Parasitic wasps related to Prays oleae (Bernard, 1788) (Lepidoptera, Praydidae) in olive orchards in Greece

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Abstract

The olive moth, Prays oleae (Bernard, 1788) (Lepidoptera: Praydidae) is categorised among the most devastating insect pests of olives, whose anthophagous and carpophagous generations can cause yield loss up to 581 and 846 kg of fruit per ha, respectively. In this study, results of the captured parasitoids in olive tree (Olea europaea Linnaeus, 1753) orchards, or infested olive plant material in Crete, Greece, is presented. Five of the six identified species captured in trap devices are related to Prays oleae, i.e., Chelonus elaeaphilus Silvestri, 1908, Chelonus pellucens (Nees, 1816), Apanteles xanthostigma (Haliday, 1834), Diadegma armillatum (Gravenhorst, 1829), and Exochus lentipes Gravenhorst, 1829. The species Eupelmus urozonus Dalman, 1820 and Pnigalio mediterraneus Ferrière & Delucchi, 1957 were reared from infested Prays oleae leaves. Chelonus pellucens is reported for the first time from Greece. According to the international literature, 59 hymenopterous and dipterous parasitoid species are associated with Prays oleae in Europe.

Keywords

Diptera, Greece, Hymenoptera, parasitoids, Prays oleae

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Introduction

Olive trees growing has been traditionally localised in the Mediterranean Basin for thousands of years, where almost 97.9% of the cultivated areas are located (Rallo et al. 2018). The list of potentially harmful organisms includes more than 255 species and the losses due to insect pests alone are estimated to be approximately 15% of production (Haniotakis 2003). Among them, the most common species are the olive fruit fly, Bactrocera oleae (Rossi, 1790) (Diptera: Tephritidae), the olive moth, Prays oleae (Bernard, 1788) (Lepidoptera: Praydidae), and the Mediterranean black scale, Saissetia oleae (Olivier, 1791) (Hemiptera: Coccidae) (Haniotakis 2003).

Prays oleae is one of the main pests infesting olives of commercial production, since larvae of the first, second, and third generations attack flowers, fruits, and leaves, respectively (Kavallieratos et al. 2005; Nave et al. 2017). The anthophagous generation can cause yield losses up to 581 kg of fruit per ha and the corresponding carpophasous up to 846 kg per ha, an issue that justifies the imposed control measures (Bento et al. 2001). In recent years, high socioeconomic pressures have forced olive growers to develop alternative control strategies in an effort to mitigate the undesirable side effects of pesticides on trophic chains and biological balances (Nave et al. 2017). In this sense, not only the economic losses due to the pest should be evaluated, but also the possible secondary effects that such control measures can have on beneficial fauna (Ramos et al. 1998).

Previous research has revealed a wide parasitoid spectrum that is related to P. oleae, resulting to biological control efforts against this pest. The first parasitoid used in biocontrol program was Trichogramma embryophagum (Hartig, 1838) (Hymenoptera: Trichogrammatidae) in former Yugoslavia (Brnetić 1988). In Spain, three species have been released against P. oleae with various levels of success; i.e., Chelonus elaeaphilus Silvestri, 1908 (Hymenoptera: Braconidae), the specialised Ageniaspis fuscicollis (Dalman, 1820) var. praysincola Silvestri, 1907 (Hymenoptera: Encyrtidae) and T. embryophagum (Civantos and Caballero 1993). Trichogramma cacaeciae Marchal, 1927 (Hymenoptera: Trichogrammatidae) has been utilised in Portugal (Bento et al. 1998) and Trichogramma evanescens Westwood, 1833 in Egypt (Agamy 2010).

Although there are previous records concerning the occurrence of P. oleae parasitoids in Greece, there are no data available from the island of Crete, the most important olive production area with almost 200,000 ha cultivated with olive trees (i.e., nearly 25% of the total island area is covered with olive plantations; Hellenic Statistical Authority 2014). Given that the knowledge of the beneficial entomofauna of the olive crop is clearly linked with the biological control of pests infesting this crop and that indigenous strains of parasitoids occurring in olive groves can be more effective against certain olive pests than the commercially available parasitoids (Herz and Hassan 2006), the objective of this study was to further investigate the parasitoid complex that is associated with P. oleae in the overlooked area of Crete by using trap devices and collecting plant material.
Materials and methods

All parasitoids were collected in olive orchards from the island of Crete, Greece from June to October 2017. A part of the material was captured in five glass McPhail trap devices, installed from June to October in an olive orchard at Messara (Crete) that covers an area of approx. 0.5 ha baited with 200 ml aqueous solution of 2% hydrolysed protein (Entomela 75 SL, 25% w/w urea; BASF Hellas, Amaroussion, Greece). Each trap device was placed with its lower part at a height of 2 m from the ground. The distances among trap devices were approx. 100 m. The solution was replaced every week. Additional specimens were reared from *P. oleae* infested plant material (*O. europaea* var. *koroneiki*). Infested leaves by *P. oleae* larvae were collected from olive trees, separately transferred into plastic vials covered with mesh, and transferred to the laboratory. Vials were maintained at 25 °C and 60% relative humidity and inspected daily for emergence of parasitoids. All parasitoid individuals, either from trap devices or plant material, were preserved in 96% alcohol. Specimens were dissected and slide mounted in Berlese medium. The identification of the captured and reared specimens was conducted under a Nikon SM2 745T binocular stereomicroscope (Nikon CEE GmbH, Wien, Austria) or an Olympus SZX9 (Olympus Corporation, Tokyo, Japan) using appropriate keys (Tobias et al. 1986; Askew and Nieves Aldrey 2000; Tolkanitz 2007; Broad 2011). Part of the specimens was deposited in the insect collection of the Laboratory of Agricultural Zoology of Entomology, Agricultural University of Athens, Greece, and a part was deposited in the insect collection of the Faculty of Sciences and Mathematics, Department of Biology and Ecology, University of Niš, Serbia.

Additional to field research, we critically reviewed all recorded parasitoids of *P. oleae* in Greece and Europe indicating the pest’s stage they attack. The synonymy among taxa was checked and adopted according to online databases (van Achterberg 2013; Fernandez Triana and Ward 2015; Noyes 2017; Tschorsnig 2017), and the database provided by Yu et al. (2012).

Results

In total, five out of six species captured in McPhail trap devices are related to *P. oleae*, i.e., *C. elaeaphilus*, *Chelonus pellucens* (Nees, 1816) (Hymenoptera: Braconidae), *Apaneles xanthostigma* (Haliday, 1834) (Hymenoptera: Braconidae), *Diadegma armillatum* (Gravenhorst, 1829) (Hymenoptera: Ichneumonidae), and *Exochus lentipes* Gravenhorst, 1829 (Hymenoptera: Ichneumonidae), while two species were reared from *P. oleae* infested olive leaves.

The exhaustive investigation of the international literature revealed 59 hymenopterous and dipterous parasitoid species that attack *P. oleae* in Europe; 14 Braconidae, 2 Chalcididae, 1 Encyrtidae, 20 Eulophidae, 1 Eupelmidae, 7 Ichneumonidae, 1 Platygastridae, 3 Pteromalidae, 2 Tachinidae, and 8 Trichogrammatidae (Table 1). Thirty-one out of
**Table 1.** Parasitoids of *Prays olete* recorded in Europe and their presence in Greece: (+) recorded, (-) not recorded.

| Family       | Species                                         | Source of host record | Host stage attacked | Recorded or not in Greece |
|--------------|-------------------------------------------------|-----------------------|---------------------|---------------------------|
| Braconidae   | *Aleiodes circumscriptus* (Nees, 1834)          | Beyarslan (2015)      | larva               | +                         |
|              | *Aleiodes gastritor* (Thunberg, 1822)           | Halperin (1986)       | larva               | +                         |
|              | *Apanteles sambustigmina* (Haliday, 1834)       | Nave et al. (2016)    | larva               | +                         |
|              | *Bracon hebetor* Say, 1836                     | Aubert (1966)         | larva               | +                         |
|              | *Bracon laetus* (Wesmael, 1838)                | Falcó et al. (1993)   | larva               | +                         |
|              | *Bracon crassicornis* Thomson, 1892             | Silvestri (1906)      | larva               | +                         |
|              | *Chelonus* (Microchelonus) *elaephila* Silvestri, 1908 | Nave et al. (2016) | larva               | +                         |
|              | *Chelonus* (Microchelonus) *silvestrii* (Papp, 1999) | Papp (1999)         | larva               | -                         |
|              | *Chelonus (Parachelonus) pellucens* (Nees, 1816) | Testeira et al. (2000) | larva               | -                         |
|              | *Clinocentrus testaceus* (Kriechbaumer, 1894)  | Testeira et al. (2000) | larva               | -                         |
|              | *Dolichogenidea dilecta* (Haliday, 1983)        | Télenda (1955)       | larva               | -                         |
|              | *Dolichogenidea ultor* (Reinhard, 1880)        | Arambourg (1969)      | larva               | -                         |
|              | *Meteorus rubens* (Nees, 1811)                 | Testeira et al. (2000) | larva               | +                         |
|              | *Phanerotoma dentata* (Panzer, 1805)           | Testeira et al. (2000) | larva               | +                         |
| Chalcididae  | *Hockeria bifasciata* Walker, 1834             | Madl (2008)          | larva               | -                         |
|              | *Hockeria unicolor* Walker, 1834                | Stavraki (1977)       | larva               | +                         |
| Encyrtidae   | *Ageniaspis fuscicolli* (Dalman, 1820) var. *praysincola* Silvestri, 1907 | Nave et al. (2016) | larva               | +                         |
| Eulophidae   | *Ascodes exrias* (Walker, 1848)                | Silvestri (1908)      | larva               | +                         |
|              | *Baryscapus nigroviolaceus* (Nees, 1834)       | Noyes (2017)          | larva               | -                         |
|              | *Chrysocharis gemma* (Walker, 1839)             | Noyes (2017)          | larva               | +                         |
|              | *Chrysocharis nipherus* (Walker, 1839)         | Noyes (2017)          | larva               | +                         |
|              | *Cirrhipus elongatus* Boucek, 1959              | Noyes (2017)          | larva               | -                         |
|              | *Dicadocerus westwoodii* Westwood, 1832        | Ramos and Panis (1975) | larva               | +                         |
|              | *Elasmus arenatus* Ferrière, 1947               | Ferrière (1947)       | larva               | -                         |
|              | *Elasmus flabellatus* (Fonscolombes, 1832)     | Nave et al. (2016)    | larva               | +                         |
|              | *Elasmus macii* Ferrière, 1929                 | Anonymous (2006)      | larva               | -                         |
|              | *Elasmus nudus* (Nees, 1834)                   | Ramos and Panis (1975) | larva               | -                         |
|              | *Elasmus steffeni* Viggiani, 1967               | Redolfi and Campos (2010) | larva               | +                         |
|              | *Elasmus westwoodii* Giraud, 1856              | Noyes (2017)          | larva               | +                         |
|              | *Euderus albitarsis* (Zetterstedt, 1838)       | Nave et al. (2016)    | larva               | -                         |
|              | *Hemiptarsenus unguicellus* (Zetterstedt, 1838) | Noyes (2017)          | larva               | -                         |
|              | *Pedobius bruchicida* (Rondani, 1872)          | Bouček (1974)         | larva/pupa          | +                         |
|              | *Prionigel aequalis* (Walker, 1839)            | Nave et al. (2016)    | larva/pupa          | +                         |
|              | *Prionigel epilobi* Bouchek, 1966               | Stavraki (1970)       | larva/pupa          | +                         |
|              | *Prionigel longulus* (Zetterstedt, 1838)       | Stavraki (1970)       | larva/pupa          | +                         |
|              | *Prionigel mediterraneus* Ferrière & Delucchi, 1957 | Stavraki (1970)     | larva/pupa          | +                         |
|              | *Prionigel pectinicornis* (Linnæus, 1758)      | Ramos and Panis (1975) | larva/pupa          | +                         |
| Eupelmidae   | *Eupelminus sozonos* Dalman, 1820              | Noyes (2017)          | larva               | +                         |
| Ichneumonidae| *Diadegona armillatum* (Gravenhorst, 1829)     | Bento et al. (1998)   | larva/pupa          | +                         |
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| Family          | Species                                | Source of host record | Host stage attacked | Recorded or not in Greece |
|-----------------|----------------------------------------|-----------------------|---------------------|---------------------------|
| Ichneumonidae   | *Diadegma semiclausum* (Hellén, 1949)  | Torres (2010)         | larva/pupa          | +                         |
|                 | *Exoxus lentipes* Geavenhorst, 1829     | Texeira et al. (2000) | larva               | -                         |
|                 | *Himetotoma superbus* Schmiedeknecht, 1900 | Vidal (1997)     | larva/pupa          | -                         |
|                 | *Itoplectus alternans* (Geavenhorst, 1829) | Silvestri (1908)    | larva/pupa          | -                         |
|                 | *Listonota superbator* Aubert, 1967     | Aubert (1969)        | larva               | +                         |
|                 | *Scambus elegans* (Woldstedt, 1877)    | Nave et al. (2017)   | larva               | -                         |
| Platygastridae  | *Platygaster apicalis* Thomson, 1859    | Stavraki (1970)      | larva               | +                         |
| Pteromalidae    | *Mesopolobus mediterraneus* (Mayr, 1903) | Bozbuğa and Elekçiçölğuler (2008) | pupa               | -                         |
|                 | *Pteromalus chrysos* Walker, 1836       | Noyes (2017)         | pupa                | -                         |
|                 | *Pteromalus seminatus* Walker, 1834     | Noyes (2017)         | pupa                | -                         |
| Tachinidae      | *Phytomyza nigra* (Meigen, 1824)        | Kara and Tschorsnig (2003) | larva          | -                         |
|                 | *Phytomyza vaccinii* Sintenis, 1897     | Tschorsnig (2017)    | larva               | -                         |
| Trichogrammatida| *Trichogramma boumanarachae* Pintureau & Babault, 1988 | Polaszek (2009)    | egg                 | -                         |
|                 | *Trichogramma kriisiceae* Bezdenko, 1968 | Polaszek (2009)    | egg                 | -                         |
|                 | *Trichogramma cordubensis* Vargas & Cabello, 1985 | Jardak (1980)    | egg                 | -                         |
|                 | *Trichogramma dendriformi* Matsunuma, 1926 | Polaszek (2009)    | egg                 | -                         |
|                 | *Trichogramma euprotidis* (Girault, 1911) | Pereira et al. (2004) | egg             | +                         |
|                 | *Trichogramma minutum* Riley, 1871      | Stavraki (1985)     | egg                 | -                         |
|                 | *Trichogramma oleae* Voegele & Pointel, 1979 | Polaszek (2009)    | egg                 | +                         |
|                 | *Trichogramma pretiosum* Riley, 1879    | Polaszek (2009)     | egg                 | -                         |

these 59 parasitoid species have been recorded in Greece: 9 Braconidae, 1 Chalcididae, 1 Encyrtidae, 13 Eulophidae, 1 Eupelmidae, 3 Ichneumonidae, 1 Platygastridae, and 2 Trichogrammatidae. All Braconidae, Chalcididae, Encyrtidae, Eupelmidae, Platygastridae, and Tachinidae which are parasitoids of *P. oleae* attack only larvae. All eulophids parasitise larvae of *P. oleae* while some of them attack both larvae and pupae. Three ichneumonids parasitise larvae exclusively and four both larvae and pupae. All pteromalids are pupal parasitoids whilst all trichogrammatids are egg parasitoids.

**Family Braconidae**

*Apanteles xanthostigma* (Haliday, 1834)

**Material examined:** 11 ♀, Messara (Crete) (35°2′20″N, 24°50′54″E), 16–23.06.2017, captured in McPhail trap device.

*Chelonus (Microchelonus) elaeaphilus* (Silvestri, 1907)

**Material examined:** 4 ♀, 4 ♂, Messara (Crete) (35°2′20″N, 24°50′54″E), 09–16.06.2017, captured in McPhail trap device.
**Chelonus (Parachelonus) pellucens** (Nees, 1816)

**Material examined:** 6 ♀, Messara (Crete) (35°2'20"N, 24°50'54"E), 09–16.06.2017, captured in McPhail trap device.

**Glyptapanteles vitripennis** (Curtis, 1830)

**Material examined:** 2 ♀, 7 ♂, Messara (Crete) (35°2'20"N, 24°50'54"E), 23–30.06.2017, captured in McPhail trap device.

**Family Eulophidae**

**Pnigalio mediterraneus** Ferrière & Delucchi, 1957

**Material examined:** 12 ♂, Heraklion, Voutes, (Crete) (35°15'54"N, 25°03'26"E), 15.03.2017 (date of host collection). Host: *Prays oleae* on *Olea europaea* var. *koroneiki*.

**Family Eupelmidae**

**Eupelmus urozonus** Dalman, 1820

**Material examined:** 8 ♀, 12 ♂, Heraklion, Voutes, (Crete) (35°15'54"N, 25°03'26"E), 15.03.2017 (date of host collection). Host: *Prays oleae* on *Olea europaea* var. *koroneiki*.

**Family Ichneumonidae**

**Diadegma armillatum** (Gravenhorst, 1829)

**Material examined:** 3 ♀, 5 ♂, Messara (Crete) (35°2'20"N, 24°50'54"E), 16–23.08.2017, captured in McPhail trap device.

**Exochus lentipes** Gravenhorst, 1829

**Material examined:** 6 ♀, 8 ♂, Messara (Crete) (35°2'20"N, 24°50'54"E), 16–23.09.2017, captured in McPhail trap device.

**Discussion**

Microgastrinae is one of the largest subfamilies of Braconidae with about 2,000 described species worldwide (Pérez Rodríguez et al. 2013). Very recently, the hymenopteran parasitoid complex of *P. oleae* was studied in Portugal where, among the 22 recorded parasitoid taxa, *A. xanthostigma* was the major natural enemy (Nave et al. 2017). Furthermore, in Egypt *A. xanthostigma* was found to parasitise the larval stage of *P. oleae* at a rate of more than 50% (Herz et al. 2005). Apart from *P. oleae*, this parasitoid species parasitises a high number of microlepidopterous species, mainly Tortricidae, Gracillariidae, and Yponomeutidae, particularly the genera *Paraswammerdamia* Friese, 1960 and *Swammerdamia* Hübner, 1825 (Yu et al. 2012). *Glyptapanteles* Ash-
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Glyptapanteles vitripennis was first reported in southern Greece in 1978 (Papp 2007) without further records since then. This parasitoid species was the second most abundant recovered from Malaise traps placed in the Artikutza forest of Pyrenees (Spain) (Pérez Rodríguez et al. 2013) while it is also known that it attacks Yponomeuta malinellus (Zeller, 1838) (Lepidoptera: Yponomeutidae) (Velcheva et al. 2012). Given that this species parasitises numerous other lepidopterous species belonging to Geometridae, Noctuidae, Plutellidae, and Tortricidae (Nixon 1973), it could be a good candidate for biological control purposes. Whether G. vitripennis parasitises P. oleae, it remains to be confirmed with additional field efforts.

The subfamily Cheloninae is formed by more than 1,300 species belonging to 15 genera, thus constituting a quite large part of Braconidae (Kittel and Austin 2014). They oviposit into eggs and larvae of various lepidopterous species, a fact that makes them valuable potential biocontrol agents (Inayatullah and Naeem 2004; Walker and Huddleston 1987; Edmardash et al. 2011). The subgenus Microchelonus Szepligeti, 1908 is even considered as a valid genus, following the standpoints of Papp (2014a, b). The genus Chelonus Panzer, 1806 counts 601 species in the Holarctic region (Papp 2014c) with M. elaeaphilus being known in the Mediterranean region, either as M. elaeaphilus or C. elaeaphilus (Papp 2012; Nave et al. 2017). This species has been introduced and established in Greece from France (Yamvrias 1998). On the other hand, C. pellucens has a wider European distribution than M. elaeaphilus (van Achterberg 2013). Chelonus pellucens is reported for the first time from Greece and although C. elaeaphilus parasitises P. oleae (Bento et al. 1998), there are no relevant records for C. pellucens, an issue that merits further investigation.

Although Eupelmidae is a relatively small family with approximately 1000 species, the genus Eupelmus Dalman, 1820 is a large taxon containing more than 300 species (Gibson and Fusu 2016) whilst Eulophidae is one of the largest families within chalcidoid wasps, with almost 5,000 species (Aguiar et al. 2013). The genus Pnigalio Schrank, 1802 is comprised by 61 valid species (Li et al. 2017). Several hosts of Pnigalio mediterraneus Ferrière & Delucchi, 1957 (Hymenoptera: Eulophidae) are major pests of plants of ornamental and agricultural importance belonging to different orders, such as B. oleae, Phyllocnistis citrella Stainton, 1856 (Lepidoptera: Gracillariidae), and Cameraria obridella Deschka & Dimić, 1986 (Lepidoptera: Gracillariidae) (Gebiola et al. 2009). Both Eupelmus urozonus Dalman, 1820 (Hymenoptera: Eupelmidae) and P. mediterraneus were found in the Greek island of Corfu as primary parasitoids of B. oleae (Kapatos and Fletcher 1986). Based on our results, these species are also parasitoids of P. oleae that occur in Greece since they were recorded from infested olive leaves.

The genus Diadegma Förster, 1869 constitutes a large group of Ichneumonid wasps with more than 200 known species worldwide (Wagener et al. 2006). Diadegma armillatum is a known parasitoid of various lepidopterous species (Velcheva et al. 2012; Fernandez Triana et al. 2014) that has been recently recorded attacking P. oleae larvae (Nave et al. 2017). The genus Exochus Gravenhorst, 1829 is the largest group of
Metopiinae including the widely distributed in Europe, *E. lentipes* that attacks various Tortricidae and Gelechiidae larvae (Yu et al. 2012).

Our original findings on associated parasitoids of *P. oleae* and the compiled information revealed could trigger further studies that deal with the management of this noxious insect species in the target area from a biological control point of view. The identified parasitoid spectrum was broad, despite the short interval of obtaining the data, indicating a potential positive impact of natural enemies to *P. oleae*, an issue however that merits further field efforts. Last but not least, given that *C. pellucens* is identified as a new member of the entomofauna of Greece during the present first attempt to record the beneficial parasitoids in olive orchards in Crete, we may expect that additional parasitoid species may occur in this agroecosystem.

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