

**DISTRIBUTION AREA AND NATURAL PARASITISM
OF *APANTELES BRUNNISTIGMA* ABDINBEKOVA
(HYMENOPTERA: BRACONIDAE)
ON THE OLIVE LEAF MOTH**

Gülay Kaçar*

* Bolu Abant İzzet Baysal University, Faculty of Natural and Agricultural Science, Department of Plant Protection, Golkoy, Bolu, TURKEY. E-mail: gulaysahan@yahoo.com

[Kaçar, G. 2020. Distribution area and natural parasitism of *Apanteles brunnistigma* Abdinbekova (Hymenoptera: Braconidae) on the olive leaf moth. Munis Entomology & Zoology, 15 (1): 66-72]

ABSTRACT: Olive leaf moth, *Palpita unionalis* Hübner (Lepidoptera: Carambidae) is an important pest in olive groves in Mediterranean countries. It feeds on leaves, shoots, and later green fruits of olive trees. The native larva parasitoid *Apanteles brunnistigma* Abdinbekova (Hymenoptera: Braconidae) was found a solitary endoparasitoid on *P. unionalis* in the eastern Mediterranean and southeastern Anatolia regions. In this study, the distribution and natural parasitism by *A. brunnistigma* on *P. unionalis* were determined in seven provinces (Adana, Gaziantep, Hatay, Kahramanmaraş, Kilis, Osmaniye, and Mersin of Turkey). *P. unionalis* larvae were collected all provinces over its period of occurrence from May to December. The collected larvae were reared until adult parasitoids or moths emerged in the laboratory. The parasitism rate and the number of *A. brunnistigma* per *P. unionalis* larva were estimated for each grove. A total of 2903 *P. unionalis* larvae were collected for two years. The highest natural parasitism rate (58.3%) was observed in Gaziantep and (30%) in Hatay. The parasitism was observed in “summer” (during the period of June through December), with a mean of 9.2-17.6% in 2014-2015, respectively. These findings highlight the need for improved biological control of *P. unionalis* through introduction, augmentation and releasing of the solitary parasitoid *A. brunnistigma*.

KEY WORDS: *Apanteles brunnistigma*, olive, *Palpita unionalis*, parasitism rate

Olive (*Olea europaea* L.), a plant species specific to the Mediterranean climate, is extensively grown along the Mediterranean coasts. It was cultivated about 8000 years ago in Anatolia from where it spreads to the Middle East, North Africa and Southern Europe (Efe et al., 2011). The olive tree has significant socio-economic importance in the Eastern basin where 98% of the world olive production was produced. Turkey holds the fourth country in world olive production with approximately 175.000 thousand trees producing 2.100 thousand ton production of olive fruits after Spain, Italy, and Greece (Faosat, 2016; Tuik, 2018). The olive trees are largely grown in the Aegean, Marmara and Mediterranean Regions, respectively (Tuik, 2018). Biological control agents have increased recently due to inadequate crop management and to avoid hazards of insecticides. Although pesticides are primarily used for olive pests' control, biological control agents are becoming a viable option. Olive trees are attacked by numerous insect pests such as olive the fruit fly, olive moth, olive scales and etc. in Turkey. Among those, the olive leaf moth *Palpita unionalis* Hüb. (Lepidoptera: Crambidae) dramatically causes reduction of green parts of trees especially leaves and green fruits of olive cultivars (*Olea europaea* L.) (Oleaceae) in the Mediterranean countries. During the last decade, *P. unionalis* has recently considered as a common pest on young olive trees and in irrigated olive groves. The olive leaf moth is one of the important olive pests in the Mediterranean region and is now distributed in Italy, Spain, Sweden, Poland, Portugal, Greece,

Turkey, and Iran, Egypt, North Africa, India, the tropical regions of the Americas, and Japan (Avidov & Harpaz, 1969; Balachowsky, 1972; Santorini & Vessiliana-Alexopoulou, 1976; Sevansson, 1988; Kitri & Rose, 1992; Saieb, 1999; Athanassiou et al., 2004; Kovancı & Kumral, 2004; Hertz et al., 2005). This species has recently been known the most common in the Mediterranean region. *P. unionalis* has four and five generations per year; the adults emerge from June to October, mate and oviposit on the fresh leaves (Kaçar & Ulusoy, 2012a). Their larvae feed first on tender leaves and shoots, later green fruits, causing the reduction of green parts.

Cultural and insecticide-based pest management is the suggested method of control (Kaçar & Ulusoy, 2012b). However, due to the ecological and health-related problems of insecticide application, alternative methods based on biological control have been recommended. The native hymenopteran parasitoid, *Apanteles brunnistigma* Abdinbekova (Braconidae) and *Trichogramama evenescens* Westwood. (Trichogrammatidae) were found to have the highest impact on *P. unionalis* populations (Kaçar et al., 2017). They are a particularly significant component in many biological control programs and have been utilized as biocontrol agents into almost every country in the world. *A. brunnistigma* was first recorded in Gökçeada for the Turkish fauna (Inanç & Beyarslandan, 2001), but host species were unknown by 2011. It was determined in Azerbaidzhan, England, Hungary, Italy, Bulgaria, Caucasus, and Western Siberia (Papp, 1980; Tobias, 1995). *A. brunnistigma* was been detected on larvae of *P. unionalis* in south Turkey in the last decade (Kaçar & Ulusoy, 2011). Solitary endoparasitoid, *A. brunnistigma* parasitizes the larvae of *P. unionalis* in the olive groves in the eastern Mediterranean and southeastern Anatolia regions.

Little is known about bio-ecological characteristics and host species of *A. brunnistigma*. The goal of this study was to estimate the parasitism rates larvae by *A. brunnistigma* on *P. unionalis* in the main olive grove producing areas of Mediterranean and southeastern Anatolia.

MATERIALS AND METHODS

Study areas and collection of insects

The study areas cover regions of the Eastern Mediterranean and South Anatolia. Provinces prolongation roughly 20-180 km at its widest point from east to west, with the working area located at an altitude of 12-907 m elevation between 30 and 45 degrees north and south of the equator. These are two of the largest commercial olive growing areas in southern Turkey, where natural surroundings and territory use traditions make the olive view. In the eastern Mediterranean region, the conventional and dense agricultural systems are widespread, whereas the surrounding olive orchards in south Anatolia region are cultivated under traditional farming systems. The generality of the areas with Mediterranean climates have relatively light winters and very warm summers. However, winter and summer temperatures can vary thoroughly different regions with a Mediterranean climate, where is characterized by warm and rainy winters under prevailing westerly winds and calm, hot and dry summer. Temperatures also change from year to year with winter temperatures falling to as low as zero and may rise to as high as over 50 °C in dry areas.

Apanteles brunnistigma was field-surveyed in seven provinces (Adana, Gaziantep, Kilis, Hatay, Kahramanmaraş, Mersin, Osmaniye of Turkey). Samples were collected from olive trees 2014 and December 2015 from 60 areas in Gaziantep (Araban, Oğuzeli, İslahiye, Karkamış, Nizip, Nurdağı, Şahinbey,

Şehitkamil and Yavuzeli), Kilis (Elbeyli, Merkez, Musabeyli and Polateli) in south Anatolia region, Adana (Aladağ, Ceyhan, Feke, Karaisalı, Karataş, Kozan, Sarıçam, Seyhan, İmamoğlu, Yüreğir and Yumurtalık), Hatay (Altınözü, Belen, Dörtiyol, Erzin, Hassa, İskenderun, Kırıkhan, Kumlu, Merkez, Reyhanlı, Samandağ and Yayladağı), Mersin (Aydıncık, Bozyazı, Çamlıyayla, Erdemli, Gülnar, Merkez, Mezitli, Mut, Silifke, Tarsus, Toroslar and Yenice), Kahramanmaraş (Andırın, Merkez, Pazarcık and Türkoğlu), and Osmaniye (Bahçe, Düziçi, Hasanbeyli, Kadirli, Merkez, Sumbas and Toprakkale) in the eastern Mediterranean region.

Olive growing locations were visited in seven provinces monthly with varying intervals between months. Olive leaves and fruits which were infested by the olive leaf moth were collected from olive groves across the provinces (Fig. 1). Sampling was performed according to Lazarov and Grigorov (1961) (Table 1). Each tree was sampled randomly from four main directions (northern, southern, western and eastern) for each sample. At least a hundred twigs with 30 cm length of 25 olive trees from each grove were sampled at each location. The samples consisted of fresh olive branches with larvae of *P. unionalis*, which were removed and placed in paper bags. Samples were transferred to the laboratory in tight closed double paper and a plastic bag in an ice box. Numbers of *P. unionalis* larvae were counted on twigs and separated in the laboratory. The larvae were supplied by clean-fresh host twigs as needed and kept in a bottle of water until emerging all adult wasps. Parasitoids were inspected daily and upon emergence, the date was noted and adults were collected. Olive twigs with *P. unionalis* larvae were reared in growth room maintained at 25±1 C, 16: 8-h light: dark photoperiod, and 70% RH for 4-6 weeks. All *A. brunistigma* adults emerging from each container were collected and recorded.

Parasitism rate and analysis

100 branch pieces of 30 cm each from 25 trees (four shots per tree) for each grove were visually inspected for the presence of *P. unionalis* larvae. The shoots were collected two per each month from all locations. The shoots bearing larvae were gently cut by using a scissor, placed in plastic bags, and were brought to the laboratory where larvae were counted. The shoots with larvae were put in 0.50 l glass cups and given water for each cup. Then they were placed into 20 l clear plastic containers and covered with firm muslin to allow air circulation and prevent escaping the parasitoid adults. Cups were checked the water in cups for two times a week and added water to cups. Cups were checked every day for the emergence of adult moths or parasitoids. After three months, when the containers weren't seen any activity, it could be checked to determine larvae occupation. Alive larvae were returned to their pods and wrapped with moist paper towels to finish development.

Parasitism rate was calculated as the percent of *Palpita* larvae that produced a parasitic wasp (*Palpita* larvae that died of unknown causes before adults emerged were not included). It was calculated according to Russell (1987).

RESULTS AND DISCUSSION

The total of 2903 *P. unionalis* larvae was collected during the study, of which 695 larvae (23.8%) were parasitized by *A. brunistigma* in seven provinces from 163 olive groves. Parasitism rates totally ranged from 30% in Kahramanmaraş and to 100% in Kilis (only one location) for two years (Table 1). The distributions of samples in the provinces and between years are listed in Table 1.

The parasitoid *A. brunnistigma* was thoroughly distributed in Mediterranean olive groves and was determined in 42% of the samples, indicating a mean parasitism rate of 46.2%. In 2015, the samples were not collected in Gaziantep, Kahramanmaraş and Kilis because parasitized larvae were slightly less than in the other four provinces in 2014. In the second year, the funding was not enough to get the samples from Gaziantep, Kahramanmaraş and Kilis. Also, only one parasitoid was determined from Kilis samples, which was not available on a study to keep going the samples from groves. The highest parasitism rate was determined in summer (from June through December), with a mean of 9.2-17.6% in 2014-2015, respectively (Fig. 1). In 2014 and 2015 the percentages of parasitism were higher from May to September in 2015.

Palpita unionalis is present across the Mediterranean area (Egypt, France, Greece, Italy, Israel, Turkey, and Spain), Asia and North Africa (Avidov & Harpaz, 1969; Balashowsky, 1972; Badawi et al., 1976; Fodale et al., 1990; Mazomenos et al., 2002; Kovancı et al., 2006; Kaçar & Ulusoy, 2012b). It is a serious pest of Oleaceae (especially *Olea europaea* L., after that comes the hosts *Jasminum* sp., *Ligustrum* sp., and *Phillyrea media* L.) and Ericaceae (*Arbutus unedo*), as its larvae attack leaves of these plant species (Badawi et al., 1976; Balachowsky, 1972; Hegazi et al., 2007). *P. unionalis* larvae may damage fresh leaves and shoots, causing a dwarf growth of plants and in severe infestations, feed the fruits, thereby heavily affecting the obtained yield. Fruit yield may also be reduced by severe infestations; the reduction rate reaches 30% in late summer and autumn during ripening (Arambourg, 1986; López-Villalta, 1999). *P. unionalis* has recently known as an important pest in commercial olive groves in Turkey. *P. unionalis* adult males and females are active from June to late October and this pest species has 4 to 5 generations per year in the Mediterranean region (Kaçar & Ulusoy, 2012a).

Apanteles brunnistigma Abdinbekova (Hymenoptera: Braconidae) and *Trichogramma evanescens* Westwood (Hymenoptera: Trichogrammatidae) were found as important parasitoids (Kaçar & Ulusoy, 2011). *A. brunnistigma*, was a larval parasitoid of *P. unionalis* activated from late May to November around six months and, the *A. brunnistigma*' parasitism rate was determined from 0.5 to 100% in four locations (Kaçar & Ulusoy, 2017). It was newly reported as a parasitoid of *A. brunnistigma* on *P. unionalis* in the world (Kaçar & Ulusoy, 2011). In the other countries were recorded as larval hymenopteran parasitoids; Eulophidae family, Perilampidae family, Formicidae family, Ichneumonidae family, Chalcididae family, *Chelonus* sp., *Apanteles* sp., *A. obscurus*, *A. xanthostigma*, *A. syleptae*, *A. lacteicolor*, *A. laevigatus*, *A. xanthostigmus* and *Dolichogenidea trachalus* from Braconidae family (Avidov & Rosen, 1961; Triggiani, 1972; Badawi et al., 1976; El-Sherif et al., 1977; El-Hakim et al., 1985; Fodale et al., 1990; Fodale & Mule, 1990; Pinto et al., 1994; Pinto et al., 1995; El-Khawas et al., 2000; Nasr et al., 2002; Shehata et al., 2002; Lababidi & Haj Hammoud, 2008).

As a result, I investigated the seasonal abundance and rate of parasitism of *A. brunnistigma* in different olive growing areas of the eastern Mediterranean region having a Mediterranean climate with hot summer and warm winter and the southeastern Anatolian region having a continental climate with rainy and cold winter. The native larval parasitoid *A. brunnistigma* was thoroughly distributed in olive produce areas and was found the mean parasitism rate of 46.2%. The highest parasitism rate was appeared in summer months, with a mean of 9.2-17.6% for two years. These findings highlight the need for improved biological

control of *P. unionalis* through introduction, augmentation and releasing of the solitary parasitoid *A. brunnistigma*.

ACKNOWLEDGEMENTS

The author wishes to express his deep thank to Dr. Papp Jenő (Department of Zoology, Hungarian Natural History Museum, Hungary) for identification of *Apanteles brunnistigma*. The author thanks is, also, due to Pınar Arıdıcı Kara for help in the computer record.

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Table 1. A total number of *Palpita unionalis* Hb. larvae collected and proportion parasitized larvae by *Apanteles brunistigma* in seven provinces, Turkey.

Location	N. of larvae	2014		2015		Total	
		N	%	N	%	N	%
Adana	Collected	298		221		519	
	Parasitized	73	24.5	93	42.1	166	33.3
Gaziantep	Collected	24		-		24	
	Parasitized	14	58.3	-	-	14	58.3
Hatay	Collected	808		253		1061	
	Parasitized	257	31.8	73	28.9	330	30.4
Kahramanmaraş	Collected	80	□	-		80	
	Parasitized	24	30	-	-	24	30
Kilis	Collected	1		-		1	
	Parasitized	1	100	-	-	1	100
Mersin	Collected	131		48		179	
	Parasitized	54	41.2	17	35.4	71	38.3
Osmaniye	Collected	243		101		344	
	Parasitized	70	28.8	19	18.8	89	33.2

*In 2015, the samples were not collected in Gaziantep, Kahramanmaraş, and Kilis.

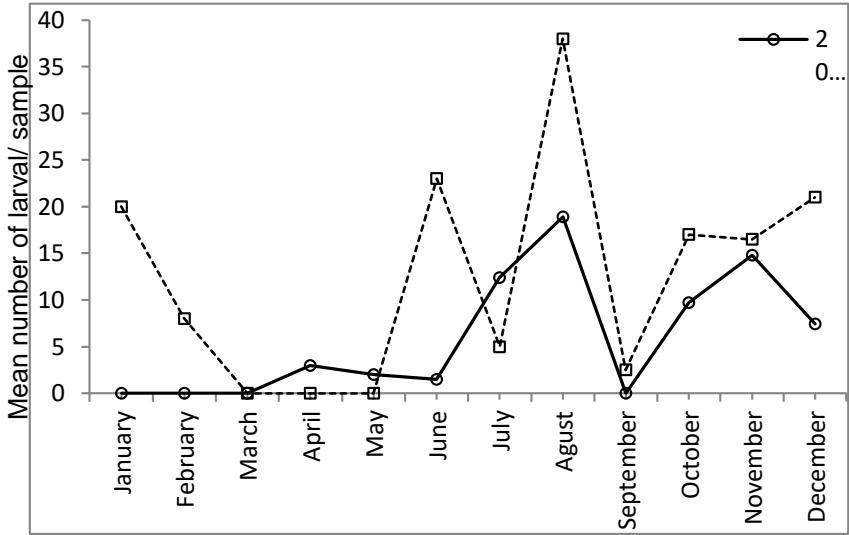


Figure 1. Mean a number of *Palpita unionalis* Hb. larval sample from olive groves in seven locations during the 2014/ 2015.