to successful development in these butterflies, and could explain the significant variability in abundance of *nufrellensis* in recent years as observed by different lepidopterists (Cuvelier 2008, Diringer, 2009, LP, pers. observation).

Secondly, we suggest an explanation for the early appearance of adults of *nufrellensis* as observed by both authors in 2012 and in a lesser extent in 2011 by LP. In terms of average temperature (Plate 5.2) or sunshine (Plate 5.3) the spring of 2012 did not differ significantly from that of previous years. However, it is a striking fact that the maximum monthly temperature (T max in Plate 5.2) and the average number of hours of sunshine (# hours sun in Plate 5.3) were significantly higher in 2012. These conditions imply that the eggs would have hatched earlier in the season, due to higher T max and more #

hours of sunshine in March, and that the development time of the caterpillars and emergence of pupae was brought forward due to higher T max and more # hours sunshine in June. Therefore, the overall cycle underwent not only one but two accelerations, which may explain the presence of worn males and females by mid July.

The above average abundance of *nufrellensis* adults in 2012, with an optimal flight period at least 2 or even 3 weeks earlier than given in literature, was probably due to these conditions. Further, the lower position in the Fango valley, surrounded by high mountain ridges (Fig. 2) could have resulted in a warmer local microclimate and probably an earlier shift in flight period compared to that in the Spasimata valley.

In the last decade, most of the years have been warmer than average for the last thirty years, a result of global warming (Plate 5.2). Possible changes in the flight period of *nufrellensis* in relation to this trend will need to be monitored in the future.

5. Update on the biology, distribution and ecology of *P. coridon nufrellensis*

5.1. Endemic butterfly-plant relationship: biology

It took 26 years following its description to get the first knowledge about *nufrellensis*' biology. Here, we discuss our new observations in the light of the complex relationship between nectar sources and *Hippocrepis* as foodplant for *nufrellensis* and other Corsican endemic *Polyommatus* species.

A. Nectar sources

In the contributions by Schurian and co-workers (2006), and Gamisans *et al.* (2011), a complex relationship between *P. coridon nufrellensis* adults and their sources of nectar is discussed, and *Doronicum corsicum* is given as the only plant known to be attractive. We confirm that *D. corsicum* is attractive in wetter places, but more importantly, we also observed *nufrellensis* feeding at the flowers of *Erica terminalis*, *Armeria leucocephala* and *Allium schoenoprasum* (described under 3.1.1). Together, they form an altitudinal gradient: at the lowest level in the valley, *D. corsicum* grows close to the riverside; higher up, *E. terminalis* occurs along the sides of the valley until just below the first *H. conradiae* foodplants, where *Armeria leucocephala* and *Allium schoenoprasum* are growing. The last two species were seen growing together with *H. conradiae* (Plate 1.d). We found that *E. terminalis* is a second, excellent food source because of the abundance of flowers and its robustness in comparison with *D. corsicum*. Conversely, the few flowers on the much smaller *A. leucocephala* and *A. schoenoprasum* offer only a small amount of food for the Blues. Even so, *nufrellensis* was strongly attracted to these flowers, and when females were feeding on them, they showed a typical “pseudo” ovipositioning behaviour. This behaviour was seen at different localities and by other lepidopterists as well (S. Cuvelier, pers. comm.); this behaviour has to be called “pseudo” or real, depending on the presence of *H. conradiae* in its proximity or not.

Next, we can have a closer look at the distribution of all known nectar sources around Mt. a Muvrella (Fig. 2). In the Spasimata valley, we discussed an altitudinal gradient of different nectar sources. In the Fango valley we observed only *D. corsicum*, but starting at a much lower altitude from as low as 510 m. It can be assumed that other nectar sources also exist in this valley, but more field work is needed to find whether there is an altitudinal gradient for nectar plants and also for *nufrellensis* in the Fango and other valleys. According to the literature, *nufrellensis* has also been observed east of a Muvrella, in the Asco valley around the ski station. In the summer of 2003, M. Gascoigne-Pees saw two specimens feeding on 'thistle' flowers (Sala *et al.* 2005). This is evidently a third nectaring place and possible breeding site for *nufrellensis*, if the food plant is located there.

To summarise, we updated the former insights (by Schurian *et al.* 2006) regarding *nufrellensis*' nectaring sites and demonstrated there are far more sites present, even next to their hostplant and breeding habitat. This is more in line with the relationship observed between *P. coridon coridon* populations and *H. comosa* growing in the mainland of Europe (Tolman & Lewington 1997, Bink 1992).

B. *Hippocrepis conradiae* as a hostplant for Lycaenidae

We have already shown (under 4.1.) that *H. conradiae* is more widespread than previously described in the Monte Cinto range, but populations of this endemic plant are patchy and very local (Schurian *et al.* 2006, Gamsans *et al.* 2011, pers. observations). Next, we brought the first evidence that *H. conradiae* is a foodplant for *P. coridon nufrellensis* and also for *P. bellieri* and demonstrated herewith that the assumed "co-evolved" unilateral relationship between *H. conradiae* and *P. coridon nufrellensis* (Gamsans *et al.* 2011) is more complex. Moreover, the extent to which this scarce plant is affected by the attentions of various caterpillars is unknown. According to the literature, *P. bellieri* is far more abundant than *nufrellensis*, but the caterpillars also feed on other (endemic) Corsican Fabaceae including *Cytisus praecox* Beauverd and *G. salzmannii*. Therefore it must be less dependent on *H. conradiae* unless when several foodplants occur together, *H. conradiae* is more attractive for the caterpillars. It is worth mentioning that *P. bellieri* was found quite commonly in the Spasimata valley, just below the TL of *H. conradiae*, feeding together with *nufrellensis* at flowers of *E. terminalis*. *P. bellieri* flies earlier in the season (Tolman & Lewington 1997, Tshikolovets 2011). If its caterpillars are attracted to *H. conradiae*, they would be ahead of those of *nufrellensis*, but as yet there is no evidence that the presence of *P. bellieri* caterpillars earlier in the season would have any detrimental effect on the survival of *nufrellensis*.

There are also other Lycaenidae caterpillars of which *H. conradiae* could be a hostplant: *Plebejus argus corsicus* Bellier de la Chavignerie and *Polyommatus icarus* Rottemburg seem to be good candidates. *P. argus*

corsicus is known to feed on *G. salzmannii* and *Anthyllis hermanniae* Linnaeus (Jutzeler & Leestmans 1999), and *H. comosa* is a well-known foodplant of *P. argus argus* Linnaeus on the European mainland (Tolman & Lewington 1997). Furthermore, it is known that *P. argus corsicus* occurs in the Spasimata valley and east of Mt. a Muvrella around 1500 m (Jutzeler & Leestmans 1999), although it is not known in the Fango valley. *P. argus corsicus* has been seen accompanying *nufrellensis* in the Spasimata valley (Schurian *et al.* 2006), and there is good reason to suppose that a third endemic taxon is also associated with *H. conradiae*. *P. icarus* is a successful species which feeds on many hostplants including Fabaceae (Tolman & Lewington 1997, Tshikolovets 2011) and it was quite plentiful scattered along Spasimata and Fango valleys, feeding at flowers in the vicinity of *H. conradiae*. More research is needed to elucidate the relationship between these Lycaenid butterflies and *H. conradiae*. In any case, it is important that the few known localities for the rare *H. conradiae* are protected.

5.2 Distribution and ecology

The populations of *nufrellensis* are closely related to the very patchy distribution of the larval foodplant. It seems that the largest population of *H. conradiae* is at the TL, but a single population of that size would be insufficient to sustain the long term survival of *nufrellensis*, and such a highly specialized butterfly is more threatened than the larval foodplant by habitat fragmentation because of its higher trophic position (Brückmann *et al.* 2010).

On the mainland of Europe, *P. coridon* is considered to be a sedentary butterfly (Bink 1992, Schmitt *et al.* 2006) that can build up populations of high density locally. Exchange of individuals, and hence genes, between fragmented populations are possible because wanderers occasionally move over longer distances (Asher *et al.* 2001, Bereton *et al.* 2007). The altitudinal gap between the first described locality of *nufrellensis*, 1900–2200 m (Schurian 1977, Tolman & Lewington 1997), and those where it was rediscovered, 1200–1450 m (Schurian 2006) suggests the presence of a wider metapopulation of this species in the Spasimata valley where the foodplant is present, which supports shifts in the core distribution of the butterfly over time, in response to local stress.

However, exchanges between metapopulations are limited by altitudinal or longitudinal barriers, and because of the low natural dispersal ability of *Hippocrepis* species (Hennenberg *et al.* 2003) and the rarity of *H. conradiae* in the Mt. Cinto mountains (see Fig. 2), these factors are likely to be a serious obstacle to the spread of *nufrellensis*.

The Fango valley locality of *nufrellensis* is about 8 km away from those in the Spasimata valley, and therefore it is not clear whether the Fango population is a separate metapopulation or conjunct with the Spasimata population. The fact that the lower locality in the Fango valley is surrounded by several high mountain peaks (Fig. 2) suggests a formidable geographical barrier for such a sedentary butterfly, but in the absence of more data this

can only be assumed. Further exploration of the Monte Cinto area and of very similar mountain ranges further south could lead to the discovery of other *nufrellensis* populations.

The discovery of the historic biotope and its rediscovery almost three decades later was greatly facilitated by the nearby presence of the GR20. Without GR20 and road leading to Haut-Asco, *nufrellensis* might very well have remained unknown. A less welcome consequence of the GR20 is that the Spasimata valley attracts many tourists who walk along the path and go sunbathing within the valley, activities which stress the biotope where the "abyssal" foodplant of *nufrellensis* might grow. A good conservation strategy for the survival of both very local endemics demands a delicate balance between their needs and human activities in the area.

6. Conclusion

Our expedition revealed the presence of *P. coridon nufrellensis* in a new locality in the Fango valley, and observed oviposition on *H. conradiae*. Eggs which were found on this endemic foodplant proved, by DNA analysis on (empty) eggs and a comparison of focus stacking pictures, to be those of *P. coridon nufrellensis*. We also found eggs of *P. bellieri*, another endemic *Polyommatus* species, on the same plant, another unique observation. The "co-evolved" relationship between *P. coridon nufrellensis* and *H. conradiae*, as suggested by Gamisans *et al.* (2011) seems not to be exclusive, as *H. conradiae* is also one of the hostplants of *P. bellieri*. This complex relationship between these restricted endemic species needs further investigation.

The new locality in the Fango valley for *P. coridon nufrellensis* is situated below 1000 m in a habitat completely different from the known places in the Spasimata valley. We conclude that the distribution area of this subspecies is wider than previously thought, because both the foodplant (*H. conradiae*) and main

nectar source (*D. corsicum*) are quite widespread and in various habitats.

The distribution of other newly discovered nectar sources, *Erica terminalis*, *Armeria leucocephala* and *Allium schoenoprasum* and that of the foodplant *H. conradiae*, at different altitudes needs further investigation in order to find possible new localities for *P. coridon nufrellensis*, perhaps in other poorly explored Corsican high mountains. It seems essential to have the foodplant and at least one nectaring plant in the same area, but adjacent contact zones would be interesting to investigate.

Finally, alterations to the habitat brought about by an increase in tourism and global climate change could be detrimental to the survival of both adult and early stages of this unique subspecies, and a broader habitat range with some hardly accessible localities would be beneficial for its long time survival. These factors should be addressed by European and national governments charged with conservation, in order to develop thoughtful conservation strategies to ensure the survival of this elusive lycaenid and its endemic hostplant.

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