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### Original article (Orijinal araştırma)

# Investigation of possible use of pheromone trap for adult population development and control of *Ips sexdentatus* (Börner, 1776) (Coleoptera: Scolytidae) damaging black pine in Başkent University Bağlıca Campus afforestation area

Başkent Üniversitesi Bağlıca Kampüsü ağaçlandırma alanında karaçamlara zarar yapan Ips sexdentatus (Börner, 1776) (Coleoptera: Scolytidae)'un ergin popülasyon gelişimi ve mücadelesine yönelik olarak feromon tuzak kullanım olanaklarının araştırılması

### Cem ERDOĞAN<sup>1,2\*</sup>

### **Abstract**

The six-toothed pine bark beetle, *Ips sexdentatus* (Börner, 1776) (Coleoptera: Scolytidae), is one of the important pests of Pinus and Picea species in the forests of Türkiye. Başkent University Bağlıca Campus was established in 1993 and approximately 5 million trees have been planted in the area to date, including those that have dried up. Pheromone traps were hung in campus for monitoring the adult population development, determining the flight activities of the insect and controlling it on IPM principles. The first adult emergence of *I. sexdentatus* in Bağlıca took place in the first half of April, the highest flight activity was in April. *Ips sexdentatus* produced three generations in the region. The highest numbers of adults caught in traps were 741 (27 July 2021) and 421 (25 April 2022), respectively. In the Bağlıca Campus, a total of over 19,000 insects were caught and eliminated. Almost 328 tC of carbon stock was preserved and prevented from being released into nature and at least 11.100 black pine trees were saved or 26.640 USD was contributed to the economy. Using pheromone traps for the control of *I. sexdentatus* is recommended as a sustainable method that protects biodiversity, without any disturbance of the ecological balance.

Key words: Bark beetles, conifers, Ips sexdentatus, pheromone trap

### Öz

On iki dişli çam kabuk böceği, *Ips sexdentatus* (Börner, 1776) (Coleoptera: Scolytidae), Türkiye'deki ormanlık alanlarda Pinus ve Picea türlerinin önemli zararlılarından biridir. Başkent Üniversitesi Bağlıca Kampüsü 1993 yılında kurulmuş ve bugüne kadar alana kuruyanlar da dahil olmak üzere yaklaşık 5 milyon ağaç dikilmiştir. Ergin popülasyon gelişiminin izlenmesi, böceğin uçuş faaliyetlerinin belirlenmesi ve IPM prensiplerine göre kontrol edilmesi için kampus alanına feromon tuzaklar asıldı. *Ips sexdentatus*'un Bağlıca'daki ilk ergin çıkışı Nisan ayının ilk yarısında gerçekleşmiş, en yüksek uçuş aktivitesi Nisan ayında olmuştur. *Ips sexdentatus* bölgede üç döl vermiştir. Tuzaklarda yakalanan en yüksek ergin sayıları sırasıyla 741 (27 Temmuz 2021) ve 421 (25 Nisan 2022) olmuştur. Bağlıca Kampüs alanında toplam 19.000'in üzerinde böcek yakalanarak bertaraf edilmiştir. Yaklaşık 328 tC karbon stoku korunarak doğaya salınması engellenmiş ve en az 11.100 karaçam ağacı kurtarılmış veya ekonomiye 26.640 USD katkı sağlanmıştır. *Ips sexdentatus* mücadelesine yönelik olarak feromon tuzakların kullanılmasının, sürdürülebilir, biyolojik çeşitliliği koruyan ve ekolojik dengeyi bozmayan bir yöntem olarak tavsiye edilmektedir.

Anahtar sözcükler: Kabuk böcekleri, iğne yapraklılar, Ips sexdentatus, feromon tuzağı

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### Introduction

The six-toothed pine bark beetle Ips sexdentatus (Börner, 1776) (Coleoptera: Scolytidae) is considered one of the most dangerous bark beetles in the world. It is a guarantine pest included in the EPPO Annex III quarantine list in the EU. In Türkiye, it is included in the EPPO A2 list (EPPO, 2016) as it is one of the most important bark beetles damaging forest areas. Ips sexdentatus has been reported on the following species; Pinus koraiensis Siebold & Zucc., Pinus leucodermis Ant., Pinus nigra J. F. Arnold, Pinus sibirica Du Tour, Pinus sylvestris L. and Picea orientalis (L.) Peterm (Pinelas: Pinaceae) in Europe, Caucasus, Anatolia, Siberia, Korea, Japan and Northern China (Pfeffer, 1995; Kolk & Starzyk, 1996; Faccoli, 2004; Gilbert et al., 2005; FAO, 2007; Sarikaya et al., 2012; Baydemir, 2016; EFSA, 2017; EPPO, 2023). Ips sexdentatus mainly attacks pines (EFSA, 2017) and was first detected in Türkiye in 1928 by Bernhard in spruce forests in the Trabzon province. Approximately one million of eastern spruce were lost due to sporadic I. sexdentatus infestations in Türkiye between 1928 and 1938 (Defne, 1954; Beşceli & Ekici, 1969). The pest was later detected in different regions of Türkiye and pheromone traps were used for its control (Defne, 1954; Chararas, 1966; Beşceli & Ekici, 1969; Tosun, 1975; Serez, 1984; Selmi, 1989; Sekendiz, 1991; Yüksel, 1996, 1998; Yüksel et al., 2000; Cebeci, 2003; Dönmez, 2006; Eyüpoğlu, 2011; Ozcan et al., 2011; Sarıkaya & Avcı, 2011; Sarıkaya et al., 2012; Akkuzu & Güzel, 2015; Anonymous, 2016; Özcan, 2017a, b; Yiğit, 2017; Özcan et al., 2018; Şahin, 2019).

This pest is mostly a secondary pest, which generally uses stumps, fallen trees and large branches as host material (EFSA, 2017). It is known that bark beetles generally cause damage to trees weakened by wind, frost, drought, etc. (Coşkun et al., 2010). Under normal conditions, bark beetles are known as secondary pests, but when conditions are favorable, they can become primary pests thanks to their ability to reproduce excessively (Çanakçıoğlu & Mol, 1998; Göktürk, 2002; Sarıkaya & Avcı, 2006). As a result of these attacks, *I. sexdentatus* can kill many trees in the same year due to the explosion on its population (Schimitschek, 1953; Christiansen et al., 1987; Jactel & Lieutier, 1987; Jactel & Gaillard, 1991; Öymen & Selmi, 1997; Seedre, 2005; Fettig et al., 2007; Ozcan et al., 2011; Özcan, 2017a). Bark beetles also play an important role in the transmission of pathogenic ophiostomatoid fungi (Levieux et al., 1991; Kirisits, 2004; Romon et al., 2008; Bueno et al., 2010; Jankowiak, 2012). They cause blue staining of the wood and some of them may lead to tree death (EFSA, 2017).

It is known that the host selection of bark beetles, their desire for population growth in their chosen hosts, and their behavior are quite complex (Graves et al., 2008; Pineau et al., 2016; Özcan, 2017a). The adults overwinter in the bark of their hosts or in the litter and disperse in spring, flying in search for new hosts, sometimes over large distances. Six-toothed pine bark beetle adults are capable of flying for miles in search of suitable host trees (Sarıkaya et al., 2012). Jactel & Gaillard (1991) found that 98% flew more than 5 km, 50% more than 20 km and 10% more than 45 km. In Türkiye, *P. sylvestris, P. nigra, Pinus brutia* Ten., *Pic. orientalis, Abies nordmanniana* (Steven) Spach and *Abies bornmulleriana* Mattf. (Pinelas: Pinaceae) are common host trees. They grow in 23 provinces and many districts of Central Anatolia, Eastern Anatolia, Black Sea, Aegean and Mediterranean Regions. The bark beetle can produce 1 to 5 offsprings per year depending on climatic conditions (FAO, 2007; EFSA, 2017). In Türkiye, it usually gives two offsprings per year, but it has been determined that depending on climatic conditions, it can also give three offsprings (Ataman, 1967; Erdem, 1968; Acatay, 1969; Beşçeli & Ekici, 1969; Tosun, 1975; Sekendiz 1984, 1985; Selmi, 1989, 1998; Yüksel, 1997; Çanakçıoğlu & Mol, 1998; Yüksel et al., 2000; Yücel, 2001; Toper Kaygın, 2007; Eyüboğlu, 2011; Güzel, 2018).

Başkent University Bağlıca campus was established in 1993 at a former excavation dumping site. The campus consisted of a rocky area, with only one or two trees on the campus. However, after the establishment of the university, following the cleaning of the excavation waste, afforestation works were conducted regularly every year. Today, more than 5 million different trees such as pine, oak, spruce, etc.

have been planted. Dead trees are replaced by new plants (Anonymous, 2022). In order to prevent possible erosion on the campus area and to create terraces, Başkent University has planted many plants including *Cupressus sempervirens* L., *Cupressus arizonica* Greene (Cupressales: Cupressaceae), *Pinus nigra* subsp. *nigra* var. *caramanica* (Loudon) Businský (Pinelas: Pinaceae), *Acer negundo* L. (Sapindales: Sapindaceae), *Robinia pseudoacacia* Mattf. (Fabales: Fabaceae), *Prunus domestica* L., *Prunus cerasifera* Ehrh., *Amygdalus communis* L. (Rosales: Rosaceae), *Betula verrucose* Ehrh. (Fagales: Betulaceae).

In addition, Başkent University was ranked the 199th Most Environmentally Friendly and Sustainable University in the World and the 13th University of Türkiye in the 2022 UI GreenMetric World Universities Ranking. A totally, 1.050 universities participated in this evaluation all around the world. Başkent University ranks as the first university among Foundation Universities in Ankara. Especially after harsh winter months, it is observed that the black pine trees on the campus area dried up. During examinations, bark beetle entry holes were detected in the dried tree trunks. The samples obtained from the dried trees were identified by the Ministry of Agriculture and Forestry, General Directorate of Forestry as *I. sexdentatus*, a six-toothed pine bark beetle. Since Başkent University Bağlıca campus has a sensitive ecological balance and as chemical control is not preferred in forest areas. The objectives of this study were to trial the use of pheromone traps in monitoring the adult flight periods of *I. sexdentatus*, tracking their population growth, determining the flight activities of the insect, and carrying out integrated control of this insect sustainably to protect biodiversity and the environment. For this purpose, studies were carried out on Başkent University Bağlıca Campus in 2021 and 2022 with the use of Scandinavian type Three Funnel Pheromone Traps.

### **Materials and Methods**

### Geographical location of the research area

The Bağlıca campus of Başkent University is located at Ankara-Eskişehir highway 18 Km in the area of Bağlıca Village (Anonymous, 2023a). It has a total area of 36 ha and an elevation ranging from 930 m to 1150 m (Töre & Erik, 2012). Başkent University Campus is located at 39°53′19.7268" latitude and 32°39′5.5944" longitude (Anonymous, 2023b). Different tree species grow on the campus area including *P. nigra* subsp. *nigra* var. *caramanica*, *Pr. domestica*, *Pr. cerasifera*, *A. communis*, *C. sempervirens*, *C. arizonica*, *A. negundo*, *Astragalus microcephalus* Willd., *Astragalus strictifolius* Boiss., *Astragalus acicularis* Bunge, *Astragalus podperae* Širj., *Astragalus nitens* Host (Fabales: Fabaceae), *Globularia orientalis* L. (Lamiales: Plantaginaceae), *Salvia cryptantha* Montbret & Aucher ex Benth. (Lamiales: Lamiaceae), *R. pseudoacacia* and *B. verrucosa* (Töre & Erik, 2012). This study was conducted out at Başkent University of Bağlıca Campus between 2021 and 2022.

### Climate

Monthly and annual temperature values provided by the nearby Etimesgut Bağlıca station from 2017 to 2022 are given in Table 1. Average temperature on Bağlıca campus is in August (22.92°C) and in January (1.89°C). The annual average temperature is 12.89°C. The maximum temperature is 31.66°C in August, while the minimum temperature is observed in January (-1.41°C). The highest precipitation is in January with 2.41 mm. The driest month is September with 0.17 mm. The average annual precipitation is 1.05 mm.

Maximum average humidity is 93.51% in June, minimum average humidity is 22.36% in August, and annual average humidity is 62.36%. Maximum wind speed is 25.32 km/h in January and minimum wind speed is 3.87 km/h in October, with an annual average wind speed of 12.18 km/h. and it is understood that the highest precipitation falls in winter (4.52 mm), followed by fall (3.38 mm), spring (3.19 mm) summer (0.72 mm) (Table 1).

Table 1. Bağlıca meteorological data (averaged data between 2017 and 2022)

Meteorological data	Months										Annual		
	I	Ш	III	IV	V	VI	VII	VIII	IX	Х	ΧI	XII	average
Average Temperature (°C)	1.89	4.93	6.98	11.98	16.25	19.55	22.07	22.92	19.11	14.45	9.45	5.42	12.89
Maximum Temperature (°C)	5.49	9.46	12.30	18.30	22.86	25.98	30.19	31.66	27.40	21.68	15.49	9.63	19.15
Minimum Temperature (°C)	-1.41	0.88	1.79	4.98	8.61	12.15	13.06	13.64	10.96	7.66	4.09	1.90	6.54
Average RH (%)	78.56	72.54	63.00	56.94	59.18	63.38	56.41	52.70	53.01	55.55	60.74	73.99	62.36
Maximum RH (%)	91.65	89.79	85.04	85.29	89.55	93.51	92.65	90.01	86.47	83.84	83.76	90.20	88.55
Minumum RH (%)	61.41	53.09	41.09	33.57	34.39	36.55	26.52	22.36	24.14	29.46	37.27	54.24	38.06
Total precipitation (mm)	2.41	1.00	1.11	0.80	1.36	1.03	0.25	0.30	0.17	0.39	1.05	1.94	1.05
Maximum wind speed (km/h)	25.32	21.68	24.48	22.11	19.99	18.49	22.92	22.46	19.74	17.28	16.40	19.03	20.70
Minumum wid speed (km/h)	6.19	4.83	5.79	5.01	4.26	4.62	5.45	4.94	4.06	3.87	3.88	4.23	4.73
Average wind speed (km/h)	15.15	13.09	14.66	13.22	12.09	11.30	13.04	12.39	11.44	10.37	10.05	10.89	12.18
Wind direction (°)	210.5	202.7	215.3	213.9	230.1	211.4	182.0	151.8	204.9	178.4	155.8	190.4	194.3

### Adult population monitoring of Ips sexdentatus

This study was conducted at Başkent University Bağlıca Campus' afforestation area between 2021 and 2022 in order to determine the basic biology, population growth, and flight periods of *I. sexdentatus* and to study possibilities of sustainable control of the pest by mass trapping. The main goal was to keep *I. sexdentatus* below the damage level by hanging pheromone traps in the study area. In this context, IPSSEX pheromone dispenser packages containing 60 mg of Ipsdienol as active substance, which is licensed in Türkiye, and Scandinavian type three funnel pheromone traps (for mass capture) were hung (Figure 1). The first bark beetle aggregation pheromone was identified in *I. paraconfusus* as a mixture of ipsdienol, ipsenol and cis-verbenol (Silverstein et al., 1966). The IUPAC name for ipsdienol, 2-methyl-6- methylene-2,7-octadien-4-ol. Detailed information about ipsdienol has documented by Blomquist et al. (2010).



Figure 1. Scandinavian type pheromone trap with three funnels.

Pheromone traps were placed in the field according to the recommendation on the label, with 4 traps per 1 ha area and the entire study area covering 2.5 ha (2021) and 5 ha (2022), respectively. The traps were hung at a height of 150-170 cm above the ground as stated in the label recommendation (Şahin, 2019), in the direction of the prevailing wind, periodically monitored and the developmental stages of the insect were observed and checked every 10 days. The study started on 17.07.2021 as soon as the pest was detected in the field (Figure 2a, b).





Figure 2. Ips sexdentatus: a) damage and b) larvae.

### Results

### Determination of adult population development of Ips sexdentatus

The flight activity of I. sexdentatus and adult population development of the pest between 2021 and 2022 on the Baskent University Bağlıca Campus are given in Figures 3 & 4. During the 2021 trials, the pheromone traps were installed on 17.07.2021. The counts showed that until the end of the season, there was a high density population. When Figure 3 is examined, two peaks are visible on 27 July and 18 October 2021 with the highest adult population recorded at 741 pieces/trap and 516 pieces/trap, respectively. In 2021, the average number of adult beetles caught in the traps during the season was determined as 1.257 per trap. In 2021, 3.120 adults were collected from traps on 27 July. On this date, the average temperature was 22.35°C, the average humidity was 57.7% and the average wind was 21.87 km/h. The peak on 27 July 2021 was understood to be the first generation of the pest and 3.120 beetles were collected from the traps. In the following period, the number of adults decreased until 10 August 2021, when the average temperature was between 18.12°C and 29.89°C, the average humidity between 35.3% and 67.3% and the average wind between 10.33 and 20.15 km/h. From 10 August to 15 September 2021, when the mean temperature ranged from 15.21 to 27.76°C, the mean humidity from 32.5% to 65.2% and the mean wind from 6.95 to 23.35 km/h, an increase in the number of adults was again observed. From 15 September to 04 October 2021, an increase in the number of adults was again detected. However, it is assumed that these increases are not due to a new generation but are probably related to the increase in temperature.

It was determined that the second generation of adults appeared on 18 October 2021, when the average temperature increased to 11.58°C and the average humidity increased to 73.2% as 2.316 adults were caught in traps. The average wind was 5.43 to 17.99 km/h. Adult flight started to decrease after October 18, 2021 and ended on November 03, 2021 in Bağlıca Campus in 2021.

In 2022, pheromone traps were hung in the campus area on 9 April and the first adult flight was observed on 16 April (Figure 4). On this date, the average temperature was 14.10°C, the average humidity was 48.29% and the average wind was 8.6 km/h. On 25 April 2022, a total of 3.199 first-generation adults were collected from the traps. On this date, the average temperature was 19.40°C and the average humidity was 49% with an average wind speed of 12.6 km/h. In the following period, there was a slight decrease in the number of adults until 23 May 2022, when the average temperature was between 8.20°C and 19.40°C, the average humidity was 39.83% to 79.13% and the average wind was 6.5 to 27.7 km/h. From 23 May to 08 June 2022, the mean temperature ranged from 8.76°C to 24.87°C, the mean humidity from 37.63% to 77.38%, the mean wind from 7.6 to 16.6 km/h and the number of adults increased again. From 08-20 June 2022, a decrease in the number of adults was observed. On 8 June and 6 September 2022, two significant population increases were detected. However, these increases were not due to a new generation but probably related to the increase in temperature.

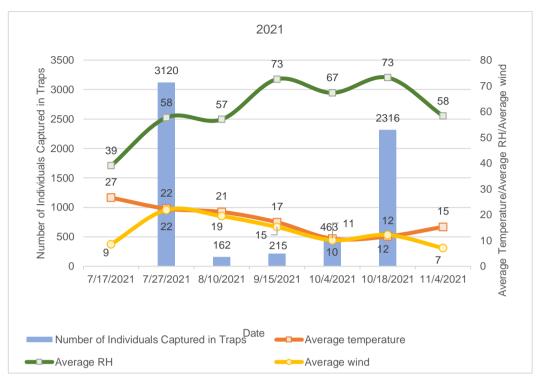


Figure 3. Adult population development and flight activity of Ips sexdentatus on Başkent University Bağlıca Campus in 2021.

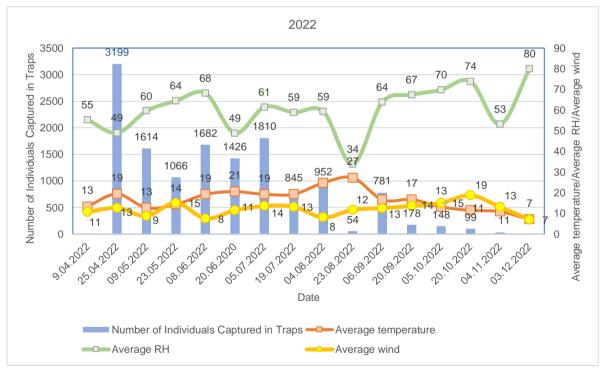


Figure 4. Adult population development and flight activity of *Ips sexdentatus* on Başkent University Bağlıca Campus in 2022.

The second generation of adults occurred on 05 July 2022, when the average temperature was 20.56°C, the average humidity 48.83% and the average wind 11.5 km/h. 1.810 beetles were caught in the traps. It was determined that a third generation of the pest occurred on 4 August 2022, when the average temperature was 24.79°C, the average humidity was 59.29% and the average wind was 8.1 km/h. 952

beetles were caught in the traps. In 2022, the highest number of adults caught for *I. sexdentatus* was on 09 May with 417 beetles per trap on average, followed by 26 June with 390 beetles per trap on average, and the number of adults caught in traps during the entire season was on average 1.892 per trap. On 26 and 30 October 2022, there were days when the minimum temperature dropped down to -0.39°C to -2.19°C and the number of adults decreased until 04 November 2022. During this period, the average humidity was 33.2% to 72.6% and the average wind was 5.43 to 17.99 km/h.

In order to determine the number of generations of the pest, Figure 4 shows that the pest population formed three main peaks on Bağlıca Campus. These peaks occurred on 25 April, 05 July and 04 August 2022. This is indicative that the pest produced three generations. To confirm, the number of generations of the pest was calculated also using meteorological data from the region. Pest development threshold and thermal constant values found by Pineau et al. (2016) were used in this calculation. Using the development threshold (10.9°C) and maximum development temperature (36°C) and thermal constant (517 day-degree) values of the pest, the daily average temperatures above 36°C were excluded from the daily average temperatures in Bağlıca district. The remaining daily average temperatures were subtracted from the development threshold temperature of 10.9°C and the remaining values were summed for the whole year and the sum of effective temperatures was calculated as 1.548,73 degree-day. When this value was divided by thermal constants, it was determined that *I. sexdentatus* could produce 2.99 generation in Bağlıca district. Considering the findings of Pineau et al. (2016), the number of generations in the Bağlıca district of Başkent University is three, which is compatible with the peaks observed in the traps.

In summary, 5.899 adults were caught in 10 traps in 2021, while 13.560 adults were caught in 20 traps in 2022. This shows how effective the control activities carried out with pheromone traps are. When the average temperature was above 10.9°C (Pineau et al., 2016), which is the development threshold of the pest, it was observed that the active flight period started. The highest number of adults caught in traps was on average 741 beetles/trap (27 July 2021) and 421 beetles/trap (25 April 2022), respectively. During the study, also the beneficial insect *Rhizophagus dispar*, the predator of *I. sexdentatus*, was caught in pheromone traps in the campus area, detected and then released back into the field.

### **Discussion**

The present study determined that the first adult emergence of *I. sexdentatus* in the Başkent University Bağlıca campus took place in the first half of April and the pest population produced three peaks on 25 April, 5 July and 04 August, equal to three generations of bark beetles. This value was confirmed by calculations using meteorological data. The active flight period spans seven to eight months a year until end of November when the weather gets colder. Selmi (1998) stated that *I. sexdentatus* normally produces two generations per year in Türkiye and can give a third generation under suitable weather conditions. Sarıkaya (2008) determined that in the Western Mediterranean Region and Yıldırım (2011) in the forests of the Isparta-Aksu region that *I. sexdentatus* gives three generations per year. In our study, it was determined that *I. sexdentatus* gives three generations per year, which is in accordance with the studies of Selmi (1998), Sarıkaya (2008) and Yıldırım (2011). The first pheromone trap trials against *I. sexdentatus* started in 1982 in oriental spruce forests in Türkiye (Serez, 2001).

Chararas (1966) states that a total of 16.000 offsprings can occur over three generations from one female insect, assuming that the sex ratio is 1:1 (Pineau et al., 2016), mortality rate is 17% due to unopened eggs and losses due to parasitoids and predators, and a maximum of 13.280 pests could occur in the 3<sup>rd</sup> generation. The number of 13.560 adult beetles caught during this study is in line with the assumption that three generations were produced. Obtained data showed the importance of ambient temperature and humidity as well as light and moderate wind speed (Anonymous, 2023c) to the spread of *I. sexdentatus*. It is known that *I. sexdentatus* beetles have the ability to fly for kilometers in search of suitable host trees (Sarıkaya et al., 2012) and it is understood that they can increase their flight distance and spread with the help of wind (EFSA, 2017).

To evaluate the environmental and economic dimension of the present study, in order to reveal the success of the mass trapping method, the carbon stock in black pine trees was determined as 65.776 (2.225-119.686) tC/ha on average by the General Directorate of Forestry of the Ministry of Agriculture and Forestry of Türkiye (Anonymous, 2015). Considering that the study area was 5 ha, it was determined that at least 328 tC of carbon stock was preserved and prevented from being released into nature. Specific economic loss is not known for *I. sexdentatus*. However, considering that there can be 2.220 to 3.100 black pine trees in 1 ha area (Anonymous, 2014), it was determined that at least 11.100 black pine trees were saved or 26.640 USD (value of one seedling tree is assessed as 12 USD). If carbon stock is included in these figures, the economic dimension will increase even more.

Another important finding was that although the pheromones in the traps were recommended to be changed at intervals of 4-6 weeks by the registered company, it was evaluated that it is better to replace the pheromones in the traps every 4 weeks during the very hot weather conditions in order to maintain the effectiveness of the pheromone traps.

*Ips sexdentatus* spends its entire developmental period in plant tissue and feeds mostly on physiologically weakened trees. Trees are host to *I. sexdentatus* as a result of storm breakage and toppling, snow breakage, fires, large-scale eating of leaves by other insects and extreme droughts (Yüksel, 1996). Although *I. sexdentatus* is generally seen as secondary pest that causes growth retardation and loss of increment in coniferous forests, it can cause large damage when appearing in clusters, especially in young affforestration areas where closure is newly formed, and in some cases, it can completely destroy the afforestration site (Anonymous 2016). Therefore, Bağlıca Campus, which is a relatively young afforestration campus, is under serious threat from *I. sexdentatus*.

In the current study, 5.899 beetles were caught with the help of pheromone traps managed in Bağlıca Campus during a 4-months period starting from 17 July 2021, while this number increased to 13.560 beetles when repeating the trials as of 09 April 2022 conducted over a doubled area with twice the number of traps. It is reported that *I. sexdentatus* females lay between 10-60 eggs. Since the sex ratio is 1:1, this control effort prevented the population from reaching higher levels and reduced the number of existing ones, and successful control was performed with non-chemical methods in the forest area (Yüksel et al., 2005).

Bağlıca Campus, which is a young afforestration area, is under serious threat from *I. sexdentatus*. Since the number of trees in the Bağlıca campus increases every year, important data were obtained in this study to determine the population density of *I. sexdentatus* in the afforestation area and whether it was possible to control it with mass trapping method. The trials also showed that the pest has a very high damage potential unless precautions are taken. As the mass trapping method with the use of pheromone traps is specific to the pest species, it has no negative effects on beneficial organisms such as parasitoids, predators, honey bees and other pollinator insects, vertebrate animals and humans. In addition, this method is compatible with the environment, and does not require special tools and equipment, while it can be reused, and help reduce the use of chemical pesticides against pests. It is understood that use of pheromone traps helps beneficial insects to resettle and develop in the area. At the same time, since it minimizes or eliminates the use of pesticides, it contributes to the re-establishment of the natural balance in that area as soon as possible, which was previously disrupted due to the use of chemical pesticides (Anonymous, 2016). In spite of the advantages mentioned above, there is a disadvantage that natural enemies may fall into the trap (Wainhouse, 2005), like the beneficial insect *Rhizophagus dispar* detected in the study area.

Pheromone traps have been used for *I. sexdentatus* control in Türkiye and this project result is supported by other researches based on field findings (Defne, 1954; Chararas, 1966; Beşceli & Ekici, 1969; Tosun, 1975; Serez, 1984; Selmi, 1989; Sekendiz, 1991; Yüksel, 1996, 1998; Yüksel et al., 2000; Cebeci, 2003; Dönmez, 2006; Eyüpoğlu, 2011; Ozcan et al., 2011, 2017a, b, 2018; Sarıkaya & Avcı, 2011; Sarıkaya et al., 2012; Akkuzu & Güzel, 2015; Anonymous, 2016; Yiğit, 2017; Şahin, 2019).

In conclusion, in order to protect forest/afforestation areas against pests like *I. sexdentatus*, it is important to monitor the pest population at regular intervals and to take timely measures for its control. It is essential to regularly monitor the population level of the six-toothed pine bark beetle, *I. sexdentatus*, and within the scope of integrated pest control, it is possible to use pheromone for mass trapping, which provides a sustainable method protecting biodiversity and does not have a negative effect on natural enemies in the environment.

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### Original article (Orijinal araştırma)

## Aphid (Hemiptera: Aphididae) species on the herbaceous host plants in the Tekirdağ Province (Türkiye)<sup>1</sup>

Tekirdağ ilindeki (Türkiye) otsu konukçu bitkilerde görülen yaprak biti (Hemiptera: Aphididae) türleri

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### **Abstract**

This research was conducted between 2018 and 2020 in Tekirdağ province and its districts in Türkiye, aiming to identify species of aphids belonging to the Aphididae family on the herbaceous plants in non-agricultural and agricultural fields, urban parks, gardens, and roadsides. The morphological diagnoses of aphids were performed, and based on that, the species from the subfamilies of the Aphididae family belonging to the Hemiptera order, namely Aphidinae, Eriosomatinae, Lachninae, and Chaitophorinae, were identified. *Echinophora tenuifolia* (L.) and *Cachyrs* sp. (L.) (Apiaceae) for *Anuraphis cachyros* (Barbagallo & Stroyan, 1982), *Malva neglecta* (Wallr.) (Malvaceae) for *Aphis* (*Aphis*) *nasturtii* (Kaltenbach, 1843), *Cichorium intybus* (L.) (Asteraceae) for *Hyperomyzus* (*Hyperomyzus*) *lactucae* (L., 1758) *Achillea* sp. (L.) (Asteraceae) for *Macrosiphoniella* (*Macrosiphoniella*) *tanecetaria* (Kaltenbach, 1843), *Amaranthus retroflexus* (L.) (Amaranthaceae) for *Macrosiphum* (*Macrosiphum*) *euphorbiae* (Thomas, 1878) and *Slybum marianum* (L.) (Asteraceae) for *Uroleucon* (*Uromelan*) *aeneum* (Hille Ris Lambers, 1939) have been identified as new host plants in Türkiye.

Keywords: Aphid fauna, Aphidoidea, new host records, Tekirdağ

### Öz

Bu araştırma, Türkiye'nin Tekirdağ ili ve ilçelerinde 2018-2020 yılları arasında tarım dışı alanlar, tarım alanları, kentsel bölgelerdeki parklar, bahçeler ve yol kenarlarındaki tek yıllık kültür bitkiler üzerinde Aphididae familyasına ait yaprakbiti türlerini belirlemek amacıyla yürütülmüştür. Yaprakbitlerinin morfolojik olarak teşhisleri yapılmış ve buna göre Hemiptera takımına bağlı Aphididae familyasının altfamilyaları olan Aphidinae, Eriosomatinae, Lachninae ve Chaitophorinae tespit edilmiştir. *Echinophora tenuifolia* (L.) ve *Cachyrs* sp. (L.) (Apiaceae) *Anuraphis cachyros* (Barbagallo & Stroyan, 1982) için; *Malva neglecta* (Wallr.) (Malvaceae), *Aphis* (*Aphis*) *nasturtii* (Kaltenbach, 1843) için; *Cichorium intybus* (L.) (Asterales: Asteraceae), *Hyperomyzus* (*Hyperomyzus*) *lactucae* (L., 1758) için; *Achillea* sp. (L.) (Asteraceae), *Macrosiphoniella* (*Macrosiphoniella*) *tanecetaria* (Kaltenbach, 1843) için; *Amaranthus retroflexus* (L.) (Amaranthaceae), *Macrosiphum* (*Macrosiphum*) *euphorbiae* (Thomas, 1878) için *ve Slybum marianum* (L.) (Asteraceae), *Uroleucon* (*Uromelan*) *aeneum* (Hille Ris Lambers, 1939) için Türkiye'de yeni konukçu kayıtları olarak belirlenmiştir.

Anahtar sözcükler: Yaprakbiti faunası, Aphidoidea, yeni konukçu kayıtları, Tekirdağ

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### Introduction

The geographical region and localization of Türkiye create an important area in terms of invasive species. Therefore, studies aimed at identifying the aphid fauna in Türkiye are of importance in terms of biodiversity, ecological, and applied sciences. Due to its unique features, such as being the homeland of many plants owing to the advantages offered by its biogeographic characteristics, the abundance of endemic plant species, the presence of different climate types, and intercontinental transitions, Türkiye constitutes an important area in terms of distribution and diversity of aphids. The biogeographic location and characteristics, diverse climatic features, microclimatic variations, acting as a transition zone between continents, richness in terms of flora, and diversity of agricultural products make aphid studies in Türkiye highly significant. The global aphid fauna currently comprises 5942 valid taxa including subspecies distributed across 538 genera and classified into 25 subfamilies (Favret, 2023).

As of now, Türkiye's aphid fauna has been documented to include 631 aphid species belonging to 169 genera (Görür et al., 2023). The first studies on the aphid fauna in Türkiye were conducted by foreign researchers such as Trotter (1903), Houard (1922), and Fahringer (1922). Researchers such as Görür et al. (2009, 2014, 2018), Kök & Özdemir (2021), Patlar et al. (2021) and Şenol et al. (2021), among others, have conducted comprehensive studies. Due to the increasing significance of agricultural production in Türkiye and the adverseeffectsof global climate change, determining the current status of Türkiye's aphid fauna is of significant importance. Therefore, this study aimed to reveal the aphid fauna of Tekirdağ province.

### **Materials and Methods**

Field outings were conducted weekly between March and June to collect aphids, and during the summer and autumn months when aphids were less abundant, the collection was carried out every 15 days in July and November. During sampling, attempts were made to collect as many winged and wingless adult individuals as possible. A code number has been assigned to each sample. Furthermore, information such as the color prior to preservation in alcohol, date, host plant, and collection location were recorded in the field notebook. The generated code numbers were placed as labels, written with a pencil, inside the cryo tubes containing the aphid samples. Plants that were found to be infested with aphids as a result of inspections on parts such as roots, stems, shoots, and leaves were first wrapped in newspaper to prevent moisture and then placed in polyethylene bags. They were subsequently transported to the laboratory in an icebox.

Aphids were collected from their host plant in the field using a small soft brush and placed in a tube containing 96 % ethyl alcohol. The collecting and preservation techniques used were based mainly on the method of Hille Ris Lambers (1950). Specimens were examined with a LEICA DM LB2 compound light microscope, and morphological features were measured using LAS version 4.1 software. The measurements of morphological characteristics were made according to Hille Ris Lambers (1945, 1947 a, b, 1949, 1969, 1973), Börner (1952), Cottier (1953), Bodenheimer & Swirski (1957), Börner & Heinze (1957), Stroyan (1957, 1961, 1963, 1977, 1984), Shaposhnikov (1964), Bissel (1978) and Blackman & Eastop (1984, 1994, 2000, 2020). All collected aphid species permanent slides have been stored at Department of Plant Protection within the Faculty of Agriculture at Tekirdağ Namık Kemal University and Plant Protection Central Research Institute in Ankara.

### **Results and Discussion**

The identification of the aphids was based on their morphological characteristics, and as a result in Tekirdağ, the subfamilies Aphidinae, Chaitophorinae, Eriosomatinae, and Lachninae, which are part of the Aphididae family, were identified along with their respective host plants. A total of 83 taxa including 1 subspecies of aphids have been diagnosed, and their descriptions are provided in more detail below.

### Family Aphididae

### Subfamily, Aphidinae

### Acyrthosiphon (Acyrthosiphon) lactucae (Passerini, 1860)

Material examined. Tekirdağ, Çorlu, Yenice, 28.V.2019, apt. 399, alt. 99, Lactuca serriola L. (Asteraceae); Tekirdağ, Kapaklı, Karlı, (41°22'09.9"N, 27°51'53.0"E), 01.VI.2019, apt. 499, alt. 299 Euphorbia sp. (Jussieu) (Euphorbiaceae).

General distribution. North America, Argentina and Chile (Blackman & Eastop, 2023).

### Acyrthosiphon (Acyrthosiphon) pisum (Harris, 1776)

General distribution. It has an almost world-wide distribution (Blackman & Eastop, 2023).

### Ammiaphis sii (Koch, 1855)

Material examined. Tekirdağ, Malkara, Güneşli, (41°01'26.1"N, 26°54'34.5"E) 22.VI.2019, apt. 3  $\bigcirc$   $\bigcirc$  F. *vulgaris* (Bernh.) (Apiaceae); Marmaraereğlisi, Sultanköy, (41°00'55.3"N, 27°58'31.7"E), 18.VIII.2018, apt. 2  $\bigcirc$   $\bigcirc$   $\bigcirc$  Falcaria sp. (Fabr.) (Apiaceae).

General distribution. South-west and central Asia, central and eastern Europe (Blackman & Eastop, 2023).

### Anuraphis cachryos (Barbagallo & Stroyan, 1982)

Material examined. Tekirdağ, Malkara, Hasköy, (40°56'00.6"N, 26°49'19.4"E), 26.VIII.2018, apt. 2  $\bigcirc$  , alt.  $\bigcirc$  , *E. tenuifolia* (Apiaceae); Saray, Pazarcık, (41° 26' 55.38 4"N, 26° 49'18.06"E), 05.IX.2018, apt. 3  $\bigcirc$  , *Cachrys* sp. (Apiaceae); Süleymanpaşa Hürriyet, (40°58'54.8"N, 27°32'41.7"E), 04.X.2019, apt.  $\bigcirc$  , *E. tenuifolia*.

General distribution. In southern Italy and recorded from Iran and Türkiye (Blackman & Eastop, 2023).

### Anuraphis subterranea (Walker, 1852)

Material examined. Tekirdağ, Muratlı, İnanlı, (41°12'09.0"N, 27°28'21.4"E), 07.V II.2019, apt. 299, alt. 299, Heracleum sp. L. (Apiaceae); Şarköy, Bulgur, (40°44'34.4"N, 27°08'23.6"E), 05.VII.2018, apt. 399, Heracleum sp.

General distribution. Western Siberia and Kazakhstan, throughout Europe, North Africa, Iran, (Blackman & Eastop, 2023).

### Aphis (Aphis) affinis (Del Guercio, 1911)

General distribution. Middle East, Central Asia, India, Pakistan, in Europe and southern Russia, (Blackman & Eastop, 2023).

### Aphis (Aphis) brotericola (Mier Durante, 1978)

Material examined. Tekirdağ, Muratlı, Aydınköy, 08.VI.2018, apt. 299, alt. 99, Euphorbia sp. (Euphorbiaceae); Saray, Kavacık, (41°29'03.6"N, 27°54'03.7"E), 21.V.2019, apt. 99, alt. 99, alt. 99, Convolvulus arvensis L. (Convolvulaceae); Süleymanpaşa, Hürriyet, 29.VI.2020, apt. 99, alt. 99, alt. 99, C. arvensis.

General distribution. In Spain, Italy, France, Greece, Türkiye, Iran and Morocco (Blackman & Eastop, 2023).

### Aphis (Aphis) craccivora (Koch, 1854)

General distribution. Worldwide, but particularly common in warm temperate and tropical regions (Blackman & Eastop, 2023).

### Aphis (Aphis) euphorbiae (Kaltenbach, 1843)

Material examined. Tekirdağ, Ergene, Esenler, 08.V.2018, apt. 4  $\ \ \,$  alt. 2  $\ \ \,$  alt. 2  $\ \ \,$  Euphorbia sp. (Euphorbiaceae); Süleymanpaşa, Naip, 06.VI.2018, apt. 3  $\ \ \,$  alt. 2  $\ \ \,$  Euphorbia sp.; Süleymanpaşa, Hürriyet, 15.VI.2020, apt. 4  $\ \ \,$  alt.  $\ \ \,$  Euphorbia sp. and Şarköy, Mürefte, 12.V.2019, apt. 3  $\ \ \,$  alt.  $\ \ \,$  Euphorbia sp.

General distribution. South-west and central Asia, in Europe, western Siberia, and introduced to USA (Blackman & Eastop, 2023).

### Aphis (Aphis) fabae (Scopoli, 1763)

Material examined. Tekirdağ, Çorlu, Yenice, 01.VIII.2018, apt. ♀, alt. ♀, Helianthus annuus L. (Asteraceae); Ergene, Esenler, (41°13'25.8"N, 27°38'49.1"E), 08.V.2018, apt. 2♀♀, alt. 2♀♀*Papaver* rhoeas L. (Papaveraceae); Kapaklı, Mimar Sinan, (41°17′11.5″N, 27°57′04.5″E), 02.VI.2018, apt. ♀, alt. ♀, Heracleum austriacum L. (Apiaceae); Malkara, Doğanköy, 15.VIII.2018, Amaranthus retroflexus L. (Amaranthaceae); Malkara, Halıç, 06.VIII.2018, apt. 3♀♀, Mentha piperita L. (Lamiaceae); Marmaraereğlisi, Bahçelievler, 09.X.2019, apt. ♀, alt. ♀, C. album (Amaranthaceae); Marmaraereğlisi, Sultanköy, 18.VIII.2018, apt. 2♀♀, alt. ♀, Cirsium arvense L. (Asteraceae); Muratlı, İnanlı, 09.V.2019, apt. 3♀♀, Anthemis arvensis L. (Asteraceae); Süleymenpaşa, Değirmenaltı, (40°559'13.1"N, 27°34'54.7"E), 11.VIII.2018, apt. ♀, alt. ♀, O. acanthium (Asteraceae); Süleymanpaşa, Hürriyet, 15.IV.2019, apt. 299, alt. 9, Rumex sp. (Polygonaceae); Süleymanpaşa, Hürriyet, 04.VI.2020, apt. Q, alt. Q, Galium sp. (Rubiaceae); Süleymanpaşa, Hürriyet, 06.V.2019, apt. 2♀♀, alt. ♀, Salvia sp. (Lamiaceae) and M. neglecta (Malvaceae): Süleymanpaşa, Hürriyet, 06.V.2019, apt. ♀, alt. ♀; Şarköy, Çengelliköy, (40°40'47.0"N, 27°09'23.6"E), 23.V.2019, apt. 2♀♀, alt. ♀, Cynara cardunculus L. (Asteraceae) and apt. ♀, alt. ♀, S. marianum (Asteraceae); Şarköy, Eriklice, (40°38'15.0"N, 27°10'55.5"E), 06.V.2019, apt. 2♀♀, alt. 2♀♀, Carduus sp. L. (Asteraceae); Şarköy, Uçmakdere, (40°47′59.0″N, 27°21′48.2″E), 03.VIII.2018, apt. 2♀♀, S. marianum (Asteraceae) and apt. 499, alt. 299, *Rumex* sp. L. (Polygonaceae).

General distribution. They are found all over the world, including Africa, Asia, Europe, India, Canada, Korea and North America (Blackman & Eastop, 2023).

### Aphis (Aphis) fabae cirsiiacanthoidis (Scopoli, 1763)

Material examined. Tekirdağ, Malkara, Gazibey, 20.V.2019, apt. 4  $\$ 0, S. *marianum* (Asteraceae); Şarköy, Uçmakdere, 03.VII.2018, apt. 2  $\$ 0, S. *marianum*; Şarköy, Ulaman, (40°42'45.9"N, 27°05'07.1"E) 14.V.2019, apt. 3  $\$ 0, alt.  $\$ 0, S0, arvense (Asteraceae).

General distribution. No information was found about its distribution, except for (Stroyan, 1984; Aslan, 2002).

### Aphis (Aphis) galiiscabri (Schrank, 1801)

Material examined. Tekirdağ, Malkara, Danişment, (40°52'15.8"N, 26°44'25.6"E), 02.VIII.2018, apt. 3♀♀, *Galium* sp. L. (Rubiaceae); Şarköy, Çengelliköy, 06.V.2019, apt. 4♀♀, alt. 2♀♀, *Galium album* Mill.

General distribution. In Europe, west Siberia, Türkiye, Iran and Central Asia (Kazakhstan), Mongolia and Canada (Blackman & Eastop, 2023).

### Aphis (Aphis) gossypii (Glover, 1877)

Material examined. Tekirdağ, Kapaklı, Mimar Sinan, 02.VI.2018, apt. 299, alt. 9, *M. neglecta* (Malvaceae); Kapaklı, Pınarca, (41°520'20.6"N, 27°54'05.3"E), 02.VI.2018, apt. 9, alt. 9, alt. 9, *P. rhoeas* (Papaveraceae); Malkara, Güneşli, 06.VII.2018, apt. 99, alt. 99, a

General distribution. They are found almost everywhere in the world and are especially common in the tropics, including the islands in the Pacific (Blackman & Eastop, 2023).

### Aphis (Aphis) intybi (Koch, 1855)

Material examined. Tekirdağ, Çerkezköy, Kızılpınar, (41°16'00.2"N, 27°57'52.3"E), 18.VII.2018, apt. 299, alt. 9, *Cichorum* sp. L. (Asteraceae); Şarköy, Bulgur, 14.VII.2019, apt. 399, *Cichorum intybus* L. (Asteraceae).

General distribution. West and central Asia east to Pakistan, in Europe, Mediterranean region (Blackman & Eastop, 2023).

### Aphis (Aphis) nasturtii (Kaltenbach, 1843)

General distribution. Now almost world-wide (but not yet in Australasia) (Blackman & Eastop, 2023).

### Aphis (Aphis) nerii (Boyer de Fonscolombe, 1841)

Material examined. Tekirdağ, Kapaklı, Pınarca, (41°520'20.6"N, 27°54'05.3"E), 01.VI.2019, apt. 3, alt. 2, Euphorbia sp.; Süleymanpaşa, Karaevli, (41°02'16.5"N, 27°39'56.7"E), 22.V.2019, apt. 2, Cynanchum acutum (L.) (Apocynaceae); Şarköy, Mürefte, 20.V.2018, apt. 6, Euphorbia sp. (Euphorbiaceae).

General distribution. Found throughout the world, especially prevalent in tropical and subtropical regions, which include a multitude Pacific islands (Blackman & Eastop, 2023).

### Aphis (Aphis) plantaginis (Goeze, 1778)

General distribution. In Europe, Central Asia, west and east Siberia, Mongolia (Blackman & Eastop, 2023).

### Aphis (Aphis) polygonata (Nevsky, 1929)

Material examined. Tekirdağ, Malkara, Danişment, 02.VIII.2018, *Polygonum* sp. L. (Polygonaceae); Saray, Kavacık, 08.VIII.2019, apt. 4\$\subseteq\$, alt. \$\subseteq\$, *Polygonum* sp.; Süleymanpaşa, Altınova, 10.X.2018, apt. 4\$\subseteq\$, alt. 2\$\subseteq\$ *Polygonum* sp.; Marmaraereğlisi, Sultanköy 18.VI.2019, apt. 4\$\subseteq\$\$\subseteq\$\$, alt. \$\subseteq\$\$, *Polygonum* sp.

General distribution. Middle East, North Africa, Central Asia, in Europe, Pakistan and also in USA (Blackman & Eastop, 2020).

### Aphis (Aphis) pomi (De Geer, 1773)

Material examined. Tekirdağ, Süleymanpaşa, Hürriyet, 19.V.2019, apt. 299, alt. 299, Hibiscus sp. L. (Malvaceae); Süleymanpaşa, Hürriyet, 07.VI.2019, apt. 499, alt. 299, Hibiscus sp.

General distribution. In Europe, north Africa, Asia eastwards to India and (Blackman & Eastop, 2023).

### Aphis (Aphis) ruborum (Börner & Schilder, 1931)

Material examined. Tekirdağ, Saray, Küçükyoncalı, 08.V.2019, apt. 2, apt. 2, alt. 2, fragaria sp. L. (Rosaceae); Saray, Küçükyoncalı, 08.V.2019, apt. 3, alt. 2, fragaria sp.; Süleymanpaşa, Hürriyet, 04.V.2018, apt. 2, fragaria sp.

General distribution. It is present in Europe, North Africa, Southwest and Central Asia, extending eastward to India and Pakistan, and has been introduced to South America (Chile, Argentina) (Blackman & Eastop, 2023).

### Aphis (Aphis) rumicis (L., 1758)

Material examined. Muratlı, İnanlı, 24.VI.2019, apt.  $3 \stackrel{\frown}{\hookrightarrow}$ , *Rumex* sp. (Polygonaceae); Şarköy, Bulgur, 01.V.2019, apt.  $4 \stackrel{\frown}{\hookrightarrow}$ , alt.  $2 \stackrel{\frown}{\hookrightarrow}$ , *Rumex* sp.

General distribution. While it is extensively documented in the Northern Hemisphere, numerous North American records exist (Blackman & Eastop, 2023).

### Aphis (Aphis) salviae (Walker, 1852)

General distribution. Europe (Switzerland, Poland, Hungary, France, Spain, Portugal, Czech Republic, Bulgaria, Serbia), Israel, Türkiye, Iran and Kazakhstan (Blackman & Eastop, 2023).

### Aphis (Aphis) solanella (Theobald, 1914)

Material examined. Tekirdağ, Malkara, Gazibey, 22.VII.2019, apt. 3  $\ \ \,$  alt.  $\ \ \,$  , *Solanum nigrum* L. (Solanaceae); Süleymanpaşa, Hürriyet, 16.V.2018, apt. 4  $\ \ \,$  , *P. rhoeas* (Papaveraceae); Süleymanpaşa, Naip, 04.IV.2019, apt. 4  $\ \ \,$  , alt. 2  $\ \ \,$  , *U. urens* (Urticaceae).

General distribution. In Europe, Asia, Africa and South America (Blackman & Eastop, 2023).

### Aphis (Aphis) spiraecola (Patch, 1914)

General distribution. Distribution is almost world-wide (Blackman & Eastop, 2023).

### Aphis (Aphis) tirucallis (Hille Ris Lambers, 1954)

Material examined. Tekirdağ, Kapaklı, Pınarca, 01.VI.2019, apt. 2 9, alt. 3 9, Euphorbia sp. (Euphorbiaceae); Süleymanpaşa, Namık Kemal, (40°59'41.2"N, 27°35'15.9"E), 22.V.2019, apt. 5 9, Euphorbia sp.; Şarköy, Mürefte, 23.VII.2018, apt. 2 9, alt. 4 9, Euphorbia sp.

General distribution. Southern Europe, Azores, Canaries, Madeira, Balearics, Yemen widely distributed in Africa (Blackman & Eastop, 2023).

### Aphis (Aphis) umbrella (Börner, 1950)

Material examined. Tekirdağ, Malkara, Develi, 05.IV. 2019, apt. 5, Malva sp. (Malvaceae); Saray, Osmanli, (41°23'05.4"N, 27°40'58.5"E), 12.VI.2018, apt. 6, M. neglecta (Malvaceae); Süleymanpaşa, Değirmenaltı, (40°59'10.5"N, 27°34'48.9"E), 10.IV.2018, apt. 3, M. neglecta (Malvaceae).

General distribution. The species is found in Europe, North Africa, and Asia and North America (Blackman & Eastop, 2023).

### Aphis (Aphis) urticata (Gmelin, 1790)

Material examined. Çorlu, Çobanceşme, (41°09'20.7"N, 27°47'54.1"E), 18.IV.2018, apt. 2 9, alt. 4 9, *U. urens* (Urticaceae); Marmaraereğlisi, Sultanköy, 19.V.2019, apt. 4 9, alt. 2 9, *U. urens*; Saray, Kavacık, 21.VI.2018, apt. 3 9, alt. 4 9, a

General distribution. In Europe, Middle East, Central Asia, Pakistan (Blackman & Eastop, 2023).

### Aphis (Aphis) vallei (Hille Ris Lambers & Stroyan, 1959)

Material examined. Tekirdağ, Kapaklı, Pınarca, 01.VI.2019, apt. 3, Euphorbia sp. (Euphorbiaceae); Süleymanpaşa, Karaevli, 22.V.2019, apt. 3, alt. 2, Euphorbia sp.; Şarköy, Bulgur, 20.VII.2018, apt. 4, Euphorbia sp.

General distribution. In Spain, Portugal, the Balearics, France, Bulgaria, Greece, Ukraine and Türkiye (Blackman & Eastop, 2023).

### Aphis (Aphis) verbasci (Schrank, 1801)

Material examined. Tekirdağ, Hayrabolu, Tatarlı, (41°08'40.0"N, 27°03'49.3"E), 22.VIII.2018, apt. 5  $\bigcirc$  alt. 2  $\bigcirc$  , *Verbascum* sp. (Scrophulariaceae); Marmaraereğlisi, Çeşmeli, (41°02'55.1"N, 27°49'47.3"E), 03.IX.2018, apt. 2  $\bigcirc$  , alt. 2  $\bigcirc$  , *Verbascum* sp.; Saray, Pazarcık, 04.VII.2019, 4  $\bigcirc$  , alt.  $\bigcirc$  , *Verbascum* sp.

General distribution. In Europe (except Scandinavia), Middle East, North Africa, Kazakhstan India and Pakistan (Blackman & Eastop, 2023).

### Aulacorthum (Aulacorthum) solani (Kaltenbach, 1843)

General distribution. Probably of European origin, now almost world-wide (Blackman & Eastop, 2023).

### Brachycaudus (Prunaphis) cardui (L., 1758)

Material examined. Tekirdağ, Çorlu, Sarılar, (41°08'38.8"N, 27°539'45.4"E), 25.V.2018, apt. 599, *Cynara* sp. (Asteraceae); Ergene, Esenler, 08.V.2018, apt. 299, alt. 499, *S. marianum* (Asteraceae); Kapaklı, Mimar Sinan 02.VI.2018, apt. 299, alt. 699, *C. cardunculus* (Asteraceae); Malkara, Develi, 02.VIII.2018, apt. 699, *Eryngium* sp. (Apiaceae); Saray, Küçükyoncalı, 08.V.2019, apt. 499, alt. 299, *C. cardunculus* (Asteraceae); Şarköy, Çengelliköy 01.V.2019, apt. 399, *Onopordium* sp. L. (Asteraceae); Şarköy, Eriklice, 06.V.2019, apt. 999, *Carduus* sp. (Asteraceae).

General distribution. In Europe, Asia, north Africa and North America (Blackman & Eastop, 2023).

### Brachycaudus (Brachycaudus) helichrysi (Kaltenbach, 1843)

Material examined. (1) *S. marianum* (Asteraceae): Süleymanpaşa, Naip, 06.VI.2018; (2) *C. arvense* (Asteraceae): Çorlu, Kazımiye, (41°09'519.3'N, 27°49'36.3"E), 27.VII.2018; (3) *Taraxum officinale* (Weber) (Asteraceae): Malkara, Doğanköy, 06.VIII.2018; (4) *Carduus pycnocephalus* (L.) (Asteraceae): Şarköy, Çengelliköy, 01.V.2019; (5) *Cynara* sp. (L.) (Asteraceae): Saray, Küçükyoncalı, 08.V.2019; (6) *Onopordium* sp. (Asteraceae): Malkara, Doğanköy, 14.V.2019; (7) *Cirsium* sp. (Mill.) (Asteraceae): Süleymanpaşa, Hürriyet, 15.VII.2019.

General distribution. World-wide, and a major pest (Blackman & Eastop, 2023).

### Brevicoryne brassicae (L., 1758)

Material examined. Tekirdağ, Malkara, Doğanköy, 14.V.2019, apt. 4  $\ \ \,$   $\ \ \ \,$   $\ \$ 

General distribution. This species can be found on various genera and species of Brassicaceae, and is considered a significant pest of field crops in all temperate and warm temperate regions worldwide (Blackman & Eastop, 2023).

### Capitophorus elaeagni (Del Guercio, 1894)

Material examined. Tekirdağ, Kapaklı, Karlı, 02.V.2018, 4  $\ \ \,$  alt.  $\ \ \,$  S. marianum (Asteraceae); Saray, Büyükyoncalı, 08.V.2019, S. marianum, Süleymanpaşa, Hürriyet, (27.IV.2018, apt. 4  $\ \ \,$  alt. 2  $\ \ \,$  cynara sp. L. (Asteraceae); Şarköy, Eriklice, 27.VI.2019, C. arvense (Asteraceae).

General distribution. Widely distributed in temperate and warm temperate regions of the world (Blackman & Eastop, 2023).

### Capitophorus hippophaes (Walker, 1852)

Material examined. Tekirdağ, Ergene, Kırkgöz, 11.VI.2018, apt. 299, alt. 2

General distribution. In Europe, North Africa, south-west and central Asia, and introduced into North America (Blackman & Eastop 2023).

### Cavariella (Cavariella) aegopodii (Scopoli, 1763)

Material examined. Tekirdağ, Malkara, Develi, 25.V.2018, 2 + 2, alt.

General distribution. Asia, North and South America, Canada, North Africa, Russia, Serbia and Montenegro, Middle East, India, Tasmania, Australia, New Zealand (Blackman & Eastop, 2023).

### Dysaphis (Dysaphis) crataegi (Kaltenbach, 1843)

Material examined.

Tekirdağ, Çerkezköy, Bağlık, (41°17'56.2"N, 28°00'35.4"E), 20.VII.2018, apt. 3  $\ \ \,$  alt. 2  $\ \ \,$   $\ \ \,$   $\ \ \,$  carota (Apiaceae); Çorlu, Kazımiye, 27.VII.2018, apt. 2  $\ \ \,$  alt. 4  $\ \ \,$   $\ \ \,$   $\ \ \,$  D. carota; Şarköy, Kirazlı, (40°42'08.7"N, 27°15'50.1"E), apt. 2  $\ \ \,$  alt. 2  $\ \ \,$   $\ \ \,$  19.IX.2018, Eryngium sp. (Apiaceae); Şarköy, Uçmakdere, 02.VIII.2019, apt. 4  $\ \ \,$   $\ \ \,$  D. carota.

General distribution. Europe, Greece, Russia, Middle East, South-West Asia and America (Blackman & Eastop, 2023).

### Dysaphis (Dysaphis) tulipae (Boyer de Fonscolombe, 1841)

Material examined. Tekirdağ, Süleymanpaşa, Hürriyet, 30.I.2019, apt. 3♀♀, *Tulipa gesneirana* L. (Liliaceae).

General distribution. Europe, Greece, Russia, Middle East, South-West Asia and America (Blackman & Eastop, 2023).

### Eucarazzia elegans (Ferrari, 1872)

Material examined. Tekirdağ, Saray, Küçükyoncalı 02.V. 2019, apt. 299, alt. 299, P. rhoeas (Papaveraceae); Süleymanpaşa, Altınova, 27.IV.2019, apt. 999, 999, 999, 999, alt. 999, alt

General distribution. Central Asia, Pakistan, northern India, in the Mediterranean area, Madeira, Middle East, and also now in southern Poland, Australia, western USA (California, Oregon) and South America (Argentina), Africa south of the Sahara (Blackman & Eastop, 2023).

### Hayhurstia atriplicis (L., 1761)

Material examined. (1) Tekirdağ, Malkara, Halıç, 21.V.2018, apt. 2  $\bigcirc$  , alt.  $\bigcirc$ , *C. album* (Amaranthaceae); Marmaraereğlisi, Sultanköy, 19.V.2019, apt. 2  $\bigcirc$  , alt. 2  $\bigcirc$  , *Chenopodium* sp. (Amaranthaceae); Saray, Kavacık, 22.IV.2018, apt. 2  $\bigcirc$  , alt. 2  $\bigcirc$  , *Atriplex* sp. L. (Amaranthaceae); Süleymanpaşa, Hürriyet, 26.VI.2020, apt. 4  $\bigcirc$  , alt.  $\bigcirc$  , *Chenopodium* sp.

General distribution. It is prevalent in Europe and Asia, as well as North and Central Africa and North and Central America (Blackman & Eastop, 2023).

### Hyadaphis coriandri (Das, 1918)

Material examined. Tekirdağ, Saray, Osmanli, 24.VI.2018, apt.  $5 \circlearrowleft \circlearrowleft$ , *Bifora radians* Bieb. (Apiaceae); Süleymanpaşa, Değirmenaltı, 16.V.2019, apt.  $3 \hookrightarrow \circlearrowleft$ , alt.  $\circlearrowleft$ , *P. crispum* (Apiaceae); Süleymanpaşa, Naip, 05.VII.2018, apt.  $5 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , *Amaranthus* sp. (Amaranthaceae); Süleymanpaşa, Yüzüncüyıl, 29.V.2020, apt.  $5 \hookrightarrow \circlearrowleft$ , *B. radians*; Şarköy, Yenikoy, 27.VIII.2018, apt.  $2 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , *Eryngium maritimum* L. (Apiaceae).

General distribution. Probably of Asian origin, now in Portugal, Spain, the Mediterranean region, the Middle East, Central Asia, India, Pakistan, Africa, USA (Florida, Hawaii) and South America (Peru, Argentina) (Blackman & Eastop, 2023).

### Hyadaphis foeniculi (Passerini, 1860)

General distribution. Widespread in Europe, especially in the north, eastward to Türkiye, Iran and Iraq, in North America (New York, New Brunswick, California), north-east Brazil (Blackman & Eastop, 2023).

### Hyalopterus pruni (Geoffroy, 1762)

Material examined. Tekirdağ, Muratlı, İnanlı, 08.05.2018, apt. 322, alt. 222, alt. 222, alt. 222, alt. 222, alt. 222, alt. 222, alt. 222, Sonchus oleraus (Asteraceae); Şarköy, Kocaali, 09.V.2018, apt. 422, Phragmites sp.

General distribution. With a wide distribution Asia, Europe and North America (Blackman & Eastop, 2023).

### Hydaphias hofmanni (Börner, 1950)

Material examined. Tekirdağ, Çorlu, Yenice, 01.VII.2018, apt. 322, *Galium* sp. (Rubiaceae); Şarköy, Demirler, 03.VII.2019, apt. 222, alt. 22, *Galium* sp.

General distribution. Throughout Europe, and across Asia to east Siberia, Korea and China (Blackman & Eastop, 2023).

### Hyperomyzus (Hyperomyzus) lactucae (L., 1758)

General distribution. It is widespread and prevalent worldwide, except in southern Africa, although it can be found in the highlands of Kenya (Blackman & Eastop, 2023).

### Lipaphis (Lipaphis) erysimi (Kaltenbach, 1843)

Material examined. Tekirdağ, Hayrabolu, Yörükler, (41°07'21.8"N, 27°14'31.0"E), 06.V.2019 apt. 3  $\bigcirc$   $\bigcirc$ , alt. 2  $\bigcirc$   $\bigcirc$ , Sisymbrium altissimum L. (Brassicaceae); Malkara, Gazibey, 12.V.2018, apt. 2  $\bigcirc$   $\bigcirc$ , alt. 2  $\bigcirc$   $\bigcirc$ , alt. 2  $\bigcirc$   $\bigcirc$ , alt. 2  $\bigcirc$   $\bigcirc$ , alt. 2  $\bigcirc$   $\bigcirc$ , alt. 2  $\bigcirc$   $\bigcirc$ , camelina sp.; Süleymanpaşa, Değirmenaltı, 13.IV.2018, apt. 3  $\bigcirc$   $\bigcirc$   $\bigcirc$ , Camelina sp.

General distribution. It is present in Northern Europe and likely in Western Siberia and Central Asia as well (Blackman & Eastop, 2023).

### Lipaphis (Lipaphidiella) lepidii (Nevsky, 1929)

Material examined. (1) Çorlu, Sarılar, 11.V.2018, apt.  $\bigcirc$ , alt.  $\bigcirc$ , *P. crispum* (Apiaceae); Malkara, Halıç, 21.V.2018, apt.  $\bigcirc$ , alt.  $\bigcirc$ , *P. crispum*; Süleymanpaşa, Köseilyas, (41°401'06.9"N, 27°34'55.6"E), 22.V.2018, apt.  $\bigcirc$ , alt.  $\bigcirc$ , *P. crispum*; Şarköy, Cumhuriyet, 23.V.2019, apt.  $\bigcirc$ , alt.  $\bigcirc$ , *P. crispum*.

General distribution. Middle East and Central Asia, in Greece, eastward to Pakistan (Blackman & Eastop, 2023).

### Macrosiphoniella (Macrosiphoniella) artemisiae (Boyer de Fonscoclombe, 1841)

Material examined. Tekirdağ, Şarköy, Uçmakdere, 5.V.2018, apt. 2♀♀, *Tanacetum artemisioides* Sch. Bip. Ex Hook.fil. (Asteraceae).

General distribution. Throughout Europe, eastward across Siberia, south-west and Central Asia, Mongolia and China, Pakistan, and introduced to North America and Argentina (Blackman & Eastop, 2023).

### Macrosiphoniella (Macrosiphoniella) sanborni (Gillette, 1908)

Material examined. Tekirdağ, Marmaraereğlisi, Yeniçiftlik, (41°00'33.8"N, 27°51'01.3"E), 07.VI.2018, apt. 399, alt. 99, *Chrysanthemum* sp. L. (Asteraceae); Süleymanpaşa, Hürriyet, 16.VI.2018, apt. 299, *Chrysanthemum* sp.

General distribution. Distribution is almost world-wide (Blackman & Eastop, 2023).

### Macrosiphoniella (Macrosiphoniella) tanacetaria (Kaltenbach, 1843)

Material examined. Tekirdağ, Çerkezköy, 24.VI.2019, apt. 2♀♀, alt. 2♀♀, Achillea sp. (Asteraceae).

General distribution. Throughout Europe, Morocco, Israel, Iran, Georgia, Kyrgyzstan, Kazakhstan, Mongolia, west and east Siberia, and introduced to North and South America (Blackman & Eastop, 2023).

### Macrosiphum (Macrosiphum) euphorbiae (Thomas, 1878)

Material examined. Tekirdağ, Çorlu, Sarılar, 03.V.2019, apt.  $3 \circlearrowleft \circlearrowleft$ , alt.  $\circlearrowleft$ , Capsella bursa pastoris L. (Brassicaceae); Malkara, Güneşli, 25.V.2019, apt.  $2 \circlearrowleft \circlearrowleft$ , alt.  $2 \circlearrowleft \circlearrowleft$ , A. retroflexus; Muratlı, Kepenekli, 24.V.2019, apt.  $4 \circlearrowleft \circlearrowleft$ , alt.  $3 \circlearrowleft \circlearrowleft$ , O. acanthium; Muratlı, Kepenekli, 24.V.2019, apt.  $3 \circlearrowleft \circlearrowleft$ , alt.  $2 \circlearrowleft \circlearrowleft$ , A. retroflexus; Saray, İnanlı, 28.VIII.2018, apt.  $2 \circlearrowleft \circlearrowleft$ , alt.  $2 \circlearrowleft \circlearrowleft$ , Alt.  $2 \circlearrowleft \circlearrowleft$ , alt.  $2 \circlearrowleft \circlearrowleft$ , alt.  $2 \circlearrowleft \circlearrowleft$ , alt.  $2 \circlearrowleft \circlearrowleft$ , alt.  $2 \circlearrowleft \circlearrowleft$ , alt.  $2 \circlearrowleft \circlearrowleft$ , alt.  $2 \circlearrowleft \circlearrowleft$ , alt.  $2 \circlearrowleft \circlearrowleft$ , alt.  $2 \circlearrowleft \circlearrowleft$ , alt.  $2 \circlearrowleft \circlearrowleft$ , alt.  $2 \circlearrowleft \circlearrowleft$ , alt.  $2 \circlearrowleft \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , A. retroflexus (Amaranthaceae); Süleymanpaşa, Hürriyet, O7.VIII.2018, apt.  $2 \hookrightarrow \circlearrowleft$ , O. acanthium (Asteraceae); Süleymanpaşa, Hürriyet, O6.V.2019, apt.  $2 \hookrightarrow \circlearrowleft$ , Euphorbia sp. (Euphorbiaceae); Süleymanpaşa, O6.V.2019, apt.  $2 \hookrightarrow \circlearrowleft$ , alt.  $2 \hookrightarrow \circlearrowleft$ , C. cardunculus (Asteraceae); Süleymanpaşa, Namık Kemal O4.VI.2019, apt.  $2 \hookrightarrow \circlearrowleft$ , P. rhoeas (Papaveraceae).

General distribution. Of North American origin, now almost world-wide (Blackman & Eastop, 2023).

### Metopolophium (Metopolophium) dirhodum (Walker, 1849)

Material examined. Tekirdağ, Muratlı, İnanlı, 08.VI.2018, apt. 3  $\bigcirc$  , *Artemisia vulgari*s L. (Asteraceae); Süleymanpaşa, Köseilyas, 16.IX.2018, apt. 5  $\bigcirc$  , *Z. mays* (Poaceae); Şarköy, Palamut,  $(40^{\circ}45'35.3"N, 27^{\circ}09'26.02"E)$ , 22.IX.2019, apt. 6  $\bigcirc$  , *Z. Mays*.

General distribution. Considered a significant pest of cereals, it has become extensively distributed, particularly in temperate regions worldwide (Blackman & Eastop, 2023).

### Microlophium carnosum (Buckton, 1876)

Material examined. Tekirdağ, Hayrabolu, Yörükler 12.V.2019, apt. 4\(\text{Q}\), *Urtica* sp.; Malkara, Doğanköy, 06.XI.2018, apt. 5\(\text{Q}\), *U. urens* (Urticaceae); Marmaraereğlisi, Sultanköy, 19.X.2019, apt. 2\(\text{Q}\), *urtica* sp.; Süleymanpaşa, Naip, 15.V.2019, apt. 2\(\text{Q}\), *Urtica* sp.

General distribution. Asia east to Pakistan and Mongolia, in Europe, Africa (Burundi, Rwanda), North America and Chile (Blackman & Eastop, 2023).

### Myzus (Myzus) lythri (Schrank, 1801)

General distribution. Middle East (Iran, Lebanon), central (Burundi) and southern Africa, throughout Europe, in North Africa (Tunisia), central Asia (Blackman & Eastop, 2023).

### Myzus (Myzus) ornatus (Laing, 1932)

Material examined. Tekirdağ, Malkara, Doğanköy, 06.VI.2018, apt. 422, *Urtica* sp. (Urticaceae); Süleymanpaşa, 01.VII.2019, apt. 322, alt. 22, *Crysanthemum* sp. (Asteraceae).

General distribution. It is present on cultivated ornamental plants worldwide, but despite being prevalent in India since 1956, only one record exists from Southeast Asia (New Guinea) (Blackman & Eastop, 2023).

### Myzus (Nectarosiphon) persicae (Sulzer, 1776)

General distribution. Probably of east Asian origin, but is now world-wide (Blackman & Eastop, 2023).

### Nasonovia (Nasonovia) ribisnigri (Mosley, 1841)

Material examined. Tekirdağ, Çerkezköy, Gaziosmanpasa, (41°17'12.3"N, 27°59'52.7"E), 04.V.2018, apt. 6\(\text{Q}\), T. officinale (Asteraceae); Malkara, Haliç, 14.VII.2019, apt. 3\(\text{Q}\), alt. 2\(\text{Q}\), Crepis sp. (Asteraceae); Süleymanpaşa, Yüzüncüyıl, apt. 2\(\text{Q}\), alt. \(\text{Q}\), 07.VII.2020 Crepis pulchra; \(\text{Q}arköy, Cumhuriyet, 23.V.2019, apt. 5\(\text{Q}\), alt. 3\(\text{Q}\), officinale.

General distribution. In Europe, Middle East, Central Asia, Africa (Algeria, Burundi, Rwanda), North and South America, New Zealand (Blackman & Eastop, 2023).

### Neomyzus (Aulacorthum) circumflexum (Buckton, 1876)

Material examined. Tekirdağ, Hayrabolu, Tatarlı, 20.VI.2018,  $4 \circlearrowleft \circlearrowleft$ , alt.  $\circlearrowleft$ , *P. sativum* (Apiaceae); Süleymanpaşa, Namık Kemal, 01.VI. 2019, apt.  $3 \circlearrowleft \circlearrowleft$ , alt.  $2 \circlearrowleft \circlearrowleft$ , *Vicia sativa* L. (Fabaceae); Süleymanpaşa, Namık Kemal, 12.VI.2020, *Vicia sativa*.

General distribution. Virtually world-wide (Blackman & Eastop, 2023).

### Protaphis terricola (Rondani, 1847)

Material examined. Süleymanpaşa, Naip, 10.VII.2019, apt. 3♀♀, *Centaurea iberica* Trevir. & Spreng. (Asteraceae).

General distribution. In southern, central and eastern Europe, Iran, Türkiye, Israel, Kazakhstan, Pakistan, Egypt, Sudan, and introduced to South America (Blackman & Eastop, 2023).

### Rhopalosiphoninus (Rhopalosiphoninus) latysiphon (Davidson, 1912)

Material examined. Hayrabolu, Kılıçlar, 12.V.2019, apt. 3♀♀, alt. 2♀♀, S. tuberosum (Solanaceae).

General distribution. Europe and in Egypt, Nepal, Sri Lanka, Japan, Kenya, South Africa, India, Pakistan, China, Australia, New Zealand, and North and South America (Blackman & Eastop, 2023).

### Rhopalosiphum maidis (Fitch, 1856)

Material examined. Tekirdağ, Hayrabolu, Yörükler, 12.V.2019, apt. 322, alt. 222, *Sorgum halepense* L. (Poaceae); Malkara, Gazibey, 05.V.2018, apt. 322, alt. 222, al

General distribution. Virtually cosmopolitan (Blackman & Eastop, 2023).

### Rhopalosiphum oxyacanthae (Schrank, 1801)

Material examined. Tekirdağ, Saray, Kavacık, 21.V.2019, apt. 3♀♀, Agrostis stolonifera L. (Poaceae).

General distribution. In Europe, North Africa (Tunisia), the Azores, south-west and central Asia, and Japan (Blackman & Eastop, 2023).

### Rhopalosiphum padi (L., 1758)

Material examined. Tekirdağ, Süleymanpaşa, Karaevli, 26.IX.2018, apt.  $4 \updownarrow \updownarrow$ , *Z. mays* (Poaceae); Süleymanpaşa, Köseilyas, apt.  $2 \updownarrow \updownarrow$ , alt.  $2 \updownarrow \updownarrow$ , 25.V.2019, *Avena sativa* L.; Süleymanpaşa, Köseilyas, 26.09.2020, apt.  $5 \updownarrow \updownarrow$ , *Z. Mays*; Süleymanpaşa, Hürriyet, 03.V.2019, apt.  $3 \updownarrow \updownarrow$ , *Hordeum murinum* L.; Şarköy, Bulgur, 01.V.2019, apt.  $2 \updownarrow \updownarrow$ , alt.  $2 \updownarrow \updownarrow$ , *H. murinum*; Şarköy, Ulaman, 15.V.2018, apt.  $3 \updownarrow \updownarrow$ , alt.  $2 \updownarrow \updownarrow$ , *Cynodon dactylon* L. (Poaceae).

General distribution. Southern Europe and America Israel, Iran, Lebanon and Egypt (Blackman & Eastop, 2023).

### Rhopalosiphum rufiabdominale (Sasaki, 1899)

Material examined. Tekirdağ, Süleymanpaşa, Naip, 08.VII.2018, apt. 5♀♀, alt. ♀, *Allium cepa* L. (Amaryllidaceae).

General distribution. It is a cosmopolitan species that occurs all over the world (Nieto Nafria, 2017).

### Schizaphis (Schizaphis) graminum (Rondani, 1852)

Material examined. Tekirdağ, Ergene, Kırkgöz, 27.V.2018, *Sorghum* sp. (Poaceae); Malkara, İbrice, (40°58'01.9"N, 26°52'24.0"E), 28.IX.2018, apt. 4\cong \tau, *Z. mays*; Muratlı, Aydınköy, 08.VI.2018, apt. 3\cong \tau, *Triticum* sp.; Saray, Kurtdere (41°24'16.4"N, 27°15'50.1"E), 02.IX.2019, apt. 5\cong \tau, *Z. mays*; Süleymanpaşa, Köseilyas, 25.V.2019, apt. 2\cong \tau, alt. 2\cong \tau, sorghum vulgare Pers.

General distribution. Southern Europe, Middle East, Central Asia, Africa, India, Nepal, Pakistan, Thailand, Korea, China, Taiwan, Japan, and North, Central and South America (Blackman & Eastop, 2023).

### Sitobion (Sitobion) avenae (Fabricius, 1775)

Material examined. Tekirdağ, Malkara, Güneşli, 25.V.2019, apt. 4  $\bigcirc$   $\bigcirc$ , alt. 2  $\bigcirc$ , *Triticum* sp.; Saray, Kadıköy, 10.V.2018, apt. 4  $\bigcirc$   $\bigcirc$ , alt.  $\bigcirc$  *T. aestivum*; Süleymanpaşa, Karaevli, apt. 3  $\bigcirc$   $\bigcirc$ , alt.  $\bigcirc$ , 06.V.2018, *T. aestivum* (Poaceae); Süleymanpaşa, Köseilyas, 22.V.2018, apt. 5  $\bigcirc$  , alt.  $\bigcirc$ , *T. aestivum*; Süleymanpaşa, Naip, 25.V.2019, apt. 3  $\bigcirc$  , alt. 2  $\bigcirc$  , *Bromus* sp. Scop.; Şarköy, Mürefte, 12.V.2019, apt. 3  $\bigcirc$  , alt. 2  $\bigcirc$  , alt. 2  $\bigcirc$  , *T. aestivum*; Şarköy, Tepeköy, (40°40'21.6"N, 27°11'05.6"E), 22.V.2019, apt. 4  $\bigcirc$  , alt. 2  $\bigcirc$  , *T. aestivum*.

General distribution. Throughout Europe, the Mediterranean, the Middle East, Central Asia, India, Nepal, Pakistan, Africa; North, Central and South America (Blackman & Eastop, 2023).

### Staegeriella necopinata (Börner, 1939)

Material examined. Tekirdağ, Çerkezköy, Bağlık, 10.VI.2019, apt.  $\bigcirc$ , alt.  $\bigcirc$ , Galium sp.; Şarköy, Eriklice, 15.VI.2018, apt.  $2\bigcirc\bigcirc$ , alt.  $2\bigcirc\bigcirc$ , alt.  $2\bigcirc\bigcirc$ , alt.  $2\bigcirc\bigcirc$ , alt.  $2\bigcirc\bigcirc$ , alt.  $2\bigcirc\bigcirc$ , alt.  $2\bigcirc\bigcirc$ 

General distribution. Throughout Europe, western Siberia, Iran, Kazakhstan and Tunisia (Blackman & Eastop, 2023).

### Uroleucon (Uroleucon) cichorii (Koch, 1855)

Material examined. Tekirdağ, Çerekezköy, Bağlık, 20.VII.2018, apt. 4 \times \times, alt. 2 \times, T. officinale (Asteraceae); Ergene, Esenler, 25.VI.2018, apt. 3 \times, alt. 2 \times, T. intybus (Asteraceae); Muratlı, Aydınköy, 08.VI.2018, apt. 5 \times, alt. 3 \times, Rumex sp. (Polygonaceae); Süleymanpaşa, Değirmenaltı, 17.VI.2019, apt. 3 \times, Cichorium sp.; Süleymanpaşa, Namık Kemal, apt. 2 \times, alt. 4 \times, 26.VI.2020, T. officinale.

General distribution. In Europe, south-west and central Asia, Eritrea, Mongolia, Korea and east Siberia (Blackman & Eastop, 2023).

### Uroleucon (Uroleucon) sonchi (L., 1767)

General distribution. It has an almost world-wide distribution (Blackman & Eastop, 2023).

### Uroleucon (Uroleucon) tanaceti (L., 1758)

Material examined. Tekirdağ, Saray, Büyükyoncalı, 08.VII.2019, apt. 5♀♀, alt. ♀, *Tanacetum* sp. L. (Asteraceae).

General distribution. In Europe, west Siberia, south-west and Central Asia, eastern Himalayas, and North America (Blackman & Eastop, 2023).

### Uroleucon (Uromelan) aeneum (Hille Ris Lambers, 1939)

Material examined. Tekirdağ, Hayrabolu, Kılıçlar, 01.VI.2018, apt. 6  $\bigcirc$  , alt.  $\bigcirc$ , *Carduus* sp. (Asteraceae); Malkara, İbrice, 22.V.2019, apt. 2  $\bigcirc$  , alt.  $\bigcirc$ , *C. arvense* (Asteraceae); Marmaraereğlisi, Sultanköy, 19.V.2019, apt. 3  $\bigcirc$  , alt. 2  $\bigcirc$  , *Urtica* sp. (Urticaceae); Saray, Kadıköy, 12.VI.2018, apt. 3  $\bigcirc$  , alt. 2  $\bigcirc$  , *Onopordium* sp. (Asteraceae); Şarköy, Ulaman, 15.V.2018, 2  $\bigcirc$  , alt.  $\bigcirc$  , *S. marianum* (Asteraceae).

General distribution. In Europe, North Africa (Algeria), Türkiye, Armenia, Iran, Central Asia, east and west Siberia, and South America (Argentina, Chile) (Blackman & Eastop, 2023).

### Uroleucon (Uromelan) carthami (Hille Ris Lambers, 1948)

Material examined. Süleymanpaşa, Namık Kemal, 29.V.2019, apt. 4  $\bigcirc$   $\bigcirc$ , alt. 2  $\bigcirc$ , Carthamus tinctorius L. (Asteraceae).

General distribution. In southern and central Europe, Algeria, Israel, Lebanon, Türkiye, and eastward to Pakistan and India (Kashmir) (Blackman & Eastop, 2023).

### Uroleucon (Uromelan) jaceae (L., 1758)

Material examined. Tekirdağ, Malkara, Gazibey, 20.V.2019, apt. 6  $\bigcirc$  , alt. 2  $\bigcirc$  ,  $Carduus\ crespis\ L$ . (Asteraceae); Malkara, Haliç, 21.V.2018, apt. 3  $\bigcirc$  , alt.  $\bigcirc$  ,  $Cynara\ sp.$  (Asteraceae); Süleymanpaşa, Naip, 06.VI.2018, apt. 2  $\bigcirc$  ,  $Acroptilon\ repens\ L$ . (Asteraceae); Şarköy, Çengelliköy, 22.V.2019, apt. 2  $\bigcirc$  , alt.  $\bigcirc$  ,  $Cynara\ sp.$ 

General distribution. In Europe, west Siberia, Middle East, Central Asia and Pakistan (Blackman & Eastop, 2023).

### Uroleucon (Uromelan) nigrocampanulae (Theobald, 1928)

Material examined. Marmaraereğlisi, Bahçelievler, 18.VIII.2018, apt. 3♀♀, *Companula trachelium* L. (Campanulaceae).

General distribution. In Europe and across Asia to east Siberia (Blackman & Eastop, 2023).

### Subfamily, Chaitophorinae

### Sipha (Rungsia) elegans (Del Guercio, 1905)

Material examined. Tekirdağ, Kapaklı, Karlı, 01.VI.2019, apt.  $2^{\circ}$ , Agropyron sp. Gaertn. (Poaceae); Süleymanpaşa, 07.VI.2018, apt.  $4^{\circ}$ , Agropyron sp.

General distribution. In Europe, across Asia to China and east Siberia, and in North America (Blackman & Eastop, 2023).

### Sipha (Rungsia) maydis (Passerini, 1860)

Material examined. Tekirdağ, Kapaklı, Pınarca, 01.VI.2019, apt. 3  $\$ 0, alt. 2  $\$ 2, Bromus sp. (Poaceae); Süleymanpaşa, Hürriyet, 05.VI.2020, apt. 2  $\$ 2, alt. 2  $\$ 2, Bromus sp.; Süleymanpaşa, Naip, 24.VI.2018, apt. 3  $\$ 2, alt. 2  $\$ 2, Bromus sp.

General distribution. In regions with arid climates beyond northwest Europe, this species has the potential to become a pest of cereal crops (Blackman & Eastop, 2023).

### Subfamily, Eriosomatinae

### Forda formicaria (von Heyden, 1837)

General distribution. In northern Europe, Iran, Siberia, North America and Central Asia (Blackman & Eastop, 2023).

### Forda marginata (Koch, 1857)

Material examined. Tekirdağ, Malkara, Develi, 11.V.2018, apt. 6, P, annua (Poaceae); Süleymanpaşa, Namık Kemal, 09.V.2019, apt. 5, P, annua; Şarköy, Tepeköy, 01.V.2019, apt. 2, H, murinum (Poaceae).

General distribution. Siberia, Central Asia, India, in northern and central Europe, North America and China (Blackman & Eastop, 2023).

### Pemphigus (Pemphigus) bursarius (L., 1758)

Material examined. Tekirdağ, Çorlu, Kazımiye, (41°09'519.3'N, 27°49'36.3"E), 24.V.2018, apt.  $5 \stackrel{\frown}{\hookrightarrow} \uparrow$ , *T. officinale* (Asteraceae); Çorlu, Yenice, 28.V.2019, apt.  $3 \stackrel{\frown}{\hookrightarrow} \uparrow$ , *T. officinale*; Saray, Kavcık, 21.V.2019, apt.  $3 \stackrel{\frown}{\hookrightarrow} \uparrow$ , *T. officinale*.

General distribution. Northern and southern Africa, in Europe, across Asia to eastern Siberia, North and South America (Blackman & Eastop, 2023).

### Rectinasus buxtoni (Theobald, 1914)

Material examined. Tekirdağ, Hayrabolu, Tatarlı, 30.VII.2018, apt. 6, X anthium strumarium; Marmaraereğlisi, Sultanköy, 18.VIII.2018, apt. 3, X anthium strumarium (L.) (Asteraceae); Muratlı, Kepenekli, 23.VIII.2019, apt. 3, X anthium sp.; Süleymanpaşa, Değirmenaltı, 12.VIII.2019, apt. 4, X Lactuca sp. (Asteraceae).

General distribution. Southern Europe, North Africa, in southwest Asia (Israel, Iran, Türkiye), northern Caucasus, Transcaucasia, Kazakhstan, Turkmenistan and Dagestan (Blackman & Eastop, 2023).

### Smynthurodes betae (Westwood, 1849)

Material examined. Tekirdağ, Muratlı, Kepenekli 02.V.2018, 3  $\bigcirc$  , Lens culinaris L. (Fabaceae); Saray, Büyükoncalı, 09.VI.2018, apt. 2  $\bigcirc$  , Orabanche sp. L. (Orobanchaceae); Süleymanpaşa, Karaevli, 22.V.2019, 5  $\bigcirc$  , T. sativum; Süleymanpaşa, Namık Kemal, 25.VI.2018, apt. 3  $\bigcirc$  , P. oleracea (Portulacaceae); Şarköy, Koçeli, 09.V.2018, apt. 5  $\bigcirc$  , Triticum sativum L. (Poaceae).

General distribution. Algeria, Morocco, Israel, Syria, Iran, southern Crimea, Transcaucasus and Pakistan (Blackman & Eastop, 2023).

### Tetraneura (Tetraneura) ulmi (L., 1758)

Material examined. Tekirdağ, Malkara, Danişment, 02.V.2018, apt. 4♀♀ *Dactylis glomerata* L. (Poaceae).

General distribution. Europe, Asia, eastern Siberia, northern Japan, and North America (Blackman & Eastop, 2020).

### Subfamily, Lachninae

### Protrama radicis (Kaltenbach, 1843)

Material examined. Malkara, Doğanköy, 15.VIII.2018,  $4 \stackrel{\frown}{}_{\downarrow}$ , *C. arvense* (Asteraceae); Marmaraereğlisi, Bahçelievler, 18.VIII.2018, apt.  $3 \stackrel{\frown}{}_{\downarrow}$ , *C. arvense*; Şarköy, Palamut, 05.VIII.2019,  $6 \stackrel{\frown}{}_{\downarrow}$ , *C. pycnocephalus* (Asteraceae).

General distribution. Excluding the Iberian Peninsula, the species is present in Europe, as well as Southwest and Central Asia (Blackman & Eastop, 2023).

Of the species found during the study, *M. persicae* and *A. fabae* were found in 16 different hosts, *A. craccivora* in 13 different hosts, *M. euphorbiae* in 10 different hosts, *A. solani, B. cardui* and *H. lactucae* in 6 different hosts. In this study, the most common *Aphis* genus was *A. gossypii*. In addition, *C. lanatus, C. sativus, P. oleracea, Carduus* sp., *A. esculentus, S. lycopersicum, O. acanthium, Chenopodium* sp., *M. piperita, A. retroflexus, M. neglecta*, and *P. rhoeas* were detected on 12 different host plants. Out of the various families, Asteraceae (Compositae) stood out as the most favored, boasting a total of 30 aphid species. Asteraceae was followed by Apiaceae (Umbelliferae) with 16 aphid species, Poaceae (Gramineae) and Amaranthaceae families with 11 aphid species. *E. tenuifolia, Cachyrs* sp., *M. neglecta, C. intybus, Achillea* sp., *A. retroflexus* and *S. marianum* have been identified as new host records.

Analysis of both the present study and previous research, such as Tuatay (1988), Akyürek (2013), Özdemir (2004), Kök et al. (2016), Görür et al. (2017), Öztürk & Muştu (2017), Kuloğlu & Özder (2017), Kök & Kasap (2019, 2022), Başer & Tozlu (2020), Özdemir (2020), Kök (2021), Görür (2022), Kök & Özdemir (2021, 2022) reveals that conducting taxonomic studies on various regions would make a significant contribution to the understanding of aphid fauna in Türkiye. This is due to the fact that Türkiye possesses rich biodiversity, diverse regions, microclimates, as well as a varied fauna and flora.

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# Original article (Orijinal araştırma)

# Host suitability of pepper cultivars to (a)virulent root-knot nematodes isolates

Biber çeşitlerinin (a)virulent kök-ur nematod izolatlarına karşı konukçuluk durumu

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D

#### **Abstract**

Root-knot nematodes (RKNs) are polyphagous and cause yield losses to crops worldwide. Using the resistant plant is the most effective and environmental method to manage RKNs. *Mi-1.2* gene in tomatoes is commonly used to control *Meloidogyne*. However, the *Mi-1.2*-virulent isolates can overcome this gene. In fields infested with *Mi-1.2*-virulent populations, plant species with different resistance genes are recommended instead of tomatoes. Therefore, investigating the host suitability of pepper cultivars to *Mi-1.2*-virulent RKNs is needed for management practices. This study was conducted in Akdeniz University Faculty of Agriculture Department of Plant Protection Nematology Laboratory in 2019. In this study, the response of resistant and susceptible pepper cultivars was investigated to *Meloidogyne incognita* (Kofoid & White,1919), Chitwood, 1949, *Meloidogyne javanica* (Treub, 1885) Chitwood, 1949, *Meloidogyne arenaria* (Neal, 1889) Chitwood, 1949, *Meloidogyne luci* Carneiro et al., 2014 (Tylenchida: Meloidogynidae), *Mi-1.2*-virulent *M. incognita* and *Mi-1.2*-virulent *M. javanica*. *Meloidogyne arenaria*, *M. incognita* and *Mi-1.2*-virulent *M. incognita* isolates multiplied very well on susceptible peppers but did not reproduce on resistant peppers. *Mi-1.2*-(a)virulent *M. javanica* isolates did not multiply on any pepper cultivars. *Meloidogyne luci* reproduced on all peppers tested. A pepper carrying *N* resistance gene was first tested with *M. luci* and did not confer resistance to *M. luci*. These results could be used to control RKNs in vegetable-growing areas.

Keywords: Capsicum annum, Meloidogyne, Mi-1.2-virulent, N gene, resistance

# Öz

Kök-ur nematodları polifag olup tüm dünyada tarım ürünlerinde verim kayıplarına neden olurlar. Dayanıklı bitki kullanımı kök-ur nematodları ile mücadelede en etkili ve çevreci yöntemdir. Ancak, *Mi-1.2-*virülent kök-ur nematod popülasyonları domatesteki dayanıklılığı kırabilmektedir. *Mi-1.2-*virülent popülasyonlarla bulaşık arazilerde domates yerine farklı dayanıklılık genleri taşıyan bitki türleri önerilmektedir. Bu nedenle biber çeşitlerinin *Mi-1.2-*virülent kök-ur nematodlarına konukçuluk durumunun araştırılması kök-ur nematodları ile mücadele için gereklidir. Bu çalışma 2019 yılında Akdeniz Üniversitesi Ziraat Fakültesi Bitki Koruma Bölümü Nematoloji laboratuvarında yürütülmüştür. Çalışmada dayanıklı ve hassas biber çeşitlerinin *Meloidogyne incognita* Kofoid & White,1919), Chitwood, 1949, *Meloidogyne javanica* (Treub, 1885) Chitwood, 1949, *Meloidogyne arenaria* (Neal, 1889) Chitwood, 1949, *Meloidogyne luci* Carneiro et al., 2014 (Tylenchida: Meloidogynidae), *Mi-1.2-*virülent *M. incognita* ve *Mi-1.2-*virülent *M. javanica*'ya karşı tepkisi incelenmiştir. *Meloidogyne arenaria*, *M. incognita* ve *Mi-1.2-*virülent *M. incognita* izolatları hassas biber çeşitlerinde ürerken dayanıklı çeşitlerde ürermemiştir. *Mi-1.2-*(a)virulent *M. javanica* izolatları hiçbir biber çeşidinde çoğalamamıştır. *Meloidogyne luci*, testlenen tüm biber çeşitlerinde çoğalmıştır. *N* dayanıklılık geni taşıyan biberler ilk kez *M. luci* ile testlenmiş ve *M. luci* ye dayanıklılık sağlamamıştır. Bu sonuçlar sebze yetiştiriciliği yapılan alanlarda kök-ur nematodlarla mücadelede faydalı olabilecektir.

Anahtar sözcükler: Capsicum annum, Meloidogyne, Mi-1.2-virülent, N geni, dayanıklılık

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#### Introduction

Root-knot nematodes (RKNs) are organisms that are difficult to manage due to their short generation times and high reproduction rates. The use of genetic resistance to RKNs provides efficient, economically competitive, and eco-friendly management (Devran et al., 2010). Some resistance genes against RKNs have been described from Solanaceae (Smith, 1944; Hendy et al., 1985; Fery et al., 1998). In tomato, *Mi-1.2* gene is the most important resistant gene for controlling RKNs and is widely used in commercial tomato cultivars (Devran & Söğüt, 2014; Seid et al., 2015). *Mi-1.2* gene provides effective protection against the three most destructive RKNs, *Meloidogyne incognita* Kofoid & White,1919), Chitwood, 1949, *Meloidogyne arenaria* (Neal, 1889) Chitwood, 1949 and *Meloidogyne javanica* (Treub, 1885) Chitwood, 1949 (Tylenchida: Meloidogynidae) (Williamson & Hussey, 1996) as well as other RKN species (Aydınlı & Mennan, 2019; Gabriel et al., 2020; Santos et al., 2020). However, the resistance mediated by *Mi-1.2* is lost at high soil temperatures and can be overcome by *Mi-1.2*-virulent RKN isolates (Dropkin, 1969; Kaloshian et al., 1996; Tzortzakakis et al., 2005; Devran & Söğüt, 2010; Özalp & Devran, 2018). Since *Mi-1.2*-virulent isolates have become common in vegetable fields, alternating the different resistance genes may be an effective management strategy against virulent populations. Thus, there is a need to understand the reactions of different resistance genes to *Mi-1.2*-virulent RKN populations

Pepper (*Capsicum annum* L.) (Solanales: Solanaceae) is an economically important member of the Solanaceae family. In pepper, several genes controlling resistance to RKNs have been determined. From these genes *N*, *Me1*, *Me3* and *Me7* confer resistance to three most destructive RKNs (Djian-Caporalino et al., 1999; Castagnone-Sereno et al., 2001). Resistant pepper cultivars are used for controlling RKNs (Barbary et al., 2015). In addition, resistant pepper cultivars are considered as rotation crops in the cultivation of susceptible vegetables for *Meloidogyne* (Thies et al., 1998). Thus, it is very important to know the responses of peppers with and without resistance genes against RKNs obtained from different locations.

Studies show that the reproductive potential of *Mi-1.2*-virulent RKN isolates on susceptible pepper cultivars is lower than that of avirulent isolates. Djian-Caporalino et al. (2011) observed that virulent RKN populations against the *Mi-1*, *Me1* and *Me3* resistance genes had lower reproduction on susceptible tomato and pepper plants. In this case, knowing the response of *Mi-1.2*-virulent isolates in different susceptible pepper cultivars can be important for crop rotation programs.

The aims of this study were i) to compare the response of pepper cultivars to three most destructive RKNs and *Meloidogyne luci* Carneiro et al., 2014 (Tylenchida: Meloidogynidae) isolates, ii) determine the effectiveness of *N* gene to (a)virulent RKN isolates, and iii) designate whether the *N* gene confers resistance to *M. luci*.

#### **Materials and Methods**

#### Plant materials

Pepper cultivars Safran F1, Mostar F1 and Mert F1 were used as susceptible to RKNs. The resistant pepper line Coralina Wonder is homozygous for the *N* gene. Responses to RKNs of pepper cultivars B5 line, B6 line and B4 F1 (B5xB6) and local accession KB401 are unknown. The seeds of KB401 were obtained from a local farmer, while other pepper varieties were provided from Yüksel Tohum (Antalya, Türkiye).

#### **Nematode isolates**

RKNs isolates used in the experiments are listed in Table 1. Pure cultures were reproduced from single egg mass. *Mi-1.2* natural virulent and avirulent isolates were reproduced on resistant and susceptible tomato cultivars, respectively.

Table 1. Meloidogyne isolates used in the experiments

Code	Nematode species	Property	Reference
S6	Meloidogyne incognita	Avirulent	Devran & Sögüt, 2009
K21	Meloidogyne javanica	Avirulent	Devran & Sögüt, 2009
K18	Meloidogyne arenaria	Avirulent	Devran & Sögüt, 2009
Tk4	Meloidogyne luci	Avirulent	Aydınlı & Mennan, 2019
V19	Meloidogyne incognita	Mi-1.2-virulent	Mıstanoğlu et al., 2020
V24	Meloidogyne incognita	Mi-1.2-virulent	Mıstanoğlu et al., 2020
V23	Meloidogyne javanica	Mi-1.2-virulent	Mıstanoğlu et al., 2020
V28	Meloidogyne javanica	Mi-1.2-virulent	Mıstanoğlu et al., 2020

### The testing of peppers

For all the nematode isolates, egg masses were collected from infected roots and hatched at 25°C. Second-stage juveniles (J2s) were collected from water every 48 hours and counted under the binocular stereo microscope. Each cultivar seedling at the four-leaf stage was transplanted into 250 cc plastic cups including autoclaved sandy soil mixture. 7 days after transplanting, each plant was separately inoculated with 1000 J2s of RKN isolates. The plants were incubated in a controlled growth chamber at 25°C, 65% relative humidity and 16:8 hours day/night cycle. The plants were gently uprooted 8 weeks after J2s inoculation and roots were cleaned from soil individually using tap water. There were five replicates for each combination (isolate x cultivar) according to a completely randomized design. Each root system was stained with Phloxine B solution for counting egg masses and investigated under a microscope (Öçal et al., 2018).

#### Data analyses

The data were analyzed using ANOVA with the statistical package SAS v. 9.0. Significant differences within nematode isolates and pepper cultivars were analyzed separately using Duncan's multiple range test (p≤0.05).

#### **Results and Discussion**

In the first experiment, pepper cultivars Safran F<sub>1</sub>, Carolina Wonder, Monstar F<sub>1</sub>, B4 F1 and KB401 were inoculated with avirulent RKN isolates, *M. incognita* (S6), *M. javanica* (K21), *M. arenaria* (K18) and *M. luci* (Tk4). Except for *M. javanica* K21 isolate, all RKN isolates reproduced on Safran F<sub>1</sub> and Mostar F<sub>1</sub> were known to be susceptible. *Meloidogyne javanica* K21 produced few or no egg masses on pepper cultivars. *M. luci* isolate produced the most egg masses Carolina Wonder genotype bearing *N* gene and B4 F<sub>1</sub>. Except *M. javanica*, all RKN isolates produced egg masses on local cultivar KB401. *Meloidogyne luci* multiplied less when compared with the other isolates on KB401, indicating that it may not be a good host for *M. luci*. As expected, three main RKN isolates did not reproduce on resistant Carolina Wonder (Table 2).

Table 2. The average number of egg masses of Mi-1.2-avirulent isolates on the whole root system in pepper cultivars

Pepper cultivars	Nematode species and their codes								
repper cultivars	M. incognita (S6)		M. javanica (K21)		M. arenaria (K18)		M. luci (Tk4)		
Safran F1	111.2±8.4	Ab <sup>x, y</sup>	0.2±0.2	Ca	32.6±1.9	Ва	38.0±7.8	Bb	
Carolina Wonder	4.4±0.9	Вс	$0.0\pm0.0$	Ba	0.6±0.6	Вс	68.4±5.9	Aa	
Mostar F1	126.2±13.9	Bab	0.2±0.2	Da	36.0±5.1	Ca	27.2±3.7	CDb	
B4 F1	7.4±3.5	Вс	0.0±0.0	Ca	0.0±0.0	Сс	31.8±2.6	Ab	
KB401	147.6±12.0	Ва	0.0±0.0	Ca	13.8±4.6	Cb	8.8±2.1	Сс	

<sup>&</sup>lt;sup>x</sup> Means (± standard error) in a column with the same lower-case letter are not significantly different in accordance with Duncan's multiple range test (p≤0.05).

y Means (± standard error) in a row with the same upper-case letter are not significantly different in accordance with Duncan's multiple range test (p≤0.05).

In the second experiment, pepper cultivars Safran F<sub>1</sub>, Monstar F<sub>1</sub>, Mert F<sub>1</sub>, Carolina Wonder, B4 F<sub>1</sub>, B5 and B6 were inoculated with *Mi-1.2*-virulent *M. incognita* isolates V19 and V24 and *Mi-1.2*-virulent *M. javanica* isolates V28 and V23. *Mi-1.2*-virulent *M. incognita* isolates caused high numbers of egg masses and galls on susceptible pepper cultivars Safran F<sub>1</sub>, Mostar F<sub>1</sub> and Mert F<sub>1</sub>, but reproduced no egg masses and galls on resistant cultivars Carolina Wonder and B4 F<sub>1</sub>. Similarly, *Mi-1.2*-virulent *M. incognita* isolates caused egg masses on B5 but did not on B6. *Mi-1.2*-virulent *M. javanica* isolates did not multiply on any of the pepper cultivars (Table 3).

	Table 3. The average	ge number of egg masse	es of <i>Mi-1</i> -virulent RKN	I isolates in pepper cultivars
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Donner cultivers	Nematode species and their codes							
Pepper cultivars	M. incognita (V19)		M. incognita (V24)		M. javanica (V28)		M. javanica (V23)	
Safran F1	64.8±9.8	Bb x, y	120.4±18.5	Aab	0.0±0.0	Ca	0.0±0.0	Ca
Mostar F1	94.8±23.9	Aab	102.6±6.0	Ab	$0.0\pm0.0$	Ва	$0.2\pm0.2$	Ва
Mert F1	107.6±11.2	Aa	135.6±16.9	Aa	$0.0\pm0.0$	Ва	$0.4 \pm 0.4$	Ва
Carolina Wonder	0.0±0.0	Ac	$0.0\pm0.0$	Ad	$0.0\pm0.0$	Aa	$0.0\pm0.0$	Aa
B4 F1	0.2±0.2	Ac	$0.0\pm0.0$	Ad	$0.0\pm0.0$	Aa	$0.0\pm0.0$	Aa
B5	65.6±7.9	Ab	70.8±11.1	Ac	$0.0\pm0.0$	Ва	$0.0\pm0.0$	Ва
B6	$0.0\pm0.0$	Ac	0.0±0.0	Ad	$0.0\pm0.0$	Aa	$0.2 \pm 0.2$	Aa

x Means (± standard error) in a column with the same lower-case letter are not significantly different in accordance with Duncan's multiple range test (p≤0.05).

Overall, in this study, avirulent *M. arenaria* and (a)virulent *M. incognita* isolates multiplied very well on susceptible peppers (Safran F1, Mostar F1 and Mert F1) but did not on Carolina Wonder. However, (a)virulent *M. javanica* isolates did not multiply all pepper cultivars tested. *M. luci* isolate was multiplied all pepper cultivars except for KB401. Pot tests showed that B4 F<sub>1</sub> were resistant to *Mi-1.2-*virulent and avirulent isolates of *M. incognita*, and *M. javanica* as well as avirulent isolate of *M. arenaria*. B4 F1 was obtained from crossing B6 with B5. Thus, the resistance in B4 F1 come from B6 line.

Castagnone-Sereno et al. (1992) reported that *Mi-1.2*-virulent RKNs lose their reproductive ability on susceptible pepper cultivars, surprisingly. A later study by Castagnone-Sereno et al. (2001) found that the *Mi-1.2*-virulent RKN populations were less able to reproduce on susceptible pepper cultivars, with few exceptions. *Mi-1.2*-virulent *M. incognita* populations collected from Greece could reproduce on resistant tomatoes but did not on susceptible pepper (Tzortzakakis & Blok, 2007). In another study, *Mi-1.2*-virulent *M. incognita* population generally failed to reproduce on susceptible pepper. In contrast to this finding, another *Mi-1.2*-virulent *M. incognita* population was able to reproduce on susceptible pepper (Tzortzakakis et al., 2016). In the present study, *Mi-1.2*-virulent *M. incognita* isolates developed on susceptible pepper cultivars but did not multiply on resistant Caroline Wonder, also B4 F<sub>1</sub> and B6 cultivars.

Caroline Wonder bearing *N* gene was resistant to *M. incognita*, *M. javanica*, *M. arenaria*, *Mi-1.2*-virulent *M. incognita* and *Mi-1.2*-virulent *M. javanica* isolates but was susceptible to *M. luci*. Our results related to *M. incognita*, *M. javanica*, *M. arenaria* are consistent with previous studies (Thies et al., 1998; Thies & Fery, 2000). To our best knowledge, no report exists on the response of *N* gene in pepper against *M. luci*. The study is the first to investigate the interaction between *M. luci* and *N* resistance gene. Results showed that *N* gene is susceptible to *M. luci* isolate tested.

As known, *M. luci* is one of the important RKNs due to its spread in Europe (Maleita et al., 2018). *Meloidogyne ethiopica* populations identified in Europe were later reidentified to be *M. luci* (Gerič Stare et al., 2017). *Meloidogyne luci* has been on the European Plant Protection Organization Warning list since 2017 (EPPO, 2017). However, there are limited studies on the host responses of plants to *M. luci* (Carneiro et al., 2014; Aydınlı & Mennan, 2016, 2019; Öçal et al., 2018; Şen & Aydınlı, 2021; Maleita et al., 2022). Carneiro et al. (2014) reported that *M. luci* can reproduce on susceptible pepper California Wonder. Şen & Aydınlı

yMeans (± standard error) in a row with the same upper-case letter are not significantly different in accordance with Duncan's multiple range test (p≤0.05).

(2021) found that one of the five pepper cultivars they tested was a poor host for *M. luci*. Maleita et al. (2022) investigated the response of ten pepper cultivars to *M. luci* and detected all of them as susceptible. In this study, *M. luci* multiplied on susceptible peppers Safran F<sub>1</sub> and Mostar F<sub>1</sub>. However, *M. luci* developed less than *M. incognita* on susceptible peppers. Also, *M. luci* developed at least on KB401 cultivar. *Meloidogyne luci* was commonly present in the northeast of Türkiye (Aydınlı & Mennan, 2016; Aydınlı, 2018). Besides, *M. luci* was found in Osmaniye, southeast of Türkiye (Gürkan et al., 2019). Our results showed that pepper cultivars that are not good hosts for *M. luci* may be used in areas infected with *M. luci*. Since tomato cultivars bearing the *Mi-1* gene and *Solanum torvum* Sw. (Solanales: Solanaceae) rootstock have been found resistant against *M. luci* in previous studies, these can be recommended for infected areas (Sargın & Devran, 2021).

It is recommended to alternate resistance genes to maintain sustainability of resistant varieties and to prevent virulent populations (Barbary et al., 2015). In this respect, results provide useful information on response of pepper cultivars to important RKNs, *M. incognita*, *M. javanica*, *M. arenaria* and *M. luci*.

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# Original article (Orijinal araştırma)

# First records of *Aulacus striatus* Jurine, 1807 (Hymenoptera Aulacidae) and its host, Xiphydria picta Konow, 1897 (Hymenoptera: Xiphydriidae), from Türkiye<sup>1</sup>

Aulacus striatus Jurine, 1807 (Hymenoptera Aulacidae) ve onun konukcusu Xiphydria picta Konow, 1897 (Hymenoptera: Xiphydriidae)'nın Türkiye'den ilk kayıtları

İlyas CAN2\*

#### Abstract

This study was performed in the Yeşilırmak delta of Samsun province located in northern Türkiye to reveal the diversity of rare wasp species. For this purpose, specimens belonging to the family Aulacidae and Xiphydriidae were collected with Malaise traps from various localities in the study area between 2022 and 2023. As a result, the occurrence of the parasitoid wasp Aulacus striatus Jurine, 1807 (Hymenoptera: Aulacidae) was reported for the first time in Türkiye. This is also the first record of genus Aulacus Jurine, 1807 (Hymenoptera: Aulacidae) from the country. Additionally, two putative hosts of A. striatus, Xiphydria picta Konow, 1897 and Xiphydria prolongata (Geoffroy, 1785) have been collected. Of these species, X. picta is new to the Turkish fauna. With these new records, the number of aulacids in Türkiye is raised up to six species in two genera, while the number of xiphydriids is raised up to three within one genus.

Keywords: Aulacidae, Aulacus striatus, new records, Xiphydriidae, Xiphydria picta

Bu calısma, Türkiye'nin kuzevinde ver alan Samsun ili Yesilırmak deltasında nadir yaban arısı türlerinin çeşitliliğini ortaya koymak amacıyla yapılmıştır. Bu amaçla 2022-2023 yılları arasında çalışma alanında çeşitli lokasyonlardan Aulacidae ve Xiphydriidae familyasına ait örnekler Malaise tuzakları ile toplanmıştır. Sonuc olarak parazitoid yaban arısı Aulacus striatus Jurine, 1807 (Hymenoptera: Aulacidae)'un Türkiye'deki varlığı ilk kez rapor edilmiştir. Bu Aulacus Jurine, 1807 (Hymenoptera: Aulacidae) cinsinin de Türkiye'deki ilk kaydıdır. Ek olarak A. striatus'un varsayılan iki konukçusu da, Xiphydria picta Konow, 1897 ve Xiphydria prolongata (Geoffroy, 1785) toplanmıştır. Bu türlerden X. picta Türkiye faunası için yenidir. Bu yeni kayıtlarla Türkiye'deki aulacid sayısı iki cinste altı türe, xiphydriid sayısı ise bir cinste üçe yükseldi.

Anahtar sözcükler: Aulacidae, Aulacus striatus, yeni kayıtlar, Xiphydriidae, Xiphydria picta

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#### Introduction

Aulacidae is a small family of Hymenoptera belonging to the superfamily Evanioidea. The extant aulacids includes two genera: *Aulacus* Jurine, 1807 with 122 species and *Pristaulacus* Kieffer, 1900 with 188 species (Turrisi et al., 2009; Turrisi, 2017). The genus *Pristaulacus* is represented in all zoogeographic regions except Antarctica, and *Aulacus* is also absent from the Afrotropical region (Jouault & Nel, 2022). Aulacids endoparasitoid of xylophagous larvae of some Hymenoptera and Coleoptera. Species of *Aulacus* Jurine, 1807 and *Pristaulacus* Kieffer, 1900 (Hymenoptera: Aulacidae), are related to the larvae of *Xiphydria* Latreille, 1803 (Hymenoptera: Xiphydriidae), beetles and borers (Coleoptera: Cerambycidae, Buprestidae) (Turrisi & Vilhelmsen, 2010; Sundukov & Lelej, 2015).

Aulacids have been the subject of limited research in Türkiye due to their rarity. The first published records of Turkish Aulacidae were those by Oehlke (1983) and a few species were subsequently recorded by Turrisi (2007, 2011). The faunistic and taxonomic aspects of the superfamily in Türkiye were recently summarized by Can (2023). According to that contribution, the family Aulacidae in the Türkiye is represented by a total of five species, all of which belong to the genus *Pristaulacus*.

Xiphydriids are a small family of wood borer sawflies that include around 150 species in 27 genera distributed worldwide, except for the Afrotropical region (Taeger & Blank, 2018). In Türkiye, this family is represented by a single genus *Xiphydria*, consisting of mid-sized to giant wasps with the pronotum characteristically stretched into an elongated (Johansson & Larsson, 2020) and only two *Xiphydria* species have been recorded so far, *Xiphydria camelus* (L., 1758) and *Xiphydria prolongata* (Geoffroy, 1785) (Hymenoptera: Xiphydriidae) (Baş, 1973; Özay, 1997).

The present work aims to contribute to the knowledge of Türkiye Aulacidae and Xiphydriidae fauna. In this context, a new genus (*Aulacus*) record has been provided for Türkiye, with the identification of *Aulacus striatus* Jurine, 1807 (Hymenoptera: Aulacidae). Additionally, one of its putative hosts, *Xiphydria picta* Konow, 1897 (Hymenoptera: Xiphydriidae), has been recorded for the first time from this country. A new distribution record of *X. prolongata* has also been added.

#### **Materials and Methods**

The material of this study was collected from three localities of Yeşilırmak Delta (Samsun province) located in the Central Black Sea region of Türkiye (Figure 1).



Figure 1. General view of collection localities in Yeşilırmak Delta of Samsun province: a) Çarşamba, Bafracalı (41°12'43.2"N, 36°44'07.7"E); b) Terme, Geçmiş village (41°15'57.4"N, 36°50'39.5"E); c) Terme, Gölyazı village (41°15'48.9"N 36°59'14.0"E).

The specimens were collected by the Malaise trap during the spring-summer of 2022 and 2023 from mid-March to late September. The specimens were removed from the traps and sorted monthly, then transferred to 70% ethanol. Subsequently, the specimens were pinned in the laboratory and turned into standard museum material. The voucher materials are deposited in the Entomology Research Laboratory, Department of Biology, Tokat Gaziosmanpaşa University (Tokat, Türkiye). Identification keys and descriptions in Oehlke (1983, 1984), Sun & Sheng (2007), and Sundukov & Lelej (2015) were used to identify the *Aulacus* specimen. The photographs of the specimens were taken using a Leica M205C (Leica Microsystems GmbH, Germany) stereomicroscope controlled by Leica Application Suite 3 software.

#### Results

Order Hymenoptera

**Suborder Apocrita** 

Family Aulacidae Shuckard, 1841

Genus Aulacus Jurine, 1807

Type species: Aulacus striatus Jurine, 1807

The species of *Aulacus* are distinct from the other aulacid genus *Pristaulacus* by the lack of the occipital carina (Figure 2b), the shape of hind coxae (Figure 2d), the simple tarsal claws (Figure 2e), and the presence of 2r-m in the forewing.

#### Aulacus striatus Jurine, 1807 (Figure 2)

Material examined. Samsun: Çarşamba, Bafracalı, 41°12'43.2"N, 36°44'07.7"E, 30 m, 28.V-19.VI.2023, ♀.

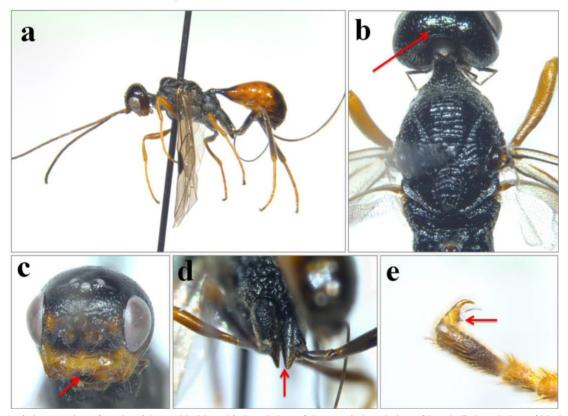


Figure 2. Aulacus striatus female: a) Lateral habitus; b) dorsal view of thorax; c) dorsal view of head; d) dorsal view of hind coxae; e) tarsal claws.

General distribution. Algeria, Austria, Belarus, Belgium, Czechia, England, Finland, France, Germany, Hungary, Kazakhstan, Netherlands, Norway, Poland, Russia (European), Slovakia, Sweden, Switzerland, Ukraine (Smith, 2001), Türkiye (new record).

Brief description. Body length (excluding ovipositor) is 7.4 mm (Figure 2a). Head mainly black (Figure 2c), with lower frons partly yellow; antenna entirely blackish-brown; clypeus dark yellow; mandible basally yellow and apically black (Figure 2c); coxae black; femora yellow except black hind femurs; tibiae and tarsi extensively reddish-orange; metasoma black with most of first tergite (except dorsal base) and second tergite reddish-brown; vertex shining, irregularly, and deeply punctured, frons between the antennal socket and ocelli oblique transverse carinulate; occiput with transverse-striate sculpture; anterior margin of clypeus with medial process (Figure 2c); propodeum weakly declivous; ovipositor 0.86 x forewing length.

#### Suborder Symphyta

Family Xiphydriidae Leach, 1815

Genus Xiphydria Latreille, 1803

Type species: Ichneumon camelus (L., 1758)

The species of *Xiphydria* are distinct from other Xiphydriidae genera with smooth and shiny vertex behind ocelli; hind wing with cell Rs; tarsal claws with distinct inner tooth; maxillary palpus with 5 or 6, labial palpus with 3 or 4 palpomeres.

#### Xiphydria picta Konow, 1897 (Figure 3a)

Material examined. Samsun: Terme, Gölyazı, 41°15′48.9″N 36°59′14.0″E, 30 m, 28.V-19.VI, 2♀♀.

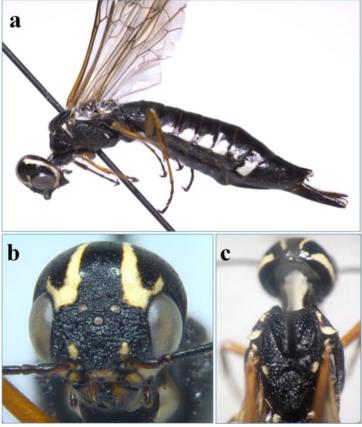


Figure 3. Xiphydria picta female: a) Lateral habitus; b) dorsal view of head; c) dorsal view of thorax.

General distribution. Austria, Croatia, Finland, France, Georgia, Hungary, Italy, Kazakhstan, Romania, Russia, Spain, Sweden, Switzerland, Ukraine (Sundukov, 2017), and Türkiye (new record).

Brief description. Body length (excluding ovipositor) is 15-17 mm. Head mainly black with two light white stripes extending from the eyes to the back; frons with white spots above antennal sockets; malar space, gena along outer genal carina up to the level of the top of eye with large creamy white mark; mandible medially yellow; clypeus yellowish-white except for black anterior edge; antenna black; thorax black with pronotal collar partly white; legs reddish brown, usually with coxae, trochanters, trochantelli blackish brown and tarsi blackish; abdomen black with white lateral spots usually on terga 2-8; that on tergum 8 largest and on terga 7 smallest. Frons and interantennal area coarsely reticulate; lateral part of clypeus, malar space and gena shallowly finely striate; vertex and upper part of gena smooth, shiny, with few shallow and irregular punctures (Figure 3b).

#### Xiphydria prolongata (Geoffroy, 1785) (Figure 4)

Material examined. Samsun: Çarşamba, Bafracalı, 41°12′43.2″N, 36°44′07.7″E, 30 m, 28.V-19.VI.2023, 2♀♀.

General distribution. This species is spread throughout Europe, Russia, Türkiye and USA (Baş, 1973; Taeger & Blank, 2019).

Distribution in Türkiye. Afyonkarahisar, Ankara, Burdur, Bursa, Kırklareli, Kocaeli, Sivas (Baş, 1973; Özay, 1997; Budak & Korkmaz, 2023).

Remarks. This species was detected for the first time in Samsun province.



Figure 4. Lateral view of Xiphydria prolongata female.

#### **Discussion**

With this study, the genus *Aulacus* was recorded for the first time from Türkiye based on a female specimen belonging to *Aulacus striatus*. This species is mostly distributed in the Palaearctic region from England to China and from Scandinavia to Algeria. However, Chen et al. (2016) stated that the record in China (Inner Mongolia) is unclear. The present note adds important data to fill the distribution gap of the species in the Western Palaearctic region.

Aulacids are not easily observed in their natural environment due to their unique biological traits and are not often collected by most conventional collection methods. As a result, many species are known from a few or just one specimen (Huflejt & Wiśniowski, 2012; Turrisi, 2017). In this study, only one specimen of *A. striatus* was collected by Malaise traps during the two-year research period.

Xiphydria camelus, Xiphydria longicollis (Geoffroy, 1785), X. picta, X. prolongata are indicated as hosts of Aulacus striatus in Europe (Sundukov & Lelej, 2015). X. camelus was known from the Marmara and Eastern Black Sea regions of Türkiye and reported in Alnus glutinosa (L.) Gaertn. and Betula spp. (Fagales: Betulaceae) trees. Xiphydria prolongata was known from the Central Anatolia, Aegean, Marmara, and Mediterranean regions of Türkiye and reported in Populus nigra L., Salix alba L., Salix fragilis L. (Malpighiales: Salicaceae) and Ulmus carpinifolia Borkh. (Rosales: Ulmaceae) trees (Baş, 1973; Özay, 1997). In this study, the specimen of A. striatus was obtained together with its putative host, X. prolongata (Figure 4), in the same Malaise trap bottle. With this study, X. prolongata has been recorded for the first time from the Central Black Sea region.

In addition to the previously known species (*X. camelus* and *X. prolongata*) in Türkiye, the existence of *X. picta* was detected for the first time in this study. The characteristics of *X. picta* are similar to the widely distributed *X. camelus* and can be easily confused with it (Johansson & Larsson, 2019). However, *X. picta* can usually be distinguished by richer pale markings on the head and face (Figure 3b).

The present note makes an important contribution to the available knowledge of the Turkish Aulacidae and Xiphydriidae families. Based on the new records, the total number of Aulacidae in Türkiye has elevated to six species, and the total number of Xiphydriidae is three.

# **Acknowledgment**

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# Original article (Orijinal araştırma)

# Description of *Podothrombium sultanae* sp. nov. (Trombidiformes: Podothrombiidae) from Türkiye and an updated key to species<sup>1</sup>

Türkiye'den *Podothrombium sultanae* sp. nov. (Trombidiformes: Podothrombiidae)'nin tanımı ve türlere ilişkin güncellenmiş bir anahtar

Firdevs ERSIN4\*

İbrahim CAKMAK<sup>3,5</sup>



#### Abstract

Members of Parasitengona belong to the suborder Prostigmata and are one of the important groups of terrestrial mites. They are ectoparasitic in their larval stage whereas their post-larval forms feed on different arthropods as freeliving predators. The genus Podothrombium Berlese, 1910 (Trombidiformes: Podothrombiidae) has 19 described species globally, with only two species currently known from Türkiye. This paper describes a new member of this genus, Podothrombium sultanae sp. nov., from Murat Mountain, Kütahya, Türkiye in 2023. Larvae of this species, collected from moss and leaf litter 1181 m a.s.l on Murat Mountain, are described, illustrated and an updated key to species of Podothrombium worldwide (larva) has been added. It is recommended that further investigations be conducted to understand the distribution of these mites and their roles in the environment in Türkiye.

Keywords: Acari, Iarva, Murat Mountain, Prostigmata, terrestrial Parasitengona

# Öz

Prostigmata alt takımında yer alan Parasitengona türleri karasal akarların önemli gruplarından biridir. Larva döneminde ektoparazittirler, larva sonrası formları ise serbest yaşayan avcılar olarak farklı eklembacaklılarla beslenirler. Podothrombium Berlese, 1910 (Trombidiformes: Podothrombiidae) cinsinin dünyada tanımlanmış 19 türü bulunmaktadır ve şu anda Türkiye'den sadece iki türü bilinmektedir. Bu çalışmada, Türkiye, Kütahya, Murat Dağı'ndan 2023 yılında bu cinsin yeni bir üyesi olan Podothrombium sultanae sp. nov. tanımlanmaktadır. Murat Dağı'ndaki 1181 m vükseklikteki vosun ve vaprak döküntülerinden toplanan bu türün larvaları, tanımlanmıs, cizilmis ve dünvadaki Podothrombium türleri (larva) için güncellenmiş bir anahtar eklenmiştir. Bu akarların Türkiye'de dağılımının ve çevre üzerindeki rollerinin anlaşılması için daha ileri araştırmaların yapılması önerilmektedir.

Anahtar sözcükler: Akar, larva, Murat Dağı, Prostigmata, karasal Parasitengona

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#### Introduction

The genus *Podothrombium* Berlese, 1910 (Trombidiformes: Podothrombiidae) was established on the basis of larval and post-larval forms. The only other genus of the family Podothrombiidae Thor, 1935 namely *Kurilothrombium* Mąkol, 1999 was similarly established solely on the basis of post-larval forms (Makol & Marusik, 1999; Makol & Wohltmann, 2012). Nineteen species of *Podothrombium* have been described worldwide based on the larvae (Saboori et al., 2015). Saboori et al. (2015) presented a key to the larval species of *Podothrombium* but *Podothrombium manolatesicus* Haitlinger, 2006 was not included. No new species from this genus has been described since 2016.

Studies on terrestrial parasitengone of Türkiye are in progress. Many parts of the country are yet to be investigated, and it seems the number of terrestrial Parasitengona is more than those reported hitherto. Only two species of *Podothrombium* are reported from Türkiye: *Podothrombium filipes* (Koch, 1837) reported by Adil & Sevsay (2014) and Sevsay (2015), and *Podothrombium macrocarpum* Berlese, 1910 reported by Doğan et al. (2015) and Sevsay (2017). Both species were reported from larval and post-larval instars (Adil & Sevsay, 2014; Doğan et al., 2015).

A new species of *Podothrombium* from Murat Mountain, Kütahya, Türkiye based on larvae is described below and incorporated into a global key to the genus.

#### **Materials and Methods**

#### Collection and preparation of specimens

Larval specimens were collected in the summer of 2023 in Murat Mountain, Gediz, Kütahya Province of Türkiye. Moss and leaf litter samples were transferred in plastic bags to the laboratory for extraction of mites by Berlese funnel. Four specimens of interest were preserved in ethanol (75%). These specimens were then cleared with Nesbitt's solution, mounted in Hoyer's medium on microscope slides (Walter & Krantz, 2009), and examined using an Axiolmager A2 microscope (Carl Zeiss AG, Jenna, Germany) equipped with differential interference and phase contrast illumination to make measurements and prepare illustrations. Measurements (in  $\mu$ m) are given for holotype followed by paratypes in parentheses, with the terminology and abbreviations used in accordance with Makol & Wohltmann (2000) and Saboori et al. (2009).

#### Results

Family Podothrombiidae Thor, 1935

Genus Podothrombium Berlese, 1910

Podothrombium sultanae sp. nov. (Figures 1-4)

Diagnosis (larva)

First row of dorsal idiosomal setae with less than 12 regularly arranged setae; IP < 1550, Ta I with 14-18 eupathidia and Ta I-II with two solenidia.

#### Description

Dorsum (Figure 1a). Idiosoma 727 (596-675) long and 543 (422-527) wide. Prodorsal scutum punctate with three pairs of normal setae (AM, AL and PL) and one pair of sensilla (S). AL finely and sparsely barbed whereas AM and PL distinctly barbed. AL and PL thicker than AM and S. Sensilla smooth in all specimens except holotype which has one barb. AL with one shaft line. Two pairs of eyes lateral to posterolateral angles of scutum, each pair situated on punctate sclerite 43 × 26 (40-45 × 25-27); diameter of anterior pair 18 (17-21) and posterior one 15 (12-15). Scutellum punctate, with two barbed setae and

deeply concave posterior border, anterior border almost straight and lateral borders slightly convex. Hysterosoma dorsally with 38 (38-39); fD = 8(+2), 8, 8-9, 6, 6. All dorsal setae arise from punctate platelets.

Venter (Figure 1b). Ventral surface with barbed setae 3a; beyond coxae III 26 (20-22) barbed setae (fV = 20-26) and anus, all ventral setae arising from platelets. Coxa I with two long barbed setae. Urstigma placed between coxae I and II. Coxae II and III each with one barbed setae. All coxae punctate.

Gnathosoma (Figure 2). Palpal setal formula (fPp) = 0-B-N-NNN-2B6N $\omega$ ζ. Palpal femur with one barbed and palpal genu with one nude seta. Palptibia with three nude setae of which one is thick, and one entire terminal claw. Palptarsus with two barbed and six nude setae, one eupathidium and one solenidion. Cheliceral base 68 (60-69) long; cheliceral blade slightly curved, 16 (15-18) long with one subterminal tooth. Seta cs nude, 8 (7-8) long and subcapitular seta (bs) long with few barbs, 35 (32-37) long.

Legs (Figures 3 & 4). Segmentation formula: 6-6-6. Leg setal formula. Leg I:  $Ta-2\omega$ ,  $0-1\epsilon$ ,  $14-18\zeta$ , 30-37n;  $Ti-2\phi$ ,  $1\kappa$ , 5n;  $Ge-2\sigma$ ,  $1\kappa$ , 4n; Fe-5n; Tr-1n. Leg II:  $Ta-2\omega$ ,  $1\epsilon$ ,  $2-3\zeta$ , 22-26n;  $Ti-2\phi$ , 5n;  $Ge-1\sigma$ ,  $1\kappa$ , 3n; Fe-4n; Tr-1n. Leg III: Ta-19-22n; Ti-5n;  $Ge-1\sigma$ , 3n; Fe-4n; Tr-1n. Only the holotype has a famulus on Ta I and three eupathidia only on one side on Ta I.

IP = 1486 (1392-1506)

The distal end of all legs has two lateral claws with a median claw-like empodium. Table 1 presents all relevant measurements.

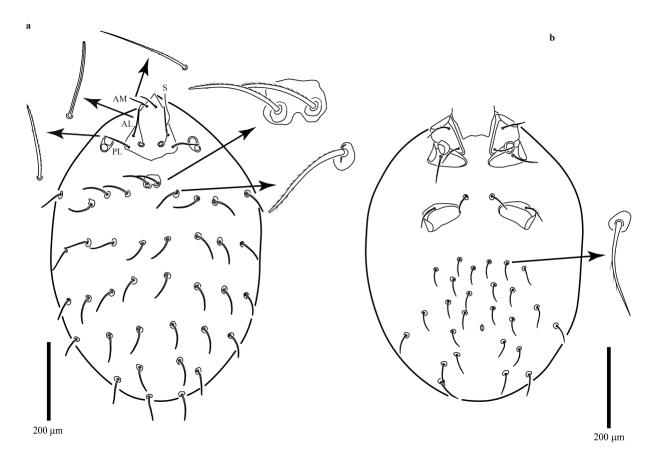


Figure 1. Idiosoma of Podothrombium sultanae sp. nov. larva: a) dorsal and b) ventral views with some setae shown in detail.

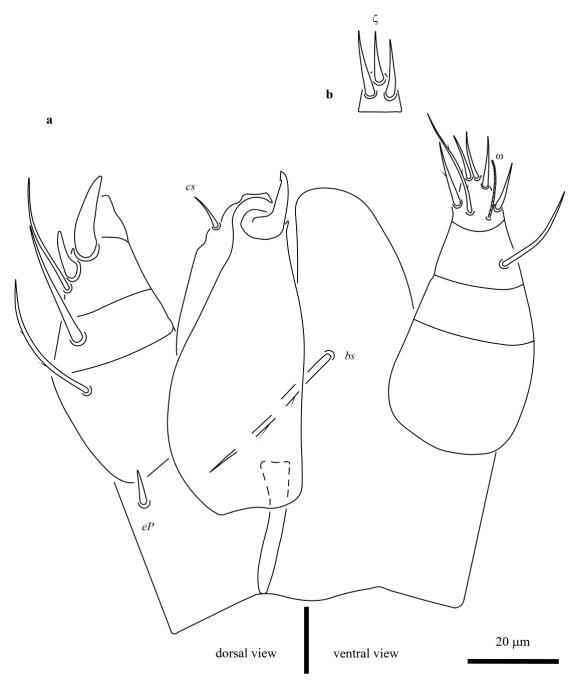


Figure 2. Gnathosoma of Podothrombium sultanae sp. nov. larva: a) dorsal (left) and ventral (right) view and b) palpal tarsus in dorsal view.

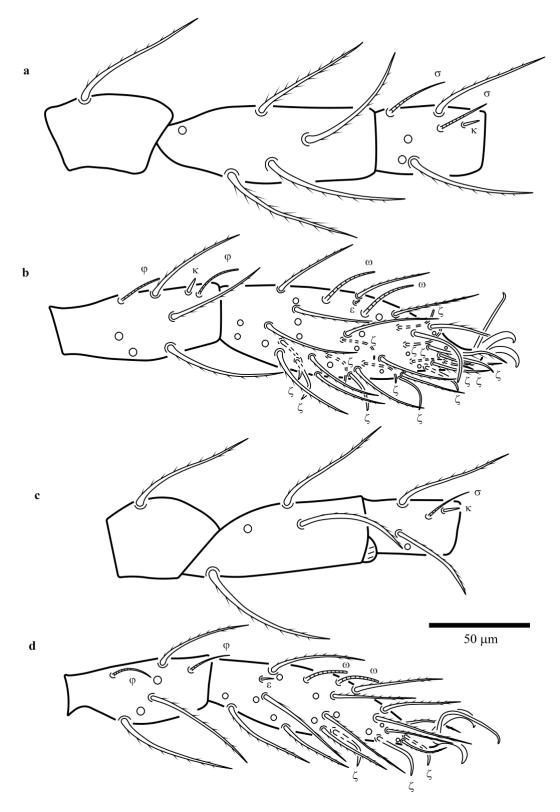


Figure 3. Legs I and II of *Podothrombium sultanae* sp. nov. larva: a) Tr-Ge I, b) Ti and Ta I, c) Tr-Ge II, and d) Ti and Ta II.

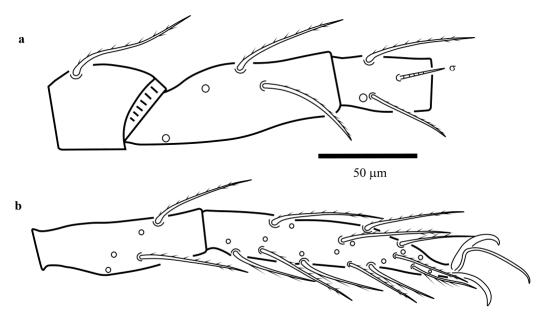


Figure 4. Leg III of *Podothrombium sultanae* sp. nov. larva: a) Tr-Ge III and b) Ti and Ta III.

Table 1. Measurements (µm) of *Podothrombium sultanae* sp. nov. larvae: a) holotype; and b-d) paratypes

Character	а	b	С	d	Range	Character	а	b	С	d	Range
IL	727	596	675	599	596-727	3a	54	52	52	54	52-54
IW	543	422	527	466	422-543	3b	61	62	60	56	56-62
AW	90	77	90	87	77-90	Ta I (L)	130	137	128	126	126-137
PW	115	107	108	107	107-115	Ta I (H)	42	36	39	39	36-42
AA	24	21	22	26	21-26	Ti I	87	85	73	77	73-87
SB	53	52	50	54	50-54	Ge I	55	59	55	51	51-59
ASB	109	118	117	115	109-118	Fe I	108	114	103	107	103-114
PSB	41	30	31	33	30-41	Tr I	51	42	38	45	38-51
SD	150	148	148	148	148-150	Cx I	85	66	73	100	66-100
W	136	128	129	126	128-136	Leg I	516	503	470	506	470-516
AP	31	24	23	25	23-31	Ta II (L)	116	122	112	111	111-122
MA	75	78	77	73	73-78	Ta II (H)	31	26	32	30	26-32
AL	65	61	58	62	58-65	Ti II	75	75	64	66	64-75
PL	72	67	69	66	66-72	Ge II	43	50	46	43	43-50
AM	55	61	51	67	51-67	Fe II	89	93	82	91	82-93
S	133	105	125	146	105-146	Tr II	44	51	45	46	44-51
SL	70	52	59	60	52-70	Cx II	94	95	86	92	86-95
SS	26	28	26	35	26-35	Leg II	461	486	435	449	435-486
HS	32	36	33	36	32-36	Ta III (L)	133	132	128	125	125-133
LSS	45	46	42	51	42-51	Ta III (H)	26	25	22	24	22-26
DS	56-73	48-67	52-68	56-77	48-77	Ti III	86	92	81	86	81-92
PDS	57-66	62-70	60-66	63-70	57-70	Ge III	51	54	53	52	51-54
cs	8	8	8	7	7-8	Fe III	102	94	92	97	92-102
bs	35	36	32	37	32-37	Tr III	48	59	46	53	46-59
1a	55	52	61	56	52-61	Cx III	89	86	87	90	86-90
1b	64	62	56	62	56-64	Leg III	509	517	487	503	487-517
2b	65	69	57	71	57-71	IP	1486	1506	1392	1458	1392-1506

Etymology. The species is named in honor of Dr. Sultan Çobanoğlu, emeritus professor of Ankara University, for her substantial contributions to the study of terrestrial Parasitengona in Türkiye.

Type material. Holotype (ARS-20231212-1a) and three paratypes (ARS-20231212-1b, 1c and 1d), came from moss and leaf litter collected by Firdevs Ersin in June 2023 in Murat Mountain, Gediz, Kütahya, Türkiye; 38° 58′ 58.9″ N, 29° 42′ 28.1″ E, 1,181 m. a.s.l., and have been deposited in the Acarological collection of Jalal Afshar Zoological Museum, Faculty of Agriculture, University of Tehran, Karaj, Iran.

#### Remarks

Podothrombium sultanae sp. nov. is a member of the species group with 7-10 setae on the first row of dorsal hysterosomal setae and IP < 1850. Five species share this feature: P. filipes occurring in all over Western Palearctic), Podothrombium paucisetarum Zhang & Xin, 1989 (occurring in China), Podothrombium verae Haitlinger, 1995 (occurring in Poland, Slovakia, and Slovenia), Podothrombium manolatesicus Haitlinger, 2006 (occurring in Greece) and *Podothrombium zlatarum* Saboori et al., 2015 (occurring in Montenegro) (Haitlinger, 1994, 1995, 2006a; Makol, 2005; Makol & Wohltmann, 2012; Saboori et al., 2015; Haitlinger & Šundić, 2016). The new species differs from P. filipes in nude seta on palpal genu (vs. barbed in P. filipes), nude setae on palpal tibia (vs. with at least one barbed seta), number of solenidia on palpal tarsus (1 vs. 2-3), placement of microseta on Ti I (prior to distal solenidion vs. after distal solenidion), and posterior border of scutellum (deeply concave vs. convex); from P. paucisetarum in number of normal setae on Ta I (30-37 vs. 14-17), on Ta II (22-26 vs. 14-16), on Ta III (19-22 vs. 14), number of eupathidia on Ta I (14-18 vs. 7-8), number of solenidia on Ta I (2 vs. 1), number of solenidia on Ta II (2 vs. 1), seta on palpal genu nude (vs. barbed in P. paucisetarum), shape of scutellum (deeply concave posterior border vs. convex posterior border), fV (20-26 vs. 34-40); from P. verae in number of barbed setae on palp tarsus (2 vs. 0), number of eupathidia on palp tarsus (1 vs. 2), number of solenidia on palp tarsus (1 vs. 2), number of eupathidia on Ta I (14-18 vs. 6-10), on Ta II (22-26 vs. 15-20), and solenidia on Ta I placed in the middle the segment (vs. near the basal 1/4 of the segment), number of solenidia on Ta I (2 vs. 1), number of solenidia on Ta II (2 vs. 1); from P. manolatesicus in number of normal setae on Ta I (30-37 vs. 19), on Ta II (22-26 vs. 17), on Ta III (19-22 vs. 17), number of solenidia on Ta I (2 vs. 1), number of solenidia on Ta II (2 vs. 1), AM (barbed vs. nude), seta on palpal femur barbed (vs. nude), number of barbed setae on palp tarsus (2 vs. 0), Ti I (73-87 vs. 104), Fe I (82-93 vs. 128), Ti II (64-75 vs. 90), Fe II (82-93 vs. 118), Ti III (81-92 vs. 112), Fe III (92-102 vs. 130); and from P. zlatarum in setae on palpal femur barbed (vs. nude), number of barbed setae on palp tarsus (2 vs. 1), number of normal setae on papal tarsus (8 vs. 6), number of setae on H row (6 vs. 9-11), shape of scutellum (deeply concave posterior border vs. slightly concave or straight), Ta I (126-137 vs. 151-173), Fe I (82-93 vs. 124-136), Ta II (111-122 vs. 129-146), Fe II (82-93 vs. 101-116), Ta III (125-133 vs. 149-161), Ti III (81-92 vs. 99-106), Fe III (92-102 vs. 114-129), and IP (1392-1506 vs. 1672-1846).

# Key to species of *Podothrombium* (larva) -IP > 1400 ......5 6. NDV 150, number of setae on first row of idiosoma 22-31 ............ P. karlovaicus Haitlinger, 2003 -NDV 120, number of setae on first row of idiosoma < 16 ........... P. svalbardense Oudemans, 1930 -First row of dorsal setae with 6-10 setae ......10 -IP < 1850 ......12 12. First row of dorsal setae with 6 setae ......13 -First row of dorsal setae with 7-10 setae ......14 -Ta I < 100, IP 1130-1234...... P. xianicum Haitlinger, 2006a

Note: *Podothrombium faeroense* Trägårdh, 1931 is not included due to insufficient data in its original description.

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# Original article (Orijinal araştırma)

# A study on the Rhyparochromidae Amyot & Serville, 1843 (Hemiptera: Heteroptera) fauna of Amasya Province (Türkiye) with new records<sup>1</sup>

Yeni kayıtlarla Amasya ili (Türkiye) Rhyparochromidae Amyot & Serville, 1843 (Hemiptera: Heteroptera) faunası üzerine bir araştırma

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#### **Abstract**

In this study, 367 adults from the Rhyparocromidae (Hemiptera: Heteroptera) family were gathered between 2020 and 2021 from 51 various locations in the Amasya Province (Türkiye). After the specimens were identified, it was discovered that 40 species of the 22 genera that make up the Rhyparocromidae family had been reported. The findings have revealed that all species are new records for the Rhyparocromidae fauna of Amasya province. Among these, the species *Drymus ryeii* Douglas & Scott, 1865, is a new record for the Heteroptera fauna of Türkiye. Additionally, 17 species are new records for the Black Sea Region while 7 species are new records for the Central Black Sea Region. The species *Emblethis amplus* Seidenstücker, 1987, are given second locality from Türkiye. In addition to all of the previously provided data, the study included a chorotype analysis of Rhyparochromidae species and updated the distribution area of these species with new locality information. Rhyparochromidae species from Amasya have been categorized into 13 groups based on chorotype analysis.

Keywords: Chorotypes analysis, Drymus ryeii, new records, Rhyparochromidae, Türkiye

#### Öz

Bu çalışmada, 2020-2021 yılları arasında Amasya ilinde (Türkiye) 51 farklı lokaliteden Rhyparocromidae (Hemiptera: Heteroptera) familyasına ait 367 ergin toplanmıştır. Örneklerin teşhis edilmesi ile Rhyparocromidae familyasını oluşturan 22 cinse ait 40 tür kaydedilmiştir. Bulgular, tüm türlerin Amasya ili Rhyparocromidae faunası için yeni kayıt olduğunu göstermektedir. Bunlardan *Drymus ryeii* Douglas & Scott, 1865 taksonu Türkiye Heteroptera faunası için yeni kayıttır. Ayrıca, 17 tür Karadeniz Bölgesi için, 7 tür Orta Karadeniz Bölgesi için yeni kayıt olarak verilmiştir. *Emblethis amplus* Seidenstücker, 1987 türüne ait Türkiye'den ikinci lokalite kaydı verilmiştir. Çalışma, daha önceki verilere ek olarak Rhyparochromidae türlerinin korotip analizini de içermekte ve bu türlerin dağılım alanlarını yeni lokalite bilgileri ile güncellemektedir. Amasya'dan elde edilen Rhyparochromidae türleri korotip analizine dayalı olarak 13 gruba ayrılmıştır.

Anahtar sözcükler: Korotip analiz, Drymus ryeii, yeni kayıtlar, Rhyparochromidae, Türkiye

<sup>&</sup>lt;sup>1</sup> This study was derivated from the MSc thesis of the first author.

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#### Introduction

Currently there are approximately 45 000 species of suborder Heteroptera (Hemiptera) in the world and 1632 genera comprising approximately 9 365 species of the Palaearctic region (Aukema et al., 2013; Henry, 2017). There are presently over 33 820 insect species reported to exist in Türkiye, according to Tezcan (2020). Of those, 1650 species belonging to suborder Heteroptera (Dursun & Fent, 2022; Çerçi & Koçak, 2023). According to these data, approximately 21% of the Heteroptera species found in the Palearctic region are distributed in Türkiye. Of these species, 239 species belonging to 23 families have type localities in Türkiye. Of those, 123 species are known to be endemic to the Turkish fauna (Önder et al., 2006; Dursun & Fent, 2015, 2017, 2018; Çerçi & Koçak, 2017, 2023).

One of the largest families within the superfamily Lygaeoidea (Heteroptera), the Rhyparochromidae Amyot & Serville, 1843, is distributed throughout the world. In the Palaearctic region, 573 species from 136 genera have been identified (Péricart, 2001). Of those, 125 species belonging 41 genera are distributed in Türkiye (Péricart, 2001; Çerçi & Koçak, 2023). The specimens of the Rhyparochromidae family are distributed especially in various reeds, swamps and wetlands (Péricart, 1999a). Surface of body colors are commonly dark brown, chestnut or black. Red parts or light patterned colors are sometimes found in certain parts of the body. The pronotum has usually narrow or wide lamelliform lateral edges. Wing polymorphism is dominated by macropterism, except for Plinthisinae. Hemelytra are dotted, especially on the clavus. Most species of this family lay their eggs of the aphanoid type, arranged individually on various plant debris. Species of this family mostly live among plant debris. It is rarely found in living plants except nymphs. They feed on the juice of various seeds by piercing them from a first Instar (Péricart, 1999a).

The geographical distribution of animals and plants can be indicated model-wise by chorotypes. The Chorotypes are known a classification based on distribution modeling obtained from comparative analysis of the geographical distributions of higher taxa, genera and species (Vigna Taglianti et al., 1999). With the chorotype study, the place of origin of faunal elements in a region can be determined. The chorotype studies can provide a useful tool for biogeographical research in a region.

Different microclimatic conditions, rich relief with fertile plans and high mountains, and transitional position between Central Anatolia and the Black Sea regions are the reasons for the rich biodiversity of Amasya. There are not faunistic and taxonomic studies on the family Rhyparochromidae from Amasya Province (Türkiye). Giving new records for the family Rhyparochromidae fauna in Amasya, evaluating ecological and chorotype data for the species that have been recorded, and opening a new way for scientific and ecological studies in the region are the aims of this first study.

#### **Materials and Methods**

The research material consists of 367 adult specimens (167 males and 200 females) collected from 51 localities with several vegetation and habitats from Amasya in the years from 2020 to 2021 (Figure 1). The specimens were collected from herbaceous vegetation with a sweep net and under stones, plants and bark of trees with the forceps. Plants on which a specimen was captured were recorded. All samples were stored in the Entomology lab in capped tubes filled with 70% ethanol. To prepare the male genitalia (pygophore, paramere, and aedeagus), used for additional identifications, the specimens were softened in hot water (80 -100°C) in the laboratory. The species identification was done using the keys of Stichel (1960) and Péricart (1999a, b) under a stereomicroscope Leica EZ4. The Palearctic region distributions of the species are given by examining the Catalogue of Aukema (2018). The map was created using Google Earth Pro (Anonymous, 2023). The material is added to the Department of Biology's collection at Amasya University's Faculty of Science and Arts (Amasya, Türkiye).

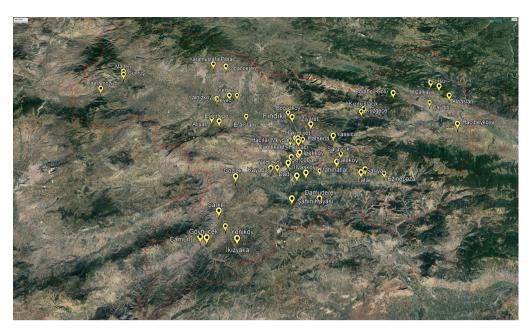


Figure 1. The study area in Amasya (Anonymous, 2023).

#### **Research Results**

Rhyparochromidae Amyot & Serville, 1843

Rhyparochrominae Amyot & Serville, 1843

Tribe: Drymini Stål, 1872

Genus: Drymus Fieber, 1860

Subgenus: Sylvadrymus Le Quesne, 1956

Drymus ryeii Douglas & Scott, 1865

Material examined. **Amasya**: Dadıköy, 40°33'44"N 35°48'28"E, 521 m, 21.IV.2021, ♂.

Distribution in Türkive. New record for the Heteroptera fauna of Türkive.

Distribution in Palaearctic Region. **Europe**: Austria, Belgium, Bulgaria, Byelorussia, Croatia, Czech Republic, Denmark, Estonia, Finland, France, Great Britain, Germany, Greece, Hungary, Ireland, Italy, Latvia, Liechtenstein, Luxembourg, Moldavia, Netherlands, Norway, Poland, Romania, Russia (Central European Territory, North European Territory, South European Territory), Serbia, Slovakia, Slovenia, Spain, Sweden, Switzerland, Ukraine. **Asia**: Georgia (?), Iran, Japan (?), Russia (East Siberia, West Siberia) (Aukema, 2018).

Note. The specimen was collected under leaf debris of *Pinus* sp., Chorotype: Sibero-European.

Redescription of male. Surface of body black and pitted (Figure 2a). Antennae black with short yellowish brown hairy. 1st antennal segment passing distal head. Lengths of antennae segments I-IV (mm): 0.48, 0.80, 0.53, 0.69 (Figure 2a). Rostrum blackish brown extends to middle coxae. Length of Proximal edge of Pronotum 1,6 mm. and median 1.00 mm. Hemelytra blackish brown, exocorium yellowish brown. Membrane smoky yellow, reaching apex of abdomen, membranous veins light, Connexivum and dorsum black. Pectus and ventral black. Lateral surface of venter yellow dense and short hairy. Femur black, distal part of fore femur with two small spines, tibia blackish brown with superficially short hairy, tarsus light brown. Paramere as in Figure (2b). Length: 4.6mm.

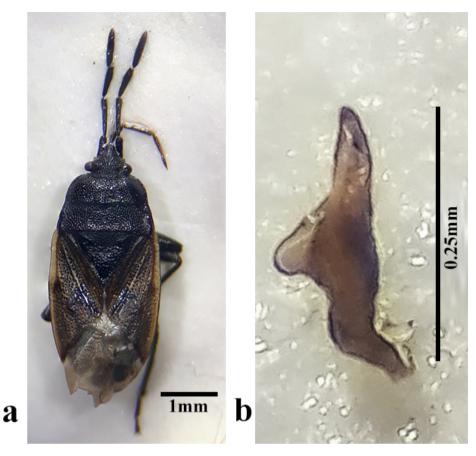


Figure 2. Drymus ryeii Douglas & Scott, 1865, a) Body (dorsal), b) Paramere.

Genus: Eremocoris Fieber, 1860

#### Eremocoris fenestratus (Herrich-Schaeffer, 1839)

Material examined. **Amasya**: Çamlıkent, 31.VIII.2021,  $\circlearrowleft$ , 2 $\circlearrowleft$  $\lozenge$ ; Hacılar Meydanı, 21.VIII.2021,  $\circlearrowleft$ ; 03.IV.2021,  $\circlearrowleft$ ; Kırklar, 02.IX.2021,  $\circlearrowleft$ , 2 $\circlearrowleft$  $\lozenge$ ; İpekköy, 24.VIII.2021,  $\circlearrowleft$ ; Kızılkışlacık, 10.IX.2021,  $\circlearrowleft$ ,  $\circlearrowleft$ ; Göynücek: İkizyaka, 03.IX.2021,  $\circlearrowleft$ .

Distribution in Türkiye. Adana, Ankara, Antalya, Bitlis, Burdur, Diyarbakır, Gaziantep, Hatay, Isparta, İzmir, Kahramanmaraş, Karaman, Kastamonu, Mersin, Muğla (Hoberlandt, 1956; Lodos et al., 1978, 1989; Önder et al., 1981, 2006; Péricart, 1999a; Fent & Japoshvili, 2012; Matocq et al., 2014; Küçükbasmacı & Kıyak, 2015; Çerçi & Koçak, 2023).

Note. This species is a new record for Central Black Sea Region. The specimens were collected under *Rosa canina* and *Junglans* sp., Chorotype: Sibero-European.

Genus: Ischnocoris Fieber, 1860

#### Ischnocoris hemipterus (Schilling, 1829)

Material examined. Gümüşhacıköy: Gümüş, 07.IX.2021, ♀.

Distribution in Türkiye. Edirne, Kayseri (Péricart, 1999a; Fent & Okyar, 2022).

Note. This species is a new record for Black Sea Region. The specimen was collected from annual herbaceous plant, Chorotype: European.

Genus: Scolopostethus Fieber, 1860

#### Scolopostethus affinis (Schilling, 1829)

Material examined. Taşova: Boraboy Gölü, 29.VIII.2021, ♀.

Distribution in Türkiye. Ankara, Antalya, Bolu, Çanakkale, Erzurum, Gaziantep, Hatay, İzmir, Karaman, Konya, Nevşehir, Ordu, Sinop, Sivas, Zonguldak (Puton & Noualhier, 1895; Hoberlandt, 1956; Lodos et al., 1978, 1989; Péricart, 1999a; Önder et al., 2006; Yazıcı et al., 2015; Çerçi & Koçak, 2023).

Note. This species is a new record for Amasya province. The specimen was collected under leaf debris of *Pinus* sp., Chorotype: Asiatic-European.

#### Scolopostethus cognatus Fieber, 1861

Material examined. **Amasya:** Serçoban, 09.IX.2021, ♀.

Distribution in Türkiye. Adana, Antalya, Gaziantep, Hatay, Mersin (Önder et al., 2006).

Note. This species is a new record for Black Sea Region. The specimen was collected under leaf debris of *Pinus* sp., Chorotype: European-Mediterranean.

According to Aukema et al. (2013) and Aukema (2018) the occurrence in Anatolia needs to be confirmed. Therefore, the specimen was redescribed: Antennae segments thick, half the body length, 1st antennal segment wheat yellow, 2nd segment wheat yellow, distal brown, 3rd segment brown, the base of 4th segment brown, distal part wheat yellow. Laterally of pronotum wheat yellow. Wings and legs conform to Péricart's description (1999a). The mesopleura without tubercles. Total length of body 3.9 mm. This finding removes any doubt about the existence of *S. cognatus* species in Türkiye.

#### Scolopostethus pictus (Schilling, 1829)

Material examined. Taşova: 29.VIII.2021, Boraboy, 3♀♀.

Distribution in Türkiye. Adana, Antalya, Balıkesir, Burdur, Bursa, Düzce, Hatay, İzmir, Kahramanmaraş, Karaman, Kocaeli, Mersin, Muş, Ordu, Osmaniye (Horváth, 1883, 1901; Puton & Noualhier, 1895; Lodos et al., 1978, 1989; Önder et al., 1983, 2006; Péricart, 1999a; Dursun & Fent, 2016; Cerci & Kocak, 2023).

Note. This species is a new record for Amasya province. The specimen was collected under leaf debris of *Pinus* sp., Chorotype: European.

Tribe: Gonianotini Stål, 1872

Genus: Aphanus Laporte, 1833

#### Aphanus rolandri (Linnaeus, 1758)

Distribution in Türkiye. Adana, Ankara, Antalya, Bolu, Balıkesir, Bingöl, Çorum, Elazığ, Kahramanmaraş, Karabük, Karaman, Kars, Kayseri, Kırşehir, Konya, Malatya, Mardin, Muğla, Niğde, Osmaniye, Samsun, Zonguldak (Horváth, 1901; Kiritshenko, 1918; Gadeau de Kerville, 1939; Çağatay, 1988; Péricart, 1999b; Lodos et al., 1989; Önder et al., 2006; Matocq et al., 2014; Yazıcı et al., 2015; Fent & Okyar, 2022; Çerçi & Koçak, 2023).

Note. This species is a new record for Amasya province. The specimens were collected under stone bottoms and leaf debris areas., Chorotype: West Palearctic.

Genus: Emblethis Fieber, 1860

#### Emblethis amplus Seidenstücker, 1987

Material examined. **Amasya**: Ziyaret, 19.IV.2021, 3.

Distribution in Türkiye. Malatya (Péricart, 1999b).

Distribution in Palaearctic Region. Asia: Iran, Irag, Türkiye (Asian part), Turkmenistan (Péricart, 2001).

Note. This species is a new record for Black Sea Region. The specimen was collected under *Verbascum* sp., Chorotype: Turanian.

#### Emblethis angustus Montandon, 1890

Material examined. **Amasya**: Ziyaret, 19.IV.2021,  $2 \circlearrowleft \circlearrowleft$ ,  $2 \hookrightarrow \circlearrowleft$ ; Yıldızköy, 25.VIII.2021,  $\hookrightarrow$ ; İlyasköy, 05.VII.2021,  $\hookrightarrow$ ; Suluova: Eraslan, 28.VI.2021,  $\circlearrowleft$ ; Taşova: Boraboy, 29.VIII.2021,  $\circlearrowleft$ ; Göynücek: Çamurlu, 03.IX.2021,  $\hookrightarrow$ .

Distribution in Türkiye. Adana, Adıyaman, Ankara, Antalya, Çanakkale, Gaziantep, Hatay, Isparta, İzmir, Kahramanmaraş, Karaman, Kayseri, Konya, Manisa, Mersin, Muğla, Niğde, Siirt (Seidenstücker, 1963; Lodos et al., 1978, 1989; Önder et al., 2006; Matocq & Özgen, 2010; Çerçi & Koçak, 2023).

Note. This species is a new record for Black Sea Region. The specimens were collected under *Verbascum* sp., Chorotype: Turano-Mediterranean.

#### Emblethis denticollis Horváth, 1878

Material examined. Göynücek: Şarklı, 03.IX.2021, ♂, ♀.

Distribution in Türkiye. Adana, Ankara, Antalya, Burdur, Çanakkale, Çorum, Edirne, Erzincan, Erzurum, Eskişehir, Gaziantep, Hatay, Kahramanmaraş, Karabük, Karaman, Kayseri, Kırşehir, Kilis, Konya, Mersin, Nevşehir, Niğde, Siirt, Yozgat (Puton & Noualhier, 1895; Horváth, 1905; Kiritshenko, 1924; Hoberlandt, 1956; Lodos et al., 1978, 1989; Önder et al., 1984, 2006; Özsaraç & Kıyak, 2001; Kıyak et al., 2004; Matocq & Özgen, 2010; Matocq et al., 2014; Yazıcı et al., 2015; Yence & Fent, 2023; Çerçi & Koçak, 2023).

Note. This species is a new record for Amasya province. The specimens were collected under *Verbascum* sp., Chorotype: Palearctic.

#### Emblethis griseus (Wolff, 1802)

Material examined. **Amasya:** Tatar, 19.V.2021, ♂; İlyasköy, 05.VII.2021, ♀.

Distribution in Türkiye. Adana, Afyonkarahisar, Ankara, Antalya, Bursa, Çankırı, Denizli, Edirne, Elazığ, Erzurum, Hatay, Isparta, İzmir, Kahramanmaraş, Karabük, Kars, Kastamonu, Kayseri, Kırşehir, Mersin, Niğde, Osmaniye, Yozgat, Zonguldak (Seidenstücker, 1963; Lodos et al., 1989; Önder et al., 2006; Fent & Japoshvili, 2012; Küçükbasmacı & Kıyak, 2015; Yence & Fent, 2023).

Note. This species is a new record for Central Black Sea Region. The specimens were collected under *Verbascum* sp. and *Prunus* sp., Chorotype: West Palearctic.

#### Emblethis latus Seidenstücker, 1966

Material examined. **Amasya**: Helvacı, 10.V.2020, 3 9 9; Sarıyar, 19.V.2021, 9; Merzifon: Alişar, 20.VIII.2021, 3, 9; Taşova: Boraboy, 29.VIII.2021, 3.

Distribution in Türkiye. Kars, Kayseri, Niğde (Seidenstücker, 1967, 1987; Yence & Fent, 2023).

Note. This species is a new record for Black Sea Region. The specimens were collected under *Verbascum* sp., Chorotype: Saharo-Turanian.

#### Emblethis nox Kiritshenko, 1912

Material examined. **Amasya**: Ziyaret, 19.IV.2021, ♂; Gümüşhacıköy: Maden, 07.IX.2021, ♂.

Distribution in Türkiye. Kayseri, Niğde (Seidenstücker, 1987; Yence & Fent, 2023).

Note. This species is a new record for Black Sea Region. The specimens were collected under *Verbascum* sp., Chorotype: Turanian.

#### Emblethis sabulosus Seidenstücker, 1963

Material examined. **Amasya**: Damudere, 08.VI.2021, ♀.

Distribution in Türkiye. İstanbul, Konya, Mersin (Önder et al., 2006).

Note. This species is a new record for Black Sea Region. The specimen was collected under *Verbascum* sp., Chorotype: Turanian.

#### Emblethis setifer Seidenstücker, 1966

Material examined. **Amasya**: Tatar, 19.V.2021,  $\circlearrowleft$ ; Kırklar, 02.IX.2021,  $\circlearrowleft$ ; Merzifon: Alişar, 20.VIII.2021,  $\circlearrowleft$ ; Taşova: Çalkaya, 27.V.2021,  $\circlearrowleft$ ,  $\diamondsuit$ .

Distribution in Türkiye. Adana, Ankara, Gaziantep, Karaman, Kayseri, Konya, Malatya, Mardin, Niğde (Seidenstücker, 1966; Péricart, 1999b; Lodos et al., 1989; Önder et al., 2006; Matocq et al., 2014; Yence & Fent, 2023; Çerçi & Koçak, 2023).

Note. This species is a new record for Black Sea Region. The specimens were collected under *Verbascum* sp. Chorotype: Turanian.

#### Emblethis verbasci (Fabricius, 1803)

Material examined. **Amasya**: Saraycık, 28.IV.2021, ♂.

Distribution in Türkiye. Adana, Afyonkarahisar, Aksaray Ankara, Artvin, Bolu, Bursa, Çankırı, Çorum, Diyarbakır, Düzce, Edirne, Hatay, Isparta, Kahramanmaraş, Karaman, Kayseri, Kırıkkale, Konya, Mardin, Nevşehir, Niğde (Horváth, 1883; Puton & Noulhier, 1895; Hoberlandt 1956; Önder & Adıgüzel, 1979; Lodos et al., 1989; Péricart, 1999b; Önder et al., 2006; Fent & Japoshvili, 2012; Matocq et al. 2014; Fent & Dursun, 2016; Çerçi & Koçak, 2023).

Note. This species is a new record for Amasya province. The specimen was collected under *Prunus* sp., Chorotype: West Palearctic.

#### Genus: Ischnopeza Fieber, 1860

#### Ischnopeza hirticornis (Herrich-Schaeffer, 1853)

Material examined. Taşova: Çaydibi, 27.V.2021, ♂.

Distribution in Türkiye. Adana, Afyonkarahisar, Ankara, Antalya, Balıkesir, Bilecik, Çanakkale, Çorum, Edirne, Erzincan, Eskişehir, Hatay, Iğdır, Isparta, İzmir, Karaman, Kahramanmaraş, Kastamonu, Kırklareli, Kırıkkale, Konya, Manisa, Mersin, Osmaniye, Uşak (Horváth, 1901; Kiritshenko, 1924; Hoberlandt, 1956; Seidenstücker, 1963; Lodos et al., 1978, 1989; Kıyak & Akar, 2010; Fent & Japoshvili, 2012; Dursun & Fent, 2016; Çerçi et al., 2018, 2022; Çerçi & Koçak, 2023).

Note. This species is a new record for Amasya province. The specimen was collected under hay plant debris. Chorotype: Turano-Mediterranean.

Genus: Neurocladus Fieber, 1860

Neurocladus brachiidens (Dufour, 1851)

Material examined. **Amasya**: Boğazköy, 17.IV.2020,  $\circlearrowleft$ , 4 $\circlearrowleft$  $\circlearrowleft$ ; Ziyaret, 19.IV.2021  $\circlearrowleft$ ,  $\circlearrowleft$ ; Gümüşhacıköy: Gümüş, 29.VIII.2020, 2 $\circlearrowleft$  $\circlearrowleft$ , 6 $\circlearrowleft$  $\circlearrowleft$ .

Distribution in Türkiye. Adana, Ankara, Diyarbakır, Elazığ, Hatay, Iğdır, Konya, İzmir, Malatya, Niğde (Puton & Noualhier, 1895; Hoberlandt, 1956; Péricart, 1999a; Lodos et al., 1989; Önder et al., 2006; Matocq et al., 2014; Yence & Fent, 2023; Çerçi et al., 2022; Çerçi & Koçak, 2023).

Note. This species is a new record for Black Sea Region. The specimens were collected under hay plant debris. Chorotype: Turano-Mediterranean.

Genus: Trapezonotus Fieber, 1860

Subgenus: Trapezonotus Fieber, 1860

Trapezonotus dispar Stål, 1872

Material examined. **Amasya**: Boğazköy, 18.V.2020, ♂.

Distribution in Türkiye. Bursa, Edirne, Hatay (Péricart, 1999b; Önder et al., 2006).

Note. This species is a new record for Black Sea Region. The specimen was collected from annual herbaceous plant. Chorotype: European-Mediterranean.

Tribe: Lethaeini Stål, 1872

Genus: Lethaeus Dallas, 1852

Lethaeus cribratissimus (Stål, 1859)

Material examined. **Amasya**: Harşena, 06.III.2020, Hacılar Meydanı, 233; 21.VIII.2021, 733, 9; İpekköy, 24.VIII.2021, 3, 399; Dadıköy, 24.VIII.2021, 3; Kayabaşı, 25.VIII.2021, 333, 299; Çamlıkent, 31.VIII.2021, 3, 299; Kırklar, 02.IX.2021, 333, 9; Kızılkışlacık, 10.IX.2021, 9, Kuzgeçe, 10.IX.2021, 9, 9, 9, 9; Gümüshacıköy: Maden, 07.IX.2021, 9, 9.

Distribution in Türkiye. Adana, Ankara, Antalya, Bursa, Çanakkale, Diyarbakır, Gaziantep, Hatay, Isparta, İstanbul, İzmir, Karaman, Kayseri, Kilis, Mersin, Muğla, Niğde, Sakarya, Sinop, Sivas, Tokat, Zonguldak (Horváth, 1883; 1901; 1905; Puton & Noualhier, 1895; Seabra, 1926; Lodos et al., 1978, 1989; Péricart, 1999a; Fent & Japoshvili, 2012; Matocq et al., 2014; Yence & Fent, 2023; Çerçi & Koçak, 2023).

Note. This species is a new record for Amasya province. The specimens were collected under *Verbascum* sp. And leaf debris of *Pinus* sp. and *Planatus* sp., Chorotype: East Mediterranean.

Tribe: Megalonotini J. A. Slater, 1957

Genus: Lamprodema Fieber, 1860

Lamprodema maura (Fabricius, 1803)

Distribution in Türkiye. Ankara, Ağrı, Antalya, Balıkesir, Bursa, Diyarbakır, Edirne, Erzincan Gaziantep, Hatay, İstanbul, İzmir, Kahramanmaraş, Karaman Kars, Kayseri, Kırşehir, Kocaeli, Konya, Sakarya, Siirt, Van (Puton & Noualhier, 1895; Horváth, 1901; Kiritshenko, 1918, 1924; Hoberlandt, 1956; Lodos et al. 1978, 1989; Önder & Adıgüzel, 1979; Önder et al., 1981, 1983, 1984, 2006; Çağatay, 1988; Péricart, 1999b; Matocq & Özgen, 2010; Çerçi et al. 2022; Çerçi & Koçak, 2023).

Note. This species is a new record for Black Sea Region. The specimens were collected under *Verbascum* sp. and leaf debris of *Pinus* sp. and *Planatus* sp., Chorotype: Turano-Mediterranean.

Genus: Lasiocoris Fieber, 1860

#### Lasiocoris anomalus (Kolenati, 1845)

Material examined. Göynücek: Çamurlu, 03.IX.2021, ♂, 2♀♀.

Distribution in Türkiye. Adana, Ağrı, Ankara, Bursa, Diyarbakır, Elazığ, Erzurum, Gaziantep, Hakkarı, Igdır, Karaman, Konya, Malatya, Niğde, Tokat, Van (Kiritshenko, 1918; Hoberlandt, 1956; Kıyak, 1990; Péricart, 1999b; Önder et al., 2006; Yazıcı et al., 2015; Özgen & Dioli, 2019; Çerçi & Koçak, 2023).

Note. This species is a new record for Amasya province. The specimens were collected under *Verbascum* sp., Chorotype: Turano-Mediterranean.

#### Lasiocoris crassicornis (Lucas, 1849)

Material examined. **Amasya:** Saraycık, 08.IV.2021, ♂, ♀.

Distribution in Türkiye. Adana, Adıyaman, Ankara, Antalya, Bursa, Edirne, Elazığ, Hatay, İstanbul, İzmir, Karaman, Kahramanmaraş, Kayseri, Konya, Kütahya, Malatya, Van (Horváth, 1883; Puton & Noualhier, 1895; Linnavuori, 1953; Lodos et al., 1989; Kıyak, 1990; Péricart, 1999b; Önder et al., 2006; Çerçi & Özgen, 2021; Çerçi & Koçak, 2023).

Note. This species is a new record for Black Sea Region. The specimens were collected under *Verbascum* sp., Chorotype: Mediterranean.

Genus: Megalonotus Fieber, 1860

#### Megalonotus sabulicola (Thomson, 1870)

Material examined. Amasya: Çamlıkent, 31.VIII.2021, ♀.

Distribution in Türkiye. Adana, Antalya, Gaziantep, Hatay, Kastamonu, Mersin (Önder et al., 2006; Küçükbasmacı & Kıyak, 2015).

Note. This species is a new record for Central Black Sea Region. The specimen was collected under *Verbascum* sp., Chorotype: Palaearctic.

## Megalonotus emarginatus (Rey, 1888)

Material examined. Amasya: 22.IV.2021, ♂; Kayabaşı, 25.VIII.2021, ♂, 2♀♀; Kızılkışlacık, 10.IX.2021, ♂.

Distribution in Türkiye. Adana, Antalya, Mersin (Önder et al., 2006).

Note. This species is a new record for Black Sea Region. The specimens were collected under *Verbascum* sp. and leaf debris of *Pinus* sp. and *Planatus* sp., Chorotype: Europeo-Mediterranean.

Tribe: Myodochini Blanchard, 1845

Genus: Paromius Fieber, 1860

#### Paromius gracilis (Rambur, 1839)

Material examined. Taşova: Çaydibi, 27.V.2021, 2♂♂.

Distribution in Türkiye. Adana, Antalya, Aydın, Çanakkale, Denizli, Hatay, İzmir, Kahramanmaraş, Mersin, Tekirdağ (Önder et al., 2006).

Note. This species is a new record for Black Sea Region. The specimens were collected from annual herbaceous plants. Chorotype: Palaearctic.

Tribe: Plinthisini J. A. Slater & Sweet, 1961

Genus: Plinthisus Stephens, 1829

Subgenus: Plinthisus Stephens, 1829

Plinthisus longicollis Fieber, 1861

Distribution in Türkiye. Adana, Afyonkarahisar, Ankara, Aydın, Bursa, Diyarbakır, Elazığ, Hatay, Isparta, Konya, Sinop (Horváth, 1901; Hoberlandt, 1956; Önder et al., 2006; Fent & Japoshvili, 2012; Matocq et al., 2014; Yazıcı et al., 2015; Fent & Dursun, 2016; Çerçi et al., 2018; Yence & Fent, 2023).

Note. This species is a new record for Amasya province. The specimens were collected under *Verbascum* sp. and leaf debris of *Pinus* sp., Chorotype: Turano-European-Mediterranean.

Tribe: Rhyparochromini Amyot & Serville, 1843

Genus: Aellopus Wolff, 1811

Aellopus atratus (Goeze, 1778)

Material examined. Göynücek, 03.IX.2021, 4∂∂, ♀; Gümüşhacıköy: Maden, 07.IX.2021, ∂, 3♀♀.

Distribution in Türkiye. Adana, Afyonkarahisar, Ankara, Aydın, Bursa, Diyarbakır, Elazığ, Hatay, Isparta, Karaman, Kayseri, Konya, Niğde, Sinop, Tokat, (Puton & Noualhier, 1895; Horváth, 1897, 1901; Hoberlandt, 1956; Péricart, 1999b; Önder et al., 2006; Fent & Japoshvili, 2012; Matocq et al., 2014; Yazıcı et al., 2015; Dursun & Fent, 2016; Özgen et al., 2021; Çerçi & Koçak, 2023; Yence & Fent, 2023).

Note. This species is a new record for Amasya province. The specimens were collected under *Verbascum* sp., *Acanholimon* sp., *Astragalus* sp., Chorotype: West Palaearctic.

Genus: Beosus Amyot & Serville, 1843

Beosus maritimus (Scopoli, 1763)

Material examined. **Amasya:** Sarıyar, 19.V.2021,  $\circlearrowleft$ ; Damudere, 08.VI.2020;  $\circlearrowleft$ ,  $\circlearrowleft$ ; Hamamözü: 07.IX.2021,  $\circlearrowleft$ ; Taşova: Boraboy, 29.VIII.2021, 2 $\circlearrowleft$ ,  $\circlearrowleft$ .

Distribution in Türkiye. Adana, Adıyaman, Ankara, Antalya, Bolu, Bursa, Çankırı, Diyarbakır, Düzce, Edirne, Elazığ, Erzurum, Gaziantep, Giresun, Hatay, Isparta, Kahramanmaraş, Karabük, Karaman, Kastamonu, Kayseri, Manisa, Mersin, Osmaniye, Van, Zonguldak (Horváth, 1883; Puton, 1892; Hoberlandt, 1956; Önder & Adıgüzel, 1979; Lodos et al., 1989; Önder et al., 2006; Fent & Japoshvili, 2012; Küçükbasmacı & Kıyak, 2015; Yazıcı et al., 2015; Kaçar & Dursun, 2022; Çerçi & Koçak, 2023; Yence & Fent, 2023).

Note. This species is a new record for Central Black Sea Region. The specimens were collected under plant debris and stones. Chorotype: West Palearctic.

#### Beosus quadripunctatus (Müller, 1766)

Material examined. **Amasya**: Damudere, 08.VI.2020,  $\circlearrowleft$ ; Fındıklı, 15.VIII.2020,  $2\circlearrowleft\circlearrowleft$ ,  $4\Lsh\supsetneq$ ; Dadıköy, 24.VIII.2021,  $\supsetneq$ ; Karasenir, 09.IX.2021,  $2\circlearrowleft\circlearrowleft$ ; Merzifon: Yalnızköy, 20.VIII.2021,  $\supsetneq$ ; Çobanören, 14.IX.2021,  $\circlearrowleft$ ; Göynücek: 03.IX.2021,  $\circlearrowleft$ ; Taşova: Boraboy, 29.VIII.2021,  $\supsetneq$ .

Distribution in Türkiye. Adana, Afyonkarahisar, Ağrı, Ankara, Antalya, Artvin, Aydın, Balıkesir, Bartın, Bilecik, Bursa, Çanakkale, Çankırı, Çorum, Denizli, Diyarbakır, Düzce, Edirne, Elazığ, Erzurum, Eskişehir, Hatay, Iğdır, İstanbul, İzmir, Kahramanmaraş, Karabük, Karaman, Kars, Kastamonu, Kayseri, Kırklareli, Konya, Kütahya, Manisa, Mardin, Niğde, Osmaniye, Sakarya, Siirt, Sinop, Tekirdağ, Uşak Zonguldak (Horváth, 1883, 1918; Puton & Noulhier, 1895; Kiritshenko, 1918, 1924; Seabra, 1926; Hoberlandt, 1956; Linnavuori, 1965; Lodos et al., 1978, 1989; Önder & Adıgüzel, 1979; Önder et al., 1984; Matocq et al., 2014; Yazıcı et al., 2015; Fent & Dursun, 2016; Çerçi & Koçak, 2023; Yence & Fent, 2023).

Note. This species is a new record for Amasya province. The specimens were collected under plant debris and stones. Chorotype: West Palearctic.

Genus: Graptopeltus Stål, 1872

#### Graptopeltus lynceus (Fabricius, 1775)

Material examined. Taşova: Çalkaya, 27.V.2021, 2♂♂, 3♀♀.

Distribution in Türkiye. Adana, Ankara, Bursa, Isparta, İstanbul, Kahramanmaraş, Muş, Niğde (Önder et al., 2006; Fent & Japhosvili, 2012; Fent & Dursun, 2016).

Note. This species is a new record for Central Black Sea Region. The specimens were collected under plant debris and stones. Chorotype: Turano-Europeo-Mediterranean.

Genus: Peritrechus Fieber, 1860

#### Peritrechus geniculatus (Hahn, 1832)

Material examined. **Amasya**: Dadıköy, 21.IV.2021, ♀.

Distribution in Türkiye. Ankara, Kars, Kastamonu, Konya (Önder et al., 2006; Fent & Dursun 2016).

Note. This species is a new record for Central Black Sea Region. The specimen was collected under debris of *Pinus* sp. Chorotype: West Palearctic.

Genus: Raglius Stål, 1872

#### Raglius alboacuminatus alboacuminatus (Goeze, 1778)

Material examined. **Amasya**: Kayabaşı, 25.VIII.2021,  $\bigcirc$ ; Gözlek, 03.IX.2021,  $2\bigcirc\bigcirc$ ; Taşova: Gürsu, 27.V.2021,  $\bigcirc$ ; Merzifon: Karamustafa Paşa, 14.IX.2021,  $\bigcirc$ ; Göynücek: Şarklı, 03.IX.2021,  $\bigcirc$ ; Gümüşhacıköy: Maden, 07.IX.2021;  $2\bigcirc\bigcirc$ ,  $\bigcirc$ .

Distribution in Türkiye. Ankara, Balıkesir, Bolu, Düzce, Edirne, Erzincan, Eskişehir, Hatay, İstanbul, Kahramanmaraş, Karabük, Kastamonu, Kars, Kayseri (Lodos et al., 1989; Önder et al., 2006; Fent & Dursun, 2016; Yence & Fent, 2023).

Note. This species is a new record for Central Black Sea Region. The specimens were collected under *Acantholimon* sp., *Astragalus* sp. *Junglans* sp. and *Verbascum* sp., Chorotype: West Palearctic.

#### Raglius confusus (Reuter, 1886)

Material examined. Gümüşhacıköy: Maden, 07.IX.2021, ♀.

Distribution in Türkiye. Ankara, Bingöl, Bursa, Hatay, İstanbul, Kars, Konya, Niğde (Puton & Noualhier, 1895; Kritshenko, 1918; Péricart, 1999b; Önder et al., 2006).

Note. This species is a new record for Black Sea Region. The specimen was collected under *Acantholimon* sp., Chorotype: Turano-European-Mediterranean.

#### Raglius zarudnyi (Jakovlev, 1905)

Material examined. **Amasya**: Ziyaret, 19.IV.2021,  $\circlearrowleft$ , 5 $\circlearrowleft$  $\circlearrowleft$ ; Sarıyar, 19.V.2021,  $\circlearrowleft$ ; Tatar, 19.V.2021,  $\circlearrowleft$ ; Yıldızköy, 25.VIII.2021, 3 $\circlearrowleft$  $\circlearrowleft$ ; Kayabaşı, 25.VIII.2021,  $\circlearrowleft$ ; Taşova: Alparslan, 27.V.2021,  $\circlearrowleft$ .

Distribution in Türkiye. Ankara, Bursa, Niğde, Konya, Eskişehir, Tokat (Seidenstücker, 1957; Tuatay et al., 1972; Péricart, 1999b).

Note. This species is a new record for Amasya province. The specimens were collected under *Acantholimon* sp., *Astragalus* sp. and *Verbascum* sp., Chorotype: Turanian.

#### Genus: Rhyparochromus Hahn, 1826

#### Rhyparochromus phoeniceus (Rossi, 1794)

Material examined. Amasya: Yassıçal, 28.IV.2021, 3♀♀; Saraycık, 28.IV.2021, ♀.

Distribution in Türkiye. Adana, Ankara, Bolu, Çankırı, Çorum, Diyarbakır, Edirne, Erzurum, Gaziantep, Hatay, Isparta, Kahramanmaraş, Karabük, Kayseri, Kırıkkale, Kütahya, Manisa, Mersin, Muş, Niğde, Zonguldak (Lodos et al., 1978, 1989; Hoberlandt, 1956; Kıyak, 1990; Önder et al., 2006; Kıyak & Akar, 2010; Fent & Dursun, 2016, Yence & Fent, 2023).

Note. This species is a new record for Amasya province. The specimens were collected under *Acantholimon* sp., *Astragalus* sp., Chorotype: European-Mediterranean.

#### Rhyparochromus sanguineus (Douglas & Scott, 1868)

Material examined. Gümüşhacıköy: Maden, 07.IX.2021, 5♂♂, 6♀♀.

Distribution in Türkiye. Adana, Ankara, Bingöl, Diyarbakır, Erzincan, Erzurum, Elazığ, Eskişehir, Gaziantep, Hatay, Iğdır, Karaman, Kastamonu, Kayseri, Konya, Mardin, Niğde, Tokat, Tunceli (Horváth 1901; Gadeau de Kerville, 1939; Péricart, 1999b; Matocq et al., 2014; Yazıcı et al., 2015; Dursun & Fent 2016; Çerçi et al. 2022; Çerçi & Koçak, 2023).

Note. This species is a new record for Amasya province. The specimens were collected under *Acantholimon* sp., *Astragalus* sp. Chorotype: Turano-European.

#### Rhyparochromus vulgaris (Schilling, 1829)

Material examined. **Amasya**: 21.VI.2021, 3  $\hookrightarrow$ ; Merzifon: Karamustafa Paşa, 14.IX.2021, 30  $\circlearrowleft$  $\circlearrowleft$ , 34  $\hookrightarrow$  $\hookrightarrow$ ; Gümüşhacıköy: Gümüş, 07.IX.2021,  $\circlearrowleft$ ; Taşova: Boraboy, 29.VIII.2021, 4  $\circlearrowleft$  $\circlearrowleft$ , 10  $\hookrightarrow$  $\hookrightarrow$ .

Distribution in Türkiye. Adana, Afyonkarahisar, Ankara, Aydın, Balıkesir, Bursa, Çanakkale, Denizli, Düzce, Edirne, Erzincan, Erzurum, Hatay, İzmir, Isparta, İzmir, Karaman, Kars, Kütahya, Manisa, Mersin, Muğla, Osmaniye, Tunceli, Uşak, Van (Horváth, 1883; Puton, 1892; Hoberlandt, 1956; Önder et al., 1983; 2006; Lodos et al., 1989; Fent & Japoshvili, 2012; Yazıcı et al., 2015; Dursun & Fent, 2016; Çerçi & Koçak, 2023).

Note. This species is a new record for Central Black Sea Region. The specimens were collected under the bark of *Juglans* sp., Chorotype: Turano-European.

#### Genus: Xanthochilus Stål, 1872

#### Xanthochilus quadratus (Fabricius, 1798)

Distribution in Türkiye. Ankara, Balıkesir, Bursa, Elazığ, Kastamonu, Konya, İstanbul, İzmir, Samsun (Çağatay, 1988; Péricart, 1999b; Önder et al., 2006; Fent & Dursun, 2016; Çerçi & Özgen, 2021).

Note. This species is a new record for Amasya province. The specimens were collected under *Astragalus* sp., Acantholimon sp., *Verbascum* sp. and debris of *Pinus* sp., Chorotype: West Palearctic.

#### Xanthochilus saturnius (Rossi, 1790)

Material examined. **Amasya:** Ziyaret, 19.IV.2021, 3♂♂; Kızılkışlacık, 10.IX.2021, 2♂♂; Taşova: Çaydibi, 27.V.2021, ♀; Gümüşhacıköy: Maden, 07.IX.2021, 3♂♂; Göynücek: Şarklı, 03.IX.2021, ♂.

Distribution in Türkiye. Adana, Ankara, Antalya, Bursa, Diyarbakır, Elazığ, Erzurum, Gaziantep, Hatay, İzmir, Karaman, Kahramanmaraş, Mersin, Osmaniye, Siirt (Gadeau de Kerville, 1939; Wagner, 1959; Kıyak, 1990; Péricart, 1999b; Lodos et al., 1989; Kıyak & Akar, 2010; Matocq & Özgen, 2010; Yazıcı et al., 2015; Cerci & Kocak, 2023).

Note. This species is a new record for Black Sea Region. The specimens were collected under *Astragalus* sp., *Acantholimon* sp., *Verbascum* sp. and leaf debris of *Pinus* sp., Chorotype: Turano-Europeo-Mediterranean.

#### **Discussion**

In this study, as a result of the identification of the 367 adult specimens collected from 51 different localities in around Amasya province between 2020 and 2021 revealed 6 species of the 4 genera belonging to tribus Drymini Stål, 1872, 13 species of the 5 genera belonging to tribus Gonianotini Stål, 1872, one species belonging to tribus Lethaenini Stål, 1872, 5 species of the 3 genera belonging to tribus Megalonotini J. A. Slater, 1957, one species belonging to tribus Myodochini Blanchard, 1845, one species belonging to tribus Plinthisini J. A. Slater & Sweet, 1961 and 13 species of the 7 genera belonging to tribus Rhyparochromini Amyot & Serville, 1843 of the Rhyparochromidae family were reported. All species are new records for the Rhyparocromidae fauna of Amasya province such as the species *Drymus ryeii* Douglas & Scott, 1865 for the Heteroptera fauna of Türkiye, 17 species for Black Sea Region and 7 species for Central Black Sea Region.

In addition, *Drymus ryeii*, is a new record for the Turkish Rhyparochromidae fauna. The species is recorded in the neighboring countries Bulgaria, Greece and Iran and it is, widely distributed in Europe. The presence of this species in Georgia, northwest neighbor of Türkiye, has not yet been clarified (Aukema, 2018). As a result of the identification of a male sample obtained from a single locality, it was determined that this species was also distributed in our country. In this study, the morphological features of the sample of the determined species were compared with the existing literature and the features that enable the diagnosis of the species were mentioned. In addition, Türkiye and Palearctic region distributions and chorotypes of the identified species were determined.

Among the species identified in the study area, *Ischnocoris hemipterus*, *Emblethis amplus*, *Emblethis latus*, *Emblethis nox*, *Emblethis sabulosus*, *Trapezonotus dispar*, *Lasiocoris crassicornis*, *Megalonotus emarginatus* and *Peritrechus geniculatus* are extremely rarely distributed species both in the research area and in Türkiye. *Emblethis amplus* was reported in Anatolia from Malatya by Péricart (1999b), but has not been found not in any other places so far. In the study, it was given a second locality from Amasya. The findings for these species in this study constitute the northernmost limit of the dispersion area and expand the distribution limits of the existing species in Türkiye.

The plants, as *Verbascum* sp., *Acantholimon* sp. and *Astragalus* sp. are very important for the family of Rhyparochromidae. In this study, the specimens *Aphanus rolandri*, *Drymus ryeii*, *Lethaeus cribratissimus* and the species belonging to the genera *Emblethis*, *Lasiocoris*, *Megalonotus*, *Rhyparochromus*, *Scolopostethus*, *Xanthochilus* has been found generally under *Verbascum* sp., *Acantholimon* sp., *Astragalus* sp., leaf debris of *Pinus* sp. and stones. According to our field studies, the population densities of Rhyparochromidae species are generally low. Therefore, it can be said that these species cannot seriously damage agricultural areas in Amasya.

According to the chorotypes analysis, Rhyparochromidae species from Amasya are divided into 13 categories: as West Palearctic (22,5%-9 spp.), Turano-Mediterranean (15%-6 spp.), Turanien (12,5%-5 spp.), Europeo-Mediterranean (10%-4 spp.), Turano-Europeo-Mediterranean (10%-4 spp.), Palearctic (7,5%-3 spp.), European (5%-2 spp.), Turano-European (5%-2 spp.), Sibero-European (2,5%-1 spp.), Asiatic-European (2,5%-1 spp.), Mediterranean (2,5%-1 spp.), East Mediterranean (2,5%-1 spp.), Saharo-Turanian (2,5%-1 spp.). According to this analysis, West Palearctic (22,5%) are a major group of Amasya, followed by Turano-Mediterranean. Sibero-European, Asiatic-European, Mediterranean, East Mediterranean, Saharo-Turanian represented by one species. These data show that richness of Faunae from Amasya. The fact that so many different chorotypes distributed in Amasya is related to the rich microclimate areas and flora of Amasya. Therefore, their richness of Rhyparochromidae in Amasya is not unexpected. This result can help future studies on ecology, biodiversity and plant protection about Heteroptera in Amasya and Türkiye.

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# Original article (Orijinal araştırma)

# An analysis of Thysanoptera associated with the flowers of some stone fruits in Mersin Province (Türkiye): composition, distribution, population dynamics and damage status

Mersin İlindeki (Türkiye) bazı sert çekirdekli meyvelerin çiçekleriyle ilişkili Thysanoptera türlerinin analizi: kompozisyon, dağılım, popülasyon dinamikleri ve zarar durumu

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Serkan PEHLİVAN<sup>2</sup>

#### **Abstract**

The distribution, abundance and damage status of Thysanoptera species were investigated in two different regions of Mersin Province, Türkiye between 2020 and 2021. A total of 18 Thysanoptera species were determined, with *Thrips major* Uzel, 1895 and *Frankliniella occidentalis* (Pergande, 1895) (Thysanoptera: Thripidae) being the most common. While nectarine had the highest diversity of habitats for thrips, plum displayed greater richness in terms of species composition. Adult thrips primarily inhabited flowers while *T. major* larvae were present during petal fall periods. No larvae of *F. occidentalis* or *Thrips tabaci* Lindeman,1889 (Thysanoptera: Thripidae) were found on the plant parts sampled throughout the samplings. Although no visible signs of damage by adult thrips were observed on the flowers, typical damage in the form of silvery scars appeared on the young fruits of nectarines and plums. The average rate of scarred fruit of nectarines or plums varied between 2-7%. Finally, *T. major* was the main pest thrips species responsible for damaging fruits of the plums and nectarines in Mersin.

Keywords: Damage, diversity, Mersin, stone fruits, Thrips major

# Öz

Mersin İlinin (Türkiye) iki farklı bölgesinde Thysanoptera türlerinin yayılışı, yoğunluğu ve zarar durumları 2020 ve 2021 yıllarında araştırılmıştır. Thysanoptera türlerinden *Thrips major* Uzel, 1895 ve *Frankliniella occidentalis* (Pergande, 1895) (Thysanoptera: Thripidae) en yaygın olmak üzere toplam 18 tür tespit edilmiştir. Thripsler için en fazla habitat çeşitliliği nektarinde görülürken, tür kompozisyonu açısından erik bahçesi daha fazla zenginlik göstermiştir. Ergin thripsler öncelikle çiçeklerde saptanmıştır. *Thips major* larvaları ise çiçek taç yapraklarının dökülme döneminde ortaya çıkmıştır. Örneklemeler boyunca örneklenen bitki kısımlarında *F. occidentalis* veya *Thrips tabaci* Lindeman, 1889 (Thysanoptera: Thripidae) türlerinin larvalarına rastlanılmamıştır. Çiçeklerde ergin thripslerin gözle görülür zararları görülmemesine karşın, nektarin ve eriklerin özellikle genç meyvelerde gümüşi lekeler şeklinde tipik zarar belirtileri (yaralar) ortaya çıkmıştır. Nektarin veya erik meyvelerinde ortalama leke oranları %2-7 arasında değişmiştir. Bu çalışma sonucunda, *T. major*'un Mersin İlinde erik ve nektarin meyvelerinde zarar oluşumundan sorumlu ana thrips türü olduğu saptanmıştır.

Anahtar sözcükler: Zarar, çeşitlilik, Mersin, sert çekirdekli meyveler, Thrips major

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#### Introduction

The cultivation of temperate climate fruits such as apples, plums, apricots, cherries, and peaches holds great significance in Türkiye as Anatolia is the origin of most fruit species and varieties (Köksal & Güneş, 2006). Depending on the cooling requirements of the temperate climate fruits, it is feasible to grow them in regions with subtropical climates (Küden et al., 2006). The Mediterranean, Aegean and Marmara coastal regions are listed in terms of ripening time of stone fruit species growing in significant proportions in our country (Küden et al., 2006). The Eastern Mediterranean Region's unique ecological and geographical characteristics make it possible to cultivate early fruit varieties and offer fresh fruits to the market for five months a year. The most commonly cultivated stone fruit species in the region are the apricot, *Prunus armeniaca* L.; nectarine, *Prunus persica* var. *nectarina* (L.) Batsch and peach, *Prunus persica* L. (Rosales: Rosaceae). In 2020, Türkiye produced approximately 3 million tons of stone fruit, with peach (892 048 tons) and cherry (724 944 tons) being the most economically important species (TUIK, 2020).

The increase in temperate climate fruit production in the region has also brought attention to the pest and mite fauna, including thrips species, which have been well studied in different fruit species and production areas across Türkiye (Aykaç, 1983; Erkılıç, 1995; Ergüden et al., 1999; Öztürk, 2003). For instance, numerous studies have identified thrips (Thysanoptera) species on fruit trees in the Mediterranean and Central Anatolian Regions, as well as the thrips fauna in cherry production areas in the Aegean Region of Türkiye (Tunç, 1989, 1996; Şahin & Tezcan, 2014; Uzun et al., 2015; Maya & Tezcan, 2016). In recent decades, thrips have become increasingly significant due to the rise in temperate climate fruit production in the Cukurova Region of Türkiye. Thysanoptera fauna has been detected in stone and pome fruit species in Adana and its surrounding regions (Hazir & Ulusoy, 2007; Atakan, 2008a). The western flower thrips, Frankliniella occidentalis (Pergande, 1895) and rose thrips, Thrips major Uzel, 1895 (Thysanoptera: Thripidae) were the main prevalent thrips species on fruit trees in the region. Although thrips fauna has been studied in the region, to date there has been little knowledge on population changes, economic importance and damage by these species. Moreover, the introduction of invasive thrips species, including Thrips hawaiiensis (Morgan, 1913) and Scirtothrips dorsalis Hood, 1913 (Thysanoptera: Thripidae), has the potential to change the composition of the Thysanoptera fauna in the region (Atakan et al., 2015; Atakan & Pehlivan, 2021). In fact, F. occidentalis entered our country in 1993 and has become the main thrips species in a short time, causing significant damage to various agricultural products, particularly in greenhouse vegetable production in the Mediterranean Region of Türkiye, T. hawaiiensis, which was the first time reported for the lemons grown in Mersin Province, Türkiye in 2015, rapidly spread to citrus production areas with leading economic losses especially in the production of citrus fruits (Atakan et al., 2021). In addition, T. hawaiiensis has been found to cause silvering damage on mature fruits of the nectarine (Atakan et al., 2015). For this reason, investigating the species of Thysanoptera in different crops including stone fruits would develop better understanding on their economic importance. Such studies are critical in the planning of effective pest control strategies. The primary objective of this paper is to assess the species of the Thysanoptera and to investigate the relationship between their population developments and damages that occurred to stone fruits.

#### **Materials and Methods**

#### Sampling areas

This study was conducted in Mersin Province (Türkiye) in 2020 and 2021. For this aim, plum, peach, nectarine and apricot trees were sampled during flowering and young fruiting periods at non-periodic intervals in 2020. Throughout 2021, thrips species were collected weekly in a plum and nectarine orchard located in the Toroslar district (Çopurlu region) and Tarsus district (Yenice region) of Mersin province. The distance between sampling sites was approximately 30 km. The Çopurlu region is characterized by the cultivation of stone fruit trees such as plum and nectarine, as well as olive and pomegranate trees. On the

other hand, the Yenice region has mostly citrus orchards around the plum and nectarine orchards, and the cultivation of faba bean, a winter vegetable, is also common. In plum and nectarine orchards, trees are generally planted with a spacing of 5 m between rows and 3 m above rows. The sampled plum, nectarine, peach and apricot varieties were 'Black diamond', Garofa, 'Transvalia' and Ninfa, respectively. Weeds were removed between rows by hoeing and plowing in all of the sampled orchards. In apricot and peach orchards, no pesticides were applied to control harmful insects, including thrips. In the plum and nectarine orchards where the thrips population was monitored, the producers applied 25% spinetoram active substance at a dose of 30 gr/100 liters of water to control thrips on March 19 and April 2. In addition, thiram 80 WP was used with 300 g/100 lt water dose, axial fan and trailed type turbo atomizer (pump capacity: 115 lt/min, air flow: 68.000 m3/h and 1000 lt capacity) to control the diseases of sampled fruit trees, monilial, *Sclerotinia laxa* Aderh. & Ruhl (Helotiales: Sclerotiniaceae and pocket plum, *Taphrina pruni* Tul. (Taphrinales: Taphrinaceae) during the flowering period.

#### Thrips samplings

Thrips species are well-known to injury especially young fruits. For this purpose, survey studies were conducted mainly from the flowering period until the fruits reached a size of 4-5 cm in diameter depending on the stone fruit variety. Sampling was performed at weekly intervals between 09:30 and 13:00. In 2020, stone fruits were sampled between March 24 and April 3 in the Karacailyas, Akdeniz, Fındıkpınarı, Cemilli, Akarca locations of Mersin Province, only to detect thrips species. In 2021, seven samplings were made between March 5 and April 16 in order to monitor the population in a nectarine and a plum orchard in the location Çopurlu in Toroslar district and in the location Yenice in Tarsus district in Mersin Province, Türkiye.

Thrips samplings were started during the flowering period in each orchard. For this aim, depending on the size of the orchards, at least six stone fruit trees (plums, nectarines, peaches, and apricots) were selected randomly in each orchard. A total of four flowering shoots (20-25 cm long) or fruiting branches were taken from the south and southeast direction of the tree and shaken into the container for 5-10 s. Thrips individuals were put into plastic Eppendorf tubes (2 cc) with 60% ethyl alcohol by collecting them with a suction tube or fine brush. The samples were examined under a stereoscopic microscope and placed in the AGA (10 parts ethyl alcohol, one-part glacial acetic acid and one-part glycerin) solution for identification. Thrips samples were preserved in this solution for one or two days were then taken into the plastic tubes with 60% ethyl alcohol and labeled (Atakan, 1998).

#### Thrips identification

In order to ease the preparation of the microscope slides, thrips specimens, which were stored in AGA liquid for 2 days after collection and then taken in alcohol (60% ethyl alcohol), were kept in 10% sodium hydroxide solution (NAOH) on the hot plate at 47°C for approximately 1 hour until a slight color change occurred in the individuals. Then, this liquid was allowed to enter the thrips body, and the body contents were cleaned. After the samples were kept in 96% ethanol for 5 minutes, they were taken to Hoyer medium and microscope slides were made. Thysanoptera species were identified by the first author by use of the key identifications published by previous studies (Priesner, 1951; Yakhontov, 1964; Nakahara, 1994; zur Strassen, 2003; Masumoto & Okajima, 2006; Vierbergen et al., 2010).

#### **Determination of thrips damage**

This study has been carried out to determine thrips damage on young fruits. Upon inspecting, 100 fruits were randomly collected from each orchard on 11 June 2021 and checked. Silver-bronze injured fruits were accepted as damaged by thrips (Figure 1) and fruits that did not show such symptoms were considered healthy. The scale of zero is rated as no damage, (1): mild scar, (2): deformation and mild scarring, (3): moderate scarring, (4): deformation and moderate scarring, and (5): severe scarring and deformation (Felland et al., 1995).



Figure 1. Heavily damaged nectarine (a) and moderate damaged plum (b) fruits due to thrips attacks.

#### Plant phenology

The flowering status of the trees sampled in the orchards were noted by observing the fruiting periods. Thus, the most intense period of the pest species according to the plant phenology was observed.

#### Species diversity indices

The parameters used and their calculation methods are given below:

Shannon-Wiener and Simpson diversity indices were used to determine species diversity.

- Shannon-Wiener diversity index (H')

$$H' = -\sum pi \ln (pi)$$

where, pi: is proportion of individuals of i-th species in a whole community;

In: is the natural logarithm (Magurran, 1988, 2004).

- Simpson diversity index (1/D)

$$1/D = 1-\sum ni (ni-1) / N (N-1)$$

where, i: species number;

ni: number of individuals of each species;

N: the total number of individuals of a species in an area (Magurran, 1988, 2004).

- Margalef species richness index (Dmg)

$$D = (S-1)/ln (N)$$

where, S: is the number of species in a sample;

N: number of organisms in the sample (Magurran, 1988, 2004).

Simpson dominancy index was used to determine dominance

- Simpson's dominance index (Sd)

$$Sd = \sum ni (ni-1)/N (N-1)$$

where, i: species number;

ni: number of individuals of each species;

N: the total number of individuals of a species in an area (Magurran, 1988, 2004).

Shannon Evenness and Simpson Evenness index were used to determine population density relationships of the species.

- Shannon Evenness (Esh)

Esh= H' / In (N)

Where, H': Shannon-Wiener diversity index;

In: is the natural logarithm;

N: the total number of individuals of a species in an area (Magurran, 1988, 2004).

- Simpson Evenness (Esm)

Esm = (1/D) / S

where, 1/D: Simpson diversity index;

S: is the number of species in a sample (Magurran, 1988, 2004).

#### Statistical analysis

The experiments were conducted in a completely randomized design. Stone fruit variety (plum and nectarine), sampling region and thrips species and various interactions between them were investigated by two-way analysis of variance. Thrips density was considered as dependent variable, fruit variety, sampling region and thrips type as fixed factors. Kolmogorov-Smirnov test and Levene's test were run to specify normality and homogeneity of the variance. Seasonal abundance, plant phenology and mean densities of three thrips species according to stone fruit species were evaluated by one-way analysis of variance (ANOVA), and significant differences were compared with Tukey HSD test at p<0.05 significance level. Since very few thrips were collected from both peach and apricot orchards in both locations in 2021, population densities of thrips species were not evaluated according to plant phenology and sampling dates. In all evaluations, larvae and adults of thrips species collected on flowers and flower petals, as well as on young fruits (0.5-1 cm in diameter) were evaluated together. All analyses were done by use of the SPSS Software (version 22).

#### Results

#### Thysanoptera composition

When the survey studies for thrips on stone fruit trees were evaluated together, 234 adults were recorded in 2020 and 728 adults in 2021 (Table 1). In general, greater numbers of adult thrips were collected in 2021 partially explained by the fact that higher number of samples were collected in this year. *Frankliniella occidentalis* and *T. major* were collected in the highest numbers in both years, and T. major was the main thrips species. This species was followed by *F. occidentalis* with a rate of 24.04% in total individuals. *Thrips tabaci* adults were recorded as the 3rd most common thrips species with 15.52%. It has been observed that *T. hawaiiensis* has a very low rate (2.61%) in stone fruit trees compared to the other three thrips species.

The total number of thrips species according to stone fruits are shown in Table 2. Plum trees had the highest number of adult thrips individuals, with 434 individuals, while apricots had the lowest number. Nectarine trees also had a high number of adult individuals, with 212 individuals. Most of *T. major* (245 adult individuals) and *F. occidentalis* (84 adult individuals) were sampled from plum trees. Adults of *T. tabaci*, which was determined as the third common species, were mostly collected from plum and nectarine. A few numbers of *T. major* and *F. occidentalis* individuals were found on peach trees. Depending on the total number of adult thrips, the highest number of species was recorded in plum (18 species), followed by nectarine with 12 species. The number of species was low and similar in peach and apricot (Table 2).

Table 1. The number of Thysanoptera species detected on some stone fruit species in Mersin, Türkiye, and their rates in total individuals (%)

			2020	2021		Total (2020+2021)	
Family	Thrips species	No of thrips	Percentage (%)	No of thrips	Percentage (%)	No of thrips	Percentage (%)
	Aeolothrips collaris	1	0.42	0	0	1	0.14
	Aeolothrips ericae	1	0.42	0	0	1	0.14
Acalethripidae	Aeolothrips intermedius	4	1.71	0	0	4	0.55
Aeolothripidae	Melanthripidae						
	Melanthrips fuscus	2	0.86	1	0.20	3	0.41
	Melanthrips pallidior	2	0.86	2	0.41	4	0.55
	Frankliniella occidentalis	50	21.36	125	25.31	175	24.04
	Mycterothrips tschirkunae	5	2.14	0	0	5	0.69
	Oxythrips ajugae	6	2.57	0	0	6	0.82
	Thrips angusticeps	1	0.42	0	0	1	0.14
	Thrips australis	6	2.57	0	0	6	0.82
Thripidae	Thrips hawaiiensis	18	7.70	1	0.20	19	2.61
	Thrips major	110	47.01	256	51.82	366	50.28
	Thrips meridionalis	16	6.84	1	0.20	17	2.33
	Thrips physaphus	1	0.42	0	0	1	0.14
	Thrips pillici	1	0.42	0	0	1	0.14
	Thrips tabaci	8	3.42	105	21.26	113	15.52
Dhlooothrinid	Haplothrips aculeatus	0	0	3	0.60	3	0.41
Phlaeothripidae	Haplothrips govdeyi	2	0.86	0	0	2	0.27
Total		234	100	494	100	728	100.00

Table 2. The number of Thysanoptera species on some stone fruits in Mersin, Türkiye, betweem 2020 and 2021

Family	Thrips species	Apricot	Nectarine	Peach	Plum	Total
	Aeolothrips collaris	0	0	0	1	1
	Aelothrips ericae	0	0	0	1	1
Acalothripidas	Aeolothrips intermedius	0	1	0	3	4
Aeolothripidae	Melanthripidae					
	Melanthrips fuscus	0	1	0	2	3
	Melanthrips pallidior	0	2	0	2	4
	Frankliniella occidentalis	4	62	25	84	175
	Mycterothrips tschirkunae	0	2		3	5
	Oxythrips ajugae	0	2	2	2	6
	Thrips angusticeps	0	0	0	1	1
	Thrips australis		3	1	2	6
Thripidae	Thrips hawaiiensis	2	7	1	9	19
	Thrips major	17	84	20	245	366
	Thrips meridionalis	0	1	0	16	17
	Thrips physaphus	0	0	0	1	1
	Thrips pillici	0	0	0	1	1
	Thrips tabaci	2	45	7	59	113
Dhlacathrinidas	Haplothrips aculeatus	0	2	0	1	3
Phlaeothripidae	Haplothrips govdeyi	0	0	1	1	2
Total		25	212	57	434	728
Species no		4	12	7	18	

#### **Distribution indices**

According to the Shannon-Wiener and Simpson diversity index, it was determined that the most diverse habitat was nectarine with the values of 1.48 and 0.72, respectively (Table 3). Margalef species richness index results were recorded as 2.80, 2.05 and 1.48 for plum, nectarine and peach trees, respectively. Increasing or decreasing dominance (Simpson) values in the opposite relationship with diversity were calculated with a maximum of 0.37 in plum and a minimum of 0.28 in nectarine depending on the results of biodiversity parameters. Different results were obtained in the Shannon, Simpson indices and species population density values. Peach was determined as the most balanced population densities between species with 0.32, 0.21 and 0.68.

Table 3. Distribution indices	of thri	ps on	stone	fruits	in	Mersin.	Türkive
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Indexes	Nectarine	Peach	Plum
Shanonn-Wiener index	1.48	1.31	1.37
Simpson diversity index	0,72	0,68	0,63
Margalef species richness index	2.05	1.48	2.80
Simpson dominancy index	0.28	0.32	0.37
Shanonn evenness index	0.27	0.32	0.22
Simpson evenness index	0.11	0.21	0.09
Species evenness index	0.59	0.68	0.47

#### Population densities of thrips species

Stone fruit species, sampling location, thrips species and interactions between them (except fruit species x sampling region) were found significant (p<0.0001; Table 4). Seasonal mean densities of the three common thrips species according to three stone fruit species were shown in Figure 2.

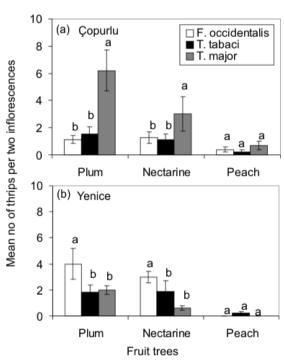


Figure 2. Seasonal mean (± SEM) numbers of three Thysanoptera species detected on some stone fruits in Çopurlu (a) and Yenice (b) located at Mersin in year 2021. The mean values with the same letter on the bars are not significant at the p<0.05 level according to the Tukey's HSD test.

Thrips major were statistically higher and more significant than the other two species on plum (F = 9.035, df = 2, 66, p <0.0001) and nectarine (F = 9.901, df = 2, 66, p <0.0001). Population densities of all the three species were low and similar in peach trees (F = 0.986, df = 2, 66, p >0.05). Contrary to location Çopurlu, high and significant numbers of F. occidentalis individuals were recorded in plum (4.00±0.64 individuals per inflorescence) and nectarine (2.97±0.45 individuals per inflorescence) in location Yenice (Plum: F = 5.286, df = 2, 66, p<0.05; Nectarine: F = 5.945, df = 2, 66, p<0.0001). Similarly, very few adult individuals of all the three species were found in peach in this location (F = 5.945, df = 2, 66, p > 0.05).

Table 4. Results of two-way analysis of variance

Source of variation	df	MS	F
Fruit species	1	228.167	193.852*
Sampling location	1	83.130	70.627*
Thrips species	2	483.227	410.513*
Fruit species × Sampling location	1	0.667	0.566**
Fruit species x thrips species	2	117.542	99.864*
Sampling location × thrips species	2	507.144	430.873*
Fruit species × Sampling location × thrips species	2	105.192	89.881*
Error		1.177	

<sup>\*</sup> p<0.0001, \*\* p>0.05.

The seasonal average densities of three common thrips species according to the different phenological periods (flowering, petal fall and fruiting period) of the three fruit species were shown in Figure 3.

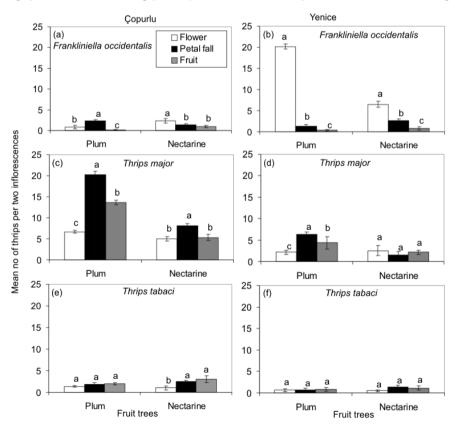


Figure 3. Seasonal mean (± SEM) numbers of Frankliniella occidentalis (a, b), Thrips major (c, d) and Thrips tabaci (e, f) according to phenology of plum and nectarine orchards in Çopurlu and Yenice located at Mersin in year 2021. The mean values with the same letter on the bars are not significant at the p<0.05 level according to the Tukey's HSD test.

In Çopurlu, significant and high numbers of F. occidentalis were found in plum trees during flower petals fall and in nectarine during flowering (Plum: F = 10.076, df = 2, 15, p < 0.05; Nectarine: F = 6.233, df = 2, 15, p < 0.05). Moreover, this harmful thrips species was recorded in significant and high numbers in both plum and nectarine flowers in Yenice (Plum: F = 724,892, df = 2, 15, p < 0.0001; Nectarine: F = 31.796, df = 2.15, p < 0.0001). Thrips major were mainly recorded in plums when trees drop their flower petals in Çopurlu (Plum: F = 160.667, df = 2, 15, p < 0.05; Nectarine: F = 7.976, df = 2, 15, p < 0.01) and Yenice (Plum: F = 5.177, df = 2,15, p < 0.05). This species was sampled in relatively lower and similar numbers in different phenological periods of nectarine trees in the sampling locations (F = 0.344, df = 2, 15, p > 0.05). The other thrips species T. tabaci were recorded in similar numbers in different phenological periods of plum and nectarine trees in Çopurlu (Plum: F = 1.327, df = 2, 15, p > 0.05; Nectarine: F = 3.824, df = 2, 15, p > 0.05) and Yenice (Plum: F = 0.048, df = 2, 15, p > 0.05; Nectarine: F = 1.287, df = 2, 15, p > 0.05).

### Population fluctuations of thrips species in stone fruit orchards

Population changes of thrips species according to the sampling dates on different fruits sampled in Copurlu in 2021 were shown in Figure 4.

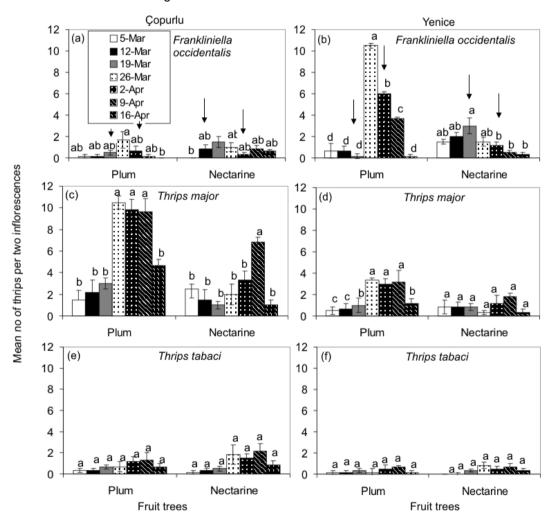


Figure 4. Mean numbers (±SEM) of Frankliniella occidentalis (a, b), Thrips major (c, d) and Thrips tabaci (e, f) according to sampling dates on plum and nectarine orchards in Çopurlu and Yenice located at Mersin in year 2021. The mean values with the same letter on the bars are not significant at the p<0.05 level according to the Tukey HSD test. Arrows show dates of the insecticides applications.

Although the population of F. occidentalis in plum blossoms in Mersin reached the highest value (1.66±0.76/2 individuals per inflorescence) on March 26, no significant difference was detected between the mean number of individuals on other sampling dates, except for April 16 (F = 3.853, df = 6, 35, p <0.05), Frankliniella occidentalis showed a short-term increase in nectarine on March 19, and the average number of individuals was found to be statistically high and significant only on the first sampling date (March 5) (F = 3.500, df = 6, 35, p <0.05). Thrips density was found to be relatively higher in both plum and nectarine in Yenice. The mean number of F. occidentalis individuals increased significantly on March 19 (3.66±0.21/2 individuals per inflorescence) in plum, which was significantly higher compared to other sampling dates, and the lowest number of individuals was recorded on April 16 on the last sampling date. Thrips major population density was quite high compared to T. tabaci and F. occidentalis on plum and nectarine flowers in Copurlu. The number of T. major individuals in this region was similar between March 26 and April 9, but significantly higher than other sampling dates (F = 20,922, df = 6, 35, p < 0.0001). In the same region, thrips population density was similar and low at all sampling dates in nectarine (F = 1.143, df = 6, 35, p > 0.05). Thrips tabaci population was found in similar densities in both plum and nectarine flowers in the Copurlu throughout the sampling date and no significant differences were observed (Plum: F = 0.885, df = 6, 35, p > 0.05; nectarine: F = 1.417, df = 6, 35, p >0.05). The density of T. tabaci was also quite low in Yenice, and they were caught in the flowers of both fruit species in similar numbers during the sampling dates (Plum: F = 0.749, df = 6, 35, p >0.05; nectarine: F = 1.542, df = 6, 35, p >0.05).

#### Thrips damage

No thrips damage was observed on both leaves and flowers of all fruit trees. Also, thrips did not damage peach and apricot fruits. But scarred fruits were mainly recorded in nectarine and plum orchards (Figure 4). The rate of scarred fruits was between 3-18% in plum orchards and 15-32% in nectarine orchards (Table 5). Generally, silvery scar tissue was formed on the fruit surfaces on plum and nectarine fruits. Moreover, large spots in the form of silvering were detected on some fruits. Cracking and gamming have occurred on nectarine fruits which are extremely damaged.

Table 5. Scarred fruit rates due to	thrips damage in some stone f	fruits in 2019 in Mersin, 7	Γürkiye
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Location	Fruit species/ orchard no	No of undamaged fruit	No of damaged (scarred) fruit	Ratio of scarred fruit	Damage scale
	Plum/1ª	93	7	7	3
	Plum/2	95	5	5	2
	Plum/3	97	3	3	2
Convelv	Plum/4	96	4	4	2
Çopurlu –	Nectarine/1 <sup>a</sup>	94	6	6	2
	Nectarine/2	68	32	32	4
	Nectarine/3	98	2	2	2
	Nectarine/4	84	16	16	3
	Plum/1 a	85	15	15	3
Yenice	Plum/2	82	18	18	3
renice	Nectarine/1 a	97	3	3	2
	Nectarine/2	96	4	4	2

<sup>&</sup>lt;sup>a</sup> Experimental areas where population densities of the thrips species were monitored.

#### **Discussion**

In this study, 18 thrips species were identified and most of them were phytophagous thrips besides a few predatory thrips (Aeolothrips). *Thrips major* and *F. occidentalis* were recorded as the two most common thrips species in fruit trees (Table 1). Atakan (2008) noted that *T. major* was the main thrips

species in apple and nectarine trees in Adana for some years. Hazır et al. (2011) found 12 thrips species in which *F. occidentalis* and *T. major* were predominant species in nectarine. Pearsall (2000) and Pearsall & Myers (2001) reported that *F. occidentalis* was the main thrips species on nectarine trees in British Columbia. Additionally, Şengonca et al. (2006) determined 21 thrips species and *F. occidentalis* was present in all sampled nectarine trees in Northern Cyprus.

The highest number of thrips individuals was recorded on plum trees, followed by nectarine flowers. The abundance and distribution of thrips can be influenced by various factors, including plant phenology, volatile compounds, pollen richness, and flower structure. In addition, plum was recorded as a plant species rich in thrips species diversity (Table 3). Since fruit trees were sampled in the same ecosystem, it has been conceived that the thrips population was similarly influenced by ecological conditions and the surrounding fauna. However, *F. occidentalis* was more prevalent in plum and nectarine flowers in Yenice, which may be attributed to the production of fruit trees and other alternative host plants like winter and summer vegetables in the area. It has been pointed out that *F. occidentalis* was a common species on vegetables both in the open field and greenhouses and has an important role in the transmission of TSWV disease, particularly in peppers around the region (Atakan et al., 2013).

Frankliniella occidentalis was generally recorded during the flowering period in fruit trees, while *T. major* was more prevalent during the petal fall (Figure 2). In Adana, *F. occidentalis* adults were collected mostly from the flowers and larvae were recorded in the period when they shed their flower petals on nectarine trees (Hazır et al., 2011). A similar pattern was observed in Güzelyurt, Northern Cyprus, where *F. occidentalis* adults were first recorded in early nectarine varieties at the end of February and in mid-March during flower petal fall in Gaziveren. In addition, the first adults were collected during the pink flower period on the late flowering variety of nectarines. In the other study, a higher number of thrips individuals were found in the petal leaf stage of apple and nectarine trees (Atakan, 2008). The results of the current study are consistent with previous findings that thrips larvae are abundant in the petal leaf stage. At the end of the flowering period, the number of adult individuals of both species decreased both in the petal leaf fall and young fruit periods. The higher occurrence of *T. major* in petal leaf falls in nectarine and plum trees in both years may be related to the fact that the larvae of this species were recorded mainly during this period. It was determined that all the collected larvae belonged to *T. major*. However, in contrast to these results, *F. occidentalis* individuals in the Mid-Atlantic region (Canada) and British Columbia were mainly found in the flowering period of nectarines according to Felland et al. (1995) and Pearsal & Myers (2001).

It seems that the timing and location of thrips species vary depending on the geographic location, climatic factors, and host plant. While F. occidentalis adults were more common in the middle or end of March in both fruit species, the number of T. major individuals (adults+larvae) was similar between the end of March and the second week of April, but significantly higher than other sampling dates (Figure 3). No larvae or adult thrips species were found on both young and mature fruits. The occurrence of thrips larvae is most commonly observed during the petal fall period, which can vary depending on climate factors, especially temperature. Frankliniella occidentalis adults lay eggs in the ovary in the flower, the larvae feed during the development period of the ovary, and the scar on the fruit surface forms when the fruit develops (Pearsall, 2000; Ready et al., 2001). Within the current study, thrips larvae (T. major) were in the last larval stage when the flower petals were shedding. This result suggests that T. major adults lay eggs in the flower organs and thus the larvae emerge in the earlier fruiting period. In other words, F. occidentalis was recorded as a pest in stone fruits, especially nectarines and peaches in North America (Yonce et al., 1990; Felland et al., 1995; Pearsall, 2000; Pearsall & Myers 2000, 2001), Spain (Lacasa et al. 1991), France (Grasselly et al. 1993), Italy (Guarino et al. 1993; Tocci, 1995; Gargani, 1996), and Türkiye (Atakan, 2008; Hazır et al., 2011; Hazır & Ulusoy, 2012). Our findings suggested that T. major is a noxious thrips species in plum and nectar as much as F. occidentalis. Although Taeniothrips meridionalis Priesner, 1926 (Thysanoptera: Thripidae) and T. major were reported to cause damage on nectarines in Italy (Cravedi & Molinari, 1984),

*T. major* was recorded in low numbers in nectarine flowers and could not be found in all sampling areas in Northern Cyprus (Şengonca et al., 2006). While both thrips species have been recorded as pests in various regions worldwide, *T. major* has been found in low numbers in some areas, indicating its presence may be influenced by local environmental factors.

Scarring and deformations in the form of silvering in both plum and nectar were recorded in fruits (Figure 4). Similarly, significant scarring on fruit surfaces has been reported in France, northern Spain, Italy, Greece, Pennsylvania, and the southeastern United States (Kourmadas et al., 1982; Cravedi et al., 1983; Cravedi & Molinari, 1984; Yonce et al., 1990; Lacasa et al., 1991; Grasselly et al., 1993; Felland et al., 1995). In this study, the scarring fruit rate was similar and low due to effect of sprayings in nectarines and plums, varying between 2-7% (Table 5). The study by Atakan (2008) reported the rate of injured fruit was 30% in the unsprayed plum and nectarine orchards located at Adana Province, Türkiye, however, it has been also determined as 74% in nectarine fruits (Hazır & Ulusoy, 2012). In our study, no damage was observed in the flower organs due to adult feedings during the periods when thrips adults were more abundant in the flowers. Possibly the damage is more related to the larvae feeding on the ovary than the egg laying. In addition, adult and larval feeding was not observed in newly formed fruits. This could be related to the nectarine and plum growers' application of insecticides in the region with active ingredients, spinosad or spinetoram insecticides, usually three times, during the flower bud period, 20% and full flowering, and the petal fall periods.

In conclusion, although *F. occidentalis* was reported as a common species in both nectarine and plum trees in the region, in the current study, *T. major* was found to be a main thrips species responsible for damage in plums and nectarines and the larvae found belonged to this thrips species. The main reason for the damage in plum and nectarine may be related to the laying of eggs by the adult thrips during the flowering period and thus the feeding of the larvae in the ovarium during the flowering period. In order to reveal this situation more clearly, further research should be undertaken to investigate the egg laying times of the adults in different generative periods of the trees, as well as the thrips species and densities. The role of meteorological factors (especially temperature and relative humidity) in egg-laying behavior of the female adults and the emergence of the first larvae should also be taken into consideration. Further studies on these issues may be important in determining the most appropriate time for spraying in early and late flowering stone fruit varieties to prevent unnecessary insecticide use in the region and also other locations sharing similar ecological conditions such as in the Mediterranean region.

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# Original article (Orijinal araştırma)

# Effect of washing method on the reduction of insecticide residues and quality characteristics of sweet cherry fruits<sup>1</sup>

Yıkama yönteminin kiraz meyvelerindeki insektisit kalıntılarının azaltılmasına ve ürün kalitesi üzerine etkisi

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#### **Abstract**

Sweet cherry trees were sprayed with 5 insecticides (acetamiprid, dimethoate, lambda-cyhalothrin, malathion, tau-fluvalinate) at the recommended field doses in this study. Fruits were harvested after the pre-harvested interval for each pesticide completed and then they were immersed into tap water and three different washing solutions (with three different concentrations) for 3 minutes (at 20°C). Insecticide concentrations were detected with a multi-residual analysis method using LC-MS/MS in Bursa Uludağ University in 2022. Following the treatments, changes in the quality characteristics of fruits were also investigated by quality (colour, texture and fruit cracking rate, water-soluble dry matter) and sensory analysis (fruit and stem colour, firmness, appearance, general acceptability). The results revealed that washing method with tap water during 3 min decreased insecticide residue level by 7-45% depending on insecticide active compound. Higher reduction rates were observed by washing with citric acid (10%), sodium bicarbonate (2.5%) and sodium hydroxide (0.5%). But significant reductions were detected only in lambda-cyhalothrin and malathion residues when compared with the newly harvested fruit samples. Processing factors (PF) of all washing methods were generally lower than 1 except for three treatments. PF values showed variations depending on the type of washing solution and the active compound of insecticides. Although washing with citric acid (10%), sodium bicarbonate (2.5%) and sodium hydroxide (0.5%) solutions caused reduction in residue levels, their negative effects on the quality and sensory characteristics of the fruits cannot be ruled out.

Keywords: Cherry, insecticide, LC-MS/MS, pesticide residues, processing factor, washing method



Bu çalışmada, kiraz ağaçlarına 5 insektisit formülasyonu (acetamiprid, dimethoate, lambda-cyhalothrin, malathion, tau-fluvalinate) önerilen dozlarda meyvelere uygulanmıştır. Hasat öncesi bekleme süresinden sonra hasat edilen meyveler musluk suyu ve üç farklı yıkama solüsyonuyla (üç farklı konsantrasyonda) 3 dakika süreyle (20°C'de) yıkanmıştır. İnsektisit kalıntıları LC-MS/MS cihazı kullanılarak çoklu kalıntı analizi yöntemiyle Bursa Uludağ Üniversitesi'nde 2022 yılında tespit edilmiştir. Yıkama uygulamaların ardından meyvelerin kalite özelliklerinde meydana gelen değişiklikler (renk, doku ve meyve çatlama oranı, suda çözünür kuru madde) ve duyusal özellikleri (meyve ve gövde rengi, sertlik, görünüm, genel kabul edilebilirlik) ayrıca araştırılmıştır. Araştırma sonuçlarına göre, musluk suyuyla 3 dakika süreyle yıkama yöntemi, insektisit etken maddesine bağlı olarak kalıntı seviyesini %7-45 oranında azalttığı ortaya konulmuştur. Diğer taraftan, sitrik asit (%10), sodyum bikarbonat (%2.5) ve sodyum hidroksit (%0.5) ile yıkamada daha yüksek etkiler gözlemlenmiştir. Ancak hasat edilen meyve örnekleriyle karşılaştırıldığında sadece lambda-cyhalothrin ve malathion kalıntılarında istatistiki anlamda önemli azalmalar tespit edilmiştir. Tüm yıkama yöntemlerinin işleme faktörleri (PF), üç işlem dışında genellikle 1'den düşük bulunmuştur. PF değerleri yıkama solüsyonunun cinsine ve insektisitlerin aktif bileşiğine bağlı olarak değişiklik göstermiştir. Sitrik asit (%10), sodyum bikarbonat (%2.5) ve sodyum hidroksit (%0.5) solüsyonları ile yıkama, kalıntı düzeylerinde azalmaya neden olsa da meyvelerin kalitesi ve duyusal özellikleri üzerinde göz ardı edilemez olumsuz etkiler oluşturmuştur.

Anahtar sözcükler: Kiraz, insektisit, LC-MS/MS, pestisit kalıntısı, işleme faktörü, yıkama metodu

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#### Introduction

Previous studies have shown that fruits have protective effects against the development of serious human diseases such as cardiovascular problems, diabetes, obesity and cancer (Ferretti et al., 2010). Their protective roles could be originated from the various nutrients such as dietary fiber, vitamins and phytonutrients. Several studies on phenols originated from fruits have demonstrated that they are bioavailable and have got a protective role against oxidative stress and free radical damages in human (Prior, 2003).

Cherries belonging to the subgenus *Cerasus*, *Prunus avium* L. of stone fruit family (Rosaceae) are known as "sweet cherry". Sweet cherries are characterized by high content of simple sugar, hydrosoluble (C) and liposoluble (A, E and K) vitamins, niacin, panthothenic acid, some carotenoids (beta carotene, lutein, zeaxanthin) and phenols. Some phenolic acids such as hydroxycinnamates, flavonols and flavan-3-ols and minerals like calcium, magnesium, phosphorous and potassium are also found in sweet cherry fruits (Gao & Mazza, 1995; Chaovanalikit & Wrolstad, 2004). Since sweet cherry fruits contain several antioxidants and particularly polyphenols, the fruits display many biological activities, such as antioxidant, anti-inflammatory and anticancer properties.

The geographic and climatic conditions in many regions of Türkiye are appropriate for sweet cherry production and Türkiye is an important sweet cherry producer with the production rate of 2.5 million tons (25.0%), followed by USA (12.0%), Uzbekistan (7.0%), Chile (6.0%), Iran (5.0%), and Italy and Spain (4.0%), respectively (TUİK, 2021; FAO, 2021). Dimethoate, tau-fluvalinate, lambda cyhalothrin and malathion are intensively used insecticides for the control of insect pests during sweet cherry growing (Hazarhun et al., 2022; BKUTARIM, 2023). The heavy use of these pesticides may end in environmental problems such as ecological imbalance, widespread pest resistance, environmental pollution, hazards to non-target organisms and wildlife (Simon, 2014). These pesticides may also cause some health problems like reproduction/ development effects, and act as endocrine disruptor, skin irritant, respiratory tract irritant, skin sensitizer, eye irritant, acetyl cholinesterase inhibitor and neurotoxicant (PPDB, 2023). Although synthetic pesticides have the significant role in crop productivity and food security, their residues in agricultural products exert serious risks for human health (Çatak & Tiryaki, 2020). Therefore, there is an increasing interest in reducing pesticide residues over the surfaces or within the tissues of vegetables and fruits using different washing methods. Success of washing treatments on residues is highly dependent on the physicochemical properties of pesticide, such as mode of action (systemic or residual), water solubility, pH sensitivity, volatility and persistence. So, due to the different mode of action of pesticides, their removal is possible only by multiple methods of decontamination depending on the effects of these methods on the quality characteristics of the target commodity. The effect of washing on pesticide residue levels of cherry fruits is still not known. Effect of washing on the concentration of pesticide residues of different commodities (Al-Taher et al., 2013; Lozowicka et al., 2016; Acoglu & Yolci Omeroglu, 2021; Duman et al., 2021; Polat, 2021; Tiryaki & Polat, 2023) were studied previously with tap water and/or solutions containing different salts such as citric acid (CA), sodium bicarbonate (SB), sodium hydroxide (SH), potassium permanganate (PP) and acetic acid (AA). Harinathareddy et al. (2014) reported 37.0-73.2% reduction in the dimethoate, chlorpyrifos, quinolphos, profenophos, phosalone, lamda-cyhalothrin, malathion and triazophos concentrations as a result of washing with tap water for 10 minutes (Harinathareddy et al., 2014). 24.0-97.41% reduction in the concentration of various pesticides were reported after washing with SB, PP, SH and CA solutions (Radwan, 2005; Harinathareddy et al., 2014; Polat & Tiryaki, 2020; Yalçın et al., 2023).

Industrial cherry packaging, involves a washing step (generally 3 minutes) for the removal of superficial dirt. For this purpose, fruits are washed with tap water during the sorting and classification processes by using automatic machines. Fresh sweet cherry fruits are very delicate, so for the protection of the fruit quality, they require painstaking processing and packaging precautions and also transportation conditions within the cold chain. Undesirable visible changes may easily occur on the fruit surface if these factors are not taken into consideration. The effects of pesticide removal applications on the quality characteristics of sweet

cherry fruits have not been studied yet. Previous studies have showed that chemical pre-treatments such as alkali solutions (sodium hydroxide) break down the wax on the cuticular surface of cherry and create microscopic cracks that increase moisture losses (Doymaz & İsmail, 2011). High amounts of dehydration (75%) were reported for sour cherry samples dipped in 1% (w/v) citric acid solution at room temperature in 50 h (Tarhan et al., 2006). The effect of SB solutions on postharvest decay of sweet cherry was investigated previously but how quality and sensory characteristics of fruits affected were not monitored concurrently (Karabulut et al., 2001). Presence of pesticide residues on agricultural commodities is an important food safety and public health issue and recently numerous studies were focused on their removal from the consumer products. The agents can safely take in low doses for human, but high concentrations could cause skin irritation or serious eye irritation (Merck, 2023). However, there is still a big gap about the reliability of these applications and how they affect the sensory and quality characteristics of the commodity. This study is conducted to fill the gap in this field by measuring the effectiveness of citric acid (CA), sodium bicarbonate (SB) and sodium hydroxide (SH) solutions for the removal of some pesticide residues and displaying their effects on the colour, texture and sensory characteristics of the sweet cherry fruits.

#### **Materials and Methods**

#### Pesticide applications

Unsprayed sweet cherry trees in an orchard in Bursa (Kestel) were used during 2022 season. Before insecticide spray, the samples collected from these trees were analysed to ensure there is no pesticide residue (Figure 1). Each plot size had four trees. The plots were with three replicates. Insecticides tested in current study were selected based on the results of our survey study during 2020–2021 seasons (Hazarhun et al. 2022). According to OECD (2008) guide, acetamiprid (Mospilan 20 SP), dimethoate (Poligor 40 EC), malathion (Malathion 65 EM), lambda-cyhalothrin (Sumosa 5 EC) and tau-fluvalinate (Mavrik 24 EW) were sprayed homogenously with an electric atomizer at the recommended field doses (Table 1). After pre-harvest interval (PHI) for each pesticide completed (7th and 14th days), 1 kg of sweet cherry sample were randomly collected and immediately transferred to laboratory in cold chain (Figure 1).

Table 1. Application dose, residue levels at the harvest day and maximum residue levels of the pesticides (BKUTARIM, 2023; EU, 2023)

Pesticides	Pesticide application dose	EU MRL	TR MRL	PHI
Pesticides	(g /100 L water)	(µg kg <sup>-1</sup> )	(µg kg <sup>-1</sup> )	(days)
Acetamiprid	5	1500	1500	7
Dimethoate	**	10	**	**
Lambda-cyhalothrin	2.5	300	300	14
Malathion	65	20	20	7
Tau-fluvalinate	7.2	400	400	14
Omethoate	*	10	**	**

<sup>\*</sup> Not used due to the restrictions in Türkiye, but determined as dimethoate metabolite,

#### **Experimental design**

Tap water and three different washing solutions (with three different concentrations) were used during the experiments (Figure 1). 75 g portions of sweet cherry samples were immersed into 1.5 L washing solution (at 20°C) for 3 min. Following washing, samples were air-dried and prepared for the pesticide residue analyses. For the simulation of transport conditions to markets, a part of the samples was stored under cold conditions (4°C) during 8 days. Then, a series of fruit quality tests were performed on them. All of the analyses were done in triplicate.

<sup>\*\*</sup> No indicated; EU: European Union, MRL: Maximum residue limit, TR, Türkiye, PHI, Post Harvest Interval.

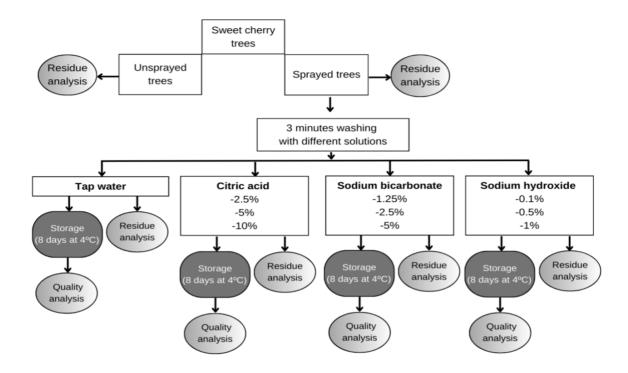


Figure 1. Processing steps and sampling details of different washing treatments.

#### Chemicals and reagents

The analytical standards used in validation studies were purchased from Dr. Ehrenstorfer GmbH (Germany), AccuStandard (USA) and CRM Labstandard (UK). The purity of all reference standards was higher than 96%. All other reagents, acetonitrile, ammonium format, formic acid and methanol were of analytical grade and obtained from different manufacturers. Quick, easy, cheap, effective, rugged, and safe (QuEChERS) extraction (Q16E15E092) and clean-up (Q2A23Z592) kits were supplied from QuE Lab (Italy) (AOAC 2007.01 method). Chemical and toxicological characteristics of each pesticide tested in this study are presented in Table S1.

#### Pesticide analysis

Pesticide analysis were performed using Agilent 6470 Triple Quad Liquid–Mass Spectrometry (Agilent Technologies, Santa Clara, CA, USA). Chromatographic separation was achieved using gradient elution with Agilent Poroshell SB-C18 analytical column (3 × 100 mm × 2.7 µm) at 40°C. The mobile phase consisted of an aqueous solution of 0.1% formic acid and 1 mM ammonium formate (A) and methanol (B). The mobile phase flow rate was 0.52 mL min<sup>-1</sup>. The gradient elution programme was as follows: 0–0.5 min 70% A, 0.5–8 min 70% A, 8-12.5 min 5% A, 8-12.6 min 5% A and 12.6-15 %70 A. The detection by the mass spectrophotometer (MS) was conducted in multiple-reaction monitoring and electrospray ionization mode. Nitrogen gas (N<sub>2</sub>) was used as nebulizing and drying gas and supplied by a nitrogen generator (Peak Scientific Scotland, UK). Gas flow, gas capillary voltage and source temperature were set at 10 psi, 3500 V and 100 °C rrespectively. Sample injection volume was 1 µL. Experimental parameters (precursor and product ions, collision energy) for LC–MS/MS analysis of pesticides were displayed at Table S2. Sweet cherry fruits for each treatment were homogenised and used for pesticide analysis. Extraction and partition of pesticides were done using QuEChERS kits according to the manufacturer's instructions (QuE Lab, Italy). Sample preparation steps for pesticide analysis were summarised in Figure 2 (Lehotay, 2007; Hazarhun et al., 2022).

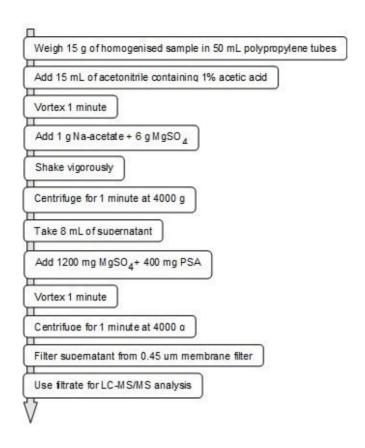


Figure 2. Sample preparation for LC-MS/MS analysis.

#### Method verification

The vertification of the analysis method was tested according to SANTE/12682/2021 guidelines (SANTE, 2021). Within the guideline's context, selectivity, linearity, limit of detection (LOD), limit of quantification (LOQ), trueness (recovery %) and precision (intra-day and inter-day) parameters were tested. The selectivity was determined by analysing fortified and blank samples. The vertification studies were performed on pesticide-free sweet cherry fruit samples. Linearity was evaluated by six levels matrix-matched calibration ranging from 2.5 to 250  $\mu$ g L<sup>-1</sup>. Trueness (recovery) and precision (repeatability and reproductively) of the method calculated for five days using five replicates for two level (10 and 50  $\mu$ g L<sup>-1</sup>). The sensitivity of the method (LOD and LOQ) was calculated using the standard deviation (SD) of the analysis results of the lowest level (5  $\mu$ g L<sup>-1</sup>) spiked samples (Tiryaki, 2016; SANTE, 2021).

#### Calculation of processing factors

The effect of washing treatments was calculated (equation 1) similar to processing factor (PF) and obtained by the division of the residue concentration of washed fruits to the residue concentration of the unprocessed fruits (Chen et al., 2013, Đorđević et al., 2013).

$$PF = \frac{PRP}{PRR} \tag{1}$$

PF= Processing factor; PRP= Pesticide residue of processed material (mg kg<sup>-1</sup>); PRR= Pesticide residue of raw material (mg kg<sup>-1</sup>)

The PF value may indicate reduction (PF<1), no change (PF=1) or concentration increase (PF>1) of the pesticide residues (Chen et al., 2013). When residues were below the limit of quantification (LOQ) after processing, the PF value was accepted as zero (Aguilera et al., 2012). Additionally, the difference between

the initial pesticide concentration and the residue level of the final product was calculated by using the following formula (equation 2).

$$RR = \frac{PRR - PRP}{PRR} *100 \tag{2}$$

RR= Residue reduction rate (%); PRR= Pesticide residue of raw material (mg kg-1); PRP= Pesticide residue of processed material (mg kg-1)

#### **Determination of quality parameters**

After performing different washing applications, the samples were divided into two equal parts. One group of samples were kept at +4°C for 8 days for the simulation of transportation stage of sweet cherries to markets. At the end of this storage period, changes in colour, texture, water soluble dry content and cracking levels were analysed. Additionally, visual and textural characteristics of the fruits were evaluated by an untrained test panel in terms of appearance, stem colour, firmness, colour and overall acceptance. Panellists were asked to evaluate samples in a random order using a numbered hedonic scale changing from 9 to 1 representing different levels from excellent to non-consumable (Kappel et al., 1996; Martínez-Romero et al., 2006).

For the determination of cracking level, 20 fruits were checked by eye and the number of fruits with cracks were noted. The percentage of cracked fruits were calculated by dividing the number of cracked fruits to the total number of fruits (Bilginer et al., 1999; Yıldırım & Koyuncu, 2010; Ozturk et al., 2018; Akkaya, 2021).

The changes in the fruit colour characteristics were measured based on the L\*, a\*, and b\* criteria determined by the International Lighting Commission (CIE). The L\* coordinate represented the lightness of the colour, a\* indicated the position between green and red, and b\* was the extent of blueness/yellowness. Measurements were made on 20 fruits using Konica Minolta CR-400 colorimeter (Tokyo, Japan). Average of minimum 2 measurements taken from the 2 opposite poles of the equatorial part of each fruit under luminous conditions (McGuire, 1992).

For the determination of fruit flesh firmness, 20 fruits were analysed using TA XT Plus (Stable Micro Systems, Surrey, UK) texture analyser. For this purpose, HDP/BS probe of the device was used in compression mode, 2 mm/s speed and 20% deformation rate (Kumral et al., 2019).

Water-soluble dry matter content of the fruits was measured using refractometer (Kem RA500, Kyoto Electronics Manufacturing Co. Ltd.) after the pitted fruits were homogenised with a household blender (Sinbo, Türkiye) and strained through a cotton cloth (Kappel et al., 1996).

#### Statistical analysis

All analyses and trials were conducted in triplicate. The insecticide concentrations and fruit quality data obtained at each treatment were subjected to one-way variance analysis using JMP 7.0 Software (SAS, Cary, NC). For the detection of different groups, Tukey's multiple comparison test was performed with a significance level of 0.05 ( $\alpha$ ) after normality testing with the Shapiro-Wilk test.

#### **Results and Discussion**

#### Verification results

The results of linearity, limit of detection and quantification, trueness and precision for each pesticide were given at Table S3. Matrix matched calibration curves of the 6 insecticides were linear ( $R^2$  = 0.997-0.999). The LOQ values (2.70 to 4.04  $\mu$ g kg<sup>-1</sup>) were quite lower than the MRLs of each insecticide (Table 1 & S3). The recovery rates of the insecticides for two spike levels were calculated between 101.391-113.74 and 100.91-112.16, respectively. For repeatability and reproducibility parameters, the highest RSDr and RSDwr did not exceed 20% for both spike levels. All verification parameters were compatible with SANTE 11312/2021 criteria (SANTE, 2021).

#### Changes in pesticide residues and processing factor

Pesticide residue levels in sweet cherries harvested at the end of PHI, the processing factors (PF) and the reduction rates (RR) are shown in Table 2. In all the washing methods, the diifferences are significant for lambda-cyhalothrin and malathion during washing with different solutions (lambda cyhalothrin:  $F_{10,36}$ =2.98, P=0.01; malathion:  $F_{10,36}$ =3.35, P=0.006). Whereas only washing with 2.5% sodium bicarbonate yielded significant reductions in residue levels of lambda-cyhalothrin. Similarly, significant declines in malathion residues were observed when washing with citric acid (10%), sodium bicarbonate (1.25%) and sodium hydroxide (0.5 and 1%) solutions compared with residue level in the raw material. Reductions in the concentrations of all active substances were detected during washing treatments, but majority of these were statistically insignificant (acetamiprid:  $F_{10,36}$ =1.09, P=0.41; dimethoate:  $F_{10,36}$ =0.94, P=0.52; fluvalinate:  $F_{10,36}$ =1.67, P=0.14; omethoate:  $F_{10,36}$ =2.12, P=0.06). Processing factors (PF) of all washing methods were generally lower than 1. PF values were showed variations depending on the washing solution and the insecticide active compound (Table 2). The lowest PFs were obtained with citric acid (10%), sodium bicarbonate (2.5%) and sodium hydroxide (0.5%) solutions complying with the previous studies reporting lower PFs for fruits washed with acid or basic solutions (Osman et al., 2014; Polat & Tiryaki, 2020).

Table 2. Insecticide residue levels, their removal rates and the processing factor after different washing treatments

					Pesticio	es		
Treat	ment		Acetamiprid (µgkg <sup>-1</sup> )	Dimethoate (µgkg <sup>-1</sup> )	Lambda- cyhalothrin (µgkg <sup>-1</sup> )	Malathion (µgkg <sup>-1</sup> )	Tau Fluvalinate (µgkg <sup>-1</sup> )	Omethoate (µgkg <sup>-1</sup> )
Initial	residue leve	el	37.67±4.05 <sup>a*</sup>	223.75±8.61 a	30.00±0.00 <sup>a</sup>	6.33±1.45 <sup>a</sup>	10.00±3.51a	10.75±0.25 <sup>a</sup>
Tap v	vater	PR (mg/kg) RR (%) PF	32.00±1.00 <sup>a</sup> 15.02 0.85	186.50±4.50° 16.65 0.83	25.00±2.88 <sup>ab</sup> 16.67 0.83	3.50±0.29 <sup>ab</sup> 44.71 0.55	6.00±1.16 <sup>a</sup> 40.00 0.60	10.00±0.00 <sup>a</sup> 6.98 0.93
Itions	2.5%	PR (mg/kg) RR (%) PF	32.00±1.73 <sup>a</sup> 15.02 0.85	191.67±9.14 <sup>a</sup> 14.34 0.86	27.50±2.50 <sup>ab</sup> 8.33 0.92	3.25±0.75 <sup>ab</sup> 48.66 0.51	6.00±1.16 <sup>a</sup> 40.00 0.60	8.67±0.33 <sup>a</sup> 19.35 0.81
Citric acid solutions	5%	PR (mg/kg) RR (%) PF	36.33±0.88 <sup>a</sup> 3.55 0.96	212.50±6.76 <sup>a</sup> 5.03 0.95	30.00±0.00 <sup>ab</sup> 0 1.00	3.33±0.33 <sup>ab</sup> 47.39 0.53	7.00±0.41 <sup>a</sup> 30.00 0.70	9.25±0.25 <sup>a</sup> 13.95 0.86
Citric	10%	PR (mg/kg) RR (%) PF	29.00±1.53 <sup>a</sup> 22.96 0.77	212.75±7.28 <sup>a</sup> 4.92 0.95	25.00±2.90 <sup>ab</sup> 16.67 0.83	2.67±0.33 <sup>b</sup> 57.82 0.42	4.25±0.63 <sup>a</sup> 57.50 0.43	10.00±0.41 <sup>a</sup> 6.98 0.93
onate	1.25%	PR (mg/kg) RR (%) PF	30.50±4.41ª 18.99 0.81	177.33±35.22° 20.75 0.79	22.50±2.50 <sup>ab</sup> 25.00 0.75	2.25±0.63 <sup>b</sup> 64.46 0.36	4.67±0.33 <sup>a</sup> 53.30 0.47	8.33±0.88 <sup>a</sup> 22.51 0.78
Sodium bicarbonate	2.5%	PR (mg/kg) RR (%) PF	29.33±2.33 <sup>a</sup> 22.09 0.78	199.25±13.83 <sup>a</sup> 10.95 0.89	20.00±0.00 <sup>b</sup> 33.33 0.67	3.00±0.57 <sup>ab</sup> 52.61 0.47	5.00±0.58 <sup>a</sup> 50.00 0.50	8.25±0.63 <sup>a</sup> 23.26 0.77
Sodiur	5%	PR (mg/kg) RR (%) PF	31.50±3.59 <sup>a</sup> 16.34 0.84	192.00±14.01 <sup>a</sup> 14.19 0.86	20.00±0.00 <sup>ab</sup> 33.33 0.67	3.67±0.88 <sup>ab</sup> 42.02 0.58	5.50±1.04 <sup>a</sup> 45.00 0.55	8.75±0.95 <sup>a</sup> 18.60 0.81
oxide	0.1%	PR (mg/kg) RR (%) PF	33.33±1.45 <sup>a</sup> 11.49 0.89	212.33±16.23 <sup>a</sup> 5.10 0.95	30.00±0.00 <sup>ab</sup> 0 1.00	4.00±0.00 <sup>ab</sup> 36.81 0.63	6.00±0.00° 40.00 0.60	9.67±0.67 <sup>a</sup> 10.05 0.89
Sodium hydroxide	0.5%	PR (mg/kg) RR (%) PF	28.00±1.00 <sup>a</sup> 25.61 0.74	202.67±16.25 <sup>a</sup> 9.42 0.91	22.50±2.50 <sup>ab</sup> 25.00 0.75	2.00±0.41 <sup>b</sup> 68.40 0.32	4.75±0.85 <sup>a</sup> 52.50 0.48	9.00±0.58 <sup>a</sup> 16.28 0.84
Sodiu	1%	PR (mg/kg) RR (%) PF	29.67±1.76 <sup>a</sup> 0 1.05	212.33±1.45 <sup>a</sup> 5.10 0.95	26.67±3.33 <sup>a</sup> 11.10 0.89	2.00±0.41 <sup>b</sup> 68.40 0.32	5.50±0.50°a 45.00 0.55	10.00±0.41 <sup>2</sup> 6.98 0.93

PR, pesticide residue; RR, reduction rate; PF, processing factor;

<sup>\*</sup>Means with different letters are significantly different at p<0.05.

Similarly, PFs as low as 0.12-0.27 were reported for neonicotonid and organophosphate insecticides for fruits treated with 2-9% citric acid solutions (Osman et al., 2014; Randhawa, 2014; Polat & Tiryaki, 2020). Radwan et al. (2005) revealed that washing with 0.1% sodium hydroxide solution exhibited PFs of 0.08-0.35 for profenofos on pepper and eggplant. Moreover, Yang (2017) reported decrease in PF of phosmet during washing of apples with sodium bicarbonate solutions.

Water solubility and octanol-water partition coefficient (Log P) are the most significant environmental fate features for removing pesticide residues from agricultural commodities (Holland et al., 1994). Especially, highly soluble pesticides with a low octanol-water partition coefficient can easily be eliminated from these commodities (Randhawa et al., 2014; Lozowicka et al., 2016). In current study, malathion displayed high reduction rate (44.67%) in tap water with a low 2.75 Log P and a high water solubility (148 mgl<sup>-1</sup>). On the contrary, acetamiprid, dimethoate and omethoate exhibited low reduction rates (15.02, 16.65, 6.98%, respectively), despite having high water solubility (2950, 25900 and 500000 mgl<sup>-1</sup>, respectively) and low logP (0.80, 0.75 and -0.9 Log P, respectively). High solubility does not always have the same impact, mode of action also plays a significant role in the removal of pesticide residues from agricultural commodities. Since acetamiprid, dimethoate and its metabolite omethoate have xylem systemic mode of action, they displayed less reduction rates (0-25.61%, 4.92-20.75%, 6.98-23.26% respectively) during different washing treatments. On the other hand, higher reduction rates were observed for contact insecticides malathion and tau-fluvalinate compared with the systemic ones. In compliance with our results, Yang et al. (2017) reported that sodium bicarbonate solutions were more effective in removing surfacecontact pesticides from apples, while it was not completely effective in removing systematic insecticide residues which have penetrated into the fruit.

#### Changes in fruit quality

Fresh cherry fruits are extremely delicate and may be easily damaged during the improper preparation or packaging steps (Goncalves et al., 2007). Fruit size, fruit colour, stem colour, firmness, sweetness, total soluble solids, dry matter content and cracking are all considered as important fruit quality traits (Kappel et al., 1996; Gonçalves et al., 2007; Kovács et al., 2009; Romano & Cittadini, 2014). Fruit firmness is the combination of skin and flesh strength and affects consumer acceptance and shelf life (Kappel et al., 1996). Losses of firmness, colour and flavour in addition to dessiccation, stem discoloration and mould growth are the major causes of product rejection by the consumer (Habib et al., 2017). Changes in fruit characteristics after washing and during storage at 4°C for 8 days are shown in Table 3. The changes in water soluble dry matter content were statistically insignificant (F<sub>9,29</sub>=1.08; p>0.05). Firmness and colour characteristics (L, a, b) of the fruits were affected by the treatments (firmness:  $F_{9,29}$ =4.42, p<0.01; L:  $F_{9,29}$ =3.31, p=0.01; a:  $F_{9,29}$ =4.98, p<0.01; b:  $F_{9,29}$ =4.06, p<0.01). Treatments of sodium hydroxide (1 and 0.5%) solutions caused slight changes in the firmness and colour characteristics of the fruits compared to washing with tap water. 5% sodium bicarbonate treatment caused a decrease in the value, that denotes a colour change towards green. The changes in cherry fruit qualities during transportation, storage and some pre-treatments were previously investigated by several researchers (Habib et al., 2017; Simsek & Sufer, 2021), but there is limited information about the effects of washing on the quality of the cherry fruits. Similar with our findings, Simsek & Sufer (2021) reported insignificant colour changes of cherries after citric acid pre-treatments compared with the control. Results of the sensory evaluation showed that (Table 4), the effects of treatments on the fruit and stem characteristics were significant and application of 5% sodium bicarbonate and 1 and 0.5% sodium hydroxide solutions caused marked decreases in the appearance, fruit colour, stem colour, texture and general acceptability scores of the samples.

Table 3. Changes in fruit quality characteristics after storage at 4°C for 8 days

Trea	ıtment	Water soluble	Firmness	Co	olour parameters	
	uneni	dry matter (%)	(kg/cm <sup>2</sup> )	L	а	b
Тар	water	14.30±1.27 <sup>a*</sup>	2986.95±79.39 <sup>a</sup>	22.31±0.08 <sup>ab</sup>	18.38±0.17 <sup>a</sup>	5.03±0.15 <sup>ab</sup>
acid	2.5%	15.37±0.84°	2553.19±0.40 <sup>ab</sup>	22.86±0.40 ab	19.09±0.43°	5.26±0.24 <sup>a</sup>
Citric acid solutions	5%	15.77±0.73°	2484.40±136.41 <sup>ab</sup>	23.47±0.05 ab	18.33±0.53ª	5.02±0.20 <sup>ab</sup>
S C	10%	16.23±0.82ª	2653.82±202.36 <sup>ab</sup>	23.46±0.14 ab	16.42±0.25 <sup>ab</sup>	4.53±0.05 <sup>ab</sup>
n iate	1.25%	16.50±1.00°	2393.97±53.43 <sup>ab</sup>	22.49±0.79 ab	14.79±0.52 <sup>ab</sup>	3.70±0.20 <sup>ab</sup>
Sodium bicarbonate	2.5%	15.10±0.82 <sup>a</sup>	2405.83±67.34 <sup>ab</sup>	23.32±0.10 ab	15.58±0.55 <sup>ab</sup>	4.04±0.25 <sup>ab</sup>
	5%	16.50±0.21ª	3018.61±172.45 <sup>a</sup>	21.62±0.39 <sup>b</sup>	12.51±2.34 <sup>b</sup>	3.46±0.44 <sup>b</sup>
⊒ ide	0.1%	16.07±0.82ª	2778.22±144.52a	22.94±0.71 ab	15.08±0.89 <sup>ab</sup>	4.12±0.66 <sup>ab</sup>
Sodium hydroxide	0.5%	14.90±0.25ª	2456.48±251.18 <sup>ab</sup>	24.48±0.35°	18.07±0.79 <sup>a</sup>	5.28±0.29 <sup>a</sup>
	1%	14.33±0.70 <sup>a</sup>	1956.72±174.85 <sup>b</sup>	23.81±0.58 ab	15.54±0.69 <sup>ab</sup>	4.93±0.36 <sup>ab</sup>

<sup>\*</sup>Means with different letters are significantly different at p<0.05.

Table 4. Sensory evaluation of textural and visual changes in sweet cherry samples

Т	reatment	Appearance	Colour	Stem colour	Texture	General acceptability
Т	ap water	6.63±0.38 <sup>a</sup> *	6.45±0.31ª	5.90±0.59 <sup>a</sup>	6.63±0.41 <sup>ab</sup>	6.27±0.38 <sup>a</sup>
- pi	2.5%	6.90±0.25 <sup>a</sup>	6.63±0.27 <sup>a</sup>	6.90±0.43 <sup>a</sup>	7.45±0.38 <sup>a</sup>	6.72±0.35 <sup>a</sup>
Citric acid solutions	5%	5.18±0.26 <sup>ab</sup>	5.45±0.41 <sup>ab</sup>	5.72±0.44 <sup>a</sup>	6.09±0.56 <sup>ab</sup>	5.36±0.33 <sup>ab</sup>
SS	10%	5.00±0.46 <sup>abc</sup>	5.54±0.43 <sup>ab</sup>	6.09±0.60 <sup>a</sup>	6.18±0.53 <sup>ab</sup>	5.36±0.43 <sup>ab</sup>
ate	1.25%	6.63±0.41 <sup>a</sup>	6.54±0.34 <sup>a</sup>	6.36±0.50 <sup>a</sup>	6.45±0.38 <sup>ab</sup>	6.36±0.38 <sup>a</sup>
Sodium bicarbonate	2.5%	5.81±0.48 <sup>a</sup>	6.09±0.36 <sup>a</sup>	5.27±0.48 <sup>a</sup>	5.90±0.54 <sup>ab</sup>	5.45±0.54 <sup>a</sup>
S	5%	3.81±0.56 <sup>bc</sup>	3.45±0.54°	2.18±0.35 <sup>b</sup>	3.09±0.51°	2.45±0.28°
	0.1%	5.90±0.41 <sup>a</sup>	5.81±0.44 <sup>ab</sup>	5.36±0.54 <sup>a</sup>	5.54±0.56 <sup>ab</sup>	5.72±0.48 <sup>a</sup>
Sodium hydroxide	0.5%	3.09±0.57 <sup>cd</sup>	3.90±0.62 <sup>bc</sup>	4.81±0.61 <sup>a</sup>	4.72±0.55 <sup>bc</sup>	3.54±0.47 <sup>bc</sup>
s y	%1	1.63±0.30 <sup>d</sup>	3.09±0.41°	4.90±0.68 <sup>a</sup>	4.36±0.65 <sup>bc</sup>	2.27±0.33°

<sup>\*</sup>Means with different letters are significantly different at p<0.05.

#### Conclusion

Pesticide residues on agricultural commodities may cause some adverse health effects for consumers but their use during agricultural practices is inevitable due to the prevention of product losses. Treatment of fruits with different washing solutions may be an alternative way of reducing pesticides residues on consumer products. Thus, applications of citric acid (10%), sodium bicarbonate (2.5%) and sodium hydroxide (0.5%) solutions resulted significant reductions in the residue concentrations of lambdacyhalothrin and malathion. However further research is necessary for the optimisation of their use due to the restrictions caused by their negative effects on the sensory and quality characteristics of delicate agricultural commodities. Instead of relying on washing methods, it is recommended to take actions to reduce pesticide residue in growing sweet cherry. However, it is a positive development that the use of some systemic insecticides (dimethoate and omethoate) has been banned in Türkiye in recent years.

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Table S1. Chemical and toxicological characteristics pesticides (PPDB, 2023)

			Toxicological features					
Pesticide	Chemical group	Mode of action	ADI (mg kg <sup>-1</sup> bw day <sup>-1</sup> )	ARfD (mg kg <sup>-1</sup> bw day <sup>-1</sup> )	Oral LD <sub>50</sub> (mg kg <sup>-1</sup> bw)	Dermal LD <sub>50</sub> (mg kg⁻¹ bw)	Inhalation $LD_{50}$ (mg $kg^{-1}$ bw)	WHO Classification
Acetamiprid	Neonicotinoids	Insecticide	0.025	0.025	146	2000	>1.15	II
Dimethoate	Organophosphorus	Insecticide	0.001	0.01	245	2000	1.68	П
Lambda-cyhalothrin	Synthetic pyrethroids	Insecticide	0.0025	0.005	56	632	0.066	П
Malathion	Organophosphorus	Insecticide	0.03	0.3	1778	2000	>5	III
Tau-fluvalinate	Synthetic pyrethroids	Insecticide	0.005	0.05	546	2000	>0.56	III
Omethoate	Organophosphorus	Insecticide	0.0003	0.002	50	145	0.3	lb

ADI: Acceptable daily intake, ARfD: Acute reference dose, Oral  $LD_{50}$ : Acute oral lethal dose for mammals, Dermal  $LD_{50}$ : Dermal lethal dose for mammals, Inhalation  $LD_{50}$ : Inhalation lethal dose for mammals, WHO: World Health Organisation, 1b: Highly hazardous; II: moderately hazardous, III: slightly hazardous.

Table S2. Pesticide information and optimized LC-MS/MS conditions

Pesticide	CAS number	Molecular weight	Molecular formula	lonization	Precursor ion	Product ion	Collision energy (V)	Retention time (min.)
Acetamiprid	135410-20-7	222.67	$C_{10}H_{11}CIN_4$	[M+H]+	223.1	126.1; 56.2	17, 11	2.67
Dimethoate	60-51-5	229.26	$C_5H_{12}NO_3PS_2$	[M+H]+	230.0	198.9; 125.0	3, 17	3.54
Lambda-cyhalothrin	91465-08-6	449.85	$C_{23}H_{19}CIF_3NO_3$	[M+H]+	467.1	450.0; 225.0	6, 14	7.88
Malathion	121-75-5	330.36	$C_{10}H_{19}O_6PS_2$	[M+H]+	330.9	285.0; 127.0	38, 4	6.33
Tau fluvalinate	102851-06-9	502.90	$C_{26}H_{22}CIF_3N_2O_3$	[M+H]+	503.1	208.1; 181.1	15, 25	8.92
Omethoate	1113-02-6	213.20	$C_5H_{12}NO_4PS$	[M+H]+	213.9	182.9; 125.0	4, 16	1.27

Table S3. Validation parameters for the tested pesticides

Pesticide	Concentration range (µg kg⁻¹)	$ m R^2$	LOD (µg kg <sup>-1</sup> )	LOQ (µg kg <sup>-1)</sup>	Spike level (µg kg <sup>-1</sup> )	Repeatability (µg kg <sup>-1</sup> )	RSD <sub>r</sub> (%)	Reproducibility (µg kg <sup>-1</sup> )	RSDwr (%)	Mean recovery (%)
Acetamiprid	2.5-250	0.9996	2.94	3.97	10	10.72-10.69	0.77-0.99	11.18	5.72	111.82
					50	54.26-54.75	0.85-0.74	56.08	5.16	112.16
Dimethoate	2.5-250	0.9993	2.64	2.98	10	10.66-10.59	1.01-0.96	11.13	11.13	111.35
Dimethoate	2.5-250	0.5555	2.04	2.30	50	51.86-52.34	0.89-0.46	51.23	51.23	102.46
Lambda-cyhalothrin	2.5-250	0.9953	2.96	4.04	10	10.19-9.28	11.41-12.18	10.14	10.14	101.39
Lambua-cyriaiouiiiii	2.5-250	0.9955	2.90	4.04	50	50.47-50.83	5.00-8.45	50.46	50.46	100.91
Malathian	2.5.250	0.0004	2.50	0.76	10	10.07-9.74	2.45-3.23	11.37	11.37	113.74
Malathion	2.5-250	0.9984	2.58	2.76	50	49.19-49.79	3.81-1.47	53.44	53.44	106.88
Tou fluvolinoto	au fluvalinate 2.5-250 0.9968 2.	2.75	3.34	10	10.63-10.45	3.39-5.21	10.52	10.52	105.24	
rau fluvalinate		0.9968	2.75	3.34	50	52.50-53.60	1.85-2.71	52.08	52.08	104.16
Omothooto	2.5.250	0.0007	0.56	2.70	10	10.17-10.10	0.84-0.59	10.65	10.65	106.55
Omethoate 2	2.5-250 0.	0.9997	2.56		50	49.35-50.15	0.61-0.72	51.88	51.88	103.76

# Original article (Orijinal araştırma)

# The role of drought in the efficacy of some entomopathogenic nematodes<sup>1</sup>

Bazı entomopatojen nematodların etkinliğinde kuraklığın rolü

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#### Abstract

Entomopathogenic nematodes (EPNs) are endoparasitic organisms commonly used in the control of agricultural pests. There are several factors that determine the efficacy of EPNs on hosts, with one of the most significant being soil moisture. The aim of this study is to determine the effectiveness of some EPNs on hosts at different doses and under different soil moisture conditions. The study utilized 1 Hybrid Strain and 3 EPN isolates, *Heterorhabditis bacteriophora* Poinar, 1976 (Rhabditida: Heterorhabditidae) HBH hybrid strain, *Steinernema carpocapsae* Weiser, 1955 TUR-S4 isolate, and *Steinernema feltiae* Weiser, 1955 (Rhabditida: Steinernematidae) TUR-S3 and S-Bilecik isolates. These species were applied to *Tenebrio molitor* L., 1758 (Coleoptera: Tenebrionidae) larvae at 5, 10, and 15 Infective Juveniles (IJs) doses, under 1, 4, 7, 10, and 13% soil moisture conditions. The study was conducted in 2024 at Bursa Uludağ University, Faculty of Agriculture, Department of Plant Protection, Nematology Laboratory. As a result, the highest mortality rates on *T. molitor larvae* were obtained at 13% soil moisture with 15 IJs, 100% for HBH, 93.33% for TUR-S4, 86.67% for TUR-S3, and 83.33% for S-Bilecik. This study carries important implications for understanding the relationship between EPN efficacy on hosts and soil moisture.

Keywords: Heterorhabditis bacteriophora, soil moisture, Steinernema carpocapsae, Steinernema feltiae

#### Öz

Entomopatojen nematodlar (EPN) tarım zararlıları ile mücadelede yaygın kullanılan endoparazitik organizmalardır. EPN'lerin konukçular üzerinde etkinliğini belirleyen birçok faktör vardır. En önemlilerinden biri ise toprak nemi olarak bilinmektedir. Bu çalışmanın amacı, farklı dozlarda ve farklı toprak nemlerinde EPN'lerin konukçular üzerinde oluşturduğu etkinliği belirlemektir. Çalışmada 1 Hibrit Irk ve 3 EPN izolatı kullanılmıştır. Bunlar, *Heterorhabditis bacteriophora* Poinar, 1976 (Rhabditida: Heterorhabditidae) HBH hibrit Irkı, *Steinernema carpocapsae* Weiser, 1955 TUR-S4 izolatı ve *Steinernema feltiae* Weiser, 1955 (Rhabditida: Steinernematidae) TUR-S3 ve S-Bilecik izolatlarıdır. Bu türler *Tenebrio molitor* L., 1758 (Coleoptera: Tenebrionidae) larvaları üzerine 5, 10 ve 15 Infektif Juvenil (IJ) dozunda, 1, 4, 7, 10 ve 13% toprak neminde uygulanmıştır. Çalışma, 2024 yılında Bursa Uludağ Üniversitesi, Ziraat Fakültesi, Bitki Koruma Bölümü, Nematoloji Laboratuvarında gerçekleştirilmiştir. Sonuç olarak *T. molitor* larvaları üzerinde görülen en yüksek ölüm oranları %13 toprak neminde 15 IJ' de HBH için %100, TUR-S4 için 93.33% TUR-S3 için 86.67% ve S-Bilecik için 83.33% şeklinde elde edilmiştir. Bu çalışma, EPN'nin konukçu üzerindeki etkinliğinin, toprak nemi ile ilişkisinin belirlenmesi açısından önemli sonuçlar taşımaktadır.

Anahtar sözcükler: Heterorhabditis bacteriophora, toprak nemi, Steinernema carpocapsae, Steinernema feltiae

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#### Introduction

In recent years, regulations implemented by the EU in the field of pest control in agriculture have led to restrictions on pesticide usage (Ewence et al., 2015; Robin & Marchand, 2019; Marchand, 2023). This limitation has emphasized the need for alternative methods in pest control, apart from the use of pesticides (Gerhardson, 2002; Baker et al., 2020; Dede et al., 2022; Erdoğan et al., 2023). One of the most significant and commonly employed alternatives is biological control. Entomopathogenic nematodes (EPNs) are widely utilized as biological agents in the agricultural sector for the control of pests (Shapiro-Ilan et al., 2006a; Susurluk & Ehlers, 2008; Dede et al., 2022; Bütüner & Susurluk, 2023; Ulu & Erdoğan, 2023).

Entomopathogenic nematodes are endoparasitic organisms belonging to the Rhabditida order, Steinernematidae and Heterorhabditidae families, requiring a host to complete their life cycle. Their life cycle consists of egg, juvenile 1, juvenile 2, juvenile 3 also called as infective juvenile, juvenile 4, and adult stages (Boemare et al., 1996; Glazer, 1996; Ehlers, 2001; Stuart et al., 2006). The first-generation adults of the Heterorhabditidae family are hermaphrodites, whereas subsequent generations exhibit a distinction between male and female forms. In contrast, in species belonging to the Steinernematidae family, this sexual dimorphism is observed from the first generation (Dix et al., 1992; Stock, 1998; Stock et al., 2002; Lewis et al., 2006).

Entomopathogenic nematodes can only infect their hosts when in the infective juvenile (IJ) stage. IJs enter hosts through natural openings such as the anus and mouth, releasing gram-negative bacteria belonging to the Enterobacteriaceae family, with which they symbiotically live inside their bodies (Stuart et al., 2006; Koppenhöfer et al., 2007; Ruan et al., 2018). This bacterial release causes the host to succumb to septicaemia. Species of the Heterorhabditidae family carry *Photorhabdus* spp. Boemare et al. (Enterobacterales: Enterobacteriaceae) mixed within their hemocoels. On the other hand, species of the Steinernematidae family carry *Xenorhabdus* spp. Thomas & Poinar (Enterobacterales: Enterobacteriaceae) within a specialized pouch in their bodies (Boemare et al., 1996; Susurluk, 2008; Bütüner & Susurluk, 2023; Ulu & Susurluk, 2024).

The underground life and effectiveness of EPNs are influenced by various factors, with soil moisture, temperature, and the physical structure of the soil being among the most crucial (Toth et al., 2022; Ulu & Susurluk, 2024). Among these, soil moisture, in particular, is known to impact the distribution of EPNs in the soil. Furthermore, it plays a significant role in the vitality, reproductive capabilities, and efficacy of these endoparasitic organisms on their hosts (Grant & Villani, 2003; Koppenhöfer & Fuzy, 2007; Salame & Glazer, 2015; Rakubu et al., 2024). Optimal soil moisture levels facilitate the movement of EPNs within the soil, enhancing their effectiveness (Bütüner & Susurluk, 2023; Garba et al., 2024). However, excessively high soil moisture levels, as in saturated soils, can have detrimental effects on the viability and development of EPNs. Therefore, it is recognized that a balanced relationship between EPNs and soil moisture is essential for their successful function and development (Koppenhöfer et al., 2020; Nouh, 2022).

In recent years, the increasing prevalence of drought globally and in our country, as well as the decrease in water usage, is of significant importance in assessing its impact on EPN effectiveness. The aim of this study is to determine the efficacy of different dose (5, 10, and 15 IJs) of EPN species, including *Heterorhabditis bacteriophora* Poinar, 1976 (Rhabditida: Heterorhabditidae) HBH Hybrid Strain, *Steinernema carpocapsae* (Weiser, 1955) (Rhabditida: Steinernematidae) TUR-S4 isolate, *Steinernema feltiae* (Filipjev, 1934) (Rhabditida: Steinernematidae) TUR-S3, and S-Bilecik isolate, on *Tenebrio molitor* L., 1758 (Coleoptera: Tenebrionidae) under varying soil moisture conditions (1, 4, 7, 10, and 13%).

#### **Materials and Methods**

#### Entomopathogenic nematode species

In this study, a hybrid strain and three distinct EPN isolates were performed. The HBH hybrid strain, selected for its high reproductive capacity, resistance to stress conditions, and prolonged durability, belongs to the *H.bacteriophora* species and is associated with the patent number TR 2013 06141 (Susurluk et al., 2013). The isolates used in the study consist of the *S. carpocapsae* TUR-S4 isolate, and *S. feltiae* TUR-S3 and S-Bilecik isolates. In this study, 3<sup>rd</sup> instar larvae (Morales-Ramos et al., 2015) of *T. molitor* were utilized in the experiment.

The *T. molitor* larvae were produced in climate chambers at Bursa Uludağ University, Department of Plant Protection, Nematology Laboratory, under controlled conditions of 26±2°C and a photoperiod of 16-8 (light-dark).

#### **Experimental design**

In the studies, the selected species of EPNs were kept in 60 ml of Ringer's solution (NaCl, KCl, CaCl<sub>2</sub> x 2H<sub>2</sub>O, NaHCO<sub>3</sub>, distilled water) within a 250 ml culture flask with a filter cap, capable of storing around 1000±20 IJs (Bütüner & Susurluk, 2023). The trial was conducted in 24 wells tissue culture plates (3 cm deep x 1.5 cm diameter). Each well was populated with a larva and subsequently filled with soil having different moisture levels of 1, 4, 7, 10, and 13%. Three doses (5, 10, and 15 IJs) of HBH, TUR-S4, TUR-S3, and S-Bilecik were then applied to the soil.

After 3 days, all plates were opened, and the mortality rate of larvae was determined. The identified deceased larvae underwent meticulous dissection and examination for the presence of IJs specific to the species, with the aim of determining whether their demise resulted from the influence the EPNs or not. Ten larvae were used for each soil moisture level and dose, and all assessments were repeated three times. Typically, no separate control group was established in the study, as previous investigations into the efficacy of the EPNs on the host had utilized a soil moisture level of 10% (Langford et al., 2014; Salame & Glazer, 2015; Dede et al., 2022; Nouh, 2022; Bütüner et al., 2023). Therefore, the 10% soil moisture employed in this study represents the control group for the experiment.

#### Statistical analyses

Mortality rates at different soil moisture (1, 4, 7, 10, and 13%) and different doses (5, 10, and 15 IJs) were analysed using one-way analysis of variance (ANOVA) and means were compared using LSD (Least Significant Differences) test (p < 0.05). ANOVA analysis was performed using JMP®Pro 16.0 software.

#### Results

#### Efficiency of HBH on Tenebrio molitor larvae

As a result of the conducted study, the highest mortality rate in the HBH hybrid strain was observed in larvae treated with 15 IJs at 13% soil moisture, reaching a rate of 100% (Table 1).

Table 1. Mortality rate of *Tenebrio molitor* larvae that were treated with HBH Hybrid Strain was analysed separately and statistically at each soil moisture value

EPN Species	Soil moisture (%)	Applied dose (IJs)	Mortality rates (%) ± SE*	F (df); p
		5	10.00 ± 5.77 h	
	1	10	23.33 ± 3.33 h	
		15	46.67 ± 3.33 fg	
		5	56.67 ± 6.77 ef	_
	4	10	56.67 ± 6.77 ef	
		15	60.00 ± 5.77 ef	
		5	56.67 ± 3.33 ef	-
Heterorhabditis bacteriophora HBH Hybrid Strain	7	10	60.00 ± 10.00 ef	F(14,30) = 24,85 p < 0.0001
		15	$70.00 \pm 5.77$ cde	p 10.0001
		5	63.33 ± 3.33 e	_
	10	10	$80.00 \pm 0.00$ bcd	
		15	93.33 ± 3.33 ab	
		5	66.67 ± 3.33 de	_
	13	10	$83.33 \pm 3.33$ bc	
		15	100 ± 3.33 a	

<sup>\*</sup> Means in columns followed by the same letters are not significantly different.

In larvae treated with 5 IJs and 10 IJs at 13% soil moisture, the mortality rates were 66.67 and 83.33%, respectively. At 10% soil moisture, the highest rate was determined in larvae treated with 15 IJs, reaching 93.33%. This rate was 63.33 and 80% for larvae treated with 5 IJs and 10 IJs, respectively. In wells covered with soil with 7% soil moisture, the highest mortality rate was observed in larvae treated with 15 IJs, reaching 70%. For larvae treated with 5 IJs and 10 IJs, the mortality rates were 56.67% and 60%, respectively. At 4% soil moisture, the highest rate was determined as 60% in larvae treated with 15 IJs, the lowest rate was obtained as 56.67% in larvae treated with 5 IJs and 10 IJs. In wells covered with soil with 1% soil moisture, the highest mortality rate was determined as 46.67% in larvae treated with 15 IJs. The lowest rate was observed as 10% in larvae treated with 5 IJs. Statistically significant differences were obtained between nearly all the values (F= 24.85; F= 14, 30; F < 0.0001) (Table 1).

#### Efficiency of TUR-S4 on Tenebrio molitor larvae

Results of the conducted study revealed that in the TUR-S4 isolate, the highest mortality rate was observed in larvae treated with 15 IJs at 10% and 13% soil moisture, reaching 93.33% (Table 2).

Table 2. Mortality rate of *Tenebrio molitor* larvae that were treated with TUR-S4 isolate was analysed separately and statistically at each soil moisture value

EPN Species	Soil moisture (%)	Applied dose (IJs)	Mortality rates (%) ± SE*	F (df); p
		5	10.00 ± 0.00 f	
	1	10	20.00 ± 5.77 f	
		15	40.00 ± 5.77 e	
		5	40.00 ± 11.54 e	_
	4	10	$53.33 \pm 3.33$ de	
		15	$56.67 \pm 3.33 \mathrm{d}$	
		5	56.67 ± 3.33 d	
Steinernema carpocapsae TUR-S4	7	10	56.67 ± 6.67 d	F(14,30) = 20,66; p < 0.0001
		15	$66.67 \pm 3.33  \text{bcd}$	ρ 10.000.
		5	63.33 ± 3.33 cd	_
	10	10	$76.67 \pm 3.33$ bc	
		15	93.33 ± 3.33 a	
		5	66.67 ± 8.81 bcd	_
	13	10	$80.00 \pm 0.00$ ab	
		15	93.33 ± 3.33 a	

<sup>\*</sup> Means in columns followed by the same letters are not significantly different.

At 13% soil moisture, the mortality rates for larvae treated with 5 IJs and 10 IJs were 66.67% and 80%, respectively. At 10% soil moisture, for larvae treated with 5 IJs and 10 IJs, the mortality rates were 63.33% and 76.67%, respectively. In wells covered with soil having 7% soil moisture, the highest mortality rate was observed as high as 66.67% in larvae treated with 15 IJs. For larvae treated with 5 IJs and 10 IJs, the mortality rates were 56.67%. At 4% soil moisture, the highest rate was determined as 56.67% in larvae treated with 15 IJs. The lowest rate was obtained in larvae treated with 5 IJs, as 40%. In wells covered with soil having 1% soil moisture, the highest mortality rate was determined as 40% in larvae treated with 15 IJs. The lowest rate was observed as 10% in larvae treated with 5 IJs. Statistically significant differences were obtained between nearly all the values (F=20.66; df=14, 30; p < 0.0001)

#### Efficiency of TUR-S3 on Tenebrio molitor larvae

The TUR-S3 isolate exhibited the highest mortality rate in larvae treated with 15 IJs at 13% soil moisture, reaching 86.67%. Under the same moisture conditions, the mortality rates for larvae treated with 5 IJs and 10 IJs were 66.67% and 70%, respectively. At 10% soil moisture, the highest rate was determined as 83.33% in larvae treated with 15 IJs. For larvae treated with 5 IJs and 10 IJs, the mortality rates were 66.67%. In wells covered with soil having 7% soil moisture, the highest mortality rate was observed as high as 63.33% in larvae treated with 15 IJs. For larvae treated with 5 IJs and 10 IJs, the mortality rates were 53.33% and 56.67%, respectively. At 4% soil moisture,

the highest rate was determined as 56.67% in larvae treated with 15 IJs. The lowest rate was obtained as 30% in larvae treated with 5 IJs. In wells covered with soil having 1% soil moisture, the highest mortality rate was determined as 26.67% in larvae treated with 15 IJs. The lowest rate was observed as 3.33% in larvae treated with 5 IJs. Statistically significant differences were obtained between nearly all the values (F=14.05; df= 14, 30; p < 0.0001) (Table 3).

Table 3. Mortality rate of *Tenebrio molitor* larvae that were treated with TUR-S3 isolate was analysed separately and statistically at each soil moisture value

EPN Species	Soil moisture (%)	Applied dose (IJs)	Mortality rates (%) ± SE*	F (df); p
Steinernema feltiae TUR-S3		5	3.33 ± 3.33 f	
	1	10	13.33 ± 3.33 ef	
		15	26.67 ± 3.33 e	
		5	30.00 ± 5.77 e	_
	4	10	$50.00 \pm 5.77 \mathrm{d}$	F (14,30) = 14,05; p <0.0001
		15	$56.67 \pm 3.33$ cd	
	7	5	53.33 ± 14.52 cd	
		10	$56.67 \pm 6.67$ cd	
		15	$63.33 \pm 8.82$ cd	
		5	66.67 ± 6.67 bcd	_
	10	10	66.67 ± 8.82 bcd	
		15	83.33 ± 3.33 ab	-
		5	66.67 ± 3.33 bcd	
	13	10	70.00 ± 5.77 abc	
		15	86.67 ± 3.33 a	

<sup>\*</sup> Means in columns followed by the same letters are not significantly different.

#### Efficiency of S-Bilecik on Tenebrio molitor larvae

As a result of the conducted study, the S-Bilecik isolate exhibited the highest mortality rate in larvae treated with 15 IJs under 13% soil moisture, reaching 83.33% (Table 4).

Table 4. Mortality rate of *Tenebrio molitor* larvae that were treated with S-Bilecik isolate was analysed separately and statistically at each soil moisture value

EPN Species	Soil moisture (%)	Applied dose (IJs)	Mortality rates (%) ± SE*	F (df); p
Steinernema feltiae S-Bilecik	1	5	6.67 ± 3.33 g	
		10	16.67 ± 6.67 fg	
		15	26.67 ± 3.33 f	
	4	5	30.00 ± 0.00 f	- F (14,30) = 17,57; p <0.0001
		10	$53.33 \pm 3.33  de$	
		15	$53.33 \pm 3.33  de$	
	7	5	50.00 ± 5.77 e	
		10	$53.33 \pm 8.82 de$	
		15	$60.00 \pm 5.77$ cde	
	10	5	66.67 ± 6.67 bcd	
		10	$63.33 \pm 8.82$ bcde	
		15	76.67 ± 3.33 ab	
	13	5	66.67 ± 6.67 bcd	-
		10	73.33 ± 3.33 abc	
		15	83.33 ± 3.33 a	

<sup>\*</sup> Means in columns followed by the same letters are not significantly different.

Under the same moisture conditions, the mortality rates for larvae treated with 5 IJs and 10 IJs were 66.67% and 73.33%, respectively. At 10% soil moisture, the highest rate was determined as 76.67% in larvae treated with 15 IJs. For larvae treated with 5 IJs and 10 IJs, the mortality rates were 66.67% and 63.33%, respectively. In wells covered with soil having 7% soil moisture, the highest mortality rate was observed as high as 60% in larvae treated with 15 IJs. For larvae treated with 5 IJs and 10 IJs, the mortality rates were 50% and 53.33%, respectively. At 4% soil moisture, the highest rate was determined as 53.33% in larvae treated with 15 IJs. The lowest rate was obtained as 30% in larvae treated with 5 IJs. In wells covered with soil having 1% soil moisture, the highest mortality rate was determined as 26.67% in larvae treated with 15 IJs. The lowest rate was observed as 6.67% in larvae treated with 5 IJs. Statistically significant differences were obtained between the values (F= 17.57; df= 14, 30; p < 0.0001) (Table 4).

## **Discussion**

In this study, the effectiveness of *H. bacteriophora* HBH Hybrid Strain and *S. carpocapsae* TUR-S4, *S. feltiae* TUR-S3, and *S. feltiae* S-Bilecik isolates at different soil moisture levels and doses on larvae of *T. molitor* has been investigated. The results indicate that the EPN isolates lead to higher mortality rates on *T. molitor* with an increase in soil moisture and applied dose. *H. bacteriophora* HBH Hybrid Strain demonstrated a significant increase in the mortality rate as soil moisture and IJs increased, reaching 100% at 13% soil moisture per Petri dish with 15 IJs. Similarly, *S. carpocapsae* TUR-S4, *S. feltiae*, and S-Bilecik isolates exhibited mortality rates of 93.33%, 86.67%, and 83.33%, respectively, at 13% soil moisture. These findings highlight the potential of these EPN isolates in controlling pests at different soil moisture levels and doses.

The effectiveness of the EPN isolates on larvae of *T. molitor* was investigated in relation to soil moisture and IJ dose, utilizing different nematode species, further supporting the findings of this study. For instance, Nouh (2022) investigated the effectiveness of different *Heterorhabditis* spp. at various doses and soil moisture levels (10, 15, and 25%) on *Agrotis ipsilon* (Hufnagel, 1766) (Lepidoptera: Noctuidae) larvae. The study revealed that an increase in soil moisture and applied IJ dose led to a higher mortality rate on larvae. Similarly, in another study conducted by Radová & Trnková (2010), the efficacy of *S. feltiae* and *S. carpocapsae* species applied at different doses was investigated on *T. molitor* larvae at soil moisture levels of 6 and 12.5%. As a result, it was determined that an increase in both dose and moisture led to an elevated mortality rate in larvae. These results seem to be compatible with the present study.

In a study conducted by Grant & Villani (2003), the efficacy of four different EPNs (*H. bacteriophora*, *Steinernema glaseri* (Steiner, 1929), *S. feltiae*, and *S. carpocapsae*) were determined on *Galleria mellonella* (L., 1758) (Lepidoptera: Pyralidae) larvae at various soil moisture levels. As a result, a reduce in soil moisture was associated with a decreased mortality rate in larvae. The other study conducted by Shapiro-Ilan et al. (2006b), the suppressive effects of different isolates of *H. bacteriophora* and *S. carpocapsae* on *Curculio caryae* G.H.Horn, 1873 (Coleoptera: Curculionidae) larvae were evaluated. It was determined that an increase in soil moisture caused an intensified suppression exerted by the EPN species on the larvae. This finding appears to be consistent with the result of the present study.

Furthermore, the structure of agricultural lands varies regionally. Among the factors that undergo changes at this point, soil moisture stands out. The soil moisture, being either too low or too high, significantly influences the efficacy of EPNs on their host insects (Molyneux & Bedding, 1984; Koppenhöfer & Fuzy, 2007; Yadav & Lamramliana, 2012; Ulu & Susurluk, 2014; Bütüner & Susurluk, 2023; Stevens et al., 2023). In their study, Koppenhöfer & Fuzy (2007) investigated the activities of *Steinernema scarabaei* Stock & Koppenhöfer, 2003, *S. glaseri, Heterorhabditis zealandica* Poinar, 1990 and *H. bacteriophora* isolates on *Popillia japonica* Newman, 1838 (Coleoptera: Scarabaeidae) larvae at different soil moisture. As a result, an increase in soil moisture was determined to correspond to an increase in larval mortality rates. However, as the soil moisture content continued to increase, the rate of larval mortality rates eventually decreased. Similarly, in a study conducted by Rohde et al. (2010), the efficacy of EPNs on *Ceratitis capitata* (Wiedemann, 1824) (Diptera: Tephritidae) larvae was determined using different isolates from the Steinernematidae and Heterorhabditidae families under various soil moisture conditions. Consequently, maximum efficacy was observed in EPNs at 75% field capacity. Mortality rates obtained at capacities exceeding or falling below this threshold were notably low. Thus, the moisture requirements of EPN isolates, which have demonstrated adaptation to the climate conditions of Türkiye based on the results obtained from the current study, have been identified.

Additionally, there is a perceived need for further research to determine the soil moisture conditions that contribute to the efficacy of EPNs on hosts, particularly for species adapted to the soil conditions of different countries or regions. Consequently, understanding the correlation between soil moisture and EPN species adapted to our country and regional conditions is crucial, especially during field studies where determining soil moisture levels can aid in identifying the potential efficacy of these species on target hosts and optimizing their impact. Furthermore, this study, conducted on the appropriateness of using EPNs in certain species at drought conditions, will shed light on which types are more suitable for the climatic conditions in our country.

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# Original article (Orijinal araştırma)

# New contributions of the family Eucharitidae (Hymenoptera: Chalcidoidea) of Türkiye, with two new records

Türkiye Eucharitidae (Hymenoptera: Chalcidoidea) familyasına iki yeni kayıtla birlikte yeni katkılar

# Emin KAPLAN<sup>1</sup>\*

#### **Abstract**

In this study, the species of the family Eucharitidae (Hymenoptera: Chalcidoidea) in Türkiye are reviewed. The adults belonging to Eucharitidae were collected from Bingöl and Diyarbakır provinces between 2021 and 2023. Two species, namely *Eucharis* (*Psilogastrellus*) acuminata Ruschka, 1924 and *Eucharis* (*Psilogastrellus*) affinis Bouček, 1956 are recorded for the first time from Türkiye. Separately, photographs of morphological features for the collected species are also provided, along with an identification key and the first checklist for Turkish Eucharitidae family.

Keywords: Checklist, Hymenoptera, Eucharitidae, new records, Türkiye

# Öz

Bu çalışmada Eucharitidae (Hymenoptera: Chalcidoidea) familyasının Türkiye'deki türleri incelenmiştir. Eucharitidae'ye ait erginler 2021 ve 2023 yılları arasında Bingöl ve Diyarbakır illerinden toplanmıştır. *Eucharis* (*Psilogastrellus*) acuminata Ruschka, 1924 ve *Eucharis* (*Psilogastrellus*) affinis Bouček, 1956 olmak üzere iki tür Türkiye'den ilk kez kaydedilmiştir. Ayrıca, toplanan türlerin morfolojik özelliklerinin fotoğrafları da verilmiş, teşhis anahtarı ve Türkiye Eucharitidae familyası için ilk kontrol listesi verilmiştir.

Anahtar sözcükler: Tür kontrol listesi, Hymenoptera, Eucharitidae, yeni kayıtlar, Türkiye

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#### Introduction

Chalcidoidea is the superfamily with the most species diversity in the order Hymenoptera (Heraty et al., 2013). Eucharitidae are a comparatively small group in Chalcidoidea, with approximately 500 species worldwide (Noyes, 2019). Eucharitidae comprise four subfamilies (Akapalinae, Gollumiellinae, Oraseminae, and Eucharitinae) and 66 genera in almost all zoogeographic regions (Noyes, 2019). The family appears to be most diversified and species-rich in tropical regions but comparatively scattered throughout the Palearctic region (Heraty, 2002). The Western Palaearctic fauna consists of 37 species belonging to the genus *Eucharis* Latreille, 1804, five species in the genus *Stilbula* Spinola, 1811, and *Hydrorhoa* Kieffer, 1904 (Lotfalizadeh et al., 2022). In previous studies, a total of 48 *Eucharis* species and 41 *Stilbula* species were listed in the world (Noyes, 2019). There are only a small number of studies of Eucharitidae in Türkiye (Ruschka, 1924; Masi, 1934a, b; Bouček, 1951, 1956; Nikol'skaya, 1952; Tudor, 1971; Doğanlar, 1984). In our fauna, we have seven species of two genera, namely *Eucharis* and *Stilbula* (Noyes, 2019).

All Turkish species belonging to Eucharitidae are rare and they are active only in limited periods of the year. Therefore, studying their biology is complicated (Lotfalizadeh, 2022). Eucharitidae are parasitoids of ants, laying their eggs distantly from the host, either in or on plant tissue (Heraty, 2002; Zhang et al., 2004). Some of the Camponotini parasitoids can have 2-3 pupae per host. The active first-instar larvae interact with foraging ant workers, then these ants transport parasitoid larvae to their nest, where they attack the ant-larvae and develop on their pupae (Heraty, 1994; Heraty & Murray, 2013). Therefore, ants are involved in the evolutionary history of these ant parasitoids (Torréns, 2013; Lachaud & Pérez-Lachaud, 2012; Murray et al., 2013).

In our present work, we discuss two new species for Türkiye's Eucharitidae fauna in details providing check list and key for Eucharitidae species of our country.

## **Materials and Methods**

Samples of Eucharitidae were collected from diverse locations on various flowering plants in Bingöl and Diyarbakır provinces between 2021 and 2023 (Figure 1).

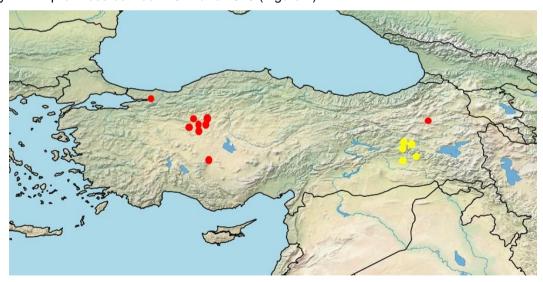


Figure 1. The maps of Eucharitidae collection indicating new localities (Yellow) and previous localities (Red) from Türkiye.

All samples were collected by sweep nets. Specimens were examined with a stereomicroscope and photographed with a digital camera. Morphological terms are based on Gibson et al. (1997) and Heraty (2002). For identification, we consulted the papers and monographs of Gussakovskij (1940), Nikol'skaya

(1952), Bouček (1956), Heraty (2002), Gadallah et al. (2013, 2017), Gadallah & Shairra (2019), and Lotfalizadeh et al. (2022). Terminology for body sculpture follows Harris (1979). The general distribution of the listed species is based mainly on Noyes (2019). The location maps of all samples were created using SimpleMappr (Shorthouse, 2010). Specimens are deposited at the Bingöl University, Faculty of Agriculture, Department of Plant Protection, Bingöl-Türkiye. In addition, an identification key and the first checklist of Turkish Eucharitidae are provided. Valid names are given according to Noyes (2019).

#### Results

In this study, all species of this family in Türkiye are listed, along with two new records.

Order: Hymenoptera L., 1758

Family: Eucharitidae Walker, 1846

Subfamily: Eucharitinae Walker, 1846

Genus: Eucharis Latreille, 1804

Subgenus: Eucharis (Psilogastrellus) Ghesquière, 1946

Eucharis (Psilogastrellus) acuminata Ruschka, 1924 (Figure 2)

Material examined. Diyarbakır, Kulp, Yayıkköyü, 38°15′44.00″N, 41°06′30.41″E, alt. 706 m, 21.V.2023, ♀.

Remarks. This species is the new record for Türkiye.

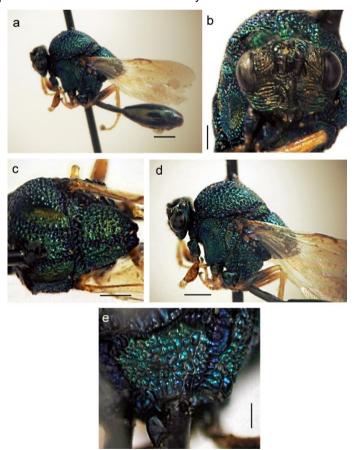


Figure 2. Eucharis (Psilogastrellus) acuminata Ruschka: female; a) lateral view of habitus; b) frontal view of head; c) dorsal view of mesosoma; d) lateral view of mesosoma; e) dorsal view propodeum. Scale bars: 0.5 mm.

## Eucharis (Psilogastrellus) affinis Bouček, 1956 (Figure 3)

Material examined. Bingöl, Çukurca, 38°57'02.16"N, 40°29'27.35"E, alt. 1546 m, 27.V.2021, ♀; Diyarbakır, Kocaköy, Suçıktı, 38°11'32.73"N, 40°32'58.57"E, alt. 743 m, 14.V.2022, ♀; Lice, Zümrüt, 38°28'02.12"N, 40°47'53.65"E, alt. 945 m, 20.V.2022, ♀.

Remarks. This species is the new record for Türkiye.

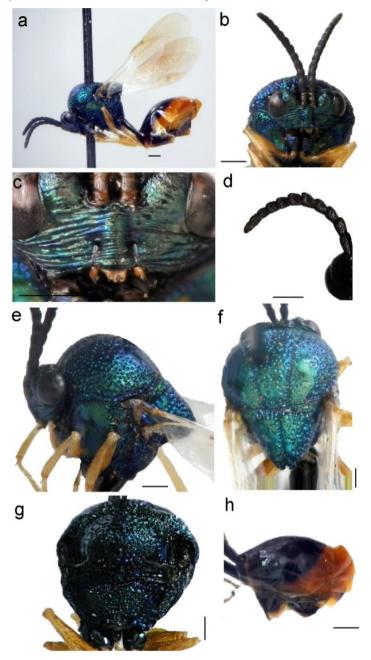


Figure 3. Eucharis (Psilogastrellus) affinis Bouček: female; a) lateral view of habitus; b) frontal view of head; c) clypeus; d) antenna; e) lateral view of mesosoma; f) dorsal view of mesosoma; g) propodeum; h) lateral view of metasoma. Scale bars: 0.5 mm.

# Stilbula cyniformis (Rossi, 1792) (Figure 4)

Material examined. Diyarbakır, Lice, Gürbeyli, 38°26′ 4.31″N, 40°42′48.20″E; alt. 854 m, 21.V.2023, ♀.

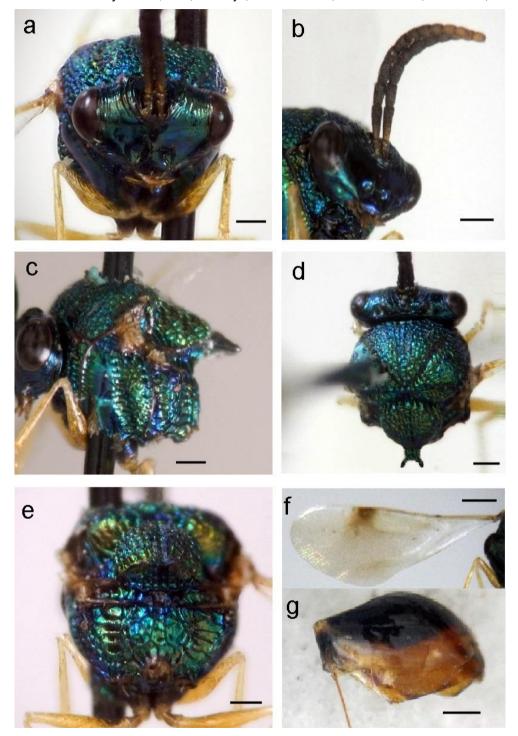


Figure 4. Stilbula cyniformis Rossi: female; a) frontal view of head; b) antenna; c) lateral view of mesosoma; d) dorsal view of mesosoma; e) dorsal view of propodeum; f) fore wing; g) lateral view of metasoma (without petiole). Scale bars: 0.5 mm.

# Key to genera, subgenera and species of Turkish Eucharitidae

	Frons shiny with vertical coarse or rarely fine carinae (Figure 4a). Mesoscutellum with apical projection arising from a single basal stalk that diverges into pair of short spines (Figure 4d)
_	Frons smooth, striate or rugose, lacking vertical carinae (Figure 2b). Mesoscutellum without prominent process, at most with posterior margin slightly emarginate (Figure 2c)
2.	First antennal flagellomere about 3.5× as long as wide (Figure 4b). Mesosoma coarsely areolate-rugose (Figure 4d). Stigmal vein of fore wing enclosed in fuscous cloud (Figure 4f). Metasomal petiole not pale throughout
_	First antennal flagellomere not more than 2.5x as long as wide. Mesosoma more finely punctate. Stigma vein of fore wing hyaline. Metasomal petiole pale throughout
3.	Mandibles without subapical tooth, projecting ventrally from oral margin. Lateral lobe of mesoscutum densely punctate
_	Mandibles with small subapical tooth, opposing and closable. Lateral lobe of mesoscutum sparsely punctate
4.	Mesoscutellum distinctly longer than wide, with deep longitudinal median furrow
_	Mesoscutellum broader than long, with shallow median furrow
5.	Mesoscutellum with two broadly rounded apical lobes, broadly separated by an emargination as broad as each lobe (Figure 2c)
_	Mesoscutellum with shorter, narrowly separated, triangular lobes
- <b>6.</b>	Mesoscutellum with shorter, narrowly separated, triangular lobes
- 6.	Body dull with metasoma brownish green. Frons punctate, lower face tranversely carinate. Mesoscutellum irregularly punctate, and projection broadly rounded posteriorly
_	Body dull with metasoma brownish green. Frons punctate, lower face tranversely carinate. Mesoscutellum irregularly punctate, and projection broadly rounded posteriorly
- 7.	Body dull with metasoma brownish green. Frons punctate, lower face tranversely carinate. Mesoscutellum irregularly punctate, and projection broadly rounded posteriorly
- 7. -	Body dull with metasoma brownish green. Frons punctate, lower face tranversely carinate. Mesoscutellum irregularly punctate, and projection broadly rounded posteriorly

#### Checklist of Turkish species of the family Eucharitidae

Genus Eucharis Latreille, 1804

Subgenus Eucharis (Eucharis) Latreille, 1804

Eucharis (Eucharis) adscendens (Fabricius, 1787)

Distribution in Türkiye. Erzurum (Doğanlar, 1984).

General distribution. Austria, Belarus, Bosnia Hercegovina, Croatia, Çekya, Germany, Hungary, Italy, Kazakhstan, Lithuania, Moldova, Poland, Romania, Russia (Bashkir ASSR, Daghestan ASSR and Perm' Oblast), Slovakia, Türkiye, Ukraine, United Kingdom (Noyes, 2019).

Host. Formica cinerea Mayr, 1853; Formica glauca Ruzsky, 1896 and Formica rufa L., 1761 (Hymenoptera: Formicidae) (Bouček, 1956; Lotfalizadeh, 2008; Szafranski, 2011).

#### Eucharis (Eucharis) reticulata Ruschka, 1924

Distribution in Türkiye. Anatolia (Tudor, 1971).

General distribution. Hungary, Türkiye (Noyes, 2019).

Host. Unknown.

#### Eucharis (Eucharis) turca Nikol'skaya, 1952

Distribution in Türkiye. Anatolia (Nikol'skaya, 1952).

General distribution. Türkiye (Noyes, 2019).

Host. Unknown.

#### Subgenus Eucharis (Psilogastrellus) Ghesquière, 1946

#### Eucharis (Psilogastrellus) acuminata Ruschka, 1924

Distribution in Türkiye. Diyarbakır (in present study).

General distribution. Iran, Russia (Daghestan, ASSR), Saudi Arabia (Noyes, 2019), Türkiye (in present study).

Host. Unknown.

#### Eucharis (Psilogastrellus) anatolica Bouček, 1951

Distribution in Türkiye. Ankara (Bouček, 1951).

General distribution. Türkiye (Noyes, 2019).

Host. Unknown.

#### Eucharis (Psilogastrellus) affinis Bouček, 1956

Distribution in Türkiye. Bingöl, Diyarbakır (in present study).

General distribution. Israel (Bouček, 1956), Saudi Arabia (Gadallah et al., 2014), Türkiye (in present study).

Host. Unknown.

#### Eucharis (Psilogastrellus) punctata Foerster, 1859

Distribution in Türkiye. Ankara, Gaziantep, Konya (Bouček, 1951, 1956; Doğanlar, 1984).

General distribution. Algeria, Austria, Bulgaria, Croatia, Egypt, Macedonia, Saudi Arabia, Tunisia, Türkiye (Noyes, 2019).

Host. Unknown.

#### Genus Stilbula Spinola, 1811

#### Stilbula cyniformis (Rossi, 1792)

Distribution in Türkiye. Ankara, Diyarbakır, Kocaeli (Ruschka, 1924; Bouček, 1951; in present study).

General distribution. Austria, Azerbaijan, Bosnia Hercegovina, China (Hebei, Heilongjiang), Croatia, Çekya, France, Germany, Hungary, Italy (Sicily), Japan, Kazakhstan, Korea, Macedonia, Moldova, Romania, Russia (Adygey AO), Slovakia, Türkiye, Ukraine (Noyes, 2019).

Host. Camponotus sp. Mayr, 1861 (Hymenoptera: Formicidae) (Fahringer & Tölg, 1912).

#### Stilbula vitripennis Masi, 1934

Distribution in Türkiye. Ankara (Masi, 1934a, b; Bouček, 1951).

General distribution. Cyprus, Greece, Israel, Türkiye (Noyes, 2019).

Host. Unknown.

#### **Discussion**

The present study evaluated three eucharitid species collected during field studies conducted between 2021 and 2023 in Diyarbakır and Bingöl provinces in eastern Türkiye. Among these samples, Eucharis (Psilogastrellus) acuminata and Eucharis (Psilogastrellus) affinis (as new records for the Turkish fauna), and Stilbula cyniformis have been identified. The sample of E. acuminata closely matches Bouček's (1956) description that the scutellum ends in two broad rounded protrusions separated by a distance as large as the length of each protrusion (Figure 2c). In addition, E. acuminata species is very similar to E. punctata. However, adults of E. acuminata can be distinguished from E. punctata by the mesoscutellum with a deep, longitudinal central groove, mesoscutellar margin posteriorly bidentate, the body being entirely green (in E. punctata, mesoscutellum, with shallow central furrow; mesoscutellar margin rounded posteriorly, only slightly projecting; the body is dark green, with red-banded metasoma). The species of E. affinis, clypeus almost smooth, metasoma mostly fulvous, apical two-thirds of the wings are grafted in females, so Gadallah et al. (2014) completely fits the definition (Figure 3a, c). The adults of E. affinis is very similar to the Eucharis (Psilogastrellus) albipennis Bouček, 1956 and Eucharis (Pachyeucaris) microcephala Bouček, 1956 except for the shape of antennae, finer sculpture of face, propodeum densely punctuate and shiny, slightly infuscate wings and longer abdominal petiole, body shiny blue and metasoma black and dark brown (in E. albipennis and E. microcephala, rough sculpture of face, propodeum sparsely punctuate and dull, infuscate wings and shorter abdominal petiole, body dark metallic green).

In previously studies, *E. acuminata* was identified by Ruschka (1924) in Iran and Russia (Dagestan, ASSR), and by Gadallah et al. (2019) in Saudi Arabia. Also, *E. affinis* was only recorded in Israel by Bouček (1956), and in Saudi Arabia by Gadallah et al. (2014). Therefore, the detection of this species in Türkiye means its first record in European continent. *E. acuminata* and *E. affinis* species are endemic species that are rarely seen only in certain parts of the world. Therefore, it is very interesting and important that these species have been detected for the first time in Türkiye. The last study on the Turkish Eucharitidae family was conducted by Doğanlar (1984), and no study has been found in the last 40 years. This study has a

special significance in revealing the Eucharitidae fauna in Türkiye. Thus, the number of eucharitid in Türkiye is increased from seven to nine. Also, the identification key and the first checklist for Türkiye Eucharitidae family are provided.

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