

## **Electrophoretic Studies in the Genus *Chrysopa* (s. l.), Evolutionary and Phylogenetic Inferences**

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The taxonomy at the genus level of the subfamily Chrysopinae, and particularly of the old genus *Chrysopa* LEACH, represents till now an open problem.

Different attempts of classification were made by several authors. TJEDER (1966, 1972) splitted the genus *Chrysopa* (s. l.) in various genera and subdivided the genus *Chrysopa* (s. str.) in a number of subgenera, on the basis of differences in male genital urites and external genital organs. This subdivision appears supported by studies on abdomen morphology of imagoes and on larval morphology, ethology and development, carried out by PRINCIPI (1977). HÖLZEL (1970) considered in the genus *Chrysopa* (s. str.) only the species with distinct 8° and 9° urosternites, including in other genera the species presenting them fused. The genus *Anisochrysa* sensu HÖLZEL (1970) comprehended two remarkably differentiated groups of species: the subgenera *Chrysoperla* and *Anisochrysa*, differing for genitalia and larval morphology, development and ethology, as pointed out by PRINCIPI (1956, 1977) and SÉMÉRIA (1977). The latter author proposed on such bases to consider *Chrysoperla* as a distinct genus. Finally, *Chrysopa* (s. str.), *Chrysoperla* and *Anisochrysa* were considered as distinct genera by ASPÖCK *et al.* (1980) in their recent revision on European Neuroptera.

The problem of the phylogenetic relationships among the species of the genus *Chrysopa* (sensu latu) was approached by us with multilocus electrophoretic techniques. Their use in taxonomy and the evidence they provide for phylogenetic interpretation were pointed out in a number of recent papers (see for instance AVISE, 1975; BULLINI and SBORDONI, 1980).

We analyzed the genetic differentiation of the following species: *Chrysopa abbreviata* CURTIS, *Ch. dorsalis* BURMEISTER, *Ch. formosa* BRAUER, *Ch. septempunctata* WES-MAEL, *Chrysoperla carnea* (STEPHENS), *Anisochrysa flavifrons* (BRAUER), *A. prasina* (BURMEISTER) and *A. clathrata* (SCHNEIDER).

Electrophoretic analysis was performed on the following 22 gene-enzyme systems: alcohol dehydrogenase (*Adh-2*),  $\alpha$ -glycerophosphate dehydrogenase ( $\alpha$ -*Gpdh*), malate dehydrogenase (*Mdh-1*, *Mdh-2*), malic enzyme (*Me*), isocitrate dehydrogenase (*Idh-1*, *Idh-2*), 6-phosphogluconate dehydrogenase (*6Pgdh*), aldehyde oxidase (*Ao-2*), glyceraldehyde-3-phosphate dehydrogenase (*G3pdh*), xanthine dehydrogenase (*Xdh*), superoxide dismutase (*Sod*), glutamate-oxaloacetate transaminase (*Got-1*, *Got-2*), hexokinase (*Hk*), adenylate kinase (*Adk*), phosphoglucomutase (*Pgm*), alkaline phosphatase (*Aph*), triose phosphate isomerase (*Tpi*), mannose phosphate isomerase (*Mpi*), phosphoglucose isomerase (*Pgi*), esterase (*Est*). Electrophoretic techniques used were, with minor modifications, those described by SHAW and PRASAD, 1970; SELANDER *et al.*, 1971; AYALA *et al.*, 1972; HARRIS and HOPKINSON, 1977.

Genetic differentiation among the considered taxa was calculated from allele frequencies, using NEI's measures of standard genetic identity (*I*) and standard genetic distance (*D*). *I* estimates the proportion of identical genes and ranges from 0 to 1, while *D* estimates the mean number of allelic substitutions and ranges from 0 to  $\infty$  (NEI, 1972).

A matrix of the values of genetic distance and genetic identity found between each possible pair of the considered species is shown in Table 1.

The values of genetic distance among the considered species range from 0.64 (between *Chrysopa dorsalis* and *Ch. formosa*) to 2.08 (between *Ch. dorsalis* and *Chrysoperla carnea*). The corresponding time of evolutionary divergence, estimated using Nei's formula :  $t$  (in years) =  $5 \cdot 10^6 D$ , ranges from about 3 to 10 million years.

The values of  $D$ , among species belonging to the same genus, according to the classification by ASPÖCK *et al.* (1980), range from 0.64 to 1.43 (average  $D = 1.03$ ), well corresponding to those observed among congeneric morphologically differentiated species of various animal groups (AYALA, 1975; BULLINI and SBORDONI, 1980).

Finally, the mean values of  $D$  among the proposed genera : 1.44 between *Chrysopa* and *Anisochrysa*; 1.54 between *Chrysopa* and *Chrysoperla*, 1.36 between *Anisochrysa* and *Chrysoperla*, clearly indicate a similar degree of genetic differentiation among these three groups.

The electrophoretic data are then in agreement with the hypothesis of a generic rank for *Chrysopa*, *Anisochrysa* and *Chrysoperla*.

	FOR	ABB	DOR	SEP	CAR	PRA	FLA	CLA
FOR	—	0.29	0.53	0.46	0.30	0.25	0.28	0.29
ABB	1.22	—	0.39	0.31	0.19	0.30	0.24	0.17
DOR	0.64	0.95	—	0.33	0.12	0.20	0.36	0.22
SEP	0.78	1.15	1.10	—	0.30	0.19	0.20	0.20
CAR	1.22	1.67	2.08	1.20	—	0.23	0.28	0.26
PRA	1.37	1.22	1.63	1.67	1.48	—	0.32	0.44
FLA	1.28	1.41	1.02	1.60	1.27	1.15	—	0.24
CLA	1.22	1.78	1.49	1.59	1.34	0.82	1.43	—

Tab. 1: Matrix of Nei's genetic identities ( $I$ , above diagonal) and genetic distances ( $D$ , under diagonal) for each possible pair comparison between species of the genus *Chrysopa* (sensu lato). FOR = *formosa*; ABB = *abbreviata*; DOR = *dorsalis*; SEP = *septempunctata*; CAR = *carnea*; PRA = *prasina*; FLA = *flavifrons*; CLA = *clathrata*.

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