Zootaxa 3721 (5): 488–494
www.mapress.com/zootaxa/

Article

ISSN 1175-5326 (print edition)
ZOOTAXA
ISSN 1175-5334 (online edition)

http://dx.doi.org/10.11646/zootaxa.3721.5.4
http://zoobank.org/urn:lsid:zoobank.org:pub:9432129B-C405-4189-8214-83F20C7FD5AC

Comparative description of larvae of the European species of *Distoleon* Banks: *D. annulatus* (Klug, 1834) and *D. tetragrammicus* (Fabricius, 1798) (Neuroptera, Myrmeleontidae)

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Abstract

The third instar larva of *Distoleon annulatus* (Klug, 1834) is described for the first time and compared with the larva of the other known species of the genus in Europe: *D. tetragrammicus* (Fabricius, 1798). Diagnostic characters of the larvae of the genus *Distoleon*, as well as the interspecific differences, are provided and illustrated. Larvae of *Distoleon* appear to be morphologically conservative and they are mainly recognized by means of the pigmentation pattern.

Key words: antlions, Nemoleontini, larval morphology, Europe

Introduction

The family Myrmeleontidae, commonly known as antlions, includes about 2000 described species in the world (Stange 2004), representing the largest family of Neuropterida. The larvae of most species develop in sandy habitats, therefore the success of the family has been attributed to this specialization (Mansell 1996). The larvae of some antlions construct pitfall traps, but most of them do not dig these remarkable structures, thus they are difficult to find in the field, which explains the scarce number of adequately described larvae (Stange & Miller 1990; Stange 2004).

The genus *Distoleon* Banks, 1910 is included in the tribe Nemoleontini by Stange & Miller (1990) and Stange (2004), a group presenting as typical common character three parallel teeth on the mandible which gradually increase in length toward the apex (with some exceptions, such as the genus *Glenurus*) (Stange & Miller 1990; Stange 2004). The genus *Distoleon* comprises at least 120 species distributed all across the Old World in tropical and temperate areas (Aspöck *et al.* 1980, 2001; Stange 2004). The larvae of this genus do not construct pitfalls traps and they are ambush predators, awaiting passing prey below the surface of sandy soils. The biology and larval stages were known only for the European *D. tetragrammicus* (Fabricius, 1798) (Brauer 1854; Hagen 1873; Redtenbacher 1884; Steffan 1975; Gepp & Hölzel 1989; Satar *et al*. 2006; Gepp 2010; Krivokhatsky 2011; Badano 2013) and for few Far Eastern species (Stange *et al.* 2003; Stange 2004). No available information exists on the larvae of the other European species *D. annulatus* (Klug, 1834), because the only alleged existing account on the larva of this taxon (Willmann 1977) is erroneous and it actually refers to *Gymnocnemia variegata* (Schneider, 1845), as clearly demonstrated by Cesaroni *et al.* (2010). *D. annulatus* is an Afro-Iranian endemic faunal element (Aspöck *et al.* 1980, 2001) reaching southernmost Europe. On the contrary, *D. tetragrammicus* is widespread in the western Palaearctic and it is particularly common in southern Europe (Aspöck *et al.* 1980, 2001).

A comparative study of the third instar larvae of the two European *Distoleon* species was conducted in order to investigate the distinctive characters useful in the identification of the genus and species, besides illustrating them by means of light microscopy.

 Accepted by A. Contreras-Ramos: 25 Sept. 2013; published: 17 Oct. 2013

Materials and methods

The larvae of both species were collected in the field, afterwards they were kept in laboratory conditions in plastic boxes filled with sandy soil and fed with the ant *Messor barbarus* (Linneus, 1767) and beetle larvae of *Tenebrio molitor* (Linneus, 1758) until pupation, thus obtaining the adults necessary for identification.

Observations and study of *D. annulatus* were made with an Olympus® SZX7 Stereomicroscope and photographed with a Olympus® Digital Camera S-C-30 mounted on it and then processed using analySIS getIT software. Measurements were conducted using an ocular micrometer and an electronic caliper.

Morphological observations of *D. tetragrammicus* were realized with a Leica® MZ9.5 stereomicroscope, while measurements and photographs were taken using a Leica® MZ16 stereomicroscope with apochromatic lens, equipped with a Leica® DFC320 digital camera. The photographs were obtained with this camera and subsequently elaborated using LAS (Leica® Application Suite) applied software Version 2.5.0 R1.

The photographs of both species were realized placing the specimens in Petri dishes filled with alcohol. Finally, the obtained images were processed and retouched using the software Adobe Photoshop® CS5 Extended Version 12.0. The measurements of the parameters of the larval head were realized following Cesaroni *et al.* (2010), and the terminology employed follows Stange & Miller (1990) and Stange (2004). The prominent structures bearing the digging setae on the sternite IX are termed here "rastra", from the Latin "rake", according to Badano (2013) due to the absence of a univocal technical term referring to this anatomical feature of diagnostic value.

Descriptions of the third instar larva

Distoleon annulatus (Klug, 1834)

Examined specimens. SE Spain, Almería: Casillas de Atochares, Rambla de Artal): 2 third instar larvae (7.V.2011, F. Acevedo and V. J. Monserrat leg., 1 third instar larva; 29.VI.2011, F. Acevedo and V. J. Monserrat leg., 1 third instar larva); Almería: San José, Cabo de Gata: 1 third instar larva (5.VII.2013, D. Badano, F. Acevedo and V. J. Monserrat leg.).

Description. Size. Average body length (without mandibles) 8.80 mm; head capsule length 2.80 mm, head capsule width (widthest part) 2.45 mm, mandible length 2.54 mm, ratio head capsule width/length 0.87, ratio mandible length/head capsule length 0.9.

General coloring. Ochre with a dark pattern (Fig. 1), ventral side paler with some dark brown spots (Fig. 5); head ochre, with two large triangular dark markings on the dorsal side, lateral side with conspicuous dark markings, ventral side pale with the anterior zone dark (Figs. 1, 2); mandibles dark brown, paler at teeth insertion and darker at the apex of the mandibles and teeth (Fig. 2).

Head. Longer than wide, trapezoidal, external margin of the labrum with a small median pale incision, delimited by two small lobes; head capsule covered with dark setae on the dorsal and ventral surface, longer on lateral sides; dorsal side bearing dolichasters (Figs. 2, 6); antennae longer than the eye tubercle, brown with the scape and pedicel darker (Figs. 2, 6); eye tubercles large and prominent, with seven black stemmata (Fig. 2); mandibles comparatively strong, shorter than the head capsule, provided with 3 pairs of equidistant teeth of which the apical one is the strongest (Fig. 2, 6); 1 seta between each pair of teeth, few (4) setae between the insertion of the mandible and the basal tooth, external margin of the mandible with black long and fine setae until the distal tooth (Figs. 2, 6); labial palpi dark in color, distal segment longer than the others (Fig. 7).

Body. Elliptical in shape, covered by black setae and provided with thoracic scoli (Figs. 1, 5).

Thorax. Ochre with two darker longitudinally lines; pronotum dorsally covered by numerous short setae, larger on lateral sides; mesothoracic spiracle subcylindrical, borne on tubercle (Fig. 3); mesothoracic scoli prominent, especially the anterior pair (Figs. 1, 3).

Legs. Pale in colour, with brown spots in the centre and distal extreme of the last tarsomere, covered by dark, large and fine setae, with two brown curved claws in each leg (Figs. 5, 8).

FIGURES 1–9. 3rd instar larva of *Distoleon annulatus* (Klug, 1834) (Spain, Almería: Casillas de Atochares, Rambla de Artal). 1. Habitus, dorsal view; 2. Head, dorsal view; 3. Detail of thorax, dorsal view; 4. Abdomen, dorsal view; 5. Habitus, ventral view; 6. Head, ventral view; 7. Detail of palpomeres; 8. Thorax and abdomen, ventral view; 9. Detail of VIII and IX sternites, ventral view.

Abdomen. Dorsal side with a median and two lateral series of dark markings creating lines (Figs. 1, 4); ventrally pale with few brown spots, sternite VIII with two spots in proximity of the odontoid processes; abdomen thickly covered by black setae (longer and broader on scoli) (Figs. 5, 8); abdominal spiracles slightly pronounced, brown; VIII sternite equipped with odontoid processes (Fig. 9); IX sternite with a ventro-posterior pair of spiniform setae and a distal pair of small but protruding rastra equipped with 4 pairs of sub-equal digging setae, of which the inner pair is slightly smaller than the others (Fig. 9).

Biological and behavioral notes. The larvae were observed in the SE of Spain, an area characterized by a semiarid climate. They were collected by sieving the dry and fine substratum contained in rock cavities, and also in loose arid soil among grasses and rocks. They can move backward and forward rapidly, but they always bury backwards. They remain buried waiting for preys; if they are disturbed, they feign death for some minutes. When the larvae are mature, they spin slightly oval cocoons covered with soil particles.

Distoleon tetragrammicus (Fabricius, 1798)

Examined specimens. Italy: Veneto, Bovolone (Verona), V.2010, F. Sanna leg., 1 third instar larva; Liguria, Pompeiana (Imperia), VII.2010, D. Badano leg., 1 third instar larva; Sardinia, Berchidda (OT),VII.2010, M. Verdinelli & S. Cossu leg., pitfall, 1 third instar larva; Sardinia, Alghero (Sassari), Capocaccia, IX.2010, D. Badano leg., 1 third instar larva; Tuscany, Elba, Portoferraio (Livorno), IX.2010, L. Forbicioni leg., 1 third instar larva; Liguria, Perinaldo (Imperia), VII.2011, D. Badano leg., 3 third instar larvae; Val d'Aosta, Aymavilles (Aosta), Pont d'Ael, VIII.2011, D. Badano leg., 1 third instar larva; Lazio, Rocca Priora (Roma), X.2011, M. Gigli leg. 1 third instar larva; Liguria, Cipressa (Imperia), I.2012, D. Badano leg., 4 third instar larvae; France: Gard, Générac, VIII.2011, D. Badano leg., 10 third instar larvae; Greece: Corfù, Kato Pauliana, V.2012, D. Badano leg., 1 third instar larva.

Description. Size. Average body length 10.60 mm; head capsule length 3.00 mm (min–max 2.41–3.33), head capsule width 2.45 mm (2.22–2.72), mandible length 2.54 mm (2.24–2.76), ratio head capsule width/length 0.82, ratio mandible length/head capsule length 0.85.

General coloring. Brown with a darker pattern (Fig. 10), ventral side paler with a dark brown marking (Fig. 14), head dark brown, lateral and ventral sides with extensive dark markings (Figs. 10, 11); mandibles dark brown (Fig. 11).

Head. Longer than wide; external margin of the labrum with a small median incision (Figs. 10, 11); antennae longer than the eye tubercle (Fig. 11); eye tubercles prominent; mandibles relatively robust, shorter than the head capsule, equipped with 3 pairs of equidistant teeth of which the apical pair is the largest (Figs. 11, 15); 1 seta between each pair of teeth, few (3–4) setae between the basal tooth and the insertion of the mandible (Figs. 11, 15); labial palpi dark, apical segment longer than the others (Fig. 16).

Body. Elliptical in shape, covered by black setae and provided with thoracic scoli (Figs. 10, 14).

Thorax. Pronotum thickly covered by short setae; mesothoracic spiracle raised on tubercle, subcylindrical (Fig. 12); thoracic scoli prominent, especially the anterior pair (Fig. 12).

Legs. Pale in color, often yellow, covered by dark setae (Figs. 14, 17).

Abdomen. Dorsal side with a median series of dark circular markings with central pale area, creating an annulated pattern (Figs. 10, 13); abdominal spiracles brown, slightly raised; VIII sternite provided with odontoid processes (Fig. 18); IX sternite with a ventral-posterior pair of spiniform setae; rastra prominent, armed with 4 pairs of sub-equal digging setae (Fig. 18).

Biological and behavioral notes. The larvae were collected in Mediterranean woods and shrublands, at the base of trees and under stones. *D. tetragrammicus* larvae are relatively common on rock escarpments and under rock overhangs, even inhabiting artificial structures, as they have been observed on stone walls and in deposits of detritus in the corners of concrete buildings. During the inspection of an arenaceous escarpement in Générac, France, 15 1st instar larvae were detected buried in close contact in a very small recess, surely representing the oviposition site of a female specimen. These larvae did not show any sign of cannibalism or aggressiveness among them. A similar behavior is reported for the larvae of Neuroptera Ascalaphidae, which rest on the stem where the oviposition occurred for a short period, before dispersing.

FIGURES 10–18. 3rd instar larva of *Distoleon tetragrammicus* (Fabricius, 1798) (Italy, Liguria: Pompeiana). 10. Habitus, dorsal view; 11. Head, dorsal view; 12. Detail of thorax, dorsal view; 13. Abdomen, dorsal view; 14. Habitus, ventral view; 15. Head, ventral view; 16. Detail of palpomeres; 17. Thorax and abdomen, ventral view; 18. Detail of VIII and IX sternites, ventral view.

Discussion

The third instar larvae of the genus *Distoleon* are distinguished by the following characters: mandible equipped with 3 pairs of teeth, pronotum with a thick covering of large setae interspersed with spiniform ones, mesothoracic spiracle protruding on tubercle, VIII sternite armed with odontoid processes, IX sternite provided with prominent rastra bearing 4 pairs of digging setae comparable in size. Both the examined species present in the mandible 1 seta between each pair of teeth, few (4) setae between the basal tooth and the insertion of the mandible, although 2 setae between the basal and the second tooth are reported by Satar *et al*. (2006) for *D. tetragrammicus*.

The larvae of the two European species of *Distoleon* are noticeably similar in morphology, however they are easily set apart by their distinctive body coloration and pattern. *D. tetragrammicus* is overall dark brown and the annulated pattern of the dorsal side of the abdomen is a diagnostic character of all larval stages while *D. annulatus* is yellowish, sand-like, and the median stripe on the dorsal side of the abdomen is simply composed by a series of dark spots. The head pigmentation is distinctive in both species: *D. tetragrammicus* is dorsally unmarked except a pair of small spots in the occipital area while the ventral side is covered by extensive dark markings, on the contrary *D. annulatus* is characterized by the presence of a pair of large dark triangular markings on dorsal side and the ventral side is very pale and unmarked, except a dark area at the insertion of the mouthparts. The coloration patterns of the ventral side of the abdomen is noticeably different in the two species, in particular *D. annulatus* shows a pair of dark spots covering the odontoid processes that are absent in the other species. Finally, the larva of *D. annulatus* is slightly smaller than the congener.

D. tetragrammicus is a common species in Mediterranean Europe. It shows a considerable ecological plasticity and its larvae are potentially found wherever a dry and fine substratum, suitable to dig, is present. The larva of *D. tetragrammicus* probably represents the first non pit-building antlion discovered, as the observations of Bonnet are assignable to this taxon (Bonnet 1780; Réaumur 1742). Nevertheless the first accurate description was realized much later by Brauer (1854) who also treated the internal anatomy (Brauer 1855). This species was also included in the works of Hagen (1873) and Redtenbacher (1884). Steffan (1975) was the first to investigate ecological requirements, reporting the presence of this species in sandy soils with a rich component of humus and in alluvial deposits; the present study confirms the observation of the above mentioned author. Finally, Satar *et al*. (2006) described the third instar larvae and the eggs.

On the contrary, *D. annulatus* remains poorly known. This species is rarer and with a significantly narrower distribution in Europe than the previous species, being exclusively reported for Spain, Sicilian islands (Pantelleria, Vulcano, Lampedusa), Crete and Dodecanese islands (Aspöck *et al.* 1980; [Bernardi] Iori *et al.*, 1995; Monserrat & Triviño, 2013). The ecology of this species is inadequately known, however it is probably associated with rocky habitats, as suggested by the actual findings.

The larvae of the species of the genus *Distoleon* are appreciably similar to the other genera included in tribe Nemoleontini by Stange & Miller (1990) such as *Creoleon* Tillyard, 1918 and especially *Neuroleon* Navás, 1909 (Steffan 1975; Devetak *et al*. 2010; Gepp 2010; Badano 2013), thus a detailed study of the larvae of the members of this tribe appears necessary to investigate the internal relationships in this complex group of Myrmeleontidae.

Acknowledgments

We wish to thank Dra. María Dolores Martínez for helping with the identification of the ants. This contribution is part of the coordinated project of I+D+i: 'Fauna Ibérica (Neuroptera) (CGL2010-22267-C07-05)'.

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