Acta Zool. Fennica 209: 255–262 ISBN 951-9481-54-0 ISSN 0001-7299
Helsinki 6 May 1998

Discrimination of the West-Palaearctic *Chrysoperla* Steinmann species of the *carnea* Stephens group by means of claw morphology (Neuroptera, Chrysopidae)

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Received 8 August 1997

A morphological study of the claws of a number of specimens of *Chrysoperla* Steinmann, from different parts of Europe, was carried out to find new characters useful for their identification, especially those of some sibling species constituting the *carnea* Stephens-complex. A discriminative factorial analysis of length, basal enlargement, tilting, and opening of claws produced usable results. The specimens from Greece constitute a distinct morph whose actual status is not yet established.

1. Introduction

The "common green lacewing" *Chrysoperla carnea* (Stephens) *sensu lato,* is probably one of the most studied chrysopid species, due to its almost worldwide distribution, its abundance, its adaptability to inhabited and cultivated areas, and the easiness of artificial production in insectaria. In the Palaearctic zone, it constitutes in many places a large part of the chrysopid populations, *sensu* Barbaut (1992). It was long considered highly polymorphic, as reflected by the numbers of varieties, sub-species, and sibling species described, e.g. 29 in Navás (1915), 16 in Aspöck *et al.* (1980), 14 in Brooks (1994), and 80 in P. Duelli (unpubl.). It was also often considered cosmopolitan and eurybiote: outside the northern temperate zones which it fully colonizes, it is able to reach northern areas up to the Polar Circle (Greve 1984), and tropical areas down to Sudan (Ohm & Hölzel 1992).

Paradoxically, *Ch. carnea* also is the Holarctic species whose taxonomic status is most problematic (Brooks 1994, Aspöck & Hölzel 1996). Recent work gave it new perspective. Modulations of courtship songs (Henry 1985, Wells & Henry

1992), morphology of larvae and adults (Thierry *et al.* 1992), enzymatic polymorphism (Cianchi & Bullini 1992), and ecophysiological variability (Thierry *et al.* 1994) lead to the conclusion that both in Europe and North America (Tauber & Tauber 1985) the so-called *Ch. carnea auctorum* is a complex composed of several sibling species.

At least six *Chrysoperla* Steinmann species of the *carnea* group *sensu* Brooks (1994) occur in western Europe. Following the nomenclature from Leraut (1991, 1992), they are, in chronological order of original descriptions: *carnea* Stephens, 1836, *lucasina* Lacroix, 1912, *kolthoffi* Navás, 1927, *renoni* Lacroix, 1933, *mediterranea* Hölzel, 1972 and *ankylopteryformis* Monserrat & Díaz-Aranda, 1989. The male and female genital structures are homogeneous within the group and do not permit easy and safe discrimination of the sibling species. These are, however, possibly separable by means of some characters appearing in the key given in the Appendix, partly following Thierry *et al.* (1992). Unfortunately some discriminant characters used in separating *carnea*, *lucasina* and *kolthoffi* often are unapplicable to old collection specimens, both pinned and stored in alcohol for a long time. Because of that we have looked for a complementary character, whose study remains possible regardless of the age and conservation of the material.

Previously, Tjeder (1966), and Yang and Yang (1989) used the claw shape to discriminate several *Chrysoperla* species from southern Africa and China, like Brooks (1994) in his determination key to *Chrysoperla* of the World, and Brooks and Barnard (1990) in their cladistic works. They all consider a triangular basal enlargement of the claw a plesiomorphic character, even if it has evolved in different ways. The basal enlargement is quadrangular in *Ch. carnea*, *Ch. lucasina* and *Ch. kolthoffi* (Fig. 4A–C) whereas it is sub-triangular in *Ch. renoni*, *Ch. mediterranea* (Fig. 4E) and *Ch. ankylopteryformis*.

A morphological study of the claws carried out on Palaearctic *Chrysoperla* by means of a discriminant factorial analysis is presented here. It involves specimens which unambiguously are referable to known species, originating from various parts of Europe. We also have studied an homogeneous population from Greece, and some specimens atypical or not easily assignable by means of the classical characters. However, *Ch. renoni* and/?or *Ch. ankylopteryformis* were not included due to the lack of a sufficient number of specimens for statistical analysis.

2. Material and methods

2.1. Material

Adult chrysopids (n = 103) were caught from June to November, by various methods, in various places in Europe. They were kept in a 9/1 mixture of 60% ethanol and glycerol. Voucher specimens, five males and five females, of each species were deposited in the collection of the Université Catholique de l'Ouest, IRFA, F-49000 Angers, France. Below are the techniques, localities, biotopes and dates of collection; altitudes were noted only when higher than 500 m a.s.l.

2.1.1. Light trap

- Skakavista, near Kjustendil, Bulgaria. Alt. 600 m. 42°17´N 22°41´E. 1.VIII.1980.
- Belasitsa, near Petric, Bulgaria. Alt. 700 m. 41°24´N 23°13´E. 25–26.VI.1981.
- Sofia, Bulgaria, 42°41´N 23°19´E. 26.VI.1982.
- 3 km south of Valencia, Spain. Conifer wood, near the seashore. 41°21´N 0°25´E. 31.VII.1987.
- Rochecorbon, France. Urban area. 47°23´N 0°41´E. 20.VIII.1995.

2.1.2. McPhail trap

- Scala, Aguistri Island (Saroniki gulf), Greece. Olive grove. 35°30´N 23°21´E. 17.VIII.1976.

2.1.3. Hand net, into the canopy of isolated deciduous trees

- L'Aquila, Italy. Cultivated area. 42°22´N 13°22´E. 10.VI.1989.
- Brindisi, Italy. Cultivated area. 40°35´N 17°40´E. 10.VII.1989.
- Rome, Italy. Suburbs, on oak. 41°54´N 12°29´E. 9.X.1990.
- Gyor, Hungary. Cultivated area. 47°27´N 17°38´E. 17.VII.1992.
- St-Rémy-sur-Durolle, France. Meadow. 45°53´N 3°36´E. 23.VII.1994.
- La Rochelle, France. Atlantic seashore. 46°10´N 1°09´W. 13.XI.1994.

— Rybina, near Gdansk, Poland. Marshland, on alder. 54°12´N 18°30´E. 2.VII.1995.
— Wieliczka, near Krakovie, Poland. Suburban fallow land. 50°03´N 19°58´E. 7.VIII.1995.

2.1.4. Hand net, into the canopy of deciduous trees, wood edges

— 2 km south-east of Trieste, Italy. 45°30´N 13°50´E. Collection date unknown.
— Niechorze, near Trzebiatow, Poland. Seashore. 54°04´N 15°14´E. 29.VII.1995.

2.1.5. Underwood, in dry leaves hung on twigs between 1 and 3 m high

— Vouvray, France. Banks of the Loire. 47°23´N 0°41´E. 20.VIII.1995.

2.1.6. Partially unknown sampling conditions

— Porto, Portugal. 41°11´N 8°36´W. Collection date unknown.
— Lisbon, Portugal. 38°43´N 9°08´W. 4.VII.1994.

2.2. Morphometry

Each pretarsus of the right hind leg of each specimen was removed and mounted on slide, so that the two claws were seen in external profile. They were measured with an optical microscope at × 500 magnification. The following reference marks were located (Fig. 1):

— the claw apex A,
— the basal point of the claw indentation B,
— the two articulation condyles C_1 and C_2,
— the distal tip of the basal enlargement D, and
— the point M, where a line perpendicular to the line C_1–C_2 at its midpoint cuts the base of the claw.

From these points the following parameters were calculated:

— the lenght of the claw, given by A–B,
— the basal enlargement, given by B–D,
— the tilting of the claw, given by the angle α, and
— the opening of the claw, given by the angle β.

2.3. Data analysis

A pair of measurements was provided by the two pretarsal claws of each specimen. The data used in the multivariate

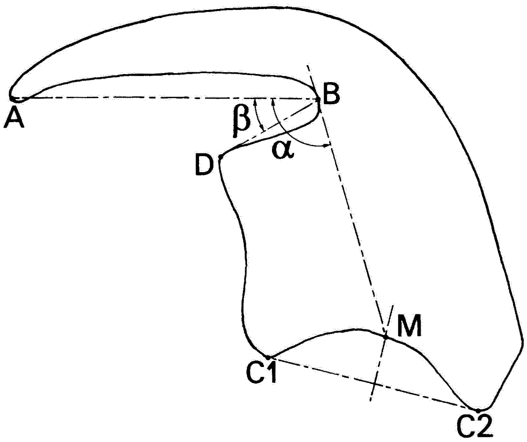

Fig. 1. Outline of a claw of *Chrysoperla* (here *Ch. lucasina* male, Rochecorbon, France, 20.VIII.1995) showing the measurements used.

analysis were the average values of these paired measurements, because a preliminary investigation carried out by means of pair-sample Student's *t*-test did not show any asymmetry in the two claws of the individuals (see Section 3.2.).

Another exploratory data analysis led us to transform the recorded measurements. The data matrix corresponding to five categories previously defined has been treated by centred reduced principal component analysis (PCA). The aim of the PCA is to reduce the space necessary to represent the multivariate data while retaining most of the variability. PCA builds up new variables from linear combinations of primary data, establishing the factorial axes (F_1, F_2, F_3 ...), allowing the representation of each individual in the planes (F_1*F_2, F_2*F_3 ...). This analysis results in a classification into five groups based on the PCA factorial coordinates by Roux's (1985) dynamic cloud method (DCM). Finally the total data set, including the atypical specimens, was analysed by discrimant function analysis (DFA). DFA tests the relevance of an *a priori* classification using quantitative variables. This analysis results in a recombination of the variables, optimizing the discrimination between each group of individuals; it then allows an assignment of the misclassified and/or undetermined specimens. STAT-ITCF software (1988) was used.

3. Results and discussion

3.1. Distribution of the specimens

Table 1 gives the numbers and the sex of the specimens assigned to six groups according to the classical characters. Four of them correspond to known species: *Ch. carnea s. str.*, *Ch. kolthoffi*, *Ch. lucasina* and *Ch. mediterranea*. The fifth group consists of the specimens collected in Greece. The sixth group contains some specimens caught in Europe sympatrically with the other above-mentioned species, but not directly assignable to them. This last group includes five specimens marked by ringed numbers in Fig. 2. Two males (nos. 1 and 2) coming from Porto (Portugal) and two females (nos. 3 and 4), coming from Rochecorbon (France) and from Krakovie (Poland), respectively, are close to *Ch. carnea s. str.*, but possess several brown setae on the abdominal sternites like *Ch. kolthoffi*. In addition, the two first specimens have a slight brown sclerotised stripe between the proximal abdominal tergites and sternites, like *Ch. lucasina*. The three other specimens, all females collected in Italy at Rome (no. 5) and Trieste (nos. 6 and 7), cannot be referred to any known species; they have numerous black setae on the pronotum and on the abdominal sternites, and they exhibit a conspicuous basal enlargement in the claws.

Table 1. Males (m) and females (f) analysed, assigned to known species (*Chrysoperla carnea, kolthoffi, lucasina, mediterranea*), Greek morph and atypical specimens.

Locality	*car* m/f	*kol* m/f	*luc* m/f	*med* m/f	Greek m/f	atyp. m/f
Aquila			–/2			
Belasitsa	–/1		2/1			
Brindisi			2/–			
Gyor		2/2	–/1			
La Rochelle			3/2			
Lisbon		2/–				
Niechorze		3/2				
Porto	1/–	3/1	3/–			2/–
Rochecorbon		3/3	1/–			–/1
Rome	3/4		–/1			–/1
Rybina		1/2				
Scala					3/7	
Skakavista		–/1				
Sofia	–/1	2/1	1/–			
St-Rémy			–/2			
Trieste						–/2
Valencia			–/2	5/4		
Vouvray	4/3					
Wieliczka	1/–	3/2	1/2			–/1

3.2. Preliminary analysis

As mentioned above (Section 2.2.), the *t*-test comparison of means conducted on the raw data (AB, BD, α and β) showed no significant difference between measurements of the two claws of the same individual (AB: $t = 1.29$, $P = 0.19$; BD: $t = 1.27$, $P = 0.20$; α: $t = 1.10$, $P = 0.23$; β: $t = 1.85$, $P = 0.06$; d.f. = 95 in each test). Therefore, the average values for the paired data were considered suitable.

Inspection of the raw data led us to log-transform the mean distances, following the suggestion by Jolicœur (1966), and to use the ratio of transformed values. The resulting variables submitted to analysis were the following: RA = $\log_n BD / \log_n AB$, α and β.

The sex variable coded 0 for males and 1 for females was also considered. It did not prove to be discriminant ($F = 0.74$; $P = 0.56$): no difference appeared due to sexual dimorphism, and consequently there is no bias due to a possible not well-balanced sex-ratio in the samples.

3.3. Multivariate analysis

3.3.1. Principal component analysis and dynamic cloud method

The two first axes of the PCA (Fig. 2), explaining 96.7% of the total variance, showed five point swarms consisting of the specimens of the five previously defined groups, namely *Ch. carnea*, *Ch. kolthoffi*, *Ch. lucasina*, *Ch. mediterranea* and the Greek morph. Only four specimens *a priori* classified as *Ch. lucasina* were located outside the *kolthoffi* swarm, three of them being on the border. Thus the preliminary assignments were confirmed.

3.3.2. Discriminant factor analysis

The variance analysis showed a high discriminant power of the variable RA ($F = 276.57$; $P < 10^{-4}$), α ($F = 174.82$; $P < 10^{-4}$) and β ($F = 264.52$; $P < 10^{-4}$). The contribution of the two first axes (82.9% and 14%) of the total variance and the values of the corresponding canonical correlations (0.9572

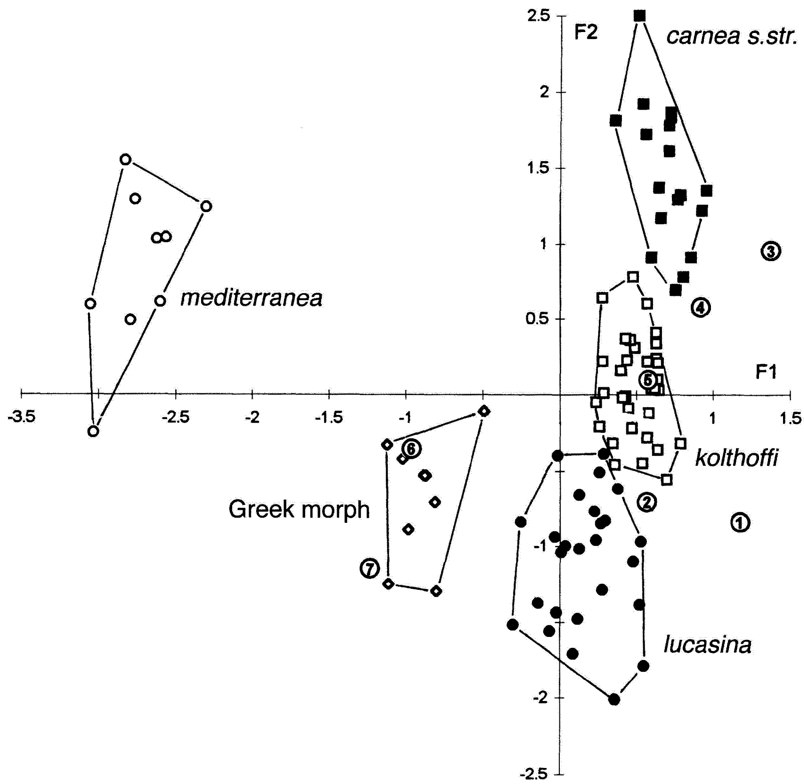

Fig. 2. Principal component analysis: grouping of specimens of *Chrysoperla* on the first two axes. Atypical specimens as ringed numbers.

and 0.8304) testify the high quality of the plane defined by these axes, even if the study of the gravity centre of the groups (Table 2) showed that the third axis allowed a better discrimination in the specimens of the Greek population.

The plot of the first two DFA factors [F_1*F_2 DFA plane] (Fig. 3) clearly shows the subdivision of the typical specimens into five homogeneous groups and the location of the atypical specimens. The F_1 axis markedly separates the *Ch. mediterranea* point swarm from those corresponding to *Ch. carnea s.str.*, *Ch. lucasina* and *Ch. kolthoffi*, whereas the Greek specimens are intermediate. This axis is related to a protuberance in the claw basal enlargement, almost lacking in *Ch. mediterranea*, light in the Greek individuals, and fairly marked in *Ch. carnea s.str.*, *Ch. lucasina* and *Ch. kolthoffi*. The DFA analysis assigned 94.8% of observations to their original group. Only five specimens were wrongly assigned (Table 3): two *Ch. carnea s.str.*, two *Ch. kolthoffi* and one *Ch. lucasina*. Such a percentage may be considered optimistic (Tomassone 1988); however, it allowed us to analyse the specimens of the sixth group, considered "atypical" and indicated by circled numbers in Fig. 2. On the basis of claw shape, the specimens nos. 1 and 2 from Porto were thus assigned to *Ch. lucasina*, and the specimens nos. 3–5, originating from central France, Krakovie and surroundings of Rome, respectively, to *Ch. kolthoffi*. These hypotheses agree with the biogeographic data available on the elements of the *carnea*-complex in Europe (Paulian *et al.* 1996, Thierry *et al.* 1996). The distribution of the *Chrysoperla* species appears to

Table 2. Discriminant function analysis: coordinates of the centres of gravity of the *Chrysoperla* groups on the first three axes.

Group	Axis 1	Axis 2	Axis 3
carnea	0.1844	**0.7842**	0.0281
kolthoffi	**0.8390**	0.0102	0.1103
lucasina	0.0204	**0.8494**	0.1255
mediterranea	**0.8531**	0.0819	0.0650
Greek morph	0.1699	0.1005	**0.7296**

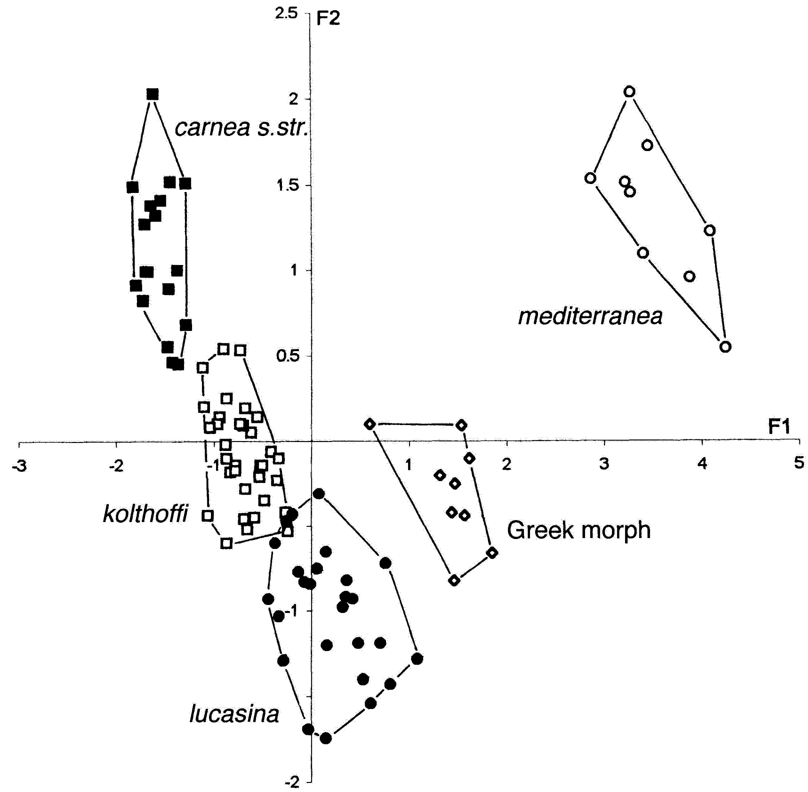

Fig. 3. Discriminant factorial analysis: grouping of specimens of *Chrysoperla* on the first two axes.

be patchy, with wide overlapping areas where they are sympatric, and some other areas where a single species dominates or even excludes all others. The two specimens nos. 6 and 7 collected in Trieste were similar to those from Greece (Fig. 2), and so that suggests an assignment to this morph.

4. Conclusion

The claw shape constitutes a valuable character in discriminating some West-Palaearctic species of the *Ch. carnea*-complex (Fig. 4A–D). The analysis of quantitative data suggests the occurrence of an additional undescribed morph, possibly a new East-Mediterranean species, occurring in the Greek Peninsula and extending up to the Adriatic coast, the status of which is not yet determined (Duelli 1995).

Table 3. Discriminant factor analysis: redistribution of the *Chrysoperla* specimens into groups.

Original group	Assignment by analysis *car*	*kol*	*luc*	*med*	Greek
carnea	**17**	1	0	0	0
kolthoffi	2	**30**	1	0	0
lucasina	0	1	**25**	0	0
mediterranea	0	0	0	**9**	0
Greek morph	0	0	0	0	**10**

Acknowledgements. We thank Dr A. Popov and J. Ganev (Sofia, Bulgaria) and C. Coelho-Matias (Alcobaça, Portugal) who kindly collected specimens. Thanks are also due to Dr R. Cianchi (Rome, Italy) and Dr M. A. Ventura (Ponta Delgada, Portugal) for valuable comments and improvement to the manuscript.

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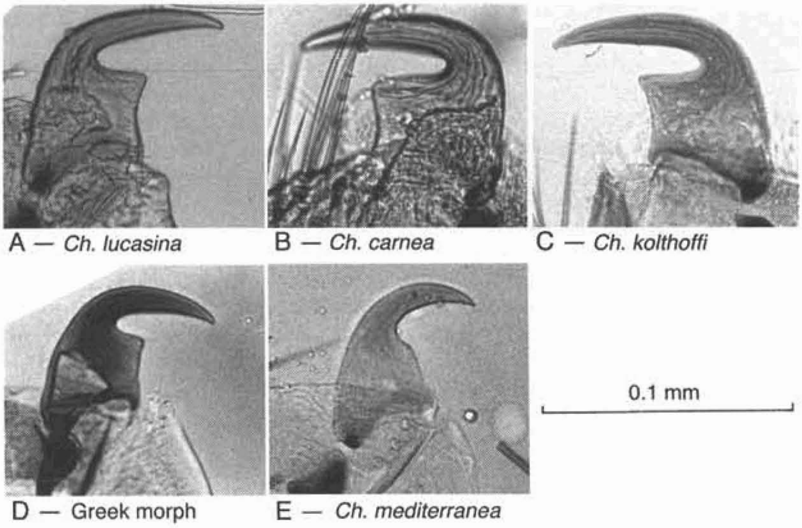

Fig. 4. Claws of *Chrysoperla* species. — *Ch. lucasina* male, Brindisi, Italy, 10.VII.1989; *Ch. carnea* female, Belasitsa, Bulgaria, 27.VI.1982; *Ch. kolthoffi* male, Krakovie, Poland, 7.VIII.1995; *Chrysoperla* sp. female (Greek population), Scala (Aguistri Island), Greece, 17.VIII.1976; *Ch. mediterranea* male, Valencia, Spain, 31.VII.1987.

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Appendix

Key to the West-European *Chrysoperla* Steinmann species of the *carnea* Stephens group *sensu* Brooks (1994). Nomenclature according to Leraut (1991, 1992). Mean values ± 2 S.D.

1. Step-like (rectangular) basal enlargement of the claws (Figs. 4A–D)) 4
— Slightly dilated (sub-triangular) basal enlargement of the claws (Fig. 4E) 2

2. Costal setae on forewings long, hyaline or brown .. 3
— Costal setae on forewings short and dark. Forewings narrow with pointed apex, mid costal margin straight, posterior margin regularly rounded ..... *mediterranea*

3. Forewings broad, costal area at base broad; costal margin regularly convex, posterior margin slightly concave beyond anal area, convex in distal part .... *ankylopteryformis*
— Forewings narrow, costal area undilated, costal and posterior margins subparallel in mid part .... *renoni*

4. Pleural membrane on second abdominal segment with thin dark brown stripe beneath sternite, this stripe often extending to segments 3–6. Costal setae short. Stipes marked laterally with brown stripe, sometimes very pale. Pronotum and abdominal sternites usually with numerous regularly distributed black setae. Basal enlargement of claws (Fig. 4A): AB/BD = 2.65 ± 0.34 (n = 26) .... *lucasina*
— Pleural membrane on second abdominal segment without any brown stripe. Costal setae on forewings long 5

5. Stipes unmarked laterally or only marked with brown distal spot located near basis of maxillary palpus. Abdominal sternites with hyaline or slightly coloured setae, rarely associated with sparse brown setae. Basal enlargement of claws broad (Fig. 4B): AB/BD = 1.93 ± 0.17 (n = 18) .... *carnea*
— Stipes marked laterally with brown longitudinal stripe. Abdominal sternites with brown or black setae 6

6. Basal enlargement of claws (Fig. 4C): AB/BD = 2.32 ± 0.24 (n = 33) .... *kolthoffi*
— Basal enlargement of claws narrower (Fig. 4D): AB/BD = 3.06 ± 0.31 (n = 10) .... Greek morph

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9082

Reference Citation:
Thierry, D.; Cloupeau, R.; Jarry, M.; Canard, M. 1998 [1998.??.??]. Discrimination of the West-Palaearctic Chrysoperla Steinmann species of the carnea Stephens group by means of claw morphology (Neuroptera, Chrysopidae). in Panelius, S. P. (ed.). Neuropterology 1997. Proceedings of the Sixth International Symposium on Neuropterology (13-16 July 1997, Helsinki, Finland). Acta Zoologica Fennica 209:255-262.

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