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Morphology and morphometry of Lycaenid eggs (Lepidoptera: Lycaenidae)

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Abstract

A morphological study with the use of scanning electron microscope of 67 species of Iberian Lycaenidae is presented. The study covers all the genera present in the area and shows an extraordinary variation in chorionic characters that allows egg diagnosis for most species. A morphometric study showed that the eggs from the sample have sizes that are correlated with adult size, but some species showed larger egg size than expected. Species hibernating at the egg stage proved to have on average larger sizes than those overwintering at other stages, probably because this trait might be favourable to endure the adverse conditions taking place during the winter. A cladistic analysis was performed using morphologic and morphometric characters from the egg with the result of poor discriminant power. However, some formal taxonomic groups such as the genera *Lycaena* and *Satyrium* were supported by our analysis due to specific apomorphic characters.

Key words: Egg size-hibernation-phylogeny-scanning electron microscope

Introduction

The family Lycaenidae has attracted the interest of scientists due to the complex life cycles of many species in which myrmecophily plays an important role (Fiedler 1991). Besides, many species or groups in the family show very complex evolutionary patterns or are currently undergoing speciation events (Lukthanov *et al.* 2005, Vila *et al.* 2011). The family groups 5201 species (van Nieukerken *et al.* 2011) from which 72 are present in the Iberian Peninsula, making this family the second of butterflies in number of species from this area, only outnumbered by the Nymphalidae (García-Barros *et al.* 2013).

Lycaenid butterflies show an extraordinary variation and morphological complexity at the egg stage. This variability is probably the effect of a response to selective pressures that are different at this stage from those operating at other stages of the life cycle (García-Barros & Martín 1995). Several papers have dealt with the task of describing the eggs of this family, but particularly classic studies such as Clark & Dickson (1971) and Downey & Allyn (1981, 1984) provided detailed accounts of the South African and North American species respectively. Following these synthesis accounts, many other life history studies included descriptions of the egg stage, some of them using the scanning electron microscope (SEM) which provides an unprecedented opportunity to study the ultrastructure of the egg. The basis of these studies has relied on the work done by Downey & Allyn (1981, 1984) that, together with species descriptions, provided SEM pictures and compiled a terminology for the chorionic structures. The lycaenid egg is spherical with flattened poles. The egg is divided in four different areas following Downey & Allyn (1984). The upper or micropylar pole has the annulus or annular zone in its centre with the micropylar rosette surrounding the micropylar openings which vary in number in different species. The annulus is surrounded by the transition zone, formed by cells which increase in size with the distance from the annulus. The tubercle-aeropyle zone covers most of the egg chorion surface and gives the egg its typical appearance. Tubercles are prominences present in a majority of species arising from the intersections of the ribs that limit the cells of the egg surface. Finally the flat area is in contact with the plant in which the egg is laid and is usually devoid of the typical chorionic sculpturing of the egg and has a very thin chorion (Downey & Allyn 1984; Thomas *et al.* 1991).

In the Iberian Peninsula the first compilation of detailed ultrastructural descriptions using Scanning Electron Microscope was made for 13 species by Munguira (1989). Some other species have been described together with natural history accounts or in monographs concerning different taxonomic groups: Munguira (1985), Munguira & Martín (1988, 1989), Munguira *et al.* (1988), Thomas *et al.* (1991), Sarto & Masó (1991) and García-Barros *et al.* (2013).

Egg morphology has been used for taxonomy and the study of evolution in different butterfly groups for example by García-Barros & Martín (1995) and Freitas & Brown (2004) for Nymphalidae, Llorente & Castro (2007), Hernández-Mejía *et al.* (2013, 2014a,b) for Pieridae and Hernández-Roldán *et al.* (2012) for Hesperiidae. Although the usefulness of characters from immature stages (including the egg) in phylogenetic reconstruction has been frequently acknowledged, the effective incorporation of such information in that context remains anecdotal. However this information may play a crucial role in revealing hidden phylogenetic relations as well as providing observable diagnostic features for clades based in molecular evidence (Wahlberg *et al.* 2005; Dinca *et al.* 2011; Cancela, Munguira & García-Barros 2014). General evolutionary relationships can be traced by the detailed study of the egg and they may also have distinctive specific characters that are useful for species identification in general.

that cannot be diagnosed on the basis of morphology are already known among the lycaenid butterflies (Talavera *et al.* 2013).

Thus some of the egg features described might be able to contribute to the diagnoses of at least two supra-specific groups. Even though the eggs are structurally simple in comparison to the more active life stages such as the larvae and the adults, it is remarkable that the exochorion surface alone provides an important amount of information. It is conceivable that, combined with other types of evidence, egg morphology may contribute efficiently not only to the diagnoses, but also to improve the phylogenetic resolution of this butterfly taxon (see *e.g.* Freitas & Brown 2004).

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