First record of *Diaeretus leucopterus* (Haliday) (Hymenoptera, Braconidae, Aphidiinae), the parasitoid of the aphid species, *Eulachnus agilis* (Kaltenbach) (Hemiptera, Aphididae) in North Africa

Monia Ben Halima Kamel, Nickolas G. Kavallieratos, Petr Starý and Ehsan Rakhshani

**Abstract**

A survey of pine-associated aphids and their parasitoids was carried out in the arboretum of *Pinus halepensis* Miller in the Higher Agronomic Institute Chott Mariem (ISA CM, Tunisia) during 2010–2011 and 2018. The survey revealed the presence of the aphid parasitoid species, *Diaeretus leucopterus* (Haliday) (Hymenoptera, Braconidae, Aphidiinae). This is the first record of *D. leucopterus* from Tunisia and the African continent. The parasitoid emerged from the spotted green pine needle aphid, *Eulachnus agilis* (Kaltenbach) (Hemiptera, Aphididae, Lachninae), on *P. halepensis*. The generalist hyperparasitoid, *Asaphes suspensus* (Nees) (Hymenoptera, Pteromalidae), was also found attacking *D. leucopterus*. The diagnosis of the primary parasitoid, on the basis of relevant illustrations, was provided, and the possible routes of its transportation into North Africa were discussed.

**Keywords:** Pine aphids, *Eulachnus agilis*, *Diaeretus leucopterus*, Hyperparasitoids, Tunisia

**Background**

*Pinus* spp. are the largest genus of the coniferous plants in the northern hemisphere. They include many important species from the economic and ecologic point of views (Gernandt et al. 2005). Three pine tree species are common in Tunisia, including the Aleppo pine, *Pinus halepensis* Miller; the maritime pine, *P. pinaster* Aiton; and the stone pine, *P. pinea* L. The first two species are naturally grown in a wide area of Tunisia. *P. halepensis* consists of the main forest element of Tunisia that encompasses about 297,000 ha, i.e., 57% of the total forest area of the country (Khouja 1997). *P. pinea* forests are found along the coasts of Tunisia, in about 27,000 ha, after plantation of dunes between Tabarka and Kelibia.

Although several insect species are associated with the pine forests globally, some of them have been primarily raised as economic pests. Aphids of the genus *Eulachnus* del Guercio (Hemiptera, Aphididae, Lachninae) have a significant anti-aesthetic impact on the urban areas, where pine species are grown as ornamental and shading plants (Morris 2006). *Eulachnus* aphids are widely distributed outside their original sources, due to the massive transportation of infested pine stock through international trade (Richardson 1998). Although *Eulachnus* spp. have a Palaeartic origin, 3 species (*Eulachnus agilis* (Kaltenbach), *E. brevipilosus* Börner, and *E. rileyi* (Williams)) have invaded North and South America (Blackman and Eastop 1994). The majority of these aphids are effectively controlled in their native areas of distribution mainly due to the activity of their natural enemies. However, they become important pests in the absence of their natural enemies in the areas...
of invasion. Therefore, once established, they cause serious damages to seedling and mature pine trees (Alford 2012).

Four Eulachnus species, i.e., E. agilis, E. nigricola (Pašek) (Ben Halima Kamel 2012; Kanturski et al. 2017), E. rileyi (Boukhris-Bouachem et al. 2007), and E. tuberculostemnatus (Theobald) (Ben Halima Kamel et al. 2019) have been recorded in Tunisia. Although the complex of natural enemies of Eulachnus spp. is well studied in the Mediterranean countries (Nuñez-Pérez and Tizado 1996; Kavallieratos et al. 2004), there are no data from Tunisia and other North African countries. Till now, only generalist coccinellids have been recorded that associated with Eulachnus spp. in this region (Ben Halima Kamel et al. 2018).

In the present study and through a survey of the pine-associated aphids and their parasitoids carried out in Tunisia, the aphid parasitoid species, Diaeretus leucopterus (Haliday) (Hymenoptera, Braconidae, Aphidiinae), was recorded for the first time. Its diagnosis and the possible routes of its transportation into North Africa were discussed.

**Material and methods**

The materials were collected from the arboretum of P. halepensis in the Higher Agronomic Institute Chott Mariem (ISA CM, Tunisia) during spring (2010, 2011, 2018). Pine needles were inspected in situ for aphid colonies (Eulachnus spp.). Once detected, needles, that carried aphids, were carefully cut off and subdivided into plastic boxes with a dimension of 100 × 150 mm. Subsequently, boxes were covered with muslin to allow ventilation of the content. Few adult aphids were separately preserved in 70% ethanol for later identification. The collected materials were maintained for 2–3 weeks under laboratory conditions at 25 °C and inspected daily for the emergence of adult parasitoids. The remaining material was kept for 2 additional weeks at the same conditions to rear any secondary parasitoids. The emerged parasitoids were carefully captured, using an aspirator and dropped into 75% ethanol. Female parasitoids were dissected and slide-mounted in the Hoyer medium. The slide specimens were studied under a Nikon® Eclipse E200 microscope (Nikon Corporation, Japan). Photos from the slides were captured by a Canon® EOS 700D camera (Canon Inc., Japan) and used to prepare line drawings in the Adobe® Illustrator CS5. A card-mounted specimen was imaged by using the Canon® EOS 700D, mounted with an adapter on Hund® stereomicroscope (Wetzlar Inc., Germany). A series of 10–15 multi-focused captured photographs were subsequently merged into a single in-focus image by using the Zerene Stacker version 1.04 software (Zerene Systems LLC, 2017). The morphological terminology used in this study follows that of Sharkey and Wharton (1997) for parasitoids and Blackman and Eastop (2016) for aphids. Parasitoid specimens are deposited in the collection of the Department of Plant Protection, University of Zabol (DPPZ), Iran.

**Results and discussion**

The aphid parasitoid species D. leucopterus emerged from mummies of E. agilis.

**Taxonomic account**

Order: Hymenoptera Linnaeus, 1758

Family: Braconidae Nees, 1811

Subfamily: Aphidiinae Haliday, 1833

*Diaeretus leucopterus* (Haliday, 1834)

* Aphidius leucopterus Haliday, 1834. Entomological Magazine, 2:93–106.

Material examined: 3♀ 1♂, ex. *Eulachnus agilis* (Kaltenbach), on *Pinus halepensis* Miller, during April 2010, 2011, and 2018, Tunisia, ISA CM (35° 54′ 58″ N, 10° 33′ 36″ E).

Diagnosis: (female—Fig. 1). Body length 1.7–1.9 mm. Head sparsely setose, wider than thorax at tegula. Eyes medium sized. Malar space 0.25–0.30 times longitudinal eye diameter. Tentorial index 0.6–0.7. Clypeus flat, with 4–6 long setae. Maxillary palpi four-segmented, labial palpi 2-segmented. Antenna filiform, 16-segmented. Mesoscutum (Fig. 2a) smooth and shiny with rows of sparse setae along the trace of effaced notaulices. Forewing (Fig. 2b) stigma triangular, its length 2.4–2.6 times its maximum width, and 1.9–2.1 times R1. Vein r&RS short, not reaching the end of stigma, its length 0.85–0.95 times R1. Propodeum (Fig. 2c) smooth with the anterolateral and central carinae which extended near the spiracles. Petiole (Fig. 2d) stout, slightly constricted after spiracles, then parallel sided, 3 times its width at spiracles. Gaster lanceolate. Ovipositor sheath (Fig. 2e) stout, sub quadrate and apically truncated. Recorded diagnostic characters agree with that reported by Kalluzhna (2014).

Coloration: Generally dark, head and mesosoma black, gaster dark brown. Antenna brown, with dark brown scape and pedicel. Legs light brown with yellow patches at tip of segments. Mumified aphids of yellowish brown color were mainly attached to the underside of the needles (Figs. 3a, b). Parasitoids were emerged 7 days post-collection by cutting a circular hole at the posterior part of the mummy above the cauda (Fig. 3c).

The hyperparasitoid Asaphes suspensus (Nees) (Hymenoptera, Pteromalidae) was also emerged from the mumified aphids (2♀ 3 ♂, same data as for the parasitoid, April 2010 and 2011). This species was already recorded in North Africa in Morocco (Vago 2002). It is a generalist hyperparasitoid of numerous aphid species excluding pine aphids (Gibson and Vikberg 1998). The emergence hole of the adult hyperparasitoid, with irregular margines, is distinctly smaller than the corresponding
Fig. 1 *Diaeretus leucopterus* (Haliday), female, lateral aspect of habitus

Fig. 2 *Diaeretus leucopterus* (Haliday), female. 

a. Mesonotum, dorsal aspect.  

b. Forewing.  

c. Propodeum, dorsal aspect.  

d. Petiole, dorsal aspect.  

e. Genitalia, lateral aspect. (After Rakhshani et al. 2019)
hole of the primary parasitoid and cutted at the anterio-
dorsal area of the mummy (Fig. 3d). Another species, *Asaphes vulgaris* Walker (Hymenoptera, Pteromalidae),
was also recorded as a hyperparasitoid of *D. leucopterus*
via *E. rileyi* (Suay Cano et al. 1998). Further records re-
garding the associations of the generalist hyperparasitoids
of the genus *Dendrocerus* Ratzeburg (Hymenoptera, Mega-
splilidae) with *D. leucopterus*, both in Eastern (Takada
1973) and Western (Starý 1977) Palaeartics, indicate the
presence of diverse food webs rooted on *Eulachnus* spp. to
be explored by subsequent studies.

*Diaeretus leucopterus* was recorded for the first time not
only in Tunisia but also in the African continent in associ-
ation with *E. agilis* (Kaltenbach). *Diaeretus leucopterus*
is a widely distributed species in the Western Palaeartic,
including some European countries (Mackauer and Starý
1967; Kavallieratos et al. 2004; Kaliuzhna 2014) and some
countries of the Eastern Palaeartic (Starý and Schlinger
1967; Takada 1968; Chen and Shi 2001; Starý et al. 2010)
or Oriental (Starý and Ghosh 1983). Several Mediterr-
anean countries, including France, Greece, and Italy, have
been recorded as common areas of distribution of *D. leu-
copterus*, from where it might be accidentally introduced
into the North African area (Tunisia). According to Sghai-
ier and Claustriaux (2015), *P. halepensis* is of mixed origin
since some *P. halepensis* forests are assumed to originate
from the nearby countries, i.e., Africa (Morocco), South-
ern Europe (Italy), and Western Asia (Israel).

*Siphonatrophia cupressi* (Swain) (Hemiptera, Aphidi-
dae, Aphidinae) has been recently recorded in Tunisia on
*Cupressus sempervirens* L. parasitized by an unknown
parasitoid species (Ben Halima Kamel and Mdellel
2017). Since both aphids (*E. agilis* and *S. cupressi*)
and their host plants (*P. halepensis, C. sempervirens*)
were found in the same area (ISA CM), the association of *D.
leucopterus* with *S. cupressi* seems expectable. It should
be noted that the 2 aphid genera, *Eulachnus* and *Schizo-
lachnus* Mordvilko (Hemiptera, Aphididae, Lachninae),
were recorded as hosts of *D. leucopterus* in the Mediterr-
anean area (Kavallieratos et al. 2004). However, further field
efforts are necessary to confirm this hypothesis. Few host
records of *D. leucopterus* including *Mindarus abietinus*
Koch (Hemiptera, Aphididae, Mindarinae) (Čapek 1960)
and *Brachycerus cardui* (L.) (Hemiptera, Aphididae,
Aphidinae) (Fulmek 1968) look doubtful and need verifi-
cation. Although some other coniferous-associated aphid
species, mainly of the genus *Cinara* Curtis are present in
Tunisia (Ben Halima Kamel 2012; Ben Halima Kamel
et al. 2019), they have not been found parasitized yet
(Rakhshani et al. 2019). One other European parasitoid of
*Eulachnus* aphids, *Praon bicolor* Mackauer (Hymenoptera,
Braconidae, Aphidiinae) (Kavallieratos et al. 2004), was
found in the Mediterranean counties (Tremblay 1975; Núñez-Perez and Tizad 1996). It is an uncommon species, while its occurrence in North Africa has not been documented yet. The Nearctic aphids of the genus *Essigella* del Guercio are biogeographically associated with pine trees in North America (Blackman and Eastop 1994; Théry et al. 2018). *Diaeretus essigellae* Starý & Zuparko (Hymenoptera, Braconidae, Aphidiinae) was described from *Essigella californica* (Essig) (Hemiptera, Aphididae, Lachninae) on *Pinus* spp. in California (Starý and Zuparko 2002). It is closely related to *D. leucopterus*, but it is immediately separated by the absence of propodeal carinae. However, it is rather unlikely to occur in Tunisia, since pine seedlings are not transported from North America to Tunisia. A comparison of both *Diaeretus* species, using DNA sequences, can provide further data about the identity of these taxa and their associated host aphids.

**Conclusion**

Both *Eulachus* aphids and their parasitoid, *D. leucopterus*, seem to be invasive species in North Africa (Tunisia). Considering the economic importance of these pest aphids, further investigation in the neighboring countries is needed to reveal the expansion of the parasitoid and to confirm its possible establishment.

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**Authors’ contributions**

MKBH reared the specimens and identified the aphid specimens. NGK identified the aphid specimens, revised the structure and manuscript and did the English grammar editing. PS identified the parasitoid specimens. ER did the aphid parasitoid and hyperparasitoid specimens, and he was the major contributor in writing the manuscript and preparation of illustrations. The authors read and approved the final manuscript.

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**Availability of data and materials**

All the mentioned specimens are deposited in the DPPZ collection and are available.

**Ethics approval and consent to participate**

Not applicable.

**Consent for publication**

Not applicable.

**Competing interests**

The authors declare that they have no competing interests.
