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REPORT OF THE LARGEST KNOWN OCCURRENCE OF MORPHOLOGICAL ANOMALIES IN A POPULATION OF AN ENDEMIC GROUND BEETLE: *CARABUS* (*CHAETOCARABUS*) *LEFEBVREI LEFEBVREI* DEJEAN, 1826

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ABSTRACT. We report the largest known occurrence of morphological anomalies in adults of a species of ground beetle. Specimens of a population of *Carabus* (*Chaetocarabus*) *lefebvrei lefebvrei* Dejean, 1826 were collected using pitfall traps in 2019 in a stand dominated by chestnut (*Castanea sativa*), near a protected area in Northeastern Sicily. More than 13% of the trapped specimens showed morphological anomalies in the pronotum or elytra. The anomalies ranged from the presence of tumours, missing or fused elytral striae, and deformed elytra to incomplete pronotum and difference in shapes of legs. These anomalies were compared with those of eight specimens collected in Eastern Sicily (Etna, Nebrodi and Peloritani). We recorded slightly more females than males with anomalies. The occurrence of so many morphological anomalies in adult beetles near a protected area is alarming.

1. Introduction

Chaetocarabus lineage has given rise to allopathic vicariants in disjointed European areas, such as *Carabus intricatus* Linnaeus, 1761 which is widespread along the Alpine and Pre-alpine chain, and southern European ones, such as *C. lefebvrei* Dejean, 1826 (present in central-southern Italy and Sicilian Apennines), *C. arcadicus* Gistl, 1850, *C. merlini* Schaum, 1861 and *C. krueperi* Reitter, 1896 in the southern Balkan Peninsula (Brandmayr *et al.* 2005). *Carabus (Chaetocarabus) lefebvrei bayardi* Solier, 1835 lives in the central-southern Italy while *Carabus (Chaetocarabus) lefebvrei lefebvrei* Dejean, 1826 is an endemic subspecies from Southern Calabria (Aspromonte) and Northern Sicily (Casale *et al.* 1982; Vigna Taglianti 2015). *C. lefebvrei* is a spring reproducing species, active in beech, chestnut, oak and pine forests, from April to September (Thiele 1977; Turin *et al.* 2003). This species hibernates as adult, often in numbers, under stones or in rotten decomposed trunks (Baviera, pers. obs.). The larvae, hatched from eggs laid in humus-rich soil, grow from June to August (Thiele 1977; Turin *et al.* 2003). Pupae moult in a cell dug, in the ground, by the 3rd instar larva (Giglio *et al.* 2009). The long time hypothesised, trophic specialisation on snails, for this species, was recently demonstrated (Giglio *et al.*

2012). Morphological anomalies in *Carabus* were quite rarely recorded, only few data were recorded by: Mocquerys (1880), Baudi (1889), Failla-Tedaldi (1897), Chinaglia (1911a,b), Dumont (1915), Ortuño and Vique (2007), Ferreira (2008), and Ortuño and Ramos Abuín (2008), with cases associated to the legs.

Cases of anomalies of the antennae, pronotum, elytral shape or position, and trends of the elytral striae have been reported (Mocquerys 1880; Della Beffa 1910; Chinaglia 1911a,b; Dumont 1915; Ćurčić *et al.* 2006; Ortuño and Vique 2007; Ortuño and Ramos Abuín 2008), pronotum (Ortuño and Vique 2007; Gandhi and Herms 2008) or to the elytral shape or position and trend of elytral striae (Mocquerys 1880; Chinaglia 1911a; Jaworska 1994; Ćurčić *et al.* 2006; Jaworska and Wiacek 2006; Ortuño and Vique 2007; Gandhi and Herms 2008). Head changes have rarely been recorded in *C. lefebvrei* until now (Mocquerys 1880; Ćurčić *et al.* 2006; Ortuño and Vique 2007; Gandhi and Herms 2008; Ferreira 2011) like in buccal pieces (Mocquerys 1880; Ćurčić *et al.* 2006). In *C. lefebrei* very few morphological anomalies were recorded until now. Failla Tedaldi in 1897 collected two females of *C. lefebrei lefebrei* with leg anomalies in July 1894 in Madonie mounts (Palermo) (Failla-Tedaldi 1897). Chinaglia collected a female of *C. lefebrei bayardi* with leg anomalies in Naples (Camaldoli) in 1901 (Chinaglia 1911a,b). We describe morphological anomalies in specimens of a population of *C. lefebvrei lefebvrei* recorded in a site of eastern Sicily and compare them with anomalies in individuals of the same species collected at other forests sites in Eastern Sicily.

2. Materials and methods

Sampling were made in five sites in Peloritani Mounts. We installed two 500 ml pitfall traps in each site, one with vinegar and salt, one with water and salt. The samples were collected from May to September 2019, with traps replaced every 15 days. The study sites were in a strongly fragmented woody area among Itala village and the Natura 2000 site (code ITA030010) "Natural Reserve of Fiumedinisi and Monte Scuderi", a 72 km2 (7198.00 ha) protected area. This protected area contains 19 types of habitats of the Habitats Directive and preserves one species of fern, two species of flowering plants, and fifteen species of animals included in Nature directives. Specimens of *C. lefebvrei lefebvrei* were collected only in one site: Tavoliere at about 250 m a.s.l. (Croce, Itala, Messina, 38° 2'42.43"N 15°25'38.14"E). The arboreal vegetation of this locality is characterised by very old chestnut trees (*Castanea sativa* Mill., 1768), along with oak (*Quercus* gr. *pubescens* Willd., 1796), hackberry (*Celtis australis* L., 1753), black mulberry (*Morus nigra* L., 1758), and walnut trees (*Juglans regia* L., 1753). The other studied specimens, with morphological anomalies, were obtained during the previous studies for obtaining a better understanding of the taxonomy and distribution of Sicilian Coleoptera (Baviera 2007, 2008, 2010; Baviera and Liberti 2010; Baviera and Magnano 2010; Bellò and Baviera 2011; Magrini and Baviera 2011; Toševski *et al.* 2011, 2014; Baviera 2015; Toševski *et al.* 2015; Bellò *et al.* 2017, 2019) and distribution (Stroscio *et al.* 2011a,b; Baviera and Audisio 2014; Baviera and Biondi 2015; Baviera *et al.* 2017; Baviera and Platia 2018; Baviera and Vienna 2019; Baviera and Caldara 2020). All specimens were collected in eastern Sicily: Nebrodi Mounts (Messina: Soro Mount), Eastern slope of Etna Mount (Catania, Milo: Cerrita Wood) and North-Eastern slope of Peloritani Mounts (Messina: Salice and Massa San Giorgio). The

images of specimens were acquired using the following instruments: Zeiss Stemi 2000C stereoscope, Nikon D3500 camera, elaborated with DigiCam Control v2.1.2 and Helicon Focus 7, and Adobe Photoshop CS6 graphic editor. All the specimens are preserved in first author collection (BC) and will be transferred to the entomological collection of Museo Cambria, Messina University, Italy.

3. Results

List of speciments

A total of 81 specimens were collected at Tavoliere, eleven (five males and six females) of these denoted morphological anomalies (13.6% of the total specimens collected). Seventy-one of the 81 specimens (87.65%) were caught in the traps containing vinegar and salt, and the other 10 (12.45%) were caught in the traps containing water and salt. The morphological anomalies were also compared with the anomalies observed in others eight specimens (four males and four females) collected in eastern Sicily by the first author.

Description of the observed morphological anomalies.

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Collection data – Messina: Peloritani, Tavoliere, pitfall traps, 14-29.VI.2019, Micali R. leg.

Description of the anomaly (Fig. I 1) – The specimen had an hemiatrophy of the right basal part of the pronotum with the lack of the right rear corner. This is associated with an hemibrachelytry of the right elytra which is corrugated, deeply embedded in the basal part and slightly raised in the distal half.

Collection data – Messina: Peloritani, Tavoliere, pitfall traps, 14-29.VI.2019, Micali R. leg.

Description of the anomaly (Figs. I 2-3) – The specimen shows an enlarged left median tibia without bristles with a constriction in the proximal portion. The left median tarsus shows a regularly conformed tarsomere.

Collection data – Messina: Peloritani, Tavoliere, pitfall traps, 13-28.VIII.2019, Micali R. leg.

Description of the anomaly (Fig. I 4) – The specimen had a tumor protuberance on the right posterior part of the pronotum.

Collection data – Messina: Peloritani, Tavoliere, pitfall traps, 30.V- 14.VI.2019, Micali R. leg.

Description of the anomaly (Fig. II 1) – The specimen has the right elytra longer than the left and superimposed on the right at the apex. Left elytra show an elongated concavity at the base.

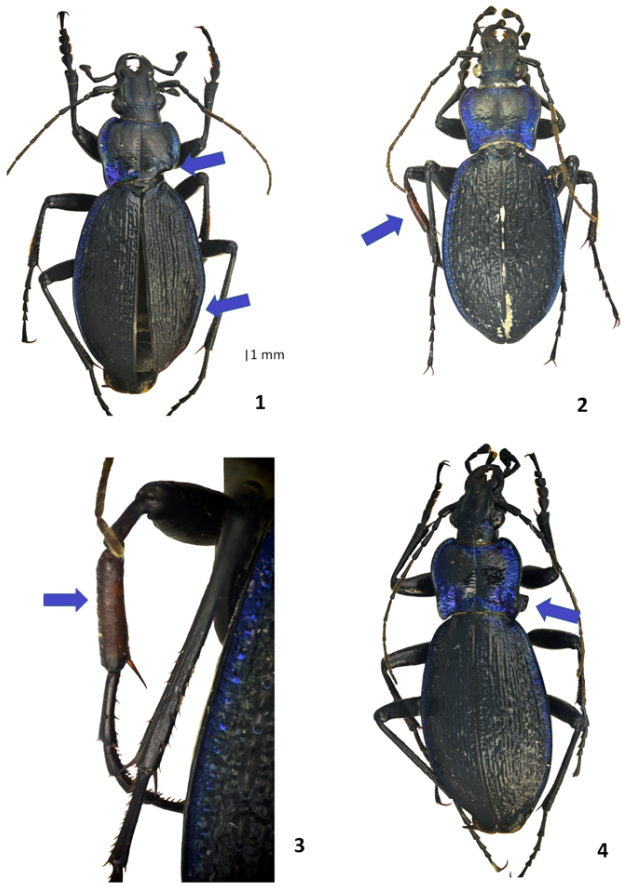

FIGURE I. *Carabus lefebvrei lefebvrei*: 1. Messina - Peloritani, Tavoliere, pitfall traps, 14-29.VI.2019, Micali R. Leg.; 2-3. Messina - Peloritani, Tavoliere, pitfall traps, 14-29.VI.2019, Micali R. Leg.; 4. Messina - Peloritani, Tavoliere, pitfall traps, 13-28.VIII.2019, Micali R. Leg.

Collection data – Messina: Peloritani, Tavoliere, pitfall traps, 29.VI-14/.VII.2019, Micali R. leg.

Description of the anomaly (Fig. II 2) – The specimen had the left elytra corrugated and raised in the distal half. At the apex, the left elytra is thus shorter than the right.

Collection data – Catania: Etna Est, Bosco Cerrita, 1500 m a.s.l., pitfall traps sub *Betula aetnensis*, IX.2002-16.V.2003, Baviera C. leg.

Description of the anomaly (Fig. II 3) – The specimen had both the elytra slightly raised at the center.

Collection data – Messina: Nebrodi, Monte Soro, 1500 m a.s.l., pitfall traps, 21.V-21.VI.2001; Baviera C. leg.

Description of the anomaly (Fig. II 4) – The specimen had both elytra deeply sunken immediately after the middle. At the suture the elytra touch each other only at the base and after the middle.

Collection data – Messina: Nebrodi, Monte Soro, 1500 m a.s.l., pitfall traps, 21.V-21.VI.2001; Baviera C. leg.

Description of the anomaly (Fig. III 1) – The specimen had both elytra slightly hollowed immediately after the middle. Elytra are slightly dehiscent at the apex. Elytral apical third is rugose and without striae. The pronotum had an elongated concavity on the left side.

Collection data – Messina: Nebrodi, Monte Soro, 1500 m a.s.l., pitfall traps, 21.V-21.VI.2001; Baviera C. leg.

Description of the anomaly (Fig. III 2) – The specimen had two concavities in the pronotum, one less wide on the left front and another, wider, on the right rear.

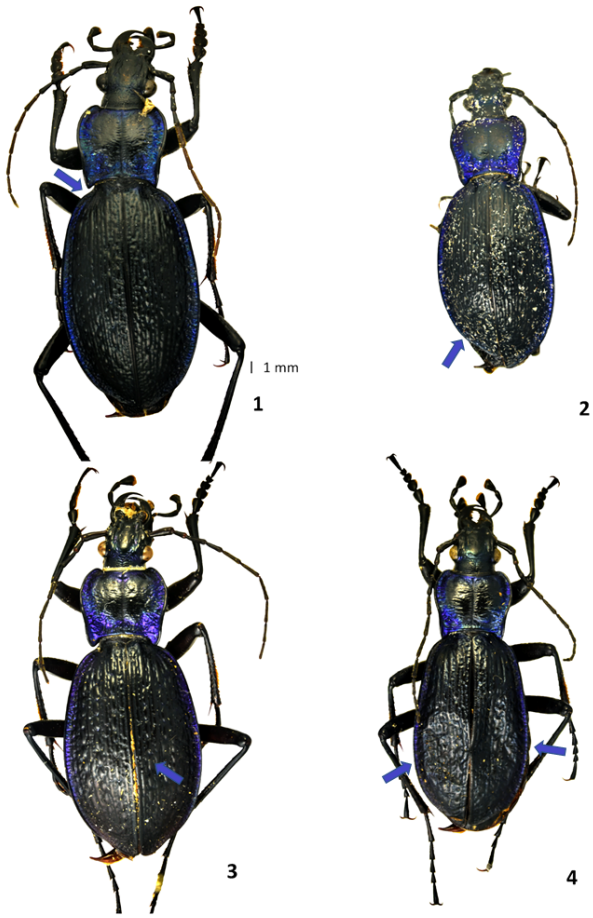

FIGURE II. *Carabus lefebvrei lefebvrei*: 1. Messina - Peloritani, Tavoliere, pitfall traps, 30.V- 14.VI.2019, Micali R. Leg.; 2. Messina - Peloritani, Tavoliere, pitfall traps, 29.VI-14/.VII.2019, Micali R. Leg.; 3. Catania - Etna Est, Bosco Cerrita, 1500 m a.s.l., pitfall traps sub *Betula aetnensis*, IX-16.V.2003, Baviera C. Leg.; 4. Messina - Nebrodi, Monte Soro, 1500 m a.s.l., pitfall traps, 21.V- 21.VI.2001; Baviera C. Leg.

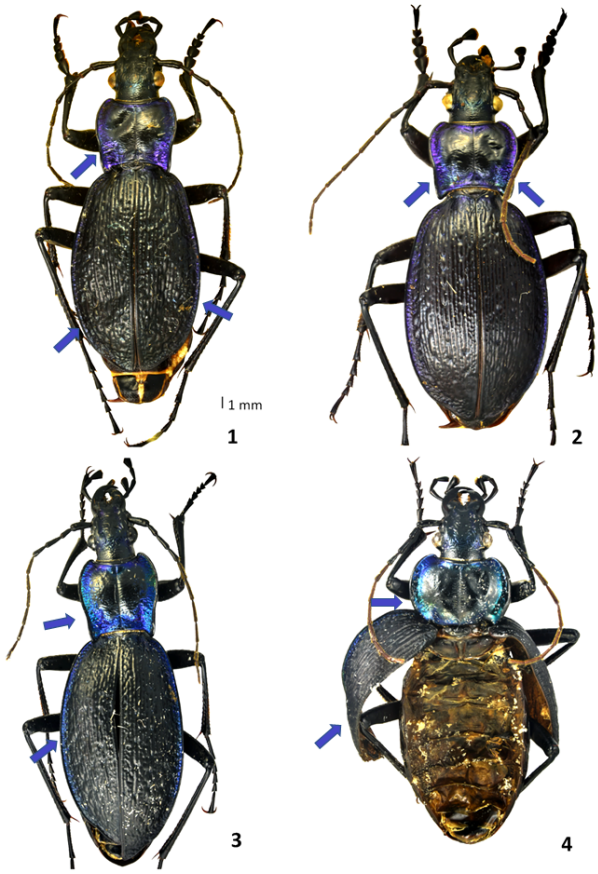

FIGURE III. *Carabus lefebvrei lefebvrei*: 1. Messina - Nebrodi, Monte Soro, 1500 m a.s.l., pitfall traps, 21.V- 21.VI.2001, Baviera C. Leg.; 2. Messina - Nebrodi, Monte Soro, 1500 m a.s.l., pitfall traps, 21.V- 21.VI.2001; Baviera C. Leg.; 3. Messina - Peloritani, Tavoliere, pitfall traps, 14-29.VI.2019, Micali R. Leg.; 4. Messina-Peloritani, Tavoliere, pitfall traps, 29.VII-13.VIII,2019, Micali R. Leg.;

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Collection data – Messina: Peloritani, Tavoliere, pitfall traps, 14-29.VI.2019, Micali R. leg.

Description of the anomaly (Fig. III 3) – The specimen had the left elytra strongly raised before the middle with the apex covered by the right elytra before the apex. The pronotum is sub-square in shape (PL / PW = 1.24)

Collection data – Messina: Peloritani, Tavoliere, pitfall traps, 29.VII-13.VIII,2019, Micali R. leg.

Description of the anomaly (Fig. III 4) – The specimen shows the elytra totally open with the abdomen completely uncovered. The left one is more widely arched above the middle and rear legs, while the right is closely to the body between the middle and the rear legs. The pronotum is distinctly transverse for the species (PL / PW = 1.63) and had many furrows.

Collection data – Messina: Peloritani, Tavoliere, pitfall traps, 23-28.VIII,2019, Micali R. leg.

Description of the anomaly (Fig. IV 1) – The specimen had both elytra folded, twisted, dehiscent at apex and joined to the suture only at the base near the scutellum.

Collection data – Messina: Peloritani, Tavoliere, pitfall traps, 23-28.VIII,2019, Micali R. leg.

Description of the anomaly (Fig. IV 2) – The specimen had, near the proximal half of the right elytra, from the ninth to the eleventh interstrie, the chains detected and significantly enlarged.

Collection data – Messina: Peloritani, Tavoliere, pitfall traps, 23-28.VIII,2019, Micali R. leg.

Description of the anomaly (Fig. IV 3) – The specimen had the elytra slightly dehiscent to the sutural middle that becomes separated at the apex.

Collection data – Messina: Peloritani, Tavoliere, pitfall traps, 29.VII-13.VIII,2019, Micali R. leg.

Description of the anomaly (Fig. IV 4) – The specimen had the left elytra proximal depressed. The catenulation of that portion of the elytra presents the striae detected and enlarged until they overlap.

Collection data – Catania: Etna Mount Est slope, Bosco Cerrita, 1500 m a.s.l., pitfall traps sub *Betula aetnensis*, 16.V-10.VII.2002, Baviera C. leg.

Description of the anomaly (Fig. V 1) – The specimen had an anomalous pronotum, sub-square in shape (PL / PW = 1.26) and with a slightly sinuated margins different from the clearly sinuated shape as normally observable in the species. The elytra appear very

weakly dehiscent along the suture.

Collection data – Messina: Peloritani, Messina, Peloritani, Salice, 500 m a.s.l., pitfall traps sub *Quercus ilex*, 13-31.VIII.2004, Baviera C. leg.

Description of the anomaly (Fig. V 2) – The specimen shows a depression in right elytra apex.

Collection data – Messina: Peloritani, Massa San Giovanni, 500 m a.s.l., 05.IV.1998, Baviera C. leg.

Description of the anomaly (Fig. V 3) – The specimen presents the elytra dehiscent at the apex. The right elytra longer than the left one.

Collection data – Messina: Nebrodi, Monte Soro, 1500 m a.s.l., 25.VI- 28.VIII.2001, pitfall traps sub *Fagus sylvatica*, Baviera C. leg.

Description of the anomaly (Fig. V 4) – The specimen had the left elytra curved and widely arched above the middle as to leave the abdomen completely uncovered. The pronotum has a concavity at the right lateral portion.

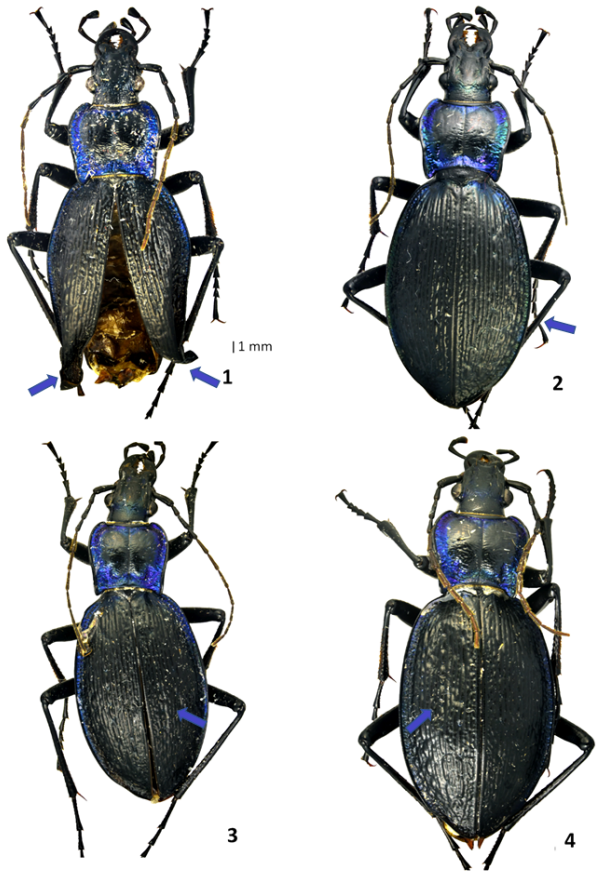

FIGURE IV. *Carabus lefebvrei lefebvrei*: 1. Messina - Peloritani, Tavoliere, pitfall traps, 23-28.VIII,2019, Micali R. Leg.; 2. Messina-Peloritani, Tavoliere, pitfall traps, 23-28.VIII,2019, Micali R. Leg.; 3. Messina - Peloritani, Tavoliere, pitfall traps, 23-28.VIII,2019, Micali R. Leg.; 4. Messina - Peloritani, Tavoliere, pitfall traps, 29.VII-13.VIII,2019, Micali R. Leg.

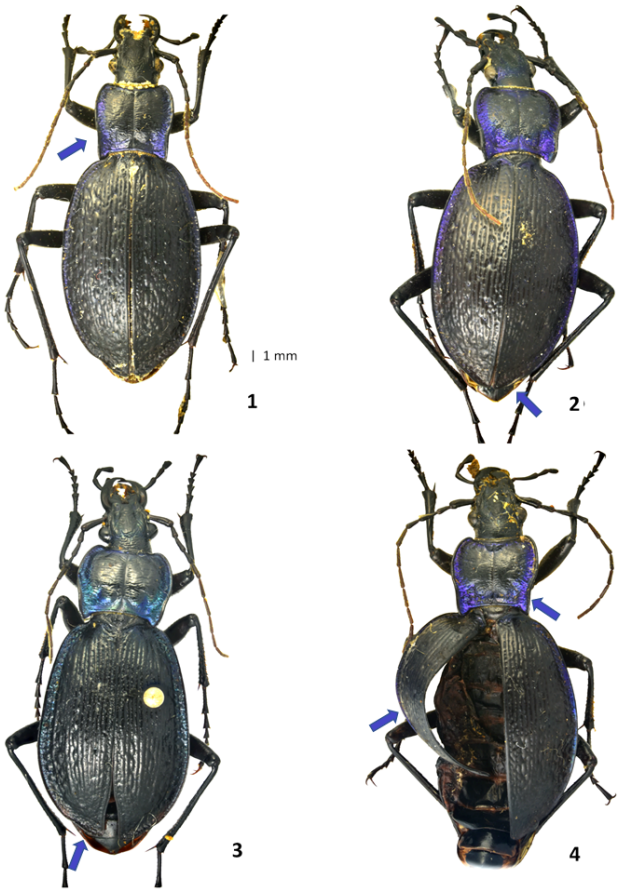

FIGURE V. *Carabus lefebvrei lefebvrei*: 1. Catania - Etna Est, Bosco Cerrita, 1500 m a.s.l., pitfall traps sub *Betula aetnensis*, 16.V-10.VII.2002, Baviera C. Leg.; 2. Messina - Peloritani, Salice, 500 m a.s.l., pitfall traps sub *Quercus ilex*, 13-31.VIII.2004, Baviera C. Leg.; 3. Messina - Peloritani, Massa San Giovanni, 500 m a.s.l., 05.IV.1998, Baviera C. Leg.; 4. Messina - Nebrodi, Monte Soro, 1500 m a.s.l., 25.VI- 28.VIII.2001, pitfall traps sub *Fagus sylvatica*, Baviera C. Leg.

TABLE 1. Morphological anomalies of studied specimens.

SITE	FIG.	SEX	ELYTRAL ANOMALIES	PRONOTAL ANOMALIES	APPENDICES ANOMALIES
Peloritani Tavoliere (Messina)	I.1	Male	hemibrachelytry of the right elytra	hemiatrophy of the right basal part, with the lack of the right rear corner	
Peloritani Tavoliere (Messina)	I.2-3	Male			enlarged left median tibia without bristles
Peloritani Tavoliere (Messina)	I.4	Male		tumor protuberance on the right posterior part	
Peloritani Tavoliere (Messina)	II.1	Male	right elytra longer than the left which show an elongated concavity at the base		
Peloritani Tavoliere (Messina)	II.2	Male	left elytra shorter than the right, corrugated and raised in the distal half		
Etna Mount (Catania)	II.3	Male	both elytra slightly raised at the center		
Nebrodi Soro Mount (Messina)	II.4	Male	both elytra deeply sunken immediately after the middle		
Nebrodi Soro Mount (Messina)	III.1	Male	both elytra with a small concavity immediately after the middle	an elongated concavity on the left side	
Nebrodi Soro Mount (Messina)	III.2	Male		two different concavities on sides	
Peloritani Tavoliere (Messina)	III.3	Female	left elytra strongly raised before the middle	pronotums sub-square in shape	

Continued on next page

Table 1 – *Continued from previous page*

SITE	FIG.	SEX	ELYTRAL ANOMALIES	PRONOTAL ANOMALIES	APPENDICES ANOMALIES
Peloritani Tavoliere (Messina)	III.4	Female	elytra totally open with the abdomen completely uncovered	pronotum is distinctly transverse for the species with many furrows	
Peloritani Tavoliere (Messina)	IV.1	Female	both eltra folded, twisted, dehiscent at apex		
Peloritani Tavoliere (Messina)	IV.2	Female	right elytra with chains detected and significantly enlarged		
Peloritani Tavoliere (Messina)	IV.3	Female	elytra slightly dehiscent to the sutural middle, separated at the apex		
Peloritani Tavoliere (Messina)	IV.4	Female	left elytra proximal depressed		
Etna Mount (Catania)	V.1	Female		anomalous pronotum, sub-square in shape	
Peloritani Salice (Messina)	V.2	Female	depression in right elytra apex		
Peloritani Masse (Messina)	V.3	Female	elytra weekly dehiscent at the suture, the right longer than the left		
Nebrodi Soro Mount (Messina)	V.4	Female	left elytra curved and widely arched above the middle as to leave the abdomen completely uncovered	a concavity at the right lateral portion	

4. Conclusions and final remarks

We examined 19 specimens of *C. lefebvrei lefebvrei* with morphological anomalies all collected in eastern Sicily. Eleven speciments (13.6% of the total specimens collected) belonged to the same population in a restricted area of Peloritani Mounts (Messina: Itala, Tavoliere). This study on morphological anomalies is thus the largest for a single species of *Carabus*. No anomalies were found in other species of Carabidae nor other Coleoptera collected at the same site and other sites during this study. As above reported, only three specimens of *Carabus lefebvrei* with morphological anomalies have previously been recorded. Two specimens of *C. lefebvrei lefebvrei* from Madonie and one of *C. lefebvrei bayardi* from Naples, all with anomalies in the legs. Considering the affected body parts in all the specimens of our study, the elytra had the most anomalies (see Table 1), in contrast with previous data of Balazuc (1948) which reported twice the number of aberrations of appendages than those of the elytra. Moreover the elytral anomalies were not associated with appendages such as antennae, mouthparts, or legs but only with pronotal anomalies. Only one specimen had pronotal hemiatrophy as observed in other Carabidae species by Balazuc (1948, 1969) and Ortuño and Peláez Aller (2004), and only another specimen had a tumour on its pronotum. Only one of the studied specimens had an enlarged tibia, the only observed anomaly in appendices. Morphological anomalies have rarely been reported in numerous specimens of the same population of a single species of carabids, particularly of the genus *Carabus*. Studies of *Carabus* morphological anomalies in Europe reported *Carabus cancellatus* Illiger, 1798 (Jaworska and Wiacek 2006) with malformations in three (two males and one female) of 187 specimens (1.6%) and *Carabus (Eucarabus) ullrichi* Germar, 1824 (Rýznar and Martiš 2014) with malformations of the elytral sculpture in three males of 132 specimens (2.27%).

Other studies of malformations in different species of various carabids have reported very low prevalence of malformations, as in (Ortuño and Vique 2007), which reported a prevalence of 0.034%. Gandhi and Herms (2008) found the highest prevalence of carabid specimens with morphological anomalies at a single site, with 109 malformed adults belonging to six genera and 12 species collected in the USA, representing 9.5% of all catches. This high prevalence involved numerous species and sites in a territory with an area of about 6000 km^2, very much larger than the area investigated in the our study. The similarities with our study were: most of the anomalies were in the elytra; anomalies were slightly less common in males than females; the sampling periods were from May to September; with traps replaced every 15 days. Gandhi and Herms (2008) provided no verified explanations for the abnormalities. In study on morphological anomalies on ground beetles, the hypothesized causing agent have been associated to: water pollution, acid rain, nitrogen deposition, genetic inbreeding due to habitat isolation and fragmentation, or the prevalence of parasites, as reported or hypothesised for other beetles (Morgan *et al.* 1986; Bengtsson *et al.* 1988; Christopher *et al.* 1996; Eeva *et al.* 1998; Gandhi and Herms 2008).

The cause of so many morphological anomalies in our population of *C. lefebvrei lefebvrei* remains unknown. We may hypothesise an effect of genetic inbreeding due to habitat isolation and fragmentation to account for some of the anomalies. Also, the unusually dry soil in the summer in this low-altitude Mediterranean forest could account for the elytral and pronotum deformations (but not the tumours or the malformed legs). Malformations

of insects after the pupal moult at too cold or too hot a temperature have been known for a long time (Balazuc (1948) and references there in). We cannot currently identify the causes of the anomalies, but we can nevertheless note that some morphological anomalies in carabid beetles can indicate changes in environmental conditions. At least some of the anomalies may be linked to environmental pollutants, even though no anomalies were found in a population of *C. lefebvrei* collected near a waste area in Calabria, with the aim of quantifying the consumption and metabolism of heavy metals. (Talarico *et al.* 2014). More studies on this morphological anomalies should identify factors responsible of the different anomalies for better define environment related alterations. The identification of the causes of the anomalies observed in this endemic subspecies would also be very important for preventing the possible reduction of its populations.

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