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Original article

Biological characteristics of the egg-larval parasitoid *Chelonus oculator* (Fabricius, 1775) (Hymenoptera: Braconidae) on the potato tuber moth *Phthorimaea operculella* (Zeller, 1873) (Lepidoptera: Gelechiidae) at different temperatures

Patates güvesi *Phthorimaea operculella* (Zeller, 1873) (Lepidoptera: Gelechiidae) üzerinde yumurta-larva parazitoiti *Chelonus oculator* (Fabricius, 1775) (Hymenoptera: Braconidae)'nin farklı sıcaklıklarda biyolojik özellikleri

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ABSTRACT

This study was conducted to determine some biological properties of the potato tuber moth *Phthorimaea operculella*, a new host of the egg-larval parasitoid *Chelonus oculator* at different temperatures. The emergence rate, development time, longevity, adult weight, and sex ratio of *C. oculator* were assessed at three different temperature levels (20±1 °C, 25±1 °C, 30±1 °C), 65±5% humidity and 16:8 light: dark conditions. Forty potato tuber moth eggs, aged 0-24 hours, adhered to the filter papers, were placed into the tubes, and presented to the parasitoids for parasitization for a day. At the end of this period, the parasitized eggs were allowed to develop at the specified temperatures. The study found the highest emergence rate was found at 30 °C, with 20.5%. The longest development period occurred at 20 °C, determined as 49.00±2.00 days in females and 52.27±1.64 days in males. The longest longevity of the parasitoid was found to be 41.71±2.29 and 45.73±3.75 days in males and females, respectively. The highest adult weight was observed at 25 °C for both males and females. The sex ratio was found to be in favor of males as the temperature decreased. It is thought that these results can be used for the biological control of potato tuber moth in the field and storage conditions for release studies.

INTRODUCTION

Potato (*Solanum tuberosum* L.) is a crucial agricultural product used in human nutrition worldwide and serves as an industrial plant. Additionally, it is utilized for animal nutrition, while its factory wastes are repurposed as fertilizer.

One of the most important pests affecting potatoes is the potato tuber moth (PTM), *Phthorimaea operculella* (Zeller) (Lepidoptera: Gelechiidae). This pest also targets primarily potatoes and other Solanaceae plants such as tomatoes,

tobacco, eggplant, and various weeds. The primary host of the PTM is the potato, causing substantial losses by feeding on tubers. Adults lay their eggs under the leaves, on shoots and buds, and around the eyes of the tubers during the harvest period. The larva emerging from the egg feeds by opening regular galleries on the leaves and branches, and by opening irregular galleries in the tuber. These galleries, which have a hard surface, are filled with white excrement. In case of infection of the tubers, the loss increases up to 100% if no control is made. Damaged tubers are infected by bacterial and fungal infections so current damage increases more. This pest deteriorates the edibility and seed properties of the potato, resulting in weight and quality loss. The pest that infects the tuber in the field before the harvest continues to reproduce if it finds suitable conditions in the stores and increases its damage (Anonymous 2008).

Survey, biology, and chemical control studies to PTM were carried out in warehouses and laboratory conditions in our country. In the control of PTM, it is desirable to apply primarily cultural control methods (such as earthing up, deep planting, irrigation and early harvest, irrigation and not leaving potato tubers in the field after harvest). However, since these cultural methods are not taken and the irrigation is stopped close to the harvest, the tubers are raised to the surface, and PTM adults lay eggs on these tubers.

In Türkiye, potatoes enter storage contaminated with pests. The main damage of the PTM occurs in storages. Therefore, it is crucial to control the PTM before the potatoes arrive in storage. Chemicals have harmful effects on the ecosystem, so alternative methods should be employed. The most up-to-date and sustainable of these methods is biological control. There have been no biological control application of this pest in our country. In the survey studies, *Bracon (Habrobracon) variegator* Spinola (Hymenoptera: Braconidae), *Temelucha decorata* (Grav) (Hymenoptera: Ichneumonidae) and *Diadegma pulchripes* (Kokujev) were identified as parasitoids of the PTM (Has et al. 1999).

The koinobiont, endoparasitoid, solitary egg-larval parasitoid *Chelonus oculator* (F.) (Hymenoptera: Braconidae) is another natural enemy of the PTM (Özkan et al. 2013). *C. oculator* was first obtained from the *Spodoptera littoralis* Bois. (Lepidoptera: Noctuidae) culture brought from the cotton cultivation areas of Adana province in 1998 (Özmen et al. 2002).

C. oculator lays one egg on the host eggs. It is known that the parasitoid lays its eggs in the egg of the PTM, and it completely consumes the host larva by feeding the first and second larval stages inside the host and the third stage

outside the host larva. In the host diversity studies, it was determined that the PTM is among the natural hosts of the parasitoid. However, little is known about the biology of *C. oculator* on its new host, the PTM (Tunca et al. 2011).

Many studies on the biology of *C. oculator* on different hosts were determined and it was concluded that this parasitoid can be used as an effective biological control agent. However, since the PTM is a new host of the parasitoid, there are gaps in the understanding of the parasitoid's biology on this pest. In this study, the effects of different temperatures on some biological properties of *C. oculator* grown on *P. operculella* were investigated.

MATERIALS AND METHODS

Rearing of insect cultures

Insect cultures used in the study were grown in climate cabinets and climate rooms (25±1 °C, 65±5% RH, and 16:8 h L:D photoperiod). The cultivation of PTM followed the method of Visser (2004) and Maharjan and Jung (2011), with modifications. Potato tubers, collected from the field during harvest, were brought to the laboratory to ensure the emergence of PTM. Potato tubers and adult moths were placed in plastic growing containers (13.5 x 18 cm), and the container was covered with gauze. Adult moths emerging from contaminated potatoes were collected with the help of an aspirator and taken into empty plastic containers for laying eggs, and the containers were covered with gauze. The filter paper was placed on the net, and glass Petri dishes of the same size were placed on the filter papers. Honey was applied to the edges of the plastic containers for feeding the adults. Subsequently, the filter papers containing the host eggs were collected, and the host eggs were used in the experiments.

The parasitoid *C. oculator* population was obtained from the culture grown in the Ankara University Plant Protection Department. 0-24 h *P. operculella* eggs were presented to the parasitoid for 24 hours. Parasitized eggs were transferred to plastic containers containing potato tuber. The emerged adult parasitoids were used for the experiments.

Biological aspects of C. oculator

The trials were carried out in the climatic chamber at three different temperatures (20±1 °C, 25±1 °C, 30±1 °C), with 65±5% RH, 16:8 L:D photoperiod at the Biological Control Laboratory of Plant Protection Central Research Institute in 2018. In the experiments, 0-24 hour parasitoids, fed with honey and mated, were used. Parasitoids were taken into glass tubes and 0-24 hour-old 40 PTM eggs on filter paper,

were presented to parasitoids to parasitize. The parasitized eggs were removed from tubes and placed into containers containing potato tubers, and their development was noted daily. The emergence rate, development time, sex ratio, adult weight, and longevity of the parasitoid were determined. To measure the adult weight, the emerging adult parasitoids were kept in aluminum foil in an oven at 60 °C for five days. The parasitoids' dry weights were measured with the help of analytical balance. Experiments were set up with 10 replications.

Statistical analysis

The analysis was performed in the Minitab 18 package program. The difference between the means was evaluated using one-way ANOVA, with the Tukey test applied within 0.05 error limits (P<0.05).

RESULTS

It was determined that the emergence rate of the parasitoid increased with the increase in temperature, and this increase was statistically found significant (df=2, F=25.02, P=0.000). The emergence rate at 30 °C was found to be higher than at other temperatures (Table 1).

Table 1. Effect of temperature on the emergence rate of the *Chelonus oculator*

Temperature (°C)	Emerged parasitoids (number)		Parasitoid emergence rate (%)
	♂	♀	
20	15	3	4.5 C*
25	31	15	11.5 B
30	57	25	20.5 A

* The difference between the means with different letters in the same column is statistically significant according to the Tukey test (P≤0.05)

Temperature effect on the development time of the parasitoid

It was concluded that temperature significantly affects the development time of both female and male individuals, with the longest development time observed at 20 °C (F_{male}=345.55, P=0.000, df=2; F_{female}=110.01, P=0.000, df=2). Although female individuals developed in a shorter time than male individuals at 20 °C, the difference was not found to be significant (df =2, F₂₀=0.37, P=0.695). The development time of both male and female individuals was found to be at least 25 °C. However, the development of female individuals emerged at 25 °C and 30 °C took longer than male parasitoids and the difference was found to be significant (F₂₅=5.87, P=0.004, df=2; F₃₀=7.92, P=0.001, df=2) (Table 2).

Table 2. Effect of temperature on the development time of *Chelonus oculator*

Temperature (°C)	Development time (days)		
	♂	♀	♂+ ♀
	Mean± SE	Mean± SE	Mean± SE
20	52.27±1.64 A*a**	49.00±2.00 Aa	51.72±1.42 Aa
25	25.71±0.31 Cb	28.00±0.63 Ca	26.46±0.33 Cb
30	29.07±0.39 Bb	31.76±0.39 Ba	29.89±0.33 Bb

* Differences between means with different capital letters in the same column are significant according to the Tukey test (P≤0.05)

**Differences between means with different lowercase letters in the same row are significant according to the Tukey test (P≤0.05)

Temperature effect on the longevity of the parasitoid

The study concluded that temperature affects the longevity of both female and male individuals (F_{male}=84.89, P=0.000, df=2; F_{female}=40.10, P=0.000, df=2). The difference between the longevity of males and females emerging at the same temperature was statistically insignificant (F₂₀=0.91, P=0.413, df=2; F₂₅= 0.46, P=0.634, df=2; F₃₀=2.9, P=0.058, df=2) (Table 3).

Table 3. Effect of temperature on the longevity of *Chelonus oculator*

Temperature (°C)	Longevity (days)		
	♂	♀	♂+ ♀
	Mean± SE	Mean± SE	Mean± SE
20	33.40±4.65 A*a**	18.67±4.71 Ba	30.94±4.13 Ba
25	41.71±2.29 Aa	45.73±3.75 Aa	43.02±1.97 Aa
30	9.26±1.15 Ba	14.28±1.65 Ba	10.79±0.97 Ca

* Differences between means with different capital letters in the same column are significant according to the Tukey test (P≤0.05)

**Differences between means with different lowercase letters in the same row are significant according to the Tukey test (P≤0.05)

Temperature effect on the adult weight of Chelonus oculator

The analysis indicates that temperature significantly affects adult weight in both males and females (df=2, F_{male}=14.16, P=0.000; df=2, F_{female}=21.15, P=0.000). According to the results, the average adult weight was higher at 25 °C. On the other hand, it was determined that the average adult weight of female parasitoids was higher than the average adult weight of male parasitoids at all three temperatures (Table 4).

Table 4. Effect of temperature on adult weight of *Chelonus oculator*

Temperature (°C)	adult weight (mg)		
	♂ Mean± SE	♀ Mean± SE	♂+ ♀ Mean± SE
20	0.89±0.20 B*a**	1.00±0.17 Ba	0.91±0.17 Ba
25	1.25±0.11 Ab	1.79±0.18 Aa	1.43±0.10 Aab
30	0.70±0.03 Ba	0.80±0.06 Ba	0.73±0.03 Ba

* Differences between means with different capital letters in the same column are significant according to the Tukey test ($P \leq 0.05$)

**Differences between means with different lowercase letters in the same row are significant according to the Tukey test ($P \leq 0.05$)

Effect of temperature on the sex ratio of *Chelonus oculator*

In the study, the sex ratio was calculated over total male and female individuals in different temperature applications. As a result of the experiments, the sex ratios of the individuals exiting from different temperatures were respectively (male:female) 5:1; 2.1:1; It was found in favor of male individuals in all three temperature degrees, 2.28:1 (Table 5).

Table 5. Effect of temperature on the sex ratio of parasitoid

Temperature (°C)	♂	♀	♂/ ♀ Male:Female (M:F)
	Number of individuals (pieces)	Number of individuals (pieces)	
20	15	3	5: 1
25	31	15	2.1: 1
30	57	25	2.28: 1

DISCUSSION

In biological control studies, determining the relationship between the parasitoid and the host is essential. It is observed that the increase in temperature leads to an increase in the emergence rate. According to the results, the higher emergence rate at 30 °C than at other temperatures suggests that the parasitoid can adapt more easily to higher temperatures.

In the study, temperature emerged as significant a factor affecting the development time of *C. oculator*. The longest development time was found at the lowest temperature, 20 °C. It was found that the development period of females was longer than males at 25 °C and 30 °C, and shorter at 20 °C,

but the difference was not significant. Different temperatures can influence the development time of *Chelonus species* in different hosts. Rao et al. (1979) reported a development time of 23.5 days for *C. blackburni* in the PTM. The development time of *C. blackburni* on *P. operculella* was found 25.8±1.6 days at 24±2 °C by Kumar and Ballal (1990). The variation may be attributed to the difference in the host nutrient utilization rate of the pre-adult stages of the parasitoid at each temperature. Although it is known that an increase in temperature shortens the development of parasitoids, it is thought that this difference between the temperature and the development time of the parasitoid is related to the compatibility of the parasitoid with the host.

The longevity of *C. oculator* at 30 °C was found to be lower in both male and female individuals compared to the other two temperatures. Medina et al. (1988) obtained that *Chelonus* sp. nr. *curvimaculatus* males live 16.5 days and females live 20 days at 20 °C. Kolaib et al. (1987) investigated the longevity of *Chelonus inanis* (Linnaeus, 1767) (Hymenoptera: Braconidae) at different temperatures (10, 15, and 20 °C). They found that female parasitoids lived 39.5 days and males lived 45.2 days at 10 °C; at 15 °C males lived 36.9 days and females lived 33.2 days; longevity was found 23.5 days for male parasitoids and 19.4 days for females at 20 °C. Qureshi et al. (2016) reported that temperature significantly affects longevity. They stated that the longevity of *C. murakatae* lasts longer at low temperatures. They reported that life expectancy is longer because metabolic activities slow down at low temperatures and that the life span of the female is especially important for the continuation of the generation. Determination of longevity is important in terms of biological control. The longer life of male parasitoids increases the encounter rate with more female individuals, so more females are provided to mate. The long life of female parasitoids means that they encounter more hosts and can parasitize more. When the obtained results are examined, the reason for the decrease in the life span of the parasitoid as the temperature increases can be explained by the increase in the metabolic activities of the parasitoid depending on the temperature. Also, it can be said that the longest longevity is found at 25 °C for both female and male parasitoids, and this temperature is the most suitable temperature for the above-mentioned reasons.

Temperature is a factor affecting adult weight in parasitoids. The adult weight of female parasitoids was found to be higher than that of male individuals at all temperatures tested. It can be said that the most suitable temperature degree in terms of the excess number of eggs in the ovarioles of females is 25 °C. Adult weight can be both an advantage and a disadvantage in

parasitoids. It may mean that female individuals have more eggs in the ovarioles. Also, the heavier the female parasitoid, the more restricted its host-seeking behavior will be. Thus, adult weight can turn into a disadvantage. Yassin Ali (2013) found that temperature increase positively affects the adult weight of *C. oculator*. Kumar and Ballal (1990) reared *C. blackburni* on six laboratory hosts (*Galleria mellonella*, *P. operculella*, *Corycra cephalonica*, *Sitotroga cerealella*, *S. litura*, *Achroia griseella*) and found no difference in terms of adult weight on *C. cephalonica*, *P. operculella* and *A. griseella*.

Another biological feature that is affected by different temperatures is the sex ratio. Özmen (2004) found that *C. oculator* sex ratio on *E. kuehniella* 2.5:1 (male:female) at 25 °C. Tunca (2005) determined the sex ratio of *C. cautella*, the host of *C. oculator*, at 15 °C, 20 °C, 25 °C, and 30 °C and found that males were more common at three temperatures except 15 °C. In our study, the sex ratio was found in favor of males at all three temperatures. In general, a high rate of male parasitoids is an undesirable situation in terms of biological control. Especially considering in terms of mass production, 20 °C is considered to be inappropriate. However, in insects that reproduce with arrhenotoky, such as *C. oculator*, the female individual must mate. The higher the number of males will increase the chance of mating, so it will be advantageous in maintaining the parasitoid generation. On the other hand, the high reproduction of *C. oculator* and the high ratio of male individuals can be considered as an advantage in biological control.

In this study, which was conducted for the first time to determine the biology of *C. oculator* on PTM at three different temperatures, it was observed that *C. oculator* could parasitize this host and develop successfully, despite its first encounter with PTM, and successfully completed its life in emerging adult parasitoids. It is thought that *C. oculator* can suppress the moth population in biological control against PTM, and it should be decided whether it can be used after adjusting the release dose.

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Author's Contributions

Authors declare the contribution of the authors is equal.

Statement of Conflict of Interest

The authors have declared no conflict of interest.

ÖZET

Bu çalışma, farklı sıcaklıklarda yumurta-larva parazitoiti *Chelonus oculator*'un yeni bir konukçusu olan patates güvesi *Phthorimaea operculella* üzerindeki bazı biyolojik özelliklerini belirlemek amacıyla yapılmıştır. Çalışmada 20±1 °C, 25±1 °C, 30±1 °C sıcaklıklarda %65±5 orantılı nem ve 16:8 aydınlık: karanlık ışıklandırma koşullarında *C. oculator*'un çıkış oranı, gelişme ve yaşam süresi, cinsiyet oranı ve ergin ağırlığı belirlenmiştir. Filtre kâğıtlarına yapışık halde bulunan 0-24 saat yaşlı kırk adet patates güvesi yumurtası tüpler içerisine konarak bir gün boyunca parazitlenmesi için parazitoidlere sunulmuştur. Bu sürenin sonunda parazitlenen yumurtalar belirtilen sıcaklıklarda gelişime bırakılmıştır. Çalışmada, en fazla çıkış oranı 30 °C'de %20.5 olarak bulunmuştur. Gelişme süresinin 20 °C'de dişi bireylerde 49.00±2.00 gün, erkek bireylerde 52.27±1.64 gün olduğu bulunmuştur. Ergin ömrü erkek ve dişilerde sırasıyla 41.71±2.29 ve 45.73±3.75 gün olarak saptanmıştır. Ergin ağırlığı en fazla 25 °C'de tespit edilmiştir. Cinsiyet oranının sıcaklık azaldıkça erkekler lehine olduğu belirlenmiştir. Elde edilen bu sonuçların patates güvesinin biyolojik mücadelesinde tarla ve depo koşullarında kullanılabileceği düşünülmektedir.

Anahtar kelimeler: biyolojik mücadele, *Chelonus oculator*, gelişme süresi, çıkış oranı, yeni konukçu

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Original article

New record *Leucodellus zagdani* (Putshkov, 1970) (Hemiptera: Heteroptera: Miridae) and updated checklist of Heteroptera on maize fields in the Central Anatolia Region

İç Anadolu Bölgesi mısır ekim alanlarında yeni kayıt *Leucodellus zagdani* (Putshkov, 1970) (Hemiptera: Heteroptera: Miridae) ve güncellenmiş Heteroptera kontrol listesi

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ABSTRACT

This study was carried out in the maize cultivation areas of Aksaray, Kırşehir, Konya, Karaman, Nevşehir, Niğde, Kayseri, Kırıkkale, Ankara, Eskişehir, Sivas, Yozgat and Çankırı provinces in the Central Anatolia Region between 2017 and 2022. Surveys were carried out in three different phenological periods of maize, according to the simple random sampling method. In each location where the research was conducted, plants on 2-meter rows at 5 points were examined using visual inspection and traps in the first two periods, and using visual inspection and a Japanese umbrella in the 3rd period. Results showed that 36 species, belonging to 25 genera from 10 families (Alydidae, Anthocoridae, Berytidae, Geocoridae, Lygaeidae, Miridae, Nabidae, Pentatomidae, Rhopalidae, Tingidae) were recorded. Among them, *Leucodellus zagdani* (Putshkov, 1970) was the second record in Türkiye, and the first record for maize. *Kalama trimaizeis* (Schrank, 1801) was reported for the third time in the fauna of Türkiye.

INTRODUCTION

Maize (*Zea mays* L., Poaceae: Poales), which has been cultivated for thousands of years, is a warm climate grain that can be cultivated in almost every climate zone in the world, except the Antarctic continent. Among the world's grain production (about 3 billion tons), maize ranks first in terms of production (about 1 billion tons) and second in terms of cultivation area (about 730 million hectares), with

approximately 196 million hectares (Anonymous 2020). Approximately 20% of world maize production is used for human food (direct consumption), 10% for processed food, 10% for other consumption and seed, and 60% for animal feed (Özcan 2009). Thirty-five percent of the maize grown in Türkiye is used in human nutrition, 30% in animal feed, and 20% in the animal feed industry (Gençtan et al. 1995).

Maize, which is grown in almost all regions of Türkiye, ranks third after wheat and barley in terms of production (Anonymous 2014), is cultivated in an area of approximately 12.900 thousand decares, and approximately 34 million tons of product is obtained (Anonymous 2021).

In Türkiye, the increase in irrigated areas in agriculture, the development of animal husbandry, agricultural support policies, etc. for these reasons, maize cultivating areas in the Central Anatolia Region have increased during the last decades and, consequently, entomological pests have raised. Nowadays, there are more than 400 more or less species harmful to maize, which cause damage in different phenological stages of the plant (Şimşek 2004). In the study conducted for this purpose, Heteroptera species found in the maize plant were also identified.

Heteroptera Latreille, 1810 is one of the largest and most diverse groups of insects, with more than 45.254 described species. Among these, 8.354 species in 1.520 genera are recorded in the Palearctic region (Henry 2017). Studies on the Heteroptera in Türkiye indicated that more than 1.500 Heteroptera species have been recorded so far, which represents about 5% of the insect fauna (Tezcan 2020). The present study aims to provide an updated checklist of Heteroptera species, in Türkiye, as well as to refer *Leucodellus zagdani* as a new record for maize.

MATERIALS AND METHODS

Heteroptera specimens were collected during the spring, summer, and autumn of 2017-2022 from Aksaray, Kırşehir, Konya, Karaman, Nevşehir, Niğde, Kayseri, Kırıkkale, Ankara, Eskişehir, Sivas, Yozgat, and Çankırı provinces of the Central Anatolia region. The surveys were conducted in 13 provinces, 77 districts and 304 locations (Figure 1) on a total of 54.720 maize plants in 3 different phenological stages of maize. Surveys were carried out by visual inspection and using a net trap in 5 points of each location in the first two phenologic periods of the maize and both visual inspection

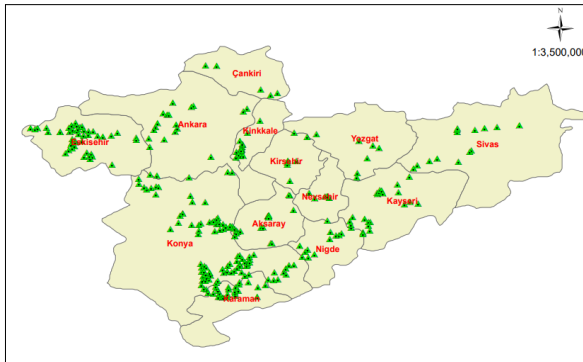


Figure 1. Survey areas of maize fields in the Central Anatolia Region

and using a Japanese umbrella in the 3rd period. Materials were deposited in the Nazife Tuatay Plant Protection Museum (Ankara).

RESULTS

In the study, 36 species, 25 genera and 10 families belonging to the Heteroptera from Türkiye are included. The species were listed below.

HETEROPTERA (HEMIPTERA)

Family ALYDIDAE Amyot and Serville, 1843

Genus: *Alydus* Fabricius, 1803

Alydus calcaratus (Linnaeus, 1758)

Material examined: Ankara, Ayaş 737 m, 40°1'887"N, 32°17'71"E, 2.07.2020, ♀, ♂.

Previous records: Adana, Amasya, Ankara, Artvin, Balıkesir, Bayburt, Çankırı, Çorum, Erzurum, Giresun, İzmir, Kars, Kastamonu, Kayseri, Tokat, Trabzon (Önder et al. 2006, Dursun and Fent 2009, Yıldırım et al. 2011, 2013, Fent and Japoshvili 2012, Küçükbasmacı and Kıyak 2015, Zengin and Dursun 2019, Akman and Dursun 2021, Kıyak and Baş 2021).

Family ANTHOCORIDAE Fieber, 1836

Genus: *Cardiastethus* Fieber, 1860

Cardiastethus nazarenus Reuter, 1884

Material examined: Kayseri, Bünyan, Büyüktuzhisar, 1208 m, 38°57'8"N, 35°50'15"E, 19.09.2019, ♀, ♂, Bünyan, Karatay, 1437 m, 38°38'67"N, 35°56'68"E, 19.09.2019, ♀, ♂, Melikgazi, Yeşilyurt, 1086 m, 38°48'73"N, 35°36'54"E, 19.09.2019, ♀, ♂, Yahyalı, İlyalı, 1081 m, 38°10'82"N, 35°17'91"E, 18.09.2019, ♀, ♂, Yeşilhisar, Musahacılı, 1077 m, 38°12'53"N, 35°17'97"E, 18.09.2019, ♀, ♂.

Previous records: Adıyaman, Antalya, Batman, Diyarbakır, Gaziantep, Hatay, İzmir, Mardin, Mersin, Muğla, Siirt, Şanlıurfa, Şırnak (Hoberlandt 1955, Tuatay et al. 1972, Önder et al. 1983, 2006, Ülgentürk et al. 2013, Yiğit and Telli 2013, Bolu 2019).

Genus: *Orius* Wolff, 1811

Orius (Heterorius) horvathi (Reuter, 1884)

Material examined: Aksaray, Çulfa, 958 m, 38°18'486"N, 33°14'19"E, 07.09.2017, ♀, ♂, Eski, Atarlar, 970 m, 38°13'466"N, 33°23'79"E, 07.09.2017, 2 ♀♀, ♂, Bayramdügün, 1002 m, 07.09.2017, ♀, ♂, Meryemağıl, 961 m, 38°18'339"N, 33°15'945"E, 07.09.2017, ♀, ♂, Ankara, Ayaş, 737 m, 40°01'887"N, 32°17'71"E, 02.09.2020, 2 ♀♀, 2 ♂♂, Gündül,

Güneyce, 735 m, 40°03'25"N, 32°11'60"E, 02.09.2020, ♀, ♂, Polatlı, Oğuzlar, 694 m, 39°47'894"N, 32°02'999"E, 02.09.2020, ♀, ♂, Şereflikoçhisar, Akin, 920 m, 39°08'35"N, 33°16'48"E, 01.09.2020, 2 ♀♀, 2 ♂♂, Şeker, 982 m, 39°08'485"N, 33°11'932"E, 01.09.2020, ♀, ♂, Çankırı, Kızılırmak, Hacılar, 565 m, 40°19'976"N, 33°51'883"E, 06.10.2021, ♀, ♂, Kızılırmak, Tepealagöz, 539 m, 40°22'00"N, 33°58'215"E, 06.10.2021, 2 ♀♀, 2 ♂♂, Eskişehir, Alpu, Aktepe, 830 m, 39°42'590"N, 30°57'508"E, 08.09.2020, 4 ♀♀, 4 ♂♂, Bahçecik, 767 m, 39°48'530"N, 30°52'57"E, 08.09.2020, 5 ♀♀, 4 ♂♂, Beylikova, Akköprü, 743 m, 39°41'445"N, 31°15'482"E, 09.09.2020, ♀, ♂, Çifteler, Emineken, 869 m, 39°22'992"N, 31°5'640"E, 09.09.2020, 4 ♀♀, 4 ♂♂, Mahmudiye, 905 m, 39°26'505"N, 31°00'513"E, 09.09.2020, ♀, ♂, 760 m, 39°47'430"N, 30°57'895"E, 08.09.2020, 4 ♀♀, 2 ♂♂, Odunpazarı, Sarıkavak, 932 m, 37°46'144"N, 34°44'567"E, 08.09.2020, ♀, ♂, Sevinç, 782 m, 39°45'870"N, 30°35'609"E, 08.09.2020, ♀, ♂, Tepebaşı, Beyazaltın-2, 836 m, 39°52'62"N, 30°51'43"E, 07.09.2020, ♀, ♂, Kayseri, Melikgazi, Yeşilyurt, 1086 m, 38°48'73"N, 35°36'54"E, 19.09.2019, ♀, 11 ♂♂, Yeşilhisar, Musahacılı, 1077 m, 38°12'53"N, 35°17'97"E, 18.09.2019, ♀, ♂, Kırıkkale, Çelebi, Karabucak-2, 749 m, 39°30'273"N, 33°24'323"E, 31.08.2020, ♀, ♂, Karakeçili, Akkoşan-1, 780 m, 39°37'562"N, 33°23'197"E, 31.08.2020, 2 ♀♀, 2 ♂♂, Keskin, Köprüköy-1, 750 m, 39°33'848"N, 33°25'989"E, 31.08.2020, ♀, ♂, Nevşehir, Avanos, Alibeyyeri, 917 m, 38°44'31"N, 34°46'48"E, 17.09.2019, ♀, ♂, Derinkuyu, 1362 m, 38°23'27"N, 34°44'70"E, 17.09.2019, ♀, ♂, Sivas, Gemerek, Eskiçubuk, 1153 m, 39°14'651"N, 36°08'625"E, 15.09.2021, ♀, ♂, Yıldızeli, Bakırcıoğlu, 1265 m, 39°49'138"N, 36°46'142"E, 15.09.2021, ♀, ♂.

Previous records: Adana, Adıyaman, Antalya, Diyarbakır, Gaziantep, Mardin, Muş, Şanlıurfa, Siirt (Çelik 1981, Zeren and Düzgüneş 1983, Karaat 1986, Yayla 1984, Akkaya 1995, Göven 1995, Büyük and Özpinar 1999, Bolu et al. 2005, Özgen and Karsavuran 2005, Büyük 2008, Bolu 2019, Pehlivan and Atakan 2020, Kaymak 2022).

Orius (Heterorius) minutus (Linnaeus, 1758)

Material examined: Ankara, Şereflikoçhisar, Şeker, 982 m, 39°08'485"N, 33°11'932"E, 01.09.2020, ♀, ♂, Eskişehir, Alpu, Gökçeoğlu, 852 m, 39°43'829"N, 30°52'866"E, 08.09.2020, ♀, ♂.

Previous records: Adana, Adıyaman, Ankara, Antalya, Artvin, Bartın, Batman, Diyarbakır, Edirne, Erzincan, Erzurum, Gaziantep, Iğdır, Karaman, Kars, Kastamonu, Konya, Mardin, Niğde, Siirt, Şanlıurfa, Tokat, Zonguldak (Hoberlandt 1955, Önder and Adıgüzel 1979, Yaşarakıncı 1991, Çam 1993, Göven 1995, Yıldırım et al. 2013, Yazıcı

2019, Bolu 2019, 2020, Pehlivan and Atakan 2020, Yazıcı 2022c).

Orius (Orius) laevigatus (Fieber, 1860)

Material examined: Ankara, Ayaş, 737 m, 40°01'887"N, 32°17'71"E, 02.09.2020, ♀, ♂, Güneyce, 735 m, 40°03'249"N, 32°11'601"E, 02.09.2020, ♀, ♂, Polatlı, Oğuzlar, 694 m, 39°47'894"N, 32°02'999"E, 02.09.2020, ♀, ♂, Eskişehir, Mihaliççık, Yunusemre, 756 m, 39°45'344"N, 31°29'84"E, 09.09.2020, ♀, ♂, Seyitgazi, Yenikent, 918 m, 39°34'281"N, 30°45'713"E, 09.09.2020, ♀, ♂, Tepebaşı, Keskin, 837 m, 39°49'963"N, 30°22'859"E, 06.07.2020, ♀, ♂, Kırıkkale, Karakeçili, Akkoşan-2, 756 m, 39°28'604"N, 33°24'361"E, 30.06.2020, ♀, ♂.

Previous records: Diyarbakır, Mardin, Şanlıurfa (Önder and Adıgüzel 1979, Bolu 2019).

Orius (Orius) niger (Wolff, 1811)

Material examined: Aksaray, Bozcamahmut, 972 m, 38°12'948"N, 33°21'52"E, 04.07.2017, 2 ♀♀, ♂, Eskil, Atarlar, 970 m, 38°13'466"N, 33°23'793"E, 07.09.2017, ♀, ♂, Eskil, Beşagül, 1005 m, 38°08'930"N, 33°18'547"E, 07.09.2017, ♀, ♂, Böget, 939 m, 38°26'432"N, 33°50'228"E, 06.09.2017, ♀, ♂, Çukuryurt, 962 m, 38°16'769"N, 33°14'933"E, 07.09.2017, ♀, ♂, Çulfa, 958 m, 38°18'486"N, 33°14'19"E, 07.09.2017, ♀, ♂, Filikçitöl, 958 m, 38°15'895"N, 33°18'262"E, 07.09.2017, ♀, ♂, Gümüşdügün, 972 m, 38°14'98"N, 33°19'944"E, 07.09.2017, ♀, ♂, Meryemağıl, 961 m, 38°18'339"N, 33°15'945"E, 07.09.2017, ♀, ♂, Koçaş-2, 950 m, 38°27'425"N, 33°49'294"E, 30.06.2017, 2 ♀♀, ♂, Koçaş, 938 m, 38°26'574"N, 33°53'196"E, 06.09.2017, 2 ♀♀, ♂, Konya, 985 m, 38°11'360"N, 33°19'702"E, 07.09.2017, ♀, ♂, Kökez, 978 m, 38°17'69"N, 33°13'309"E, 07.09.2017, ♀, ♂, Koçaş, 940 m, 38°27'927"N, 33°50'133"E, 06.09.2017, 2 ♀♀, ♂, Ortaköy, Sarıkaraman-1, 992 m, 38°46'837"N, 34°09'989"E, 08.09.2017, ♀, ♂, Ortaköy, Sarıkaraman-1, 992 m, 38°46'837"N, 34°09'989"E, 29.06.2017, ♀, ♂, Yenikent, 919 m, 38°17'202"N, 33°44'743"E, 07.09.2017, ♀, ♂, Yenikent, 919 m, 38°16'888"N, 33°42'960"E, 06.09.2017, 2 ♀♀, ♂, Yeşiltömek, 945 m, 35°28'430"N, 33°50'225"E, 06.09.2017, ♀, ♂, Ankara, Bala, Kesikköprü-1, 770 m, 39°24'723"N, 33°22'200"E, 01.09.2020, ♀, 3 ♂♂, Bala, Tigem-Hacıbekir, 867 m, 39°22'32"N, 33°19'471"E, 01.09.2020, ♀, ♂, Gölbaşı, Karacaören, 1057 m, 39°22'613"N, 32°56'220"E, 01.09.2020, 2 ♀♀, 2 ♂♂, Gündül, Güneyce, 735 m, 40°03'249"N, 32°11'601"E, 02.09.2020, ♀, 2 ♂♂, Haymana, Kerpiçlik, 990 m, 39°03'737"N, 32°35'730"E, 01.09.2020, 2 ♀♀, ♂, Kalecik, 733 m, 40°07'746"N, 33°29'926"E, 01.07.2020, 2 ♀♀, ♂, Polatlı, Kıranharmanı, 690 m, 39°66'266"N, 31°96'912"E, 02.07.2020, ♀, ♂, Oğuzlar, 694 m, 39°47'894"N,

32°02'99"E,02.09.2020, ♀, 2 ♂♂, Şereflikoçhisar, Akin, 920 m, 39°08'35"N, 33°16'48"E, 01.09.2020, ♀, ♂, Çankırı, Germece, 612 m, 40°25'18"N, 33°42'59"E, 06.10.2021, ♀, ♂, Eskişehir, Alpu, Bozan, 783 m, 39°46'51"N, 31°6'43"E, 08.09.2020, 2 ♀♀, ♂, Gökçeoğlu, 852 m, 39°43'82"N, 30°52'86"E, 08.09.2020, ♀, ♂, Beylikova, Yalınli, 740 m, 39°42'28"N, 31°23'37"E, 09.09.2020, 2 ♀♀, ♂, Çifteler, Hayriye, 892 m, 39°22'62"N, 30°57'83"E, 09.09.2020, 3 ♀♀, 2 ♂♂, Mahmutiye, Tokathan, 908 m, 39°36'26"N, 30°49'73"E, 09.09.2020, ♀, ♂, Odunpazarı, Karacahüyük, 780 m, 39°45'87"N, 30°35'60"E, 08.09.2020, ♀, ♂, Sarıkavak, 932 m, 37°46'14"N, 34°44'56"E, 08.09.2020, ♀, 2 ♂♂, Seyitgazi, Çukurağıl, 925 m, 39°30'66"N, 30°46'86"E, 09.09.2020, ♀, ♂, Doğançayır, 915 m, 39°32'56"N, 30°49'44"E, 09.09.2020, ♀, ♂, Yenikent, 918 m, 39°34'28"N, 30°45'71"E, 09.09.2020, ♀, ♂, Sivrihisar, İlören, 688 m, 39°54'13"N, 32°46'54"E, 09.09.2020, ♀, ♂, Tepebaşı, Beyazaltın-2, 836 m, 39°52'62"N, 30°51'43"E, 07.09.2020, ♀, ♂, Karagözler, 814 m, 39°45'90"N, 30°24'52"E, 06.07.2020, ♀, ♂, Kayseri, Develi, Sarca, 1090 m, 38°14'38"N, 35°24'36"E, 18.09.2019, ♀, ♂, Soysallı, 1074 m, 38°21'89"N, 35°22'93"E, 18.09.2019, ♀, ♂, Melikgazi, Yeşilyurt, 1086 m, 38°48'73"N, 35°36'54"E, 19.09.2019, ♀, ♂, Yeşilhisar, 1092 m, 38°26'56"N, 35°8'27"E, 18.09.2019, ♀, ♂, Yeşilhisar, Musahacılı, 1077 m, 38°12'53"N, 35°17'97"E, 18.09.2019, 2 ♀♀, ♂, Kırnkale, Balıseyh, 838 m, 39°56'36"N, 33°42'64"E, 31.08.2020, ♀, ♂, Çelebi, Aliciyeniyapan, 747 m, 39°29'31"N, 33°25'12"E, 30.06.2020, ♀, ♂, Kaldırım-1, 750 m, 39°25'34"N, 33°23'81"E, 31.08.2020, ♀, ♂, Karakeçili, Akkoşan-1, 780 m, 39°37'56"N, 33°23'19"E, 31.08.2020, ♀, ♂, Keskin, Köprükoy-3, 724 m, 39°34'66"N, 33°25'49"E, 31.08.2020, ♀, ♂, Köprükoy-1, 750 m, 39°33'84"N, 33°25'98"E, 31.08.2020, ♀, 2 ♂♂, Kırşehir, Boztepe, Külhöyük, 1147 m, 39°18'81"N, 34°15'85"E, 08.09.2017, ♀, ♂, Konya, Altnekin, Nasuh kuyusu, 977 m, 38°22'68"N, 32°59'33"E, 07.09.2018, ♀, ♂, Oğuzeli, 995 m, 38°20'14"N, 33°07'41"E, 29.05.2018, ♀, 4 ♂♂, Yenikuyu, 984 m, 38°14'91"N, 33°04'61"E, 07.09.2018, ♀, ♂, Yeniayla, 959 m, 38°20'65"N, 32°46'89"E, 10.09.2018, ♀, ♂, Çumra, Güvercinlik, 1002 m, 37°36'36"N, 32°49'77"E, 06.09.2018, ♀, ♂, Okçu-1, 992 m, 37°31'78"N, 32°48'81"E, 06.09.2018, ♀, ♂, Üçhöyük, 988 m, 37°54'98"N, 32°54'91"E, 06.09.2018, ♀, ♂, Ereğli, Çiller, 994 m, 37°38'95"N, 34°06'08"E, 03.09.2018, ♀, ♂, Karapınar, Akkuyu, 983 m, 37°42'90"N, 33°23'98"E, 04.09.2018, ♀, ♂, Küçükaşlama, 992 m, 37°36'52"N, 33°11'46"E, 23.05.2018, ♀, ♂, Nevşehir, Acıgöl, Karapınar, 1301 m, 38°33'22"N, 34°13'10"E, 17.09.2019, ♀, ♂, Avanos, Ayhanlar, 933 m, 38°46'14"N, 34°44'57"E, 17.09.2019, ♀, ♂, Gülşehir, Yeşilöz, 915 m, 38°45'88"N, 34°42'58"E, 17.09.2019, ♀, ♂, Niğde,

Bor, Balcı, 1099 m, 37°56'23"N, 34°26'37"E, 27.06.2019, ♀, ♂, Sivas, Gemerek, Yenidoğan, 1150 m, 39°11'80"N, 36°05'12"E, 15.09.2021, ♀, ♂, Şarkışla, Cemel, 1258 m, 39°21'54"N, 36°27'20"E, 15.09.2021, 2 ♀♀, 2 ♂♂, Yozgat, Boğazlıyan, 1148 m, 39°07'66"N, 35°12'63"E, 08.09.2021, ♀, ♂.

Previous records: Adana, Adıyaman, Ankara, Antalya, Artvin, Bayburt, Batman, Diyarbakır, Edirne, Elazığ, Erzincan, Erzurum, Gaziantep, Iğdır, Karaman, Kars, Kastamonu, Konya, Mardin, Niğde, Siirt, Şanlıurfa (Hoberlandt 1955, Önder and Adıgüzel 1979, Önder et al. 1984, Karaat 1986, Yıldırım et al. 2013, Kaplan 2014, Matocq et al. 2014, Yazıcı 2019, 2022c, Bolu 2019, 2020, Pehlivan and Atakan 2020).

Family: BERYTIDAE Fieber, 1851

Genus: *Berytinus* Kirkaldy, 1900

Berytinus (Lizinus) montivagus (Meyer-Dür, 1841)

Material examined: Karaman, Ayrancı, Böğecik, 1003 m, 38°28'54"N, 33°49'61"E, 26.06.2019, ♀, ♂.

Previous records: Ankara, Aydın, Diyarbakır, İzmir (Hoberlandt 1955, Lodos et al. 1984, Önder et al. 2006, Bolu 2020, Yazıcı 2022b).

Family: GEOCORIDAE Baerensprung, 1860

Genus: *Geocoris* Fallen, 1814

Geocoris (Geocoris) megacephalus (Rossi, 1790)

Material examined: Konya, Karapınar, Yirce, 996 m, 37°33'19"N, 33°31'57"E, 28.06.2018, ♀, ♂.

Previous records: Adana, Adıyaman, Amasya, Antalya, Aydın, Denizli, Diyarbakır, Elazığ, İzmir, Kahramanmaraş, Kayseri, Malatya, Manisa, Mardin, Mersin, Muğla, Nevşehir, Niğde, Siirt, Tekirdağ, Şanlıurfa, Uşak (Hoberlandt 1955, Aysev 1974, Çakır and Önder 1990, Yazıcı et al. 2015, Çerçi and Özgen 2021).

Family: LYGAEIDAE Schilling, 1829

Genus: *Nysius* Dallas, 1852

Nysius cymoides (Spinola, 1837)

Material examined: Ankara, Polatlı, Karapınar, 690 m, 39°38'90"N, 32°12'60"E, 2.07.2020, ♀, ♂, Karaman, Ayrancı, Böğecik, 1003 m, 38°28'54"N, 33°49'61"E, 26.06.2019, 2 ♀♀, ♂, 1117 m, 37°21'24"N, 33°40'2"E, 20.08.2019, ♀, ♂, Bölük yazı, 1047 m, 37°12'21"N, 33°45'8"E, 25.06.2019, ♀, ♂, 27.05.2019, ♀, ♂, Burunoba-2, 999 m, 37°25'98"N, 33°22'87"E, 26.06.2019, ♀, ♂, Kazımkarabekir, Kızılkuyu,

1052 m, 37°20'11"N, 32°49'58"E, 24.06.2019, ♀, ♂, Karaman, İslıhisar, 1011 m, 37°20'48"N, 33°1'59"E, 21.08.2019, ♀, ♂, 25.06.2019, 5 ♀♀, 2 ♂♂, Kaşoba, 1007 m, 37°25'69"N, 33°1'15"E, 25.06.2019, 4 ♀♀, 6 ♂♂, Kılbasan-3, 1004 m, 37°15'8"N, 33°11'66"E, 27.05.2019, ♀, ♂, Mesudiye-1, 1010 m, 37°29'59"N, 33°8'11"E, 25.06.2019, ♀, 3 ♂♂, Ortaoba, 1011 m, 37°27'35"N, 33°2'71"E, 27.05.2019, ♀, ♂, Kayseri, Bünyan, 1424 m, 38°50'1"N, 35°50'99"E, 10.07.2019, ♀, 3 Melikgazi, 19.06.2019, 6 ♀♀, 6 ♂♂, Yeşilyurt, 1086 m, 38°48'73"N, 35°36'54"E, 10.07.2019, ♀, 2 ♂♂, 19.06.2019, ♀, 2 ♂♂, Pınarbaşı, Pazarören, 1437 m, 38°39'1"N, 36°9'44"E, 20.06.2019, 3 ♀♀, 3 ♂♂, Sarioğlan, 1176 m, 39°4'44"N, 36°1'16"E, 20.06.2019, 2 ♀♀, 2 ♂♂, Yahyalı, 19.06.2019, 4 ♀♀, 7 ♂♂, Yeşilhisar, Kılcan, 1100 m, 38°16'88"N, 35°7'2"E, 9.07.2019, ♀, ♂, Nevşehir, Acıgöl, Karapınar, 1301 m, 38°33'22"N, 34°13'10"E, 30.05.2019, ♀, ♂, Nevşehir, Derinkuyu, 1362 m, 38°23'27"N, 34°44'70"E, 28.06.2019, 2 ♀♀, ♂, Gülşehir, Eğrikuyu, 976 m, 38°44'9"N, 34°33'6"E, 28.06.2019, ♀, ♂, Yeşilöz, 915 m, 38°45'88"N, 34°42'58"E, 30.05.2019, ♀, ♂, Yeşilyurt, 935 m, 38°49'6"N, 34°27'76"E, 17.09.2019, ♀, ♂, Niğde, Altunhisar, Yakacık, 1180 m, 37°59'95"N, 34°18'99"E, 29.05.2019, ♀, ♂, Niğde, Bor, Emen, 1074 m, 37°48'91"N, 34°27'7"E, 29.05.2019, ♀, ♂, Konaklı, 1351 m, 38°9'74"N, 34°52'80"E, 27.06.2019, ♀, ♂, Ovacık-1, 1342 m, 38°6'19"N, 34°47'7"E, 27.06.2019, ♀, ♂, Sivas, Çayboyu, 1326 m, 39°47'651"N, 37°2'249"E, 28.06.2021, ♀, ♂, Ulaş, Ekincioglu, 1367 m, 39°28'428"N, 36°59'75"E, 28.06.2021, ♀, ♂, Yozgat, Boğazlıyan, 1148 m, 39°7'661"N, 35°12'636"E, 23.06.2021, ♀, ♂, Sarıkaya, Çıkrıkçı, 1097 m, 39°33'303"N, 35°27'243"E, 8.09.2021, ♀, ♂, Mescitli, 1125 m, 39°31'17"N, 35°33'35"E, 23.06.2021, ♀, ♂, Sorgun, Karaveli, 1088 m, 39°37'540"N, 35°14'319"E, 10.06.2021, ♀, ♂.

Previous records: Adana, Adıyaman, Ankara, Antalya, Aksaray, Artvin, Aydın, Balıkesir, Bayburt, Burdur, Bursa, Çanakkale, Çorum, Denizli, Diyarbakır, Edirne, Elazığ, Erzincan, Erzurum, Eskişehir, Gaziantep, Hatay, Iğdır, Isparta, İstanbul, İzmir, Kahramanmaraş, Karaman, Kars, Kayseri, Kırklareli, Kırşehir, Kilis, Kocaeli, Konya, Nevşehir, Manisa, Mardin, Mersin, Muğla, Niğde, Osmaniye, Sivas, Şanlıurfa, Şırnak, Tekirdağ, Tokat, Yalova, Yozgat, Zonguldak (Aysev 1974, Lodos et al. 1999, Önder et al. 2006, Abacıgil et al. 2010, Matocq et al. 2014, Yazıcı et al. 2015, Çerçi et al. 2018, Yazıcı 2022a, b).

Nysius graminicola graminicola (Kolenati, 1845)

Material examined: Konya, Çumra, Okçu-1, 992 m, 37°31'781"N, 32°48'817"E, 28.06.2018, ♀, ♂, Üçhöyük, 988 m, 37°54'985"N, 32°54'919"E, 6.09.2018, ♀, ♂, Karapınar, Akkuyu, 983 m, 37°42'908"N, 33°23'98"E, 27.06.2018, ♀, ♂.

Previous records: Adana, Afyonkarahisar, Ankara, Artvin, Aydın, Balıkesir, Bayburt, Bilecik, Burdur, Çanakkale, Çorum, Denizli, Edirne, Erzincan, Erzurum, Eskişehir, Gaziantep, Hatay, Isparta, İstanbul, İzmir, Kahramanmaraş, Karaman, Kars, Kastamonu, Kayseri, Kocaeli, Konya, Manisa, Mersin, Muğla, Nevşehir, Şanlıurfa, Tekirdağ, Uşak, Zonguldak (Hoberlandt 1955, Aysev 1974, Önder et al. 2006, Yazıcı et al. 2015, Dursun 2016, Aksu and Çıkman 2019, Sarı and Yıldırım 2021, Yazıcı 2022b).

Nysius senecionis senecionis (Schilling, 1829)

Material examined: Karaman, Ayrancı, Böğecik, 1003 m, 38°28'54"N, 33°49'61"E, 26.06.2019, ♀, ♂, Nevşehir, Derinkuyu, 1362 m, 38°23'27"N, 34°44'70"E, 28.06.2019, ♀, ♂, Niğde, Konaklı, 1351 m, 38°9'74"N, 34°52'80"E, 27.06.2019, ♀, ♂.

Previous records: Adana, Ankara, Artvin, Aydın, Bayburt, Çanakkale, Denizli, Edirne, Erzincan, Erzurum, Hatay, İzmir, Kars, Muğla (Hoberlandt 1955, Lodos et al. 1978, Fent 2011, Yazıcı et al. 2015).

Genus: *Oxycarenus* Fieber, 1837

Oxycarenus (Euoxycarenus) pallens (Herrich-Schäffer, 1850)

Material examined: Ankara, Kahramankazan, Aydın, 862 m, 40°10'89"N, 32°39'927"E, 5.06.2020, ♀, ♂, Eskişehir, Odunpazarı, Sevinç, 782 m, 39°45'870"N, 30°35'609"E, 9.06.2020, ♀, ♂, Sivas, Zara, Şehitler, 1321 m, 39°52'30"N, 37°43'985"E, 14.06.2021, ♀, ♂.

Previous records: Adana, Ankara, Antalya, Balıkesir, Bayburt, Bolu, Çorum, Diyarbakır, Edirne, Elazığ, Erzincan, Erzurum, Gaziantep, Hatay, Iğdır, Isparta, İzmir, Kahramanmaraş, Karabük, Karaman, Kastamonu, Kayseri, Kırkkale, Kırşehir, Kilis, Konya, Nevşehir, Mardin, Mersin, Nevşehir, Niğde, Sinop, Sivas, Zonguldak (Hoberlandt 1955, Tuatay et al. 1972, Aysev 1974, Lodos et al. 1999, Kiyak et al. 2004, Önder et al. 2006, Abacıgil et al. 2010, Matocq et al. 2014, Yazıcı et al. 2015, Dursun 2016, Çerçi and Özgen 2021, Çerçi et al. 2022, Yazıcı 2022b).

Oxycarenus (Oxycarenus) hyalinipennis (A. Costa, 1843)

Material examined: Eskişehir, Alpu, Gökçeoğlu, 852 m, 39°43'829"N, 30°52'866"E, 10.06.2020, ♀, ♂, Kayseri, Pınarbaşı, Pazarören, 1437 m, 38°39'1"N, 36°9'44"E, 20.06.2019, ♀, ♂, Konya, Çumra, İnli, 1055 m, 37°27'419"N, 32°51'15"E, 28.06.2018, ♀, ♂.

Previous records: Adana, Ankara, Antalya, Çanakkale, Gaziantep, Hatay, İstanbul, İzmir, Karaman, Kastamonu, Kilis, Konya, Mersin, Muğla, Niğde, Osmaniye, Sinop

(Hoberlandt 1955, Aysev 1974, Lodos et al. 1999, Şerban 2010, Yazıcı et al. 2015, Yazıcı 2022b).

Genus: *Raglius* Stål, 1872

Raglius alboacuminatus (Goeze, 1778)

Material examined: Kırıkkale, Çelebi, Kaldırım-2, 749 m, 39°26'242"N, 33°23'135"E, 30.06.2020, ♀, ♂.

Previous records: Ankara, Balıkesir, Edirne, Erzincan, Eskişehir, Hatay, İstanbul, Kars, Kayseri, Kahramanmaraş, Muş (Lodos et al. 1999, Önder et al. 2006, Abacıgil et al. 2010, Dursun 2016, Çerçi et al. 2018, Yazıcı 2022b).

Family: MIRIDAE Hahn, 1833

Genus: *Adelphocoris* Reuter, 1896

Adelphocoris lineolatus (Goeze, 1778)

Material examined: Niğde, Bor, Emen, 1074 m, 37°48'91"N, 34°27'7"E, 27.06.2019, ♀, ♂.

Previous records: Adana, Ankara, Artvin, Bayburt, Edirne, Erzincan, Erzurum, Elazığ, Iğdır, Kars, Kayseri, Manisa, Mersin, Niğde, Tokat, Tunceli (Hoberlandt 1955, Yazıcı and Yıldırım 2016a, Çerçi et al. 2018, Yazıcı 2015).

Genus: *Campylomma* Reuter, 1878

Campylomma verbasci (Meyer-Dür, 1843)

Material examined: Karaman, Ayrancı, Saraybüyükburun, 1102 m, 37°12'56"N, 33°39'37"E, 20.08.2019, ♀, ♂.

Previous records: Adana, Ankara, Antalya Aydın, Balıkesir, Bayburt, Bolu, Burdur, Bursa, Çanakkale, Çankırı, Denizli, Diyarbakır, Edirne, Erzincan, Erzurum, Eskişehir, Gaziantep, Hatay, Iğdır, Isparta, İzmir, Kahramanmaraş, Karaman, Kars, Kastamonu, Kayseri, Kırıkkale, Kırşehir, Konya, Mardin, Mersin, Nevşehir, Niğde, Sakarya, Siirt, Yozgat, Zonguldak (Hoberlandt 1955, Önder 1976, Önder et al. 1981, Özkan 1984, Önder et al. 1995, Lodos et al. 2003, Ayyıldız and Atlıhan 2006, Önder et al. 2006, Matocq et al. 2014, Yazıcı 2015, Yazıcı and Yıldırım 2016b).

Genus: *Deraeocoris* Kirschbaum, 1856

Deraeocoris (*Camptobrochis*) *pallens* (Reuter, 1904)

Material examined: Ankara, Bala, Tigem-Hacıbekir, 867 m, 39°22'32"N, 33°19'471"E, 1.07.2020, ♀, ♂, Şereflikoçhisar, Akin, 920 m, 39°8'35"N, 33°16'48"E, 1.09.2020, ♀; Eskişehir, Alpu, Karakamış, 787 m, 39°50'237"N, 30°53'997"E, 8.09.2020, ♀, ♂; Konya, Altınekin, Oğuzeli, 995 m, 38°20'147"N, 33°7'414"E, 2.07.2018, ♀, ♂; Yeniyayla, 959 m, 38°20'654"N, 32°46'898"E, 3.07.2018, ♀, ♂; Karapınar,

Erozyon, 1009 m, 37°42'282"N, 33°30'175"E, 27.06.2018, ♀, ♂; Sekizli Yolu, 995 m, 37°44'427"N, 33°32'179"E, 5.09.2018, ♀, ♂; Sivas, Ulaş, Ekincioglu, 1367 m, 39°28'428"N, 36°59'75"E, 28.06.2021, ♀, ♂.

Previous records: Adana, Antalya, Artvin, Diyarbakır, Gaziantep, Hatay, İzmir, Kahramanmaraş, Kilis, Mardin, Mersin, Muğla, Osmaniye, Şanlıurfa (Hoberlandt 1955, Önder 1976, Özkan 1984, Önder et al. 1995, Atakan 2000, Lodos et al. 2003, Önder et al. 2006, Tezcan et al. 2010, Yazıcı 2015).

Deraeocoris (*Camptobrochis*) *serenus* (Douglas and Scott, 1868)

Material examined: Kayseri, Kocasinan, Kızık, 1079 m, 38°51'75"N, 35°33'69"E, 19.06.2019, ♀, ♂.

Previous records: Adana, Afyon, Ağrı, Ankara, Antalya, Artvin, Aydın, Balıkesir, Bilecik, Bolu, Burdur, Bursa, Çanakkale, Çankırı, Çorum, Denizli, Diyarbakır, Edirne, Elazığ, Erzurum, Eskişehir, Gaziantep, Hakkâri, Hatay, Iğdır, İçel, İstanbul, İzmir, Kahramanmaraş, Karabük, Karaman, Kastamonu, Kayseri, Kırklareli, Kırşehir, Kilis, Kocaeli, Konya, Kütahya, Malatya, Manisa, Mardin, Mersin, Muğla, Nevşehir, Niğde, Sakarya, Sinop, Tekirdağ, Uşak, Van, Yozgat (Hoberlandt 1955, Önder 1976, Önder et al. 1981, Özkan 1984, Önder et al. 1995, Yıldırım et al. 1999, Yaşarakıncı and Hincal 2000, Lodos et al. 2003, Gençer et al. 2004, Ayyıldız and Atlıhan 2006, Önder et al. 2006, Matocq et al. 2014, Küçükbasmacı and Kiyak 2015, Yazıcı 2015, Çerçi and Özgen 2021).

Genus: *Leptopterna* Fieber, 1858

Leptopterna dolabrata (Linnaeus, 1758)

Material examined: Niğde, Edikli, 1358 m, 38°11'55"N, 34°58'4"E, 27.06.2019, ♀, ♂.

Previous records: Adana, Ankara, Bursa, Çankırı, Edirne, Giresun, Kars, Kahramanmaraş, Karabük, Kastamonu, Kocaeli, Zonguldak (Hoberlandt 1955, Önder et al. 2006).

Genus: *Leucodellus* Reuter, 1906

Leucodellus zagdani (Putshkov, 1970)

Material examined: Aksaray, Yenikent, 919 m, 38°16'888"N, 33°42'960"E, 4.07.2017, ♂; Ankara, Polatlı, Oğuzlar, 694 m, 39°47'894"N, 32°2'999"E, 2.07.2020, ♀; Kayseri, Kocasinan, Elagöz, 1061 m, 38°48'96"N, 35°31'3"E, 19.06.2019, ♂; Kırıkkale, Karakeçili, Akkoşan-2, 756 m, 39°28'604"N, 33°24'361"E, 2.06.2020, ♀; Konya, Çumra, Alemdar, 1052 m, 37°39'755"N, 32°47'530"E, 28.06.2018, ♀.

Previous records: Ordu (Çerçi and Koçak 2017).

Note: This species was reported in 2017 by Çerçi and Koçak as a new record for Türkiye from Ordu province. In our study, we are listed as the second record for Türkiye. In addition, *Leucodellus zagdani* is reported as a new record for maize in the study.

Genus: *Lygus* Hahn, 1833

Lygus gemellatus gemellatus (Herrich-Schäffer, 1835)

Material examined: Aksaray, Yenikent, 919 m, 38°17'202"N, 33°44'743"E, 5.07.2017, 2 ♀♀, 2 ♂♂, Eskil, Bozcamağmut, 972 m, 38°12'948"N, 33°21'52"E, 7.09.2017, ♀, ♂, Koçaş, 940 m, 38°27'927"N, 33°50'133"E, 30.06.2017, ♀, ♂, Ankara, Bala, Tigem-Hacıbekir, 867 m, 39°22'32"N, 33°19'471"E, 1.07.2020, ♀, ♂, Eskişehir, Beylikova, Akköprü, 743 m, 39°41'445"N, 31°15'482"E, 10.06.2020, ♀, ♂, Seyitgazi, Doğançayır, 915 m, 39°32'567"N, 30°49'44"E, 9.09.2020, ♀, ♂, Kırşehir, Boztepe, Külhöyük, 1147 m, 39°18'819"N, 34°15'851"E, 3.07.2017, ♀, ♂, Konya, Altınekin, Akıncılar, 990 m, 38°21'922"N, 32°54'918"E, 2.07.2018, ♀, ♂, Dedeler, 958 m, 38°19'222"N, 32°44'821"E, 2.07.2018, ♀, ♂, Cihanbeyli, Sırtağıl, 950 m, 38°24'874"N, 33°1'502"E, 3.07.2018, ♀, ♂, Çumra, Kuzucu, 1055 m, 37°23'609"N, 32°47'212"E, 28.06.2018, ♀, ♂, 1055 m, 37°23'609"N, 32°47'212"E, 6.09.2018, ♀, ♂, Çumra, Uzunkuyu-Feriz, 1002 m, 37°34'960"N, 33°8'213"E, 29.06.2018, ♀, ♂, Üçhöyük, 988 m, 37°54'985"N, 32°54'919"E, 6.09.2018, ♀, ♂, Konya, Karapınar, Yassıca, 1002 m, 37°51'628"N, 33°36'207"E, 28.06.2018, ♀, ♂, Sarayönü, Gözlü, 954 m, 38°28'67"N, 32°27'516"E, 10.07.2018, ♀, ♂, 38°28'67"N, 32°27'516"E, 28.05.2018, ♀, ♂, Sarayönü, Karatepe, 1026 m, 38°21'613"N, 32°25'885"E, 10.07.2018, ♀, ♂, Selçuklu, Kaleköy, 975 m, 38°14'459"N, 32°44'918"E, 10.09.2018, ♀, ♂, 975 m, 38°14'459"N, 32°44'918"E, 3.07.2018, ♀, ♂, Yunak, Yolçatı, 918 m, 38°54'716"N, 32°7'371"E, 9.07.2018, ♀, ♂, Yozgat, Yerköy, Sekili, 692 m, 39°45'311"N, 34°13'836"E, 23.06.2021, ♀, ♂.

Previous records: Adana, Ağrı, Amasya, Ankara, Antalya, Artvin, Bayburt, Bitlis, Bursa, Çanakkale, Denizli, Diyarbakır, Edirne, Elazığ, Erzincan, Erzurum, Eskişehir, Iğdır, İçel, İzmir, Kahramanmaraş, Kars, Kayseri, Konya, Kütahya, Mardin, Mersin, Muş, Nevşehir, Siirt, Tekirdağ, Uşak, Yozgat (Hoberlandt 1955, Önder 1976, Altınayar 1981, Kıyak 1990, Önder et al. 1995, Önder et al. 2006, Fent 2011, Matocq et al. 2014, Yazıcı 2015, Yazıcı and Yıldırım 2016a).

Lygus pratensis (Linnaeus, 1758)

Material examined: Aksaray, Yenikent, 919 m, 38°17'202"N,

33°44'743"E, 5.07.2017, ♀, ♂, Koçaş, 940 m, 38°27'927"N, 33°50'133"E, 30.06.2017, ♀, ♂, Ankara, Gündül, Güneyce, 735 m, 40°3'249"N, 32°11'601"E, 2.07.2020, ♀, ♂, Polatlı, Kiranharmanı, 690 m, 39°66'266"N, 31°96'912"E, 2.07.2020, ♀, ♂, Eskişehir, Alpu, Bozan, 783 m, 39°46'513"N, 31°6'438"E, 7.07.2020, ♀, ♂, Mahmudiye, Fahriye, 897 m, 39°37'66"N, 30°55'768"E, 8.07.2020, ♀, ♂, Kayseri, Bünyan, Köprübaşı, 1433 m, 38°40'95"N, 36°1'82"E, 19.09.2019, ♀, ♂, 1424 m, 38°50'1"N, 35°50'99"E, 10.07.2019, ♀, ♂, Yeşilhisar, Musahacılı, 1077 m, 38°12'53"N, 35°17'97"E, 18.09.2019, ♀, ♂, Konya, Altınekin, Soyhan, 995 m, 38°17'903"N, 33°10'215"E, 29.05.2018, ♀, ♂, Kadınhanı, Altınova, 978 m, 38°48'13"N, 32°4'874"E, 8.06.2018, ♀, ♂, Karapınar, Yirce, 996 m, 37°33'191"N, 33°31'572"E, 5.09.2018, ♀, ♂, Sarayönü, Gözlü-2, 1027 m, 38°29'877"N, 32°29'855"E, 11.09.2018, ♀, ♂, Nevşehir, Gülşehir, Yeşilöz, 915 m, 38°45'88"N, 34°42'58"E, 17.09.2019, ♀, ♂, Niğde, Bor, Kayı, 1082 m, 37°54'62"N, 34°24'19"E, 27.06.2019, 2 ♀♀, ♂, Sivas, Çukurbel, 1284 m, 39°49'387"N, 37°11'352"E, 14.09.2021, ♀, ♂, Ulaş, Karacalar, 1384 m, 39°27'267"N, 36°57'563"E, 14.09.2021, ♀, ♂, Karacalar, 1384 m, 39°27'267"N, 36°57'563"E, 28.06.2021, ♀, ♂, Yıldızeli, Mumcu, 1268 m, 39°47'678"N, 36°45'762"E, 29.06.2021, ♀, ♂.

Previous records: Adana, Adıyaman, Afyonkarahisar, Ağrı, Amasya, Ankara, Antalya, Ardahan, Artvin, Aydın, Balıkesir, Bartın, Bayburt, Bilecik, Bingöl, Bitlis, Bolu, Burdur, Bursa, Çanakkale, Çankırı, Çorum, Denizli, Diyarbakır, Düzce, Edirne, Elazığ, Erzincan, Erzurum; Eskişehir, Gaziantep, Hakkâri, Hatay, Iğdır, Isparta, İstanbul, İzmir, Kahramanmaraş, Karabük, Karaman, Kars, Kastamonu, Kayseri, Kırıkkale, Kırklareli, Kırşehir, Kilis, Kocaeli, Konya, Kütahya, Malatya, Manisa, Mardin, Muğla, Muş, Nevşehir, Niğde, Osmaniye, Sakarya, Samsun, Siirt, Sinop, Şanlıurfa, Trabzon, Tunceli, Uşak, Van, Yozgat, Zonguldak (Hoberlandt 1955, Önder 1976, Altınayar 1981, Önder et al. 1981, Önder et al. 1984, Karaat 1986, Kıyak 1990, Önder et al. 1995, Yaşarakıncı and Hincal 2000, Lodos et al. 2003, Önder et al. 2006, Tezcan et al. 2010, Matocq et al. 2014, Yazıcı 2015, Yazıcı and Yıldırım 2016a, Yazıcı 2022c).

Lygus rugulipennis Poppius, 1911

Material examined: Aksaray, Bozcamağmut, 972 m, 38°12'948"N, 33°21'52"E, 4.07.2017, ♀, ♂, Yenikent, 919 m, 38°17'202"N, 33°44'743"E, 5.07.2017, ♀, ♂, Yenikent, 919 m, 38°17'202"N, 33°44'743"E, 7.09.2017, ♀, 2 ♂♂, Eskil, Böğet, 939 m, 38°26'432"N, 33°50'228"E, 6.09.2017, ♀, ♂, Koçaş-4, 933 m, 38°28'226"N, 33°49'690"E, 30.06.2017, 3 ♀♀, 2 ♂♂, Ortaköy, Sarıkaraman-1, 992 m, 38°46'837"N, 34°9'989"E, 29.06.2017, ♀, ♂, Ortaköy, Sarıkaraman-2, 993 m,

38°46'920"N 34°8'834"E, 29.06.2017, ♀, ♂, Ankara, Gündül, Güneyce, 735 m, 40°3'249"N, 32°11'601"E, 2.07.2020, ♀, ♂, Gökçeoğlu, 852 m, 39°43'829"N, 30°52'866"E, 7.07.2020, ♀, ♂, Eskişehir, Alpu, Yeşildon, 772 m, 39°46'407"N, 31°2'114"E, 7.07.2020, ♀, ♂, Beylikova, Akköprü, 743 m, 39°41'445"N, 31°15'482"E, 10.06.2020, 2 ♀♀, 2 ♂♂, Çifteler, 871 m, 39°23'158"N, 32°2'816"E, 10.06.2020, ♀, ♂, Sakaryabaşı, 877 m, 39°20'544"N, 31°3'451"E, 8.07.2020, ♀, ♂, İnönü, Oklubalı, 820 m, 39°49'454"N, 30°14'588"E, 7.09.2020, ♀, ♂, Odunpazarı, Karacahüyük, 780 m, 39°45'877"N, 30°35'609"E, 9.06.2020, ♀, ♂, Sarıkavak, 932 m, 37°46'144"N, 34°44'567"E, 9.06.2020, ♀, ♂, Karaman, Ayrancı, Saray-Hüyükburun, 1102 m, 37°12'56"N, 33°39'37"E, 26.06.2019, ♀, ♂, Kızık, 1009 m, 37°15'90"N, 33°18'64"E, 28.05.2019, ♀, ♂, Kayseri, Bünyan, Karatay, 1437 m, 38°38'67"N, 35°56'68"E, 10.07.2019, ♀, ♂, Köprübaşı, 1433 m, 38°40'95"N, 36°1'82"E, 20.06.2019, ♀, ♂, Develi, Sindelhöyük, 1107 m, 38°21'93"N, 35°24'80"E, 18.09.2019, 3 ♀♀, 4 ♂♂, Yahyalı, İlyaslı, 1081 m, 38°10'82"N, 35°17'91"E, 18.09.2019, ♀, ♂, Yeşilhisar, Çıtık, 1092 m, 38°19'26"N, 35°6'71"E, 31.05.2019, ♀, ♂, Kırıkkale, Keskin, Köprüköy-2, 767 m, 39°32'107"N, 33°25'320"E, 30.06.2020, ♀, ♂, Kırşehir, Kaman, Bügüz, 831 m, 39°23'683"N, 33°26'394"E, 29.06.2017, ♀, ♂, Güzler, 910 m, 38°21'261"N, 34°81'813"E, 3.07.2017, ♀, ♂, Konya, Altunekin, Akıncılar, 990 m, 38°21'922"N, 32°54'918"E, 2.07.2018, ♀, ♂, Dedeler, 958 m, 38°19'222"N, 32°44'821"E, 5.06.2018, ♀, ♂, Yenikuyu, 984 m, 38°14'917"N, 33°4'613"E, 2.07.2018, ♀, ♂, Cihanbeyli, Böğürdelik, 961 m, 38°46'664"N, 32°37'222"E, 10.07.2018, ♀, ♂, Kavaklı, 960 m, 38°26'58"N, 33°2'739"E, 3.07.2018, ♀, ♂, Çeltik, Gökpinar, 864 m, 39°2'122"N, 31°48'854"E, 30.05.2018, ♀, ♂, Çeltik, Odabaşı, 832 m, 38°54'310"N, 31°53'825"E, 13.09.2018, ♀, 2 ♂♂, Çumra, İnlı, 1055 m, 37°27'419"N, 32°51'15"E, 28.06.2018, ♀, ♂, Çumra, Türkmencamili, 999 m, 37°32'389"N, 32°55'754"E, 29.06.2018, ♀, ♂, Uzunkuyu-Feriz, 1002 m, 37°34'960"N, 33°8'213"E, 29.06.2018, ♀, ♂, Üçhöyük, 988 m, 37°54'985"N, 32°54'919"E, 6.09.2018, ♀, ♂, 998 m, 37°33'590"N, 32°51'174"E, 6.09.2018, ♀, ♂, Ereğli, Melicek, 1014 m, 37°28'538"N, 33°58'581"E, 26.06.2018, ♀, ♂, Sazgeçit, 1010 m, 37°35'451"N, 33°55'4418"E, 3.09.2018, ♀, ♂, Taşağıl, 1005 m, 37°28'108"N, 33°54'690"E, 26.06.2018, ♀, ♂, Kadınhanı, Bakırpinarı, 1058 m, 38°15'746"N, 32°18'448"E, 9.07.2018, ♀, ♂, Karapınar, Akkuyu, 983 m, 37°42'908"N, 33°23'98"E, 27.06.2018, ♀, ♂, Karapınar, Yassıca, 1002 m, 37°51'628"N, 33°36'207"E, 28.06.2018, ♀, ♂, Sarayönü, Başhöyük, 950 m, 38°19'270"N, 32°38'345"E, 5.06.2018, ♀, ♂, Gözli, 954 m, 38°28'67"N, 32°27'516"E, 10.07.2018, ♀, ♂, 1027 m, 38°29'877"N, 32°29'855"E, 28.05.2018, ♀, ♂, 11.09.2018, ♀, ♂, Yunak, Altınöz, 1027 m, 38°29'877"N, 32°29'855"E,

28.05.2018, ♀, ♂, Hacifaklı, 896 m, 38°52'327"N, 31°56'315"E, 13.09.2018, 3 ♀♀, 3 ♂♂, Yolçatu, 918 m, 38°54'716"N, 32°7'371"E, 9.07.2018, ♀, ♂, Niğde, Bor, Aliyer, 1090 m, 37°52'4"N, 34°32'65"E, 27.06.2019, ♀, ♂, Sivas, Altınyayla, Deli İlyas, 1450 m, 39°18'224"N, 36°46'913"E, 14.09.2021, ♀, ♂, Deli İlyas, 1450 m, 39°18'224"N, 36°46'913"E, 29.06.2021, ♀, ♂, Şarkışla, Cemel, 1258 m, 39°21'54"N, 36°27'204"E, 29.06.2021, ♀, ♂, Ulaş, Karacalar, 1384 m, 39°27'267"N, 36°57'563"E, 28.06.2021, ♀, ♂, Sorgun, Karaveli, 1088 m, 39°37'540"N, 35°14'319"E, 10.06.2021, ♀, ♂.

Previous records: Adana, Adapazarı, Adıyaman, Afyonkarahisar, Ağrı, Aksaray, Ankara, Antalya, Artvin, Bartın, Bayburt, Bilecik, Bingöl, Bitlis, Bolu, Burdur, Bursa, Çankırı, Çorum, Denizli, Düzce, Edirne, Elazığ, Erzincan, Erzurum, Eskişehir, Gaziantep, Giresun, Hakkâri, Iğdır, İskenderun, İstanbul, İzmir, Kahramanmaraş, Karabük, Karaman, Kars, Kastamonu, Kayseri, Kırıkkale, Kırklareli, Kırşehir, Kocaeli, Konya, Kütahya, Malatya, Mersin, Muş, Nevşehir, Niğde, Sakarya, Samsun, Sinop, Şanlıurfa, Tekirdağ, Trabzon, Tunceli, Uşak, Van, Yozgat, Zonguldak (Önder 1976, Önder et al. 1981, Önder et al. 1984, Karaat 1986, Özbek and Alaoğlu 1987, Kıyak 1990, Önder et al. 1995, Yıldırım et al. 1999, Lodos et al. 2003, Gençer et al. 2004, Önder et al. 2006, Tezcan et al. 2010, Yazıcı 2015, Yazıcı and Yıldırım 2016a, Çerçi et al. 2018, Yazıcı 2022c).

Family: NABIDAE A. Costa, 1853

Genus: *Nabis* Latreille, 1802

Nabis (Nabis) ferus ferus (Linnaeus, 1758)

Material examined: Ankara, Polatlı, Kiranharmanı, 690 m, 39°66'266"N, 31°96'912"E, 2.07.2020, ♀, ♂, Kayseri, Kocasinan, Salur, 1080 m, 38°51'38"N, 35°35'33"E, 10.07.2019, ♀, ♂, Niğde, Bor, Emen, 1074 m, 37°48'91"N, 34°27'7"E, 27.06.2019, ♀, ♂.

Previous records: Adana, Ağrı, Aksaray, Amasya, Ankara, Antalya, Bursa, Çanakkale, Diyarbakır, Edirne, Elazığ, Erzincan, Erzurum, Eskişehir, Giresun, Iğdır, Isparta, İzmir, Karaman, Kars, Kayseri, Kırklareli, Konya, Mardin, Mersin, Niğde, Şanlıurfa, Tokat, Trabzon (Hoberlandt 1955, Kıyak 1990, Önder et al. 2006, Dursun 2011, Yıldırım et al. 2013, Kaplan 2014, Asal 2015, Yazıcı 2023).

Nabis (Nabis) pseudoferus pseudoferus Remane, 1949

Material examined: Ankara, Ayaş, Tekke, 774 m, 39°52'636"N, 32°23'758"E, 2.07.2020, ♀, ♂, Ankara, Bala, Tigem-Hacıbekir, 867 m, 39°22'32"N, 33°19'471"E, 1.07.2020, 2 ♀♀, 2 ♂♂, 850 m, 39°52'17"N, 32°5'820"E, 2.07.2020, 2 ♀♀, ♂, 850 m, 39°52'17"N, 32°5'820"E, 2.09.2020, ♀, ♂, Polatlı,

Kıranharmanı, 690 m, 39°66'26"N, 31°96'91"E, 2.07.2020, ♀, ♂, Sarioba, 669 m, 39°50'57"N, 32°4'59"E, 2.07.2020, ♀, ♂, Uzunbeyli, 853 m, 39°6'33"N, 31°59'60"E, 2.09.2020, ♀, ♂, Sincan, Malıköy, 737 m, 39°46'116"N, 32°22'831"E, 3.07.2020, ♀, ♂, Şereflikoçhisar, Akin, 920 m, 39°8'35"N, 33°16'48"E, 1.07.2020, ♀, ♂, Eskişehir, Alpu, Gökçeoğlu, 852 m, 39°43'829"N, 30°52'866"E, 7.07.2020, ♀, ♂, Gökçeoğlu, 852 m, 39°43'829"N, 30°52'866"E, 8.09.2020, 2 ♀♀, ♂, Osmaniye, 781 m, 39°50'964"N, 30°56'90"E, 8.09.2020, ♀, ♂, Beylikova, Akköprü, 743 m, 39°41'445"N, 31°15'482"E, 8.07.2020, ♀, ♂, 743 m, 39°41'445"N, 31°15'482"E, 9.09.2020, ♀, ♂, Çifteler, 871 m, 39°23'158"N, 32°2'816"E, 9.09.2020, ♀, ♂, İnönü, Oklubalı, 820 m, 39°49'454"N, 30°14'588"E, 7.09.2020, ♀, ♂, Mahmudiye, 905 m, 39°26'505"N, 31°0'513"E, 9.09.2020, ♀, ♂, Odunpazarı, Karaçay, 770 m, 39°46'753"N, 30°51'8"E, 8.09.2020, ♀, 2 ♂♂, Sarıkavak, 932 m, 37°46'144"N, 34°44'567"E, 8.09.2020, ♀, ♂, Sevinç, 782 m, 39°45'870"N, 30°35'609"E, 8.09.2020, ♀, ♂, Seyitgazi, Çukurağıl, 925 m, 39°30'663"N, 30°46'861"E, 9.09.2020, ♀, ♂, Kalkanlı, 915 m, 39°35'927"N, 30°46'671"E, 9.09.2020, ♀, ♂, Yenikent, 918 m, 39°34'281"N, 30°45'713"E, 9.09.2020, ♀, ♂, Sivrihisar, Gülçayır, 865 m, 39°15'523"N, 31°23'956"E, 9.09.2020, ♀, 2 ♂♂, Ortaklar, 721 m, 39°38'559"N, 31°46'540"E, 10.06.2020, ♀, ♂, Tepebaşı, Çukurhisar, 932 m, 38°46'144"N, 34°44'567"E, 7.09.2020, ♀, ♂, Keskin, 837 m, 39°49'963"N, 30°22'859"E, 6.07.2020, ♀, ♂, Satılmışoğlu, 804 m, 39°48'231"N, 30°120'166"E, 7.09.2020, 2 ♀♀, ♂, Karaman, Ayrancı, Böğecik, 1003 m, 38°28'54"N, 33°49'61"E, 20.08.2019, ♀, ♂, Saray-hüyükburun, 1102 m, 37°12'56"N, 33°39'37"E, 20.08.2019, ♀, 2 ♂♂, Kâzımkarabekir, Kızılkuyu, 1052 m, 37°20'11"N, 32°49'58"E, 24.06.2019, ♀, ♂, 1025 m, 37°15'37"N, 32°58'48"E, 19.08.2019, ♀, ♂, Özyurt, 1056 m, 37°16'42"N, 32°53'65"E, 24.06.2019, ♀, ♂, 1056 m, 37°16'42"N, 32°53'65"E, 19.08.2019, ♀, ♂, Sinci, 1036 m, 37°19'48"N, 32°51'91"E, 24.06.2019, ♀, ♂, Çakırbağ, 1018 m, 37°11'96"N, 33°7'24"E, 20.08.2019, ♀, ♂, Dinek, 1003 m, 37°21'36"N, 33°12'57"E, 20.08.2019, ♀, ♂, 1003 m, 37°21'36"N, 33°12'57"E, 25.06.2019, ♀, ♂, Mesudiye-1, 1010 m, 37°29'59"N, 33°8'11"E, 21.08.2019, ♀, ♂, Salur, 1004 m, 37°17'48"N, 33°18'53"E, 25.06.2019, ♀, ♂, Kayseri, Bünyan, Köprübaşı, 1433 m, 38°40'95"N, 36°1'82"E, 19.09.2019, ♀, 2 ♂♂, 1433 m, 38°40'95"N, 36°1'82"E, 20.06.2019, ♀, ♂, Develi, Soysallı, 1074 m, 38°21'89"N, 35°22'93"E, 18.09.2019, ♀, 2 ♂♂, Kocasinan, Buğdaylı, 1059 m, 38°48'8"N, 35°31'61"E, 19.09.2019, 2 ♀♀, 2 ♂♂, Salur, 1080 m, 38°51'38"N, 35°35'33"E, 10.07.2019, ♀, Yazır, 1063 m, 38°49'93"N, 35°32'86"E, 19.09.2019, ♀, ♂, Melikgazi, Bağpınar, 1106 m, 38°49'9"N, 35°38'14"E, 4.07.2018, ♀, ♂, Yeşilyurt, 1086 m, 38°48'73"N, 35°36'54"E, 19.09.2019, ♀, ♂, Yeşilhisar, Kesik, 1105 m, 38°22'63"N, 35°5'59"E, 9.07.2019, ♀, ♂, Musahacılı,

1077 m, 38°12'53"N, 35°17'97"E, 18.09.2019, ♀, 4 ♂♂, Kırıkkale, Çelebi, Alıcıyeniapan, 747 m, 39°29'318"N, 33°25'121"E, 30.06.2020, ♀, ♂, Kaldırım-1, 750 m, 39°25'344"N, 33°23'812"E, 30.06.2020, ♀, ♂, 31.08.2020, ♀, ♂, Karabucak, 732 m, 39°30'50"N, 33°24'800"E, 31.08.2020, ♀, ♂, Keskin, Köprüköy-2, 767 m, 39°32'107"N, 33°25'320"E, 30.06.2020, ♀, ♂, Köprüköy-3, 724 m, 39°34'660"N, 33°25'491"E, 31.08.2020, ♀, ♂, Nevşehir, Avanos, Ayhanlar, 933 m, 38°46'14"N, 34°44'57"E, 28.06.2019, ♀, ♂, Gülşehir, Eğrikuyu, 976 m, 38°44'9"N, 34°33'6"E, 30.05.2019, ♀, ♂, 17.09.2019, ♀, ♂, Niğde, Bor, Balcı, 1099 m, 37°56'23"N, 34°26'37"E, 22.08.2019, ♀, ♂, Kayı, 1082 m, 37°54'62"N, 34°24'19"E, 22.08.2019, ♀, ♂, Sivas, Yıldızeli, Bakırcıoğlu, 1265 m, 39°49'138"N, 36°46'142"E, 15.09.2021, ♀, ♂, Yozgat, Sarıkaya, Mescitli, 1125 m, 39°31'17"N, 35°33'35"E, 8.09.2021, 2 ♀♀, ♂, Yerköy, 737 m, 39°40'994"N, 34°25'930"E, 23.06.2021, ♀, ♂, 8.09.2021, ♀, ♂.

Previous records: Adana, Adapazarı, Adıyaman, Amasya, Ankara, Artvin, Bartın, Bayburt, Bursa, Çanakkale, Diyarbakır, Düzce, Edirne, Elazığ, Erzincan, Erzurum, Giresun, Iğdır, Isparta, İzmir, Kars, Kastamonu, Kayseri, Konya, Sivas, Şırnak, Tokat, Trabzon (Hoberlandt 1955, Tuatay et al. 1972, Önder et al. 1981, 1983, Alaoğlu and Özbek 1987, Önder et al. 2006, Tezcan et al. 2010, Dursun 2011, Fent 2011, Fent and Japoshvili 2012, Yıldırım et al. 2013, Küçükbasmacı and Kıyak 2015, Asal 2015, Dirik and Kivan 2016, Yazıcı 2023).

Nabis (Nabis) punctatus punctatus A. Costa, 1847

Material examined: Aksaray, Eskil, Bayramdügün, 1002 m, 38°8'938"N, 33°18'551"E, 7.09.2017, ♀, ♂, Aksaray, Yenikent, 919 m, 38°16'888"N, 33°42'960"E, 4.07.2017, ♀, ♂, Ortaköy, Sarıkaraman-2, 993 m, 38°46'920"N, 34°8'834"E, 29.06.2017, ♀, ♂, Kırşehir, Çoğun-1, 1064 m, 39°17'415"N, 34°7'385"E, 3.07.2017, 2 ♀♀, 4 ♂♂.

Previous records: Adıyaman, Afyonkarahisar, Amasya, Ankara, Batman, Burdur, Bursa, Çankırı, Diyarbakır, Edirne, Elazığ, Erzincan, Erzurum, Gaziantep, Giresun, Iğdır, Isparta, İzmir, Karaman, Kayseri, Kırşehir, Konya, Malatya, Mardin, Niğde, Şanlıurfa, Siirt, Sivas, Şanlıurfa, Tokat, Van, Yozgat, Zonguldak (Gençer et al. 2004, Dursun 2011, Yıldırım et al. 2013, Kaplan 2014, Matocq et al. 2014, Asal 2015, Bolu 2020, Yazıcı 2023).

Family: PENTATOMIDAE Leach, 1815

Genus: *Eurydema* Laporte, 1833

Eurydema (Rubrodorsalium) ventralis Kolenati, 1846

Material examined: Konya, Çumra, Kuzucu, 1055 m,

37°23'609"N, 32°47'212"E, 6.09.2018, ♀, ♂, Karapınar, Apak, 977 m, 37°41'512"N, 33°29'384"E, 27.06.2018, ♀, ♂.

Previous records: Adana, Afyon, Ağrı, Amasya, Ankara, Antalya, Artvin, Balıkesir, Bayburt, Bilecik, Bolu, Burdur, Bursa, Çanakkale, Çankırı, Çorum, Denizli, Diyarbakır, Düzce, Edirne, Erzincan, Erzurum, Giresun, Hakkâri, Hatay, Iğdır, Isparta, İstanbul, İzmir, Kahramanmaraş, Karaman, Kastamonu, Kırıkkale, Kırklareli, Kırşehir, Kütahya, Kocaeli, Konya, Manisa, Mersin, Muğla, Muş, Niğde, Sakarya, Sinop, Sivas, Şırnak, Tekirdağ, Tokat, Trabzon, Tunceli, Uşak (Hoberlandt 1955, Lodos et al. 1978, Fent and Aktaş 1999, Önder et al. 2006, Külekçi et al. 2009, Şerban 2010, Dursun and Fent 2011, Fent 2011, Matocq et al. 2014, Yazıcı et al. 2014, Küçükbasmacı and Kıyak 2015, Dirik and Kıvanç 2016, Bulak and Yıldırım 2021, Fent and Dursun 2022).

Genus: *Eysarcoris* Hahn, 1834

Eysarcoris ventralis (Westwood, 1837)

Material examined: Konya, Ereğli, Sazgeçit, 1010 m, 37°35'451"N, 33°55'418"E, 26.06.2018, ♀, ♂, Karapınar, Kurtbasan, 976 m, 37°43'968"N, 33°30'51"E, 27.06.2018, ♀, ♂.

Previous records: Ağrı, Adana, Adıyaman, Amasya, Ankara, Antalya, Artvin, Aydın, Balıkesir, Bartın, Bolu, Burdur, Bursa, Çanakkale, Çorum, Diyarbakır, Düzce, Edirne, Elazığ, Erzincan, Erzurum, Gaziantep, Hatay, Isparta, İçel, İstanbul, İzmir, Kahramanmaraş, Karaman, Kastamonu, Kırklareli, Kocaeli, Konya, Manisa, Mardin, Muğla, Muş, Nevşehir, Niğde, Ordu, Osmaniye, Rize, Sakarya, Samsun, Siirt, Sinop, Sivas, Şanlıurfa, Şırnak, Tekirdağ, Tokat, Trabzon, Uşak, Zonguldak (Lodos et al. 1978, Önder et al. 1981, 1983, 1984, 1995, Kıyak 1990, Fent and Aktaş 1999, Özsaraç and Kıyak 2001, Külekçi et al. 2009, Abacıgil et al. 2010, Şerban 2010, Dursun and Fent 2011, Gözüaçık et al. 2011, Tezcan et al. 2013, Yazıcı et al. 2014, Bulak and Yıldırım 2021, Çerçi and Özgen 2021, Fent and Dursun 2022).

Genus: *Graphosoma* Laporte, 1833

Graphosoma (Graphosoma) italicum italicum (O.F. Müller, 1766)

Material examined: Ankara, Kahramankazan, Aydın, 862 m, 40°10'89"N, 32°39'927"E, 3.07.2020, ♀, ♂.

Previous records: Adana, Adıyaman, Ağrı, Ankara, Antalya, Artvin, Aydın, Balıkesir, Bayburt, Burdur, Bilecik, Bolu, Burdur, Bursa, Çanakkale, Çankırı, Çorum, Denizli, Düzce, Edirne, Erzincan, Erzurum, Eskişehir, Gaziantep, Hatay, Iğdır, Isparta, İstanbul, İzmir, Kahramanmaraş, Karabük, Kars, Kastamonu, Kayseri, Kırklareli, Konya, Kütahya,

Manisa, Mersin, Muğla, Osmaniye, Sakarya, Sinop, Sivas, Tunceli, Yalova, Yozgat, Zonguldak (Hoberlandt 1955, Lodos et al. 1978, Fent and Aktaş 1999, Önder et al. 1995, Özsaraç and Kıyak 2001, Önder et al. 2006, Külekçi et al. 2009, Şerban 2010, Fent 2011, Fent and Japoshvili 2012, Yazıcı et al. 2014, Küçükbasmacı and Kıyak 2015, Fent and Dursun 2022).

Genus: *Peribalus* Mulsant and Rey, 1866

Peribalus (Peribalus) strictus strictus (Fabricius, 1803)

Material examined: Kırıkkale, Çelebi, Alıcıyeniapan, 747 m, 39°29'318"N, 33°25'121"E, 30.06.2020, ♀, ♂, Konya, Karapınar, Hotamış, 995 m, 37°39'112"N, 33°20'729"E, 5.09.2018, ♀, ♂.

Previous records: Adana, Antalya, Aydın, Bayburt, Burdur, Çanakkale, Çorum, Denizli, Diyarbakır, Edirne, Elazığ, Gaziantep, Giresun, Iğdır, İzmir, Karabük, Kastamonu, Manisa, Mardin, Mersin, Muğla, Samsun, Sivas, Tekirdağ, Tokat, Zonguldak (Hoberlandt 1955, Önder et al. 1995, Fent and Aktaş 1999, Awad 2000, Dursun and Kartal 2008, Şerban 2010, Tezcan et al. 2010, 2013, Dursun and Fent 2011, Matocq et al. 2014, Çerçi et al. 2018, Bulak and Yıldırım 2021, Fent and Dursun 2022).

Family: RHOPALIDAE Amyot and Serville, 1843

Genus: *Brachycarenum* Fieber, 1860

Brachycarenum tigrinus (Schilling, 1829)

Material examined: Niğde, Bor, Balcı, 1099 m, 37°56'23"N, 34°26'37"E, 27.06.2019, ♀, ♂.

Previous records: Adana, Adıyaman, Amasya, Ankara, Aydın, Balıkesir, Bayburt, Çanakkale, Çorum, Denizli, Diyarbakır, Edirne, Elazığ, Erzincan, Erzurum, Iğdır, İstanbul, Karabük, Kars, Kastamonu, Kayseri, Kırşehir, Malatya, Nevşehir, Yozgat (Tuatay et al. 1972, Önder et al. 1984, Kıyak et al. 2004, Abacıgil et al. 2010, Şerban 2010, Yıldırım et al. 2011, 2013b, Matocq et al. 2014, Dursun 2016, Çerçi et al. 2018, Fent and Dursun 2019, Zengin and Dursun 2019, Akman and Dursun 2021, Kıyak and Baş 2021, Çerçi and Özgen 2021, Çerçi et al. 2022, Yazıcı 2022b).

Genus: *Drymus* Fieber, 1860

Drymus (Sylvadrymus) brunneus confinis Reuter, 1893

Material examined: Karaman, Kâzımkarabekir, Kızılkuyu, 1052 m, 37°20'11"N, 32°49'58"E, 24.06.2019, ♀, ♂.

Previous records: Adana, Diyarbakır, Erzincan, Erzurum, Hatay, Mardin, Mersin (Matocq et al. 2014, Yazıcı et al. 2015, Bolu 2020).

Genus: *Liorhyssus* Stål, 1870

Liorhyssus hyalinus (Fabricius, 1794)

Material examined: Karaman, Kâzımkarabekir, Sinci, 1036 m, 37°19'48"N, 32°51'91"E, 28.05.2019, ♀, ♂; Kayseri, Develi, Sarıca, 1090 m, 38°14'38"N, 35°24'36"E, 18.09.2019, ♀, ♂.

Previous records: Adana, Adıyaman, Amasya, Ankara, Antalya, Artvin, Batman, Bursa, Çankırı, Çorum, Denizli, Diyarbakır, Elazığ, Erzincan, Erzurum, Gaziantep, Giresun, Hatay, Iğdır, Isparta, İstanbul, İzmir, Kars, Kastamonu, Konya, Mardin, Nevşehir, Ordu, Samsun, Siirt, Sivas, Şanlıurfa, Tokat, Tunceli (Hoberlandt 1955, Kıyak et al. 2004, Dursun and Fent 2009, Yıldırım et al. 2011, 2013, Matocq et al. 2014, Küçükbasmacı and Kıyak 2015, Çerçi et al. 2018, Zengin and Dursun 2019, Bolu 2020, Akman and Dursun 2021, Bulak and Yıldırım 2021, Çerçi and Özgen 2021).

Genus: *Maccevethus* Dallas, 1852

Maccevethus corsicus corsicus Signoret, 1862

Material examined: Konya, Karapınar, Hotamış, 995 m, 37°39'112"N, 33°20'729"E, 5.09.2018, ♀, ♂.

Previous records: Amasya, Çanakkale, Çorum, Erzurum, Giresun, Sivas, Tokat (Dursun and Fent 2009, Yıldırım et al. 2013, Zengin and Dursun 2019, Akman and Dursun 2021).

Genus: *Rhopalus* Schilling, 1827

Rhopalus (Rhopalus) parumpunctatus Schilling, 1829

Material examined: Ankara, Şereflikoçhisar, Akin, 920 m, 39°8'35"N, 33°16'48"E, 1.07.2020, ♀, ♂; Karaman, K.karabekir, Kızılkuyu, 1052 m, 37°20'11"N, 32°49'58"E, 24.06.2019, ♀, ♂.

Previous records: Adana, Adıyaman, Amasya, Ankara, Balıkesir, Batman, Bolu, Çankırı, Çorum, Diyarbakır, Edirne, Elazığ, Erzurum, Gaziantep, Giresun, Kastamonu, Kayseri, Mardin, Nevşehir, Siirt, Sivas, Şanlıurfa, Tokat (Hoberlandt 1955, Lodos et al. 1984, Kıyak et al. 2004, Dursun and Fent 2009, Fent and Japoshvili 2012, Yıldırım et al. 2013, Matocq et al. 2014, Küçükbasmacı and Kıyak 2015, Çerçi et al. 2018, Zengin and Dursun 2019, Bolu 2020, Akman and Dursun 2021, Çerçi and Özgen 2021, Kıyak and Baş 2021).

Family: **TINGIDAE** Laporte, 1832

Genus: *Kalama* Puton, 1876

Kalama trimaizeis (Schrank, 1801)

Material examined: Sivas, Zara, Şehitler, 1321 m, 39°52'30"N, 37°43'985"E, 14.06.2021, ♀, ♂

Previous records: Diyarbakır (Bolu 2020, Maral et al. 2020).

Note: This species is the third record for Türkiye.

DISCUSSION

With the increase in the area under maize cultivation in Türkiye due to the expansion of irrigated areas in agriculture, the development of livestock farming, policies to support agriculture, etc., the area under maize cultivation in the Central Anatolia region has increased day by day, and continues to increase.

Individual studies have been conducted in the Central Anatolia region, which has a wide host range and where the corn cultivation area has increased rapidly in recent years and constitutes 24% of Türkiye's total corn cultivation area (Ayrancı and Sade 2005, Elmalı 1996). Except for the study by Ercan (2006), no study has been realized to date. This study is the first and fundamental for our region, the provinces of the Central Anatolia region.

The research material for this study is 54.720 maize plants in three different phenological stages that were sampled from 304 locations across 77 districts in the provinces of Aksaray, Kırşehir, Konya, Karaman, Nevşehir, Niğde, Kayseri, Kırıkkale, Ankara, Eskişehir, Sivas, Yozgat, and Çankırı in Central Anatolia between 2017 and 2022. The result of the determination of the collected material was 883 samples belonging to 36 species from 25 genera from 10 families, including Alydidae (2 samples), Anthocoridae (302 samples), Berytidae (2 samples), Berytidae (2 samples), Geocoridae (2 samples), Lygaeidae (139 samples), Miridae (243 samples), Nabidae (163 samples), Pentatomidae (14 samples), Rhopalidae (14 samples), Tingidae (2 samples) (Figure 2). Among them, *Leucodellus zagdani* (Putshkov, 1970) is recorded for the second time and *Kalama trimaizeis* (Schrank, 1801) for the third time for the fauna of Türkiye. In addition, *Leucodellus zagdani* is reported as a new record for maize.

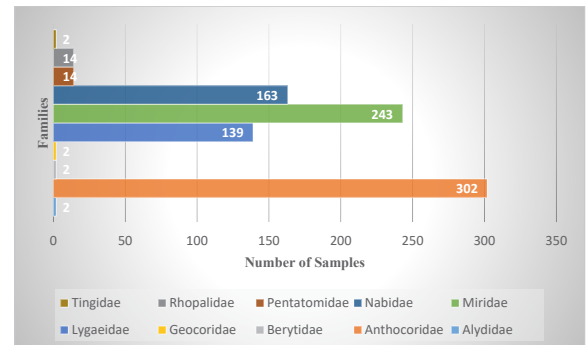


Figure 2. Numbers of families' specimens

Species recorded from Çankırı belong to 10 families (Figure 2). The majority of them belong to the families Miridae (25%, 9 species), Lygaeida (16%, 6 species), Anthocoridae (14%, 5 species), Rhopalidae (14%, 5 species), Pentatomidae (11%, 4 species), and Nabidae (8%, 3 species). Berytidae (3%, 1 sample), Alydidae (3%, 1 sample), Geocoridae (3%, 1 sample), and Tingidae (3%, 1 species) were underrepresented in the Heteroptera fauna of the Central Anatolia region when compared to the fauna of Türkiye (Figure 3). The underrepresentation of these families is probably due to the methods that we used to collect specimens.

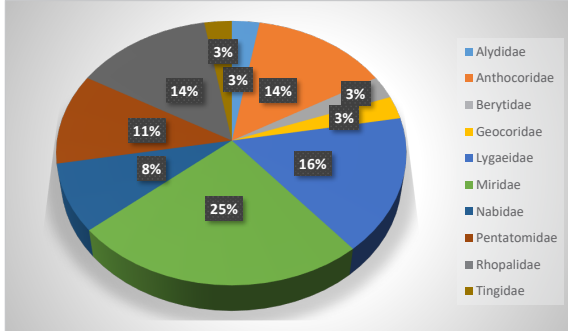


Figure 3. The percentage number of species recorded from the Central Anatolia Region sorted into families

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Author's Contributions

Authors declare the contribution of the authors is equal.

Statement of Conflict of Interest

The authors have declared no conflict of interest.

ÖZET

Bu çalışma, 2017-2022 yılları arasında İç Anadolu Bölgesi'nde yer alan Aksaray, Kırşehir, Konya, Karaman, Nevşehir, Niğde, Kayseri, Kırıkkale, Ankara, Eskisehir, Sivas, Yozgat ve Çankırı illerinin mısır ekim alanlarında yürütülmüştür. Araştırmalar basit tesadüfi örnekleme yöntemine göre mısırın üç farklı fenolojik döneminde gerçekleştirilmiştir. Araştırmanın yapıldığı her lokasyonda 5 noktada 2 metrelik sıralar üzerinde bulunan bitkiler, ilk iki dönemde görsel inceleme ve tuzak kullanılarak, 3. dönemde ise görsel inceleme ve Japon şemsiyesi kullanılarak sürveyler yapılmıştır. Bu çalışmada 10

familyaya bağlı (Alydidae, Anthocoridae, Berytidae, Geocoridae, Lygaeidae, Miridae, Nabidae, Pentatomidae, Rhopalidae, Tingidae) 25 cinsine ait 36 tür kaydedilmiştir. Bunlardan *Leucodellus zagdani* (Putshkov, 1970) Türkiye faunası için ikinci kez ve mısır için yeni kayıt, *Kalama trimaizeis* (Schrank, 1801) ise üçüncü kez kaydedilmiştir.

Anahtar kelimeler: mısır, sürvey, Heteroptera, yeni kayıt, İç Anadolu Bölgesi, Türkiye

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Original article

Morphological and molecular identification of plant parasitic nematodes in wheat fields of Eastern Anatolian Region (Türkiye)

Doğu Anadolu Bölgesi (Türkiye) buğday alanlarında bitki paraziti nematodların morfolojik ve moleküler teşhisi

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ABSTRACT

The aim of this study is to identify plant-parasitic nematodes molecularly and morphologically in wheat (*Triticum* spp.) fields in the Eastern Anatolia Region (Türkiye) between 2017-2019. For this purpose, a total of 258 soil samples were collected from 7 provinces (Erzincan, Elazığ, Erzurum, Iğdır, Kars, Malatya, and Sivas) in the Eastern Anatolia Region. Nematodes were morphologically identified using a light microscope at the genus (some of them species) level. DNA extraction was performed and PCR products were used to DNA sequencing and nucleotide analysis for 28S ribosomal DNA region by comparing the results with the database. According to the obtained data, a total of 20 genera and 7 species belonging to 2 orders and 9 families were identified: *H. digonicus* Perry, 1959; *H. canadensis* Waseem, 1961; *H. vulgaris* Yuen, 1964 (Nematoda: Hoplolaimidae); *Ditylenchus myceliophagus* Goodey, 1958 (Nematoda: Anguinidae); *Amplimerlinius macrurus* (Goodey, 1932) (Nematoda: Dolichodoridae); *Scutylenchus quadrifer* (Andrassy, 1954); (Nematoda: Dolichodoridae), and *Pratylenchoides alkani* Yüksel, 1977 (Nematoda: Pratylenchidae). *H. vulgaris* was identified at the species level using molecular techniques. The rates of presence of economically important plant parasitic nematodes were determined as 73%, 43%, 36%, 33% and 28% for *Ditylenchus* spp., *Pratylenchus* spp., *Aphelenchus* spp., *Xiphinema* spp. and *Helicotylenchus* spp. respectively. It is thought that the results obtained will help to plan nematode control methods in the region.

INTRODUCTION

Türkiye's wheat, *Triticum* L. (Poales: Poaceae) production in 2021 was 17 million tons. (TUIK 2021). Approximately 10% of the wheat cultivated areas that are great importance

in Türkiye's agriculture are located in the Eastern Anatolian Region. It is reported that plant parasitic nematodes cause a 10% loss in wheat production worldwide (Bongers and

Ferris 1999, Gaugler and Bilgrami 2004, Nicol et al. 2011, Sasser and Freckman 1987). There are 25.043 species identified in Nematoda phylum (Zhi-Qiang 2013) and 4305 plant parasitic nematodes species were identified (Maggenti 1991). In 48 distinct regions of Türkiye, 240 plant parasitic nematodes have been identified on 66 plant host species (Kepenekçi 2014). Plant parasitic nematodes can live in diverse habitats. The majority of nematodes cause damage to the root system of a plant and fewer nematodes cause damage to above-ground parts such as leaves and flowers (Nicol 2002). Nematodes create symptoms that resemble nutrient deficiencies in various parts of the plant. The feeding of plant parasitic nematodes results in a significant reduction in a plants root density and as a result, the plant turns yellow and becomes dwarf. Therefore, the areas with very serious nematode infection in the field are observed in distinct or visible patches (Kort 1972, Lung 1992).

Nematode species that cause economic losses in wheat cultivated areas worldwide. Although there are many plant parasitic nematode species that cause yield losses in wheat, the main species are cereal cyst nematodes (CCN), root lesion nematodes (RLN), root-knot nematodes (RKN), wheat gal nematode, and the stem-bulb nematode (Nicol et al. 2002). Although many studies have been carried out on plant parasitic nematodes damaging different hosts in different regions of Türkiye, studies in Eastern Anatolia are very limited. In the Eastern Mediterranean Region, *Geocnamus brevidens* and *Pratylenchus thornei* were identified as the most common of the 9 nematode species in the Eastern Mediterranean Region (Elekcioglu 1996). In another study, *Heterodera ciceri*, *Pratylenchoides erzurumensis*, *Pratylenchoides leiocauda* and *Pratylenchus mediterraneus*, *P. penetrans*, *P. thornei* species were reported in chickpea and lentils in Türkiye (Di Vito et al. 1994). In a survey of plant parasitic nematodes associated with chickpea conducted in Türkiye, *Ditylenchus dipsaci*, *Pratylenchus neglectus*, *P. penetrans* and *P. thornei*, were found to be the most common plant parasitic nematodes (Behmand et al. 2019). In a study carried out by İmren (2007), nematodes belonging to 8 families, 10 subfamilies, 12 genera and 23 species were reported in vegetable and vineyard areas of Diyarbakır province. In another study, among the 39 nematode species determined in the wheat, barley, vegetable and fruit production areas of the Southeastern Anatolia Region, 6 species including *Ditylenchus longicauda*, *Filenchus hamatus*, *Helicotylenchus crassatus*, *H. goodi*, *H. oleae* and *Rotylenchus echelimae* were newly recorded in Türkiye (Uludamar Kasapoğlu et

al. 2018). It was determined that the vineyards of Malatya, Şanlıurfa and Mardin provinces were infected with Dagger nematode, *Xiphinema* spp. (Öztüzün 1970). In the Eastern Anatolia Region, *Pratylenchus thornei*, *P. neglectus*, *P. penetrans* and *P. crenatus* were identified (Yüksel 1974). It has been reported that the highest infection rate in root lesion nematodes is in Erzurum region with a rate of 42.50% and in Sivas region with a low rate of 17.14% (Toktay et al. 2015). In the study by Toktay et al. (2021) *Heterodera cruciferae* populations were identified and characterized using molecular techniques for the first time in Türkiye. It was revealed that there were no polymorphisms in *H. cruciferae* populations in Niğde according to ribosomal DNA region (rDNA-ITS) and cytochrome oxidase subunit 1 (mtDNA-COI) gene regions.

Although there are local studies conducted in Eastern Anatolia wheat cultivation areas, there is no comprehensive study covering the whole region. Within the scope of this study, sampling was carried out in seven different provinces covering the whole region and the plant parasitic nematode fauna in wheat cultivated areas was determined.

MATERIALS AND METHODS

Material collection

Soil samples from 258 different wheat fields from Elazığ, Erzincan, Erzurum, Iğdır, Kars, Malatya and Sivas provinces in the Eastern Anatolia Region of Türkiye between 2015 and 2016 within the scope of TUBITAK project (112O565) were used in the study (Table 1, Figure 1). Each soil sample was taken according to a zigzag pattern in each field with a soil auger at a depth of 30 cm (Southey 1986) and final weight per sample was 1.5–2 kg soil. For each sampled field, GPS coordinates of sampling sites were recorded. Soil samples were stored in a cold storage at 4 °C for further morphological and molecular evaluations.

Table 1. The location of soil samples collected from the Eastern Anatolia Region of Türkiye

Provinces	Number of soil samples
Elazığ	29
Erzincan	29
Erzurum	29
Iğdır	28
Kars	34
Malatya	35
Sivas	74
Total	258

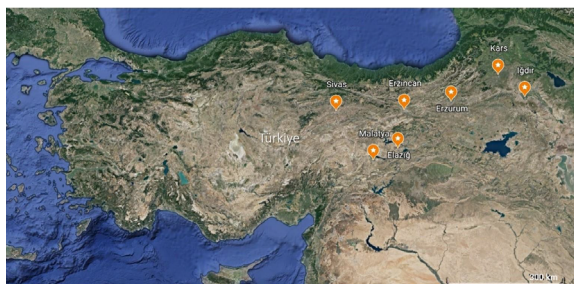


Figure 1. Provinces where the samples are taken in Türkiye

Morphological identification

Each soil sample was thoroughly mixed and a 100 g of sub-sample was processed by a Petri Sieving Method, which is a modification of Baermann Funnel Method, to extract migratory nematodes from the samples (Barker 1985, Southey 1986). Permanent slides were prepared to identify the plant parasitic nematodes at the species level. For this purpose, nematodes extracted from the soil were fixed in TAF solution [7 ml of formalin (40% formaldehyde) at 65 °C, 2 ml of triethanolamin and 91 ml of pure water] (Hooper 1986). After the fixation process, the nematodes were incubated in solution 1 (1 part of glycerol and 79 parts of pure water) at 35-40 °C for 10-12 h. Then, they were transferred to solution 2 (5 parts of glycerol and 95 parts of 96% ethanol) and incubated at 40 °C for 3 h. Fixed nematodes were put in a desiccator for the required period of time for all remaining water to evaporate (Seinhorst 1959). The nematodes divided into groups according to their genus under a dissecting light microscope (Leica DM 5500 B, Germany). Then, each genus was permanently fixed on glass slides using the wax-ring method (Hooper 1986), and the specimens were examined under a compound light microscope (Leica, Germany). Measurements of L, a, b, c, c', V (%), stylet length and tail length, and taxonomic identification were done according to the formula and keys cited by Siddiqi (1986) from the second stage juveniles, females or cysts. L value measurement was taken as 'mm' and other measurements as 'µm' (Siddiqi 2000). Finally, taxonomic classification of the nematode species of Nematoda and Dorylaimida (Longidoridae family) order was done according to De Ley and Blaxter (2002).

Molecular identification

DNA isolation was performed for molecular identification of morphologically undetectable species. The DNA was isolated from a single individual from the populations of wheat nematodes collected from wheat fields after extraction freshly, according to the protocol of Holterman

et al. (2006). Each individual was transferred into an Eppendorf tube containing 25 µl of double distilled water (ddH₂O) and was kept at -80 °C for a 10 min. Then, 950 µl of Worm Lysis Buffer (WLB (-)), 10 µl of beta-mercaptoethanol and 40 µl of proteinase K (20 mg/ml) were added to each tube. Tubes were incubated at 65 °C for 2 h, after that successively incubated at 95 °C for 10 min using a thermal cycler. After incubation, tubes were centrifuged for 1 min at 10.000 rpm, then stored at -20 °C until the samples were used.

For molecular identification of the nematodes, the LSU-rDNA region (1050 bp) was amplified by using LSU primers (11F and 21R) in a PCR reaction (Holterman et al. 2006). Two µl of DNA was added to the PCR reaction mixture containing 21 µl of ddH₂O, 25 µl of 2× DreamTaq PCR Master Mix (Thermo Scientific, Belgium) and 1 µM of each forward (11F: 5'GTCGTGATTACCCGCTGAACTTA3') and reverse primers (21R: 5'TCGGAAGGAACCAGCTACTA3'). For the PCR amplification, the thermal cycler program was set up to 1 cycle for 5 min at 94 °C followed by 35 cycles of incubation at 94 °C for 30 s, then at 54 °C for 30 s and finally at 72 °C for 110 s for elongation. For final elongation, reaction was incubated at 72 °C for 5 s. Following the PCR amplification, 5 µl of each PCR product was mixed with 1 µl of 6x loading buffer (Thermo Scientific, Belgium) and loaded on a 1.5% standard agarose gel in 1x TAE buffer. After the electrophoresis (at 120 V for 40 min), the gel was stained with ethidium bromide (0.1 µg/ml) for 15-20 min, photographed and visualized under UV-light. The remaining PCR products were stored at -20 °C. The products obtained from the PCR amplification using LSU primers were sent to a company (MacroGen Inc., Ankara) to obtain the DNA sequences in a bidirectional Sanger sequencing. After sequencing, the LSU sequences were blasted in GeneBank for identification. Phylogenetic trees are constructed from sequences *H. vulgarens* from a range of countries available in GeneBank. The sequences of *Helicotylenchus multicinctus* (MT321731, Colombia), *H. caudatus* (MN 764335, South Korea), *H. pseudorobustus* (MG653533, Poland), *H. microlobus* (MN 764322, South Korea), *Heterodera schachtii* (MH790255, USA), *H. vulgarens* (MK825777, Iran and MG770483, Greece), *Globodera rostochiensis* (MG994942, UK), *G. pallida* (JN712219, UK), *Rotylenchus goodeyi* (MW960041, Poland), *Pratylenchus thornei* (MZ 956971, Türkiye), *P. penetrans* (MW720692, Netherlands), were included for analyses of LSU locus.

The evolutionary history was inferred by using the Maximum Likelihood method and Tamura-Nei model

(Tamura and Nei 1993). The tree with the highest log likelihood (-4277,83) is shown. The percentage of trees in which the associated taxa clustered together is shown next to the branches. Initial tree(s) for the heuristic search were obtained automatically by applying Neighbor-Join and BioNJ algorithms to a matrix of pairwise distances estimated using the Tamura-Nei model, and then selecting the topology with superior log likelihood value. A discrete Gamma distribution was used to model evolutionary rate differences among sites [5 categories (+G, parameter = 0,4228)]. This analysis involved 15 nucleotide sequences. There were a total of 1164 positions in the final dataset. Evolutionary analyses were conducted in MEGA X (Kumar et al. 2018).

RESULTS AND DISCUSSION

Plant parasitic nematodes negatively impact the plant growth and yields. It is essential to identify the nematode species in the field before deciding the best strategy to control. Identification of nematodes in the wheat fields from the Eastern Anatolia Region, which is one of the important winter wheat production centers of Türkiye, is limited. Therefore, it was followed a systematic sampling method to collect the soils from monoculture wheat cultivated areas in 7 provinces of the Eastern Anatolia Region of

Türkiye (Bora and Karaca 1970). As a result of study 20 genera (Table 2) and 7 species (Table 3) were identified in 9 families in the orders of Rhabditida and Dorylaimida in the Nematoda phylum. In previous studies, 12 plant parasitic nematodes *Anguina tritici*, *Meloidogyne incognita*, *Xiphinema* spp., *Heterodera avenae*, *Pratylenchus thornei*, *P. neglectus*, *P. penetrans*, *P. crenatus*, *Pratylenchoides alkani*, *P. erzurumensis*, *Heterodera filipjevi* and *H. latipons* were detected in Eastern Anatolian Region. *Xiphinema* spp. was observed in vineyards in Malatya, Şanlıurfa, Mardin while *Anguina tritici* Steinbuch, 1799 was observed in wheat fields in Şanlıurfa, Mardin, Van and Bitlis. *Meloidogyne incognita* Kofoid and White, 1919 was observed in Malatya and Elazığ (Öztüzün 1970).When the distribution of nematode genera was evaluated in the soil samples, the highest soil infestation was observed in the genus of *Ditylenchus* (73.25%) followed by *Paratylenchus*, (43.02%), *Aphelenchus* (36.82%), *Xiphinema* (33.33%) and *Helicotylenchus* (28.29%). In this study, the lowest rate of contamination was found in the genera of *Bitylenchus* (0.77%), *Zygotylenchus* (1.16%), *Amplimerlinius* (1.93%), *Telotylenchus* (1.93%) and *Tylenchorhynchus* (2.71%) (Table 2). In a previous survey performed in wheat fields in Adıyaman province, a total of 17 species, 7 families and 9 subfamilies were identified. The most common nematodes

Table 1. Distribution of plant parasitic nematodes in the soil samples collected from seven provinces of the Eastern Anatolia

Nematode Genera	Provinces							Total	Soil infestation (%)
	Sivas*	Erzurum	Erzincan	İğdır	Kars	Elazığ	Malatya		
<i>Ditylenchus</i> spp.	65	14	23	9	27	23	28	189	73.25
<i>Pratylenchus</i> spp.	29	12	13	21	25	4	7	111	43.02
<i>Aphelenchus</i> spp.	30	6	15	9	3	9	23	95	36.82
<i>Xiphinema</i> spp.	27	13	13	7	6	10	10	86	33.33
<i>Helicotylenchus</i> spp.	22	11	7	9	8	8	8	73	28.29
<i>Merlinius</i> spp.	29	10	5	6	11	12	8	71	27.51
<i>Scutylenchus</i> spp.	14	7	7	6	6	2	9	51	19.76
<i>Tylenchus</i> spp.	23	4	8	2	7	4	6	49	18.99
<i>Trophurus</i> spp.	9	5	8	0	5	5	3	35	13.56
<i>Aphelenchoides</i> spp.	9	3	5	1	3	6	5	32	12.41
<i>Filenchus</i> spp.	12	2	1	6	2	2	5	30	11.62
<i>Pratylenchoides</i> spp.	13	0	5	1	4	2	5	30	11.62
<i>Paratylenchus</i> spp.	10	2	2	0	4	4	7	29	11.24
<i>Rotylenchus</i> spp.	2	0	0	1	4	1	1	9	3.48
<i>Geocenamus</i> spp.	5	0	2	0	0	1	0	8	3.10
<i>Tylenchorhynchus</i> spp.	3	1	1	1	0	1	0	7	2.71
<i>Telotylenchus</i> spp.	2	0	1	0	1	0	1	5	1.93
<i>Amplimerlinius</i> spp.	2	0	2	0	0	1	0	5	1.93
<i>Zygotylenchus</i> spp.	0	0	0	3	0	0	0	3	1.16
<i>Bitylenchus</i> spp.	0	0	0	1	0	0	1	2	0.77

* Number of soil samples including nematodes

were *Aphelenchus avenae*, *H. latipons*, *Merlinius brevidens*, *P. thornei* and *Scutylenchus quadrifer* (Öcal and Elekçioğlu 2015). Many genera known to cause significant economic losses have been identified as a result of our survey study. Because the provinces where the samples were taken in the Eastern Anatolia region are close to each other in terms of geographical height and climatic characteristics, similar genera were identified in many of the provinces. Therefore, our results show soil infestation of different nematode genera than the ones identified in Adiyaman province.

Considering the distribution of plant parasitic nematodes in each province, the two most abundant genera in Sivas province were *Ditylenchus* spp. and *Aphelenchus* spp. with 87.8% and 39.2%, respectively, while *Zygotylenchus* spp. or *Bitylenchus* spp. were not detected (Table 2). *Ditylenchus* spp. and *Xiphinema* spp. were the top two contaminating nematode genera in soil samples collected from Erzurum province. In this province, the soil infestation of *Ditylenchus* spp. and *Xiphinema* spp. were calculated as 48.3, 44.8%, respectively. In soil samples collected from Erzincan province, *Ditylenchus* spp. and *Aphelenchus* spp. were the top two contaminating nematode genera with soil infestation of 79.3 and 51.7%, respectively. Interestingly, *Pratylenchus* spp. was the top contaminating nematode genera in the soil samples collected from Iğdır province with a soil infestation of 75.0%. Finally, *Ditylenchus* spp. was the nematode genera with the highest soil infestation in the soil samples collected from Kars, Elazığ and Malatya provinces, where the rates reached to 79.4, 79.3 and 80.0%, respectively, in each province. *Pratylenchus thornei*, *P.*

neglectus, *P. penetrans* and *P. crenatus* were found in wheat fields of Eastern Anatolian Region in a previous survey study (Yüksel 1974). *P. alkani* and *P. erzurumensis* were reported for the first time in Eastern Anatolian Region by Yüksel (1977). These results are parallel with our observations. The highest infection rate of root lesion nematodes occurred in Erzurum region with the rate of 42.50% and the lowest rate was reported in Sivas region with 17.14% by Toktay et al. (2015). Moreover, *Heterodera filipjevi* and *H. latipons* were also identified in wheat fields of Eastern Anatolian Region in the same study. These previous studies combined with our observations suggest that these nematode species are widely distributed in the region. As opposed to our findings, *Heterodera*, *Pratylenchus*, *Pratylenchoides*, *Paratylenchus*, *Merlinius*, *Helicotylenchus* and *Tylenchorhynchus* genus were detected in soil samples collected from Bolu in North West Black Sea Region (İmren et al. 2015). In this study, the most harmful plant parasitic nematodes were determined as *Heterodera* (82.6%) and *Pratylenchus* (73.3%). Finally, the root-lesion nematodes (*Pratylenchus neglectus* and *P. thornei*) and cereal cyst nematodes (*Heterodera avenae*, *H. filipjevi* and *H. latipons*) were found to be economically important in wheat fields in the Eastern Mediterranean and Central Anatolian Regions (Kasapoğlu et al. 2015). In a study conducted in wheat fields in the Eastern Mediterranean Region, nine nematode species were identified, and it was reported that *Geocenamys brevidens* and *P. thornei* were wide-spread and could be of economic importance (Elekçioğlu 1996).

Table 3. Plant parasitic nematode species identified in this study

Genus	Species	L	a	b	c	c'	V (%)	stylet length	tail
<i>Ditylenchus Filipjev, 1936</i>	<i>Ditylenchus myceliophagus</i> Goodey, 1968	0,750 ± 0,16	40,00 ± 4,06	5,59 ± 0,30	15,00 ± 1,13	4,10 ± 0,63	81,12 ± 1,50	7,00 ± 1,05	50,00 ± 12,06
<i>Helicotylenchus steiner, 1945</i>	<i>Helicotylenchus canadensis</i> Waseem 1961	0,953 ± 0,46	26,00 ± 6,26	5,12 ± 1,03	52,94 ± 5,16	not detected	66,11 ± 3,40	29,18 ± 1,05	18,00 ± 4,06
<i>Helicotylenchus steiner, 1945</i>	<i>Helicotylenchus vulgaris</i> Yuen, 1964	0,807 ± 0,91	26,03 ± 2,03	5,02 ± 2,88	62,78 ± 8,02	0,65 ± 0,67	61,02 ± 1,21	30,12 ± 2,04	13,69 ± 0,15
<i>Pratylenchoides winslow, 1958</i>	<i>Pratylenchoides alkani</i> Yüksel, 1977	0,872 ± 0,06	30,00 ± 1,02	5,40 ± 0,16	16,50 ± 1,09	3,20 ± 0,03	56,70 ± 3,90	24,60 ± 0,05	47,12 ± 6,16
<i>Amplimerlinius Siddiqi, 1976</i>	<i>Amplimerlinius macrurus</i> Goodey, 1932 Siddiqi, 1976	0,852 ± 0,17	27,12 ± 5,01	5,20 ± 0,93	17,00 ± 1,09	2,40 ± 0,96	56,37 ± 1,80	26,60 ± 7,00	55,12 ± 1,09
<i>Scutylenchus Jairajpuri, 1971</i>	<i>Scutylenchus quadrifer</i> Andrassy, 1954: Siddiqi, 1979	0,772 ± 0,97	27,00 ± 2,56	5,23 ± 1,93	13,50 ± 4,03	2,34 ± 0,01	48,37 ± 3,50	21,60 ± 0,95	50,12 ± 3,06

Morphological identification of seven species isolated from sample collection sites was performed. Accordingly, *Ditylenchus myceliophagus* was isolated from wheat fields in Erzincan in our survey (Figure 2, Table 4). Previously it was reported by Goodey (1958) in mushrooms in England. It was first reported by Ağdacı et al. (1990) in mushrooms in Türkiye.

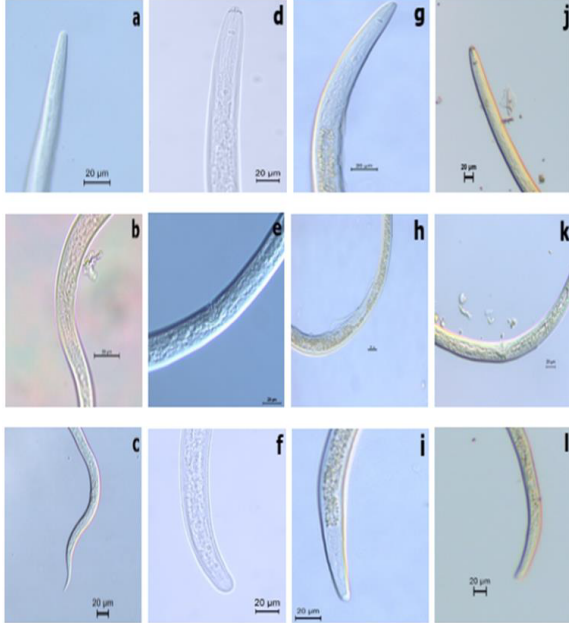


Figure 2. *Ditylenchus myceliophagus*, a. anterior end of the female of *D. myceliophagus*, b. vulva of the female of *D. myceliophagus*, c. tail of the female of *D. myceliophagus*. *Pratylenchoides alkani*, d. anterior end of the female of *P. alkani* e. vulva of the female of *P. alkani* f. tail of the female of *P. alkani*. *Scutylenechus quadrifera*, g. anterior end of the female of *S. quadrifera*, h. vulva of the female of *S. quadrifera*, i. tail of the female of *S. quadrifera*, *Amplimerlinius macrurus*, j. anterior end of the female of *A. macrurus*, k. anterior end of the female of *A. macrurus*, l. tail of the female of *A. macrurus*

Pratylenchoides alkani was first reported by Yüksel (1977) in beans in Türkiye (Erzurum). In this study, it was reported in wheat fields of Sivas (Center and Gemerek) (Figure 2, Table 4). *Scutylenechus quadrifera* was found in wheat areas of Elazığ-Center of Türkiye (Figure 2, Table 4). Ercan (1976) found this species in ornamental plants in Istanbul in Türkiye. *Amplimerlinius macrurus* was identified in wheat areas of Sivas-Center (Figure 2, Table 4). It was described for the first time by Saltukoğlu (1973) in watermelons in Istanbul of Türkiye. *Helicotylenchus canadensis* was identified in wheat cultivated areas in Erzurum (Karaçoban-Duman), Kars (Çıldır-Çanaksu) and Sivas (Center - Yıldızeli and Yavru-Ekecik)

(Figure 3, Table 4). This species was previously reported by Waseem (1961) in vineyards in Canada. Previously reported by Kepenekçi (1999) in lentil in Türkiye (Nevşehir and Yozgat). *Helicotylenchus digonicus* was identified in wheat areas of Sivas (Kayseri Road) and Kars (Susuz-Arpaçay road-Akçalar) (Figure 3, Table 4). This species was first described by Yuen (1964) in grass in England and was detected by Saltukoğlu (1974) in grass and garlic in Istanbul. Finally, *Helicotylenchus vulgaris* was detected in samples collected from Kars (Çıldır road-Çanaksu) and Malatya (Arguvan-Bozburun) in wheat fields (Figure 3, Table 4). This species was first described by Yuen (1964) in grass in England and was found by Ertürk et al. (1973) in potato cultivated areas of Türkiye (Çanakkale and İzmir). The morphological and morphometric measurements of the species identified in the study were found to be compatible with the reference values (Table 3).

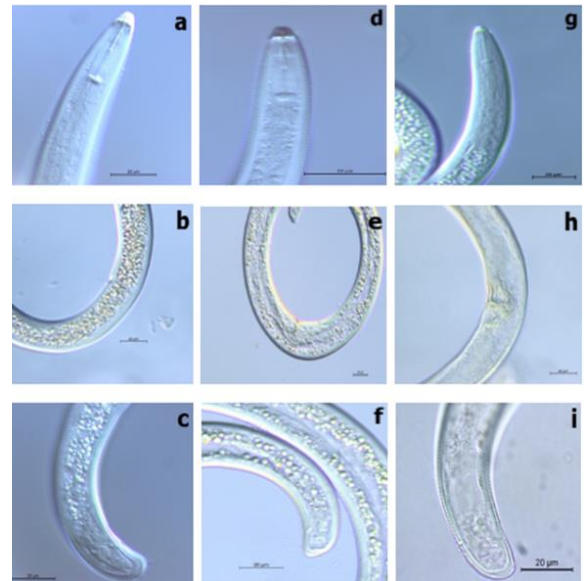


Figure 3. *Helicotylenchus canadensis* a. anterior region, b. vulval region, c. tail region, *Helicotylenchus digonicus* d. anterior region, e. vulval region, f. tail region, *Helicotylenchus vulgaris*, g. anterior region, h. vulval region, i. tail region

For the first time in Eastern Anatolia Region (Türkiye), nematode species have been identified by molecular methods in this study. Molecular diagnostic of *H. vulgaris* is not been reported in the literature in Türkiye. Nematode species were determined by molecular methods for the first time in the wheat fields in the provinces of Erzincan (Kemaliye), Sivas (Center) and Sivas (Gemerek) (Figure 4). After the females were identified at the genus and species levels under light microscope, some species were identified molecularly by using LSU primers to determine the species. After DNA isolation, bands of 1050 bp in length

Table 4. Locations of nematodes identified by morphological or molecular methods at the species level

Species	*PPNs/F	Molecular	Morphology	Cities/District	Location
<i>Ditylenchus myceliophagus</i> Goodey, 1968	F		X	Erzincan/Center	N 39°47'91.4." E38°58'81.3"
<i>Helicotylenchus canadensis</i> Waseem 1961	PPNs		X	Erzurum / Karaçoban-Duman Kars / Çıldır Sivas/ Yıldızeli Sivas/ Yavru- Ekecik	N 39°30'71.8." E 41°93'14.6" N 41°03'96.8." E 43°30'51.8" N 39°86'97.4." E 36°61'93.1" N 39°80'76.4." E 36°14'33.2"
<i>Helicotylenchus vulgaris</i> Yuen, 1964	PPNs	X	X	Kars/ Çıldır-Çanaksu Malatya/Arguvan-Bozburun Sivas/ Center Sivas/ Kayseri-Road Erzincan/ Kemaliye-İliç	N 40°99'94.7." E 43°30'27.9" N 38°66'00.1." E 38°33'03.3" N 39°69'66.6." E 37°00'72.0" N 39°57'27.8." E 37°00'89.2" N 39°44'58.8." E 38°47'22.0"
<i>Helicotylenchus digonicus</i> , Perry, Darlind and Thorne, 1959	PPNs		X	Sivas/ Kayseri-Road Kars/ Susuz-Arpaçay	N 39°51'23.3." E 36°84'93.4" N 40°75'15.3." E 43°24'64.9"
<i>Pratylenchoides alkani</i> Yüksel, 1977	PPNs		X	Sivas/ Center Sivas/ Gemerek	N 39°55'68.6." E 37°02'72.2" N 39°25'72.9." E 36°12'11.1"
<i>Amplimerlinius macrurus</i> Goodey, 1932 Siddiqi, 1976	PPNs		X	Sivas/ Center	N 39°24'51.5." E 37°40'58.7"
<i>Scutylenechus quadrifer</i> Andrassy, 1954:Siddiqi, 1979	PPNs		X	Elazığ/ Center	N 38°67'83.3." E 39°15'38.6"

*PPNs (Plant parasitic nematode, F (Fungivore nematode))

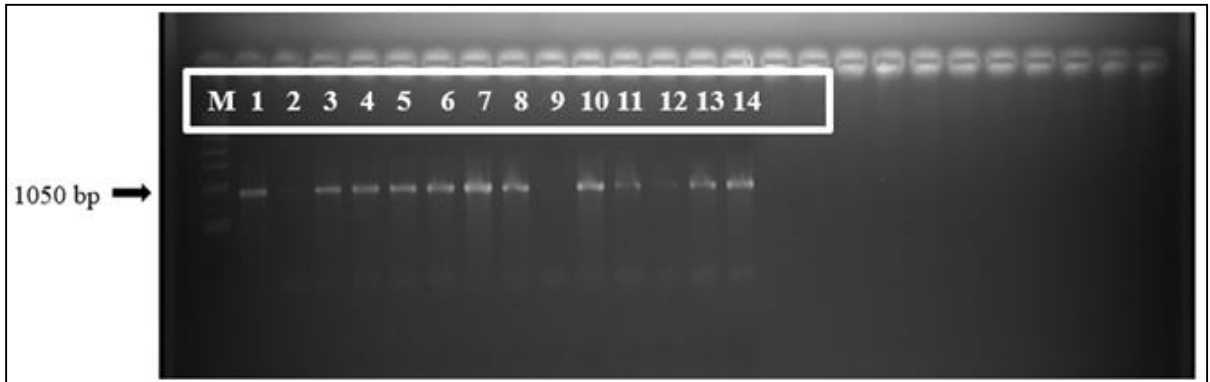


Figure 4. Band image obtained by PCR using LSU primers. M: 100bp DNA ladder (Thermo Scientific).3. *Helicotylenchus vulgaris* (N 39°57'27.8." E 37°00'89.2"), (Sivas, Gemerek): 4. *H. vulgaris* (N 39°69'66.6." E 37°00'72.0"), (Sivas, merkez): 10. *H. vulgaris* (N 39°44'58.8." E 38°47'22.0"), (Erzincan, Kemaliye)

were obtained from the samples by using LSU (11F and 21R) primers. PCR products obtained from PCR using LSU primers and were sent for sequence analysis. Then, the species of these genera were determined with BLAST (Wageningen University- Netherlands Database) analysis. *H. vulgaris* was determined in 3 locations after sequencing. Among the samples sent for sequencing, 4 populations did not a match with any species in the database.

Based on phylogenetic analysis, three *H. vulgaris* populations obtained from this study separately clustered into a group which indicates nucleotide differences of them. The sequences obtained from Erzincan and Sivas (Gemerek) provinces are highly similar than the sequences of Sivas (Merkez) (Figure 5). The LSU gene region sequences of the two Turkish population were similar to the Greece population, an example is similar to the Iranian population.

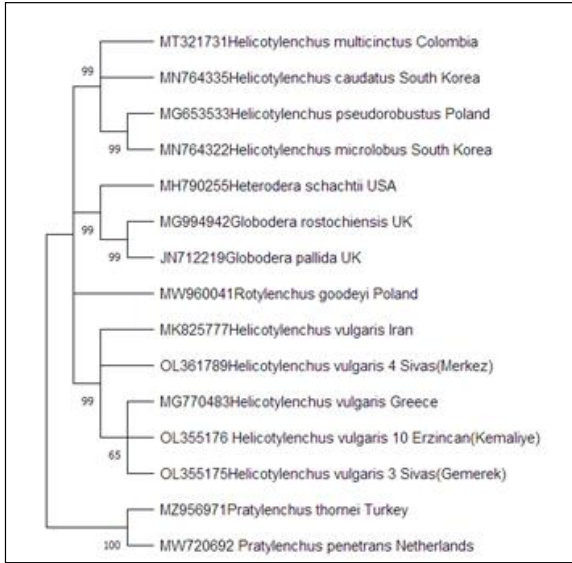


Figure 5. Molecular phylogenetic status of *Helicotylenchus vulgaris* LSU sequences

Considering the species that were not determined in the database search, further molecular characterization studies are suggested from this region. Overall, *H. digonicus* (from the samples of Sivas and Kars), *S. quadrifer* (from the samples of Elazığ), *D. myseliophagus* (from the samples of Erzincan), *A. macrurus* (from the samples of Sivas), *H. canadensis* (from the samples of Erzurum, Kars and Sivas), *H. vulgaris* (from the samples of Kars, Erzincan, Sivas and Malatya), and *P. alkani* (from the samples of Sivas) were reported for the first time in Eastern Anatolia Region according to the molecular identification studies.

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Author's Contributions

Authors declare the contribution of the authors is equal.

Statement of Conflict of Interest

The authors have declared no conflict of interest.

ÖZET

Bu çalışmanın amacı, Doğu Anadolu Bölgesi'nde (Türkiye) 2017-2019 yılları arasında buğday (*Triticum* spp.) alanlarında bulunan bitki paraziti nematod türlerinin moleküler ve morfolojik olarak belirlenmesidir. Bu amaçla, Doğu Anadolu Bölgesi'ne ait toplam 7 ilden (Erzincan, Elazığ, Erzurum, Iğdır, Kars, Malatya ve Sivas) toplam 258 toprak örneği alınmıştır. Elde edilen nematodlar cins veya tür düzeyinde ışık mikroskobu kullanılarak morfolojik olarak tanımlanmıştır. Tanımlanamayan nematod türlerinden bazılarının 28S ribosomal DNA bölgesi kullanılarak dizi analizi oluşturulmuş ve veribankasında karşılaştırmaları yapılmıştır. Elde edilen verilere göre; toplam 2 takım 9 familyaya ait 20 cins ve 7 tür, *Helicotylenchus digonicus* Perry, 1959, *Helicotylenchus canadensis* Waseem, 1961, *Helicotylenchus vulgaris* Yuen, 1964 (Nematoda: Hoplolaimidae), *Ditylenchus myceliophagus* Goodey, 1958 (Nematoda: Anguinidae), *Amplimerlinius macrurus* (Goodey, 1932) (Nematoda: Dolichodoridae), *Scutylenchus quadrifer* (Andrassy, 1954) (Nematoda: Dolichodoridae) ve *Pratylenchoides alkani* Yüksel, 1977 (Nematoda: Pratylenchidae) morfolojik olarak teşhis edilmiştir. *Helicotylenchus vulgaris* moleküler tekniklerle tür düzeyinde belirlenmiştir. Ekonomik açıdan önemli bitki paraziti nematodların bulunuş oranları *Ditylenchus* spp., *Pratylenchus* spp., *Aphelenchus* spp., *Xiphinema* spp. ve *Helicotylenchus* spp. için sırasıyla %73, %43, %36, %33 ve %28 olarak belirlenmiştir. Elde edilen sonuçların bölgedeki nematod mücadele yöntemlerinin planlanmasına yardımcı olacağı düşünülmektedir.

Anahtar kelimeler: ektoparazit nematodlar, teşhis, nematod, *Triticum* spp., Türkiye

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Original article

Assessment of thrips species diversity and their impact on lemon orchards in the Eastern Mediterranean Region of Türkiye

Doğu Akdeniz Bölgesindeki limon bahçelerindeki trips türlerinin çeşitliliğinin ve etkilerinin araştırılması

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ABSTRACT

Thrips are polyphagous, and some species cause serious damage to citrus fruits worldwide including Türkiye. This study was conducted to determine thrips species on different lemon varieties in the Eastern Mediterranean Region in Türkiye. Surveys were conducted on lemon orchards in 2017, 2018 and 2019. Thrips species and damage on lemon varieties in the Eastern Mediterranean Region is not fully examined therefore this study helped to understand the biodiversity and damage of thrips species on lemon orchards in this region. Eight thrips species were determined. *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae) was found to be the most common species on Kütdiken, Interdonate and Mayer lemon varieties in Adana and Hatay. *Thrips hawaiiensis* (Morgan) (Thysanoptera: Thripidae) was detected as the most common thrips species in Mersin on Kütdiken and Interdonate. *T. hawaiiensis* was first detected in Türkiye in 2015, and it has become the dominant species in lemon orchards in Mersin province. Moreover, the Shannon-Wiener values ($H=0,99525$, $EH=0,478614$), and the Simpson Biodiversity values ($D=0,44972$, $Sd=0,5502$) were calculated to determine thrips species diversity in the Eastern Mediterranean Region in this study. During the late flowering period, *Thrips hawaiiensis* had a higher population density in Kütdiken, Interdonate and Mayer varieties. This resulted severe damage to fruit. It is important to note that *F. occidentalis* does not cause any damage to lemon fruits. In this study, the distribution and biodiversity of thrips species based on lemon varieties were studied in Türkiye, with a focus on their distribution across provinces.

INTRODUCTION

Citrus is one of the most economically important agricultural products grown in the Eastern Mediterranean Region (Adana, Mersin, Hatay). Many pests, diseases, and weeds cause significant yield loss in citrus vegetation areas (TÜİK 2022, Uygun et al. 2010). More than 89 pests have been determined on citrus in Türkiye since 1990 (Uygun et al. 1992, Uygun et al. 2010). Thysanoptera is an order that includes economically important polyphagous pest species causing severe damage to various crops (Lewis 1973). Thrips species (Insecta: Thysanoptera) are invasive due to their high adaptation capability (Marullo and Grazia 2017). Besides, thrips are known as virus vectors. These pests usually feed on leaves and flowers (Marullo and Grazia 2013).

Faunistic studies on Thysanoptera were carried out in several regions of Türkiye (Özsemerci et al. 2006, Tunç 1991, 1992). Tekşam and Tunç (2009) found 36 species in Antalya in their study on thrips species. In addition, thrips species were studied and reported on various crops in the Eastern Mediterranean Region (Atakan 2007a, 2007b, 2010, 2011, Hazır et al. 2011, Nas et al. 2007, Ölçülü 2014). According to those studies, *Frankliniella occidentalis* (Pergande) and *Thrips hawaiiensis* (Morgan) (Thysanoptera: Thripidae) were the most common Thrips species on vegetables and field crops in the Eastern Mediterranean Region (Pehlivan and Atakan 2017). *Thrips hawaiiensis* was first reported in Çukurova in 2015 and quickly spread to the region (Atakan et al. 2015, Pehlivan and Atakan 2017). In addition, Hazır et al. (2022) studied the effectiveness of some insecticides against *T. hawaiiensis* and the efficacy of *Orius laevigatus* (Fieber) (Hemiptera: Anthocoridae) in the lemon orchard in Mersin.

This study was conducted to determine different thrips species in lemon orchards of Adana, Mersin, and Hatay provinces located in the Eastern Mediterranean Region. In addition, the distribution and density of thrips species were determined in this study between 2017 and 2019. This study helps to determine the damage of thrips species in lemon varieties separately in the Eastern Mediterranean Region. Moreover, it contributes to IPM strategies to control thrips species in lemon orchards in the region.

MATERIALS AND METHODS

Sampling of thrips species from fruits and flowers

Surveys were conducted in Adana, Mersin and Hatay provinces in the Eastern Mediterranean Region in Türkiye between 2017, 2018 and 2019 (Figure 1). A random selection of 100 fruits and flowers from each citrus group in an orchard was examined individually. Any fruit with silvery or bronzed spots or scars on surface, indicating irregular

holy damage, was considered damaged. Conversely, fruits without such symptoms were considered healthy. This enabled the determination of the damage ratio in each orchard based on the percentage of damaged fruits within the sample of 100 (Atakan and Pehlivan 2020).

The thrips adults were collected during flowering and



Figure 1. Locations of thrips collections in the Eastern Mediterranean Region of Türkiye in 2017, 2018, and 2019

fruiting periods in lemon orchards and were recorded with collected lemon varieties for determining the distribution of thrips species on different lemon varieties in three different provinces (Adana, Mersin and Hatay) in the Eastern Mediterranean Region. The adults were preserved in alcohol in Eppendorf tubes and brought to the laboratory for identification. Furthermore, fruit samples were gathered to identify the species of thrips. Samplings were done on randomly selected lemon flowers and fruits. Thrips samples were collected using a fine brush and then put into 50 ml Eppendorf tubes filled with 70% ethanol (Atakan et al. 2015).

Thrips identification

Thysanoptera (Thrips) species collected from the flower and the fruit samples were brought to the Çukurova University, Faculty of Agriculture, Plant Protection Department Industrial Plant Pests Laboratory in Eppendorf tubes (50 ml). The following method was used to identify thrips: (Atakan et al. 2015) the samples were extracted from flowers and fruits into Petri dishes and placed in Eppendorf tubes consisting of 60% ethanol. These were transferred to AGA medium (10:1:1 60% ethyl alcohol, glycerin and glacial acetic acid) for two days to facilitate their preparation and, for this purpose, to soften their bodies before returning them to 60% alcohol. Samples were placed separately into glass Petri dishes and kept in 10% KOH for approximately one hour at 48 °C. Body contents of thrips specimens were evacuated by entering the hind leg bases of thrips individuals with a very fine-tipped needle (maceration). The samples were cleaned by passing through an alcohol series and transferred to the Hoyer medium to prepare their microscopic slides (Atakan et al.

2015). The third co-author carried out the identifications.

Thrips species diversity index in the Eastern Mediterranean Region

The Shannon Diversity Index is a tool used to measure the variety of species in a given ecosystem. It is calculated using the formula $H = \sum p_i \times \ln(p_i)$. A higher value of H indicates a greater diversity of species in that ecosystem, whereas a lower value of H indicates a lower diversity within that ecosystem. In summary, the Shannon Diversity Index is a useful method for quantifying the level of biodiversity within a particular community. (Shannon et al. 1948).

The Shannon Equitability Index is a metric used to evaluate the uniformity of species in a particular community. The term "evenness" highlights how closely related the frequencies of different species are in that community. The Shannon Equitability Index is calculated by dividing the Shannon Diversity Index (H) by the natural logarithm of the total number of unique species (S). The resulting value ranges from 0 to 1, where 1 indicates perfect evenness. The Shannon Diversity Index is categorized into low ($H < 2$), moderate ($2 < H < 4$), and high ($H > 4$) species of gastropods and nematodes in some studies. In summary, the Shannon Equitability Index is a valuable tool for assessing the degree of evenness among species in a given ecosystem (Keçici et al. 2022, Miller et al. 2015, Shannon et al. 1948).

The Simpson diversity index was used to determine thrips biodiversity in lemon orchards in Adana, Mersin, and Hatay provinces in the Eastern Mediterranean Region. Simpson diversity (D) dominance (Sd) indexes and Simpson Evenness (Esm) were used to detect biodiversity values in this study (Magurran 1988, 2004).

RESULTS

Thrips species on lemon flowers in the Eastern Mediterranean Region

Surveys were conducted in three provinces (Adana, Mersin and Hatay) in the Eastern Mediterranean Region in Türkiye in 2017, 2018 and 2019. During the survey studies, eight different thrips species belonging to three different families were determined in lemon orchards. According to the results, *Frankliniella occidentalis* Pergande 1895 (Thysanoptera: Thripidae), *Thrips hawaiiensis* Morgan 1913 (Thysanoptera: Thripidae), *Thrips tabaci* Lindeman 1889 (Thysanoptera: Thripidae), *Thrips major* Uzel 1895 (Thysanoptera: Thripidae), *Thrips meridionalis* Priesner 1926 (Thysanoptera: Thripidae), *Haplothrips reuteri* Karny 1907 (Thysanoptera: Phlaeothripidae), *Melanthrips fuscus* Sulzer 1776 (Thysanoptera: Melanthripidae), *Haplothrips vuillei* Priesner 1920 (Thysanoptera: Phlaeothripidae) were

detected in the study.

The most widespread species was found as *T. hawaiiensis* with the highest percentage in Mersin between 2017 and 2019, while *F. occidentalis* was found in Adana and Hatay in lemon orchards (Table 1). *T. hawaiiensis* has caused damage to lemons in Adana, Mersin and Hatay since 2015. It has been spreading more rapidly than other species. The damage of *T. hawaiiensis* can be seen easily on fruit and negatively affects lemon exports (Figure 2). A small percentage of *T. tabaci*, *T. major* and *T. meridionalis* were detected as pests in lemon orchards in Adana, Mersin and Hatay provinces. In addition to these five species, *H. reuteri*, *H. vuillei*, and *M. fuscus* were also found in lemon orchards in Mersin within this study (Table 1). *Thrips hawaiiensis* was first reported in lemon orchards in Mersin in 2015. In a short time, it was also found in Adana and Hatay due to its faster rate of spread than other thrips species since 2015. The easily distinguishable thrips damage on the fruit adversely affected the export of lemons (Figure 2). A small percentage of *T. tabaci*, *T. major* and *T. meridionalis* were detected to feed together with others as a species complex in lemon orchards in Adana, Mersin and Hatay. In addition to these three species, small amounts of *H. reuteri*, *H. vuillei* and *M. fuscus* were found in Mersin. *F. occidentalis* was the dominant thrips species in all three lemon varieties (Kütüden, Interdonate and Mayer) of the individuals sampled in Adana in 2017, 2018 and 2019. It was followed by *T. hawaiiensis* and *T. major* (Table 1). In Hatay, similar to Adana, *F. occidentalis* was the dominant thrips species which was followed by *T. hawaiiensis* and *T. major* (Table 1). The species composition in Mersin was formed differently from other provinces. In Mersin, *T. hawaiiensis* was the dominant thrips species followed by *F. occidentalis* and *T. tabaci* in Kütüden and Interdonate lemon varieties. In this study, The Shannon Diversity Index (H) was 0,99525 in 8 different thrips species in citrus orchards and thrips diversity can be identified as a low diversity for thrips species in the Eastern Mediterranean Region (Adana, Mersin and Hatay provinces). According to Shannon et al. (1948): low means $H < 2$, moderate means $2 < H < 4$, and $H > 4$ means high for gastropods. In addition, evenness ($E_H = 0,478614$) is not close to 1 therefore it can be identified as low in citrus (Table 2). The Simpson Diversity index ranges from 0 to 1, 1 represents infinite diversity, and 0, no diversity (Simpson 1949). Table 2 shows the results of The Simpson Diversity Index, and Simpson diversity index (D) was 0,44972, the Simpson dominance index (Sd) was 0,5502, and the Simpson evenness index (E_{sm}) was 0,27794 for thrips species in lemon orchards in Adana, Mersin and Hatay in the Eastern Mediterranean Region. The biodiversity results for the two different indexes were similar to each other and low in the lemon orchards in Adana, Mersin and Hatay (Table 2).

Table 1. The percentage of thrips species detected on flowers and fruits in lemon orchards in Adana, Mersin and Hatay Provinces in 2017, 2018 and 2019

Thrips species	2017			2018			2019		
	Adana	Mersin	Hatay	Adana	Mersin	Hatay	Adana	Mersin	Hatay
Melanthripidae									
<i>Melanthrips fuscus</i> Sulzer	0,00	0,00	0,00	0,00	11,52	0,00	0,00	0,00	0,00
Phlaeothripidae									
<i>Haplothrips reuteri</i> Karny	0,00	0,40	0,00	0,00	0,00	0,00	0,00	0,00	0,00
<i>Haplothrips vuilleti</i> Priesner	0,00	3,24	0,00	0,00	0,00	0,00	0,00	0,00	0,00
Thripidae									
<i>Frankliniella occidentalis</i> Pergande	61,02	27,94	55,88	37,23	8,68	40,00	33,88	26,69	49,18
<i>Thrips major</i> Uzel	4,60	1,74	5,88	8,76	1,86	6,67	10,74	2,79	13,11
<i>Thrips meridionalis</i> Priesner,	0,00	2,27	0,00	0,00	0,00	0,00	0,00	0,00	0,00
<i>Thrips hawaiiensis</i> Morgan,	30,51	61,76	33,82	48,18	88,84	48,33	45,45	63,75	34,43
<i>Thrips tabaci</i> Lindeman	3,87	5,61	4,41	5,84	0,41	5,00	9,92	6,77	3,28



Figure 2. *Thrips hawaiiensis* and its damage; a) thrips on lemon flower, b, c, d: damage symptoms on young lemon fruits due to *T. hawaiiensis*

DISCUSSION

According to EPPO (2022), the most common thrips species in citrus species were found as *Pezothrips kellyanus* Bagnall (Thysanoptera: Thripidae), *T. major* Uzel, *F. occidentalis* and *Heliothrips haemorrhoidalis* Bouché 1833 (Thysanoptera: Thripidae) in the Mediterranean basin. Vono et al. (2022) reported that approximately 20 different thrips species were determined in the Mediterranean geographical distribution. The most common are *F. occidentalis*, *H. haemorrhoidalis*, *P. kellyanus*, *Scirtothrips dorsalis* Hood, 1919 (Thysanoptera: Thripidae), *T. hawaiiensis* Morgan, and *T. major*. Childers and Nakahara (2006) conducted a study about determining thrips species on citrus in Florida, and 36 thrips species were determined in citrus canopies. The most widespread species were *Aleurodothrips fasciapennis* Franklin 1908 (Thysanoptera: Phlaeothripidae), *Frankliniella bispinosa* Morgan, *Chaetanaphothrips orchidii* Moulton 1907, *Karnyothrips flavipes* Jones 1912, and *Danothrips trifasciatus* Sakimura 1975 (Thysanoptera: Thripidae) within that study.

Table 2. Diversity of thrips species in citrus orchards by using the Shannon diversity index and the Simpson Diversity Index results in the Eastern Mediterranean Region

Thrips species	Total Samples/100 fruits+flowers (n.)	Pi	ln(pi)	pi*ln(pi)
1 <i>Frankliniella occidentalis</i>	754	0,321809646	-1,1338	-0,36486619
2 <i>Haplothrips reuteri</i>	3	0,00128041	6,66058	0,008528265
3 <i>Haplothrips vuilleti</i>	2	0,000853606	7,06604	0,006031618
4 <i>Melanthrips fuscus</i>	1	0,000426803	7,75919	0,003311646
5 <i>Thrips hawaiiensis</i>	1372	0,58557405	0,53516	0,313377349
6 <i>Thrips major</i>	89	0,037985489	3,27055	0,124233481
7 <i>Thrips meridionalis</i>	17	0,007255655	4,92597	0,035741169
8 <i>Thrips tabaci</i>	105	0,044814341	3,10523	0,139158704
Total(N)	2343	1	34,4565	0,995248423
Shannon diversity index	H= 0,99525 EH= 0,478614			
Simpson diversity index i	D=0,44972 Sd= 0,5502 E _{sm} =0,27794			

Although Scirtothrips sp. and *T. hawaiiensis* were found lower than abundant species, these species can potentially be a major pest on citrus in Florida. Elimem and Chermiti (2013) studied thrips species distribution in organic citrus orchards, and 12 thrips species were determined within that study, and the most abundant was determined as *F. occidentalis* (32.97% in 2010 and 27.93% in 2011). In addition, *T. hawaiiensis* was common in Mediterranean countries and found in lemon orchards in Italy, France, and Spain as well (Goldarazena 2011, Marullo and De Grazia 2012, Reynaud et al. 2008). Atakan and Pehlivan (2020) studied thrips species on citrus in Adana and Mersin. *Frankliniella occidentalis* was the most common in Adana, *T. hawaiiensis* was also most common in Mersin, specifically in lemon orchards. Atakan et al. (2016) determined different thrips species on lemon in Yenice and Tarsus/Mersin between 2013 and 2014. According to the results of this study, ten different thrips species were observed, and most species were determined as *F. occidentalis* on flowers. Although 6-7 thrips individuals per flower were detected, this study did not record the damage to flower parts and fruits.

Belaam-Kort et al. (2020) studied thrips fauna (pest and predator species), and a total of 21 species were found, *F. occidentalis*, *P. kellyanus* and *T. major* were detected as the most abundant species in citrus orchards within this study. Moreover, *T. major* was found in navel oranges in Italy and Tunisia (Belaam-Kort et al. 2020). In addition, Costa et al. (2006) revealed that the thrips genera on lemon orchards in Portugal and the most common genera were found as *Pezothrips* spp. (44%), *Aeolothrips* spp. (30%) and *Thrips* spp. (14%) respectively within this study. Xu et al. (2012) studied the population fluctuation of thrips species on citrus. According to the results of that study, *F. intonsa* (46.28%) and *F. occidentalis* (48.46%) were found to be the most abundant on navel orange and ponkan mandarin orange. In addition, *Thrips hawaiiensis*, *Thrips palmi* and *Thrips andrewsi* were seen as a pest on navel oranges and ponkan mandarin oranges. The species composition and diversity in communities are estimated using the Shannon diversity index. Some studies are carried out in terms of thrips diversity on different crops (Amoozadeh et al. 2019, De Breuil et al. 2021, Mirab-blaou et al. 2017; Mirab-blaou et al. 2019, Wang et al. 2014). In this study, thrips diversity was found low in citrus orchards and it may occur due to the monoculture of citrus in the region (Table 2).

This study determined the composition and distribution of thrips species in the citrus groves in the Eastern Mediterranean Region (Adana, Mersin and Hatay provinces). According to the results of this study, eight

thrips species were identified as harmful pest thrips in the lemon orchards. In addition, *F. occidentalis* and *T. hawaiiensis* were determined as the most abundant species in lemon orchards in the study. The population levels of thrips species on different lemon varieties were detected separately during the study. Especially, *T. hawaiiensis* causes damage to flowers and early stages of lemon fruits, affecting the economic value of lemon fruit in terms of citrus exportation.

The tendency of lemons to blossom lasts all year, considering the variety. As far as lemon varieties are concerned, *T. hawaiiensis* was more common in Interdonate and Kütdiken in Mersin and Hatay. The variety Kütdiken blooms and bears fruit throughout the year; therefore, *T. hawaiiensis* causes more damage to this variety in Mersin and Hatay. *Frankliniella occidentalis* was more common in Kütdiken, Interdonate, and Mayer in Adana and Hatay. As can be seen, from the above result, thrips species showed corresponding differences in lemon varieties and climatic conditions.

It is important to have a clear understanding of the types and distribution of thrips species on citrus plants to develop effective Integrated Pest Management (IPM) strategies. Since thrips often become resistant to insecticides quickly, more studies should be done on biological control management options.

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Author's Contributions

Authors declare the contribution of the authors is equal.

Statement of Conflict of Interest

The authors have declared no conflict of interest.

ÖZET

Trips, polifag bir zararlı olup, bazı türleri hem dünya hem de Türkiye'de çeşitli turuncgil meyvelerinde ciddi zararlara neden olmaktadır. Bu çalışma, Türkiye'nin Doğu Akdeniz Bölgesi'nde farklı limon çeşitlerindeki trips türlerini belirlemek amacıyla gerçekleştirilmiştir. Sürveyler, 2017, 2018 ve 2019 yıllarında limon bahçelerinde yapılmıştır. Bu çalışma Doğu Akdeniz Bölgesindeki limon bahçelerinde trips türlerinin biyoçeşitliliği ve limon çeşitlerindeki zarar durumu hakkında bilgi sahibi olmamızı sağlayacaktır. Bu çalışma sonucunda sekiz trips türü belirlenmiştir; *Frankliniella occidentalis* (Pergande) (Thysanoptera:

Thripidae), Adana ve Hatay'daki Kütdiken, Interdonate ve Mayer limon çeşitlerinde en yaygın bulunan tür olarak tespit edilmiştir. *Thrips hawaiiensis* (Morgan) (Thysanoptera: Thripidae), Mersin'de Kütdiken ve Interdonate üzerinde en yaygın trips türü olarak belirlenmiştir. *T. hawaiiensis*, Türkiye'de 2015 yılında ilk kez tespit edilmiş olup, bu tarihten itibaren Mersin ilinde limon bahçelerinde baskın tür haline gelmiştir. Ayrıca, bu çalışmada Doğu Akdeniz Bölgesi'ndeki trips tür çeşitliliğini belirlemek amacıyla Shannon-Wiener değerleri ($H=0,99525$, $EH=0,478614$) ve Simpson Biyoçeşitlilik değerleri ($D=0,44972$, $Sd=0,5502$) hesaplanmıştır. *Thrips hawaiiensis*, Kütdiken, Interdonate ve Mayer çeşitlerinde geç çiçeklenme döneminde daha yüksek bir popülasyon yoğunluğuna sahip olduğu ve meyvelere ciddi zarar verdiği tespit edilmiştir. Buna ek olarak, *F. occidentalis*'in limon meyvelerine zarar vermediği bilinmektedir. Bu çalışmada, Türkiye'de limon çeşitlerindeki trips türleri ve bu türlerin illere göre dağılımı ve biyoçeşitliliği incelenmiştir.

Anahtar kelimeler: biyoçeşitlilik, Doğu Akdeniz, limon, Thysanoptera, Türkiye

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Original article

Insecticidal effects of some plant extracts against Khapra beetle [*Trogoderma granarium* Everts (Coleoptera: Dermestidae)]

Bazı bitkisel ekstraktların Khapra böceği [*Trogoderma granarium* Everts (Coleoptera: Dermestidae)]'ne karşı insektisidal etkileri

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ABSTRACT

The study aims to determine the toxicity of extracts in three different solvents (methanol, hot water, and cold water) obtained from 10 different plants [*Rosmarinus officinalis* L. (Lamiaceae), *Nigella sativa* L. (Ranunculaceae), *Laurus nobilis* L. (Lauraceae), *Anethum graveolens* L. (Apiaceae), *Origanum onites* L. (Lamiaceae), *Lavandula angustifolia* Mill. (Lamiaceae), *Foeniculum vulgare* Mill. (Apiaceae), *Hypericum perforatum* L. (Clusiaceae), *Mentha piperita* L. (Lamiaceae), and *Nicotiana tabacum* L. (Solanaceae)] against the larvae of the third instar of *Trogoderma granarium* Everts (Coleoptera: Dermestidae) collected from different provinces of Türkiye. The results of the study varied depending on the plant species and the solvent used. Based on the observations, methanol was found to be the most effective solvent, followed by hot water and then cold water. On the 14th day of application, the highest mortality rate of 100% was observed when methanol was used as a solvent at a concentration of 20% (w/v) of the plant extracts. In contrast, this rate was 44% when cold water was used and 56% when hot water was used. According to the research results, extracts of *A. graveolens*, *N. tabacum*, and *N. sativa* showed a highly toxic effect on the pest, suggesting that these extracts are promising for the control of storage pests. However, more extensive studies are still needed to confirm the applicability and feasibility of these applications on an industrial scale.

INTRODUCTION

Ensuring adequate nutrition for every newborn is a critical challenge in the context of a growing world population, and Türkiye is a major player in the global production and export of stored products, especially cereals (Erdem 2020). Neglecting the crucial aspects of food storage can lead to

diseases and pests in warehouses, resulting in significant losses in stored products. Storage pests are one of the main biotic factors that cause losses in the products produced by growers. The FAO reports that annual crop losses due to stored product pests during post-harvest are 10-30%

worldwide (Kiaya 2014). Pests in stored products can cause direct or indirect damage by feeding on the infested items. Their consumption leads to weight loss, adverse plant quality, changes in nutritional value, and a decline in seed quality and commercial value (Boyer et al. 2012, Rosentrater 2022).

The Khapra beetle [*Trogoderma granarium* Everts (Col.: Dermestidae)] poses a significant threat to stored wheat in Türkiye and is one of the 100 most invasive species worldwide (Athanassiou et al. 2019, Yadav et al. 2021). It is classified as a primary pest and is subject to post-harvest quarantine measures due to its ability to cause direct damage to cereals (Hagstrum et al. 2012). The population density of this species increases significantly in environmental conditions above 30 °C (Kavallieratos et al. 2017), which can lead to the plants infested by it becoming completely unusable. The Khapra beetle, which can cause losses of up to 30% in post-harvest crops (Honey et al. 2017), causes damage primarily through its larvae. These larvae feed on the embryo and endosperm of cereal grains, effectively turning the grains into husks (Ahmedani et al. 2007). The rashes caused by these larvae significantly affect product quality. In addition, the body parts of the larvae can cause severe allergic reactions and respiratory problems.

In studies conducted in Türkiye and other countries, attempts have been made to control this pest species using various control methods. However, these control methods have not achieved the desired goal of maintaining the pest and it has been reported that the pest has developed considerable resistance to phosphine, malathion, and some pyrethroids used for control (Ahmedani et al. 2007). Given the increasing damage attributed to conventional fumigants and preservative insecticides in recent years, many researchers have turned to exploring alternative strategies beyond chemical control measures (Regnault-Roger et al. 2005, Saifi et al. 2023, Yiğit et al. 2023). Recent studies on the control of stored product pests have begun to emphasize the use of natural products of plant origin.

Plants have evolved various defense mechanisms to protect themselves from potential threats in their natural environment (War et al. 2012). These defense mechanisms range from physical barriers within the plant to chemicals synthesized by the plants themselves. Natural insecticidal compounds found in plants have been shown to have a lethal effect on insects (Boulogne et al. 2012, Mann and Kaufman 2012). Researchers have identified nearly 2000 different plants that have insecticidal properties (Grainge and Ahmed 1988, Prakash and Rao 2018).

Although the use of plant extracts for pest control in agriculture has been known for 3000 years, these studies have been intensified, especially in the last 30 years (Pavela 2016). In recent years, more and more studies have been carried out on insecticides from plants. Using various methods, researchers extract plant compounds from different parts of plants, including flowers, leaves, and seeds. These studies investigate the efficacy of these herbal extracts against agricultural pests and show successful results in various studies in controlling numerous pest species, including *Callosobruchus maculatus* F. (Coleoptera: Bruchidae) (Dessenbe et al. 2022, Karunaratne and Karunaratne 2012, Kasinathan et al. 2014), *Rhyzopertha dominica* (F.) (Coleoptera: Bostrychidae) (Guruprasad and Akmal 2014, Guruprasad and Pasha 2015), *Sitophilus oryzae* L. (Coleoptera: Curculionidae) (Hematpoor et al. 2022, Rajashekar et al. 2014), *Sitophilus granarius* L. (Coleoptera: Curculionidae) (Jawalkar and Zambare 2020, Kisa et al. 2018), and *T. granarium* (Derbalah 2012, Musa et al. 2009, Omar et al. 2012).

The studies have significantly increased our knowledge of the use of herbal extracts to control agricultural pests. However, conventional research methods rely heavily on organic solvents such as methanol, ethanol, acetone, and ethyl acetate to test these extracts. However, the widespread use of these solvents poses a health risk to researchers and contributes to environmental problems (Dirar et al. 2019). Therefore, the selection of a suitable extraction solvent is of utmost importance.

In the existing literature, there are remarkably few studies using hot and cold water as extraction solvents, so the comprehensive knowledge in this area remains incomplete and fragmented. To fill this critical gap, this research aims to evaluate toxic effects of extracts from 10 different plants [rosemary (*Rosmarinus officinalis*), black cumin (*Nigella sativa*), bay laurel (*Laurus nobilis*), dill (*Anethum graveolens*), Izmir thyme (*Origanum onites*), lavender (*Lavandula angustifolia*), Fennel (*Foeniculum vulgare*), St. John's wort (*Hypericum perforatum*), peppermint (*Mentha piperita*), tobacco (*Nicotiana tabacum*)] prepared in three different solvents (methanol, hot water, and cold water) against the larvae of the third instar of the Khapra beetle [*Trogoderma granarium* Everts (Col.:Dermestidae)].

MATERIALS AND METHODS

Cultivation of Trogoderma granarium used in bioassay

In this study, the 3rd larval stage of the Khapra beetle, *Trogoderma granarium* Everts (Col.: Dermestidae), one

of the most common pests of stored grain in Türkiye, was used. The larvae used for the biological tests were obtained from the stock culture in the Entomology Laboratory of Kahramanmaraş Sütçü İmam University, Faculty of Agriculture, Department of Plant Protection.

Soft bread wheat served as food for the breeding of *T. granarium*. To prevent contamination by insects, the wheat was stored in a freezer at -20 °C for one week (Tefera et al. 2010). To extract the insect eggs, 100-200 adult insects were placed in jars containing 300-400 g of wheat and 5% dry yeast. These jars were then placed in an air-conditioned chamber for 3-4 days to allow the adult insects to lay their eggs. After this period, the jars containing the adult *T. granarium* were sieved using 500 µm and 212 µm sieves. The larger sieve collected the wheat, the smaller sieve retained the insects and eggs, while the flour was collected in a separate container.

The eggs and insects collected in the 212 µm sieve were subjected to a further sieving process in order to separate them. The isolated eggs were transferred to 650 ml glass bottles filled with prepared wheat. These glass containers were covered with breathable gauze to allow air circulation, and incubated in the dark at 30±1 °C and 65±5% humidity. Their development was monitored regularly. When a new generation of adults was observed, they were screened for contamination and relocated to uncontaminated wheat to ensure the continuity of the culture. This procedure was maintained carefully throughout the study.

Collection of plants and preparation of extracts

The plants whose efficacy was determined in the study, their families, the plant parts used, and the types of solvents used for extraction are listed in Table 1.

The seeds of fennel (*Foeniculum vulgare*), dill (*Anethum graveolens*), and black cumin (*Nigella sativa*) used in this study were obtained from a commercial market in June 2021. The flowers of St. John's wort (*Hypericum perforatum*) (during the flowering period of the plant) and the leaves of laurel (*Laurus nobilis*) were collected from Samsun, Atakum Çakırlar district between June-July 2020. The lavender flowers (*Lavandula angustifolia*) (during the flowering period) were collected in Çaltıbozkır district of Mersin Silifke district. The flowers and leaves of Izmir thyme (*Origanum onites*) were collected during the flowering period from Balandız village in Mersin Silifke district. The leaves of medicinal mint (*Mentha piperita*) were collected in Kahramanmaraş 12 Şubat Gayberli district. The leaves of rosemary (*Rosmarinus officinalis*) were collected from Yeni Mahalle district of Samsun Atakum. The leaves of tobacco (*Nicotiana tabacum*) were collected from the village of Sarıkaya in the Samsun Bafra Hacı Hafızlar district.

The relevant plant parts of the tested plants were collected from the indicated locations and brought to the laboratory, then placed on blotting paper in dark rooms without direct sunlight and high humidity, and dried at room temperature (23-24 °C) for about one week. The dried plant materials were mechanically crushed using a blender (Fakir Mr. Chef Quadro). The plant powders were then filled into glass jars, labeled, and stored in the dark until used in the study.

Methanol (Merck 99.5%), hot water (100 °C), and cold water (25 °C) were used as three different solvents in biological tests.

Obtaining methanol extraction

The method described in de Souza Tavares et al. (2009) was followed for the extraction of methanol extracts from the selected plants. Each plant material was weighed exactly 100 grams using a precision balance (OHAUS Pioneer,

Table 1. Information about the plants used in the study.

	Scientific name	Common name	Family	Part used	Solvent used
1.	<i>Foeniculum vulgare</i> Mill.	Fennel	Apiaceae	Seed	
2.	<i>Anethum graveolens</i> L.	Dill	Apiaceae	Seed	
3.	<i>Nigella sativa</i> L.	Black cumin	Ranunculaceae	Seed	
4.	<i>Hypericum perforatum</i> L.	St. John's Wort	Clusiaceae	Flower	Methanol
5.	<i>Lavandula angustifolia</i> Mill.	Lavender	Lamiaceae	Flower	Hot water
6.	<i>Origanum onites</i> L.	Izmir thyme	Lamiaceae	Flower+Leaf	Cold water
7.	<i>Mentha piperita</i> L.	Medicinal mint	Lamiaceae	Leaf	
8.	<i>Rosmarinus officinalis</i> L.	Rosemary	Lamiaceae	Leaf	
9.	<i>Laurus nobilis</i> L.	Laurel	Lauraceae	Leaf	
10.	<i>Nicotiana tabacum</i> L.	Tobacco	Solanaceae	Leaf	
11.	Azadirachtin	Nimbecidine		790 g/l Neem oil + 0.3 g/l	

Merck KGaA, Darmstadt, Germany). These weighed plant materials were then placed into 1000 ml autoclave bottles, to which 600 ml of methanol (Merck 99.5%) was added as an organic solvent.

The samples that were prepared were subjected to a 24-hour shake at room temperature at a speed of 120 rpm on an orbital shaker (Daihan SHO-2D, Hanoi, Vietnam). After the shake period, the suspensions of each plant were filtered separately using filter paper (Whatman Filter Paper No. 1) to remove the liquid part of the suspension and discard the pulpy residue. After filtration, the methanol in the resulting liquid was removed using a vacuum rotary evaporator (Heidolph Rotovap, Shanghai, China) at 170 rpm for 1 hour at 40 ± 2 °C. The extracts obtained were placed in a water bath at 42 °C for 24 hours to ensure complete evaporation of the residual methanol, so that a pure extract was obtained after these procedures.

The plant extracts were stored in amber-colored vials sealed with plastic lids, where in the methanol was evaporated separately for each plant. These vials were stored in the refrigerator at a temperature of +4 °C until use. When needed, the solid extracts were dissolved with 10% acetone (Sigma-Aldrich) in water (v/v) to reach the target concentration (20% w/v) established for the study.

Obtaining cold and hot water extractions

To prepare cold water extracts, 20 g of each plant material was placed in an Erlenmeyer for 20% (w/v) solution and 80 ml of pure water at 25 °C. These solutions were then shaken in a shaker at 100 rpm for 24 hours at 4 °C. The resulting plant-water mixtures were successively sieved through cheesecloth and a 38-micron sieve (400 mesh), and collected in a beaker. These solutions were then transferred to tubes of 15 ml volume, centrifuged at 5000 rpm for 10 minutes and the supernatant of the solutions was passed through Whatman filter paper (No. 1). The extracts thus obtained were filled into white 500 ml plastic bottles and stored in a refrigerator at +4 °C until use (Dura and Kepenekçi 2022, Parwinder 1989).

For the hot water extracts, the plant-water mixtures were boiled at 100 °C for 10 minutes in the indicated ratio of dry plant material and pure water. After boiling, these solutions were successively filtered through cheesecloth and Whatman filter paper (No. 1). The resulting hot water extracts were carefully poured into white 500 ml plastic bottles and stored in a refrigerator at +4 °C until use.

Determination of insecticidal effects of plant extracts against *Trogoderma granarium* larvae

The insecticidal activity of the extracts of the plants used in the study, obtained at a concentration of 20% in 3 different solvents, was tested against 3rd instar *T. granarium* larvae (8-10 days old).

A soft wheat variety (*Triticum aestivum* L. Poaceae) with a moisture content of $11 \pm 1\%$ was used for the biological tests. Before the experimental units were set up, the insect feed (common wheat variety) was sterilized by storing it in a freezer at -20 °C for one week to prevent possible contamination by insects. All experiments were performed randomly, with 5 replicates and 10 larvae in each replicate. A control group was formed for each treatment. Two separate control groups were formed for the extract experiments. The preparation of Nimbecidin (790 g/l neem oil + 0.3 g/l Azadirachtin) was used as a positive control and pure water as a negative control.

To test the effect of the plant extracts on insect mortality, plastic containers of a volume of 100 ml were used. For both pests, 10 g of wheat was weighed into each container using a precision balance and made available for feeding. The solution at the target concentration was mixed with a vortex device (WiseMix VM-10, Wertheim, Germany) for 1 minute before use. 2 ml of the extract solution taken from the target concentration solution was sprayed evenly onto the feed in all jars except the control group. The solution was then stirred with a glass cylinder to ensure uniform mixing of the extracts with the wheat grains. For the control group, 2 ml of pure water was sprayed onto 10 g of feed in plastic jars. After 10 larvae were placed in each jar, the plastic jars were labeled and covered with a muslin cloth to prevent the larvae from escaping. The jars were placed in a climatic cabinet with a temperature of 30 °C and a relative humidity of $70 \pm 5\%$ (Panezai et al. 2019).

After the biological tests, the dead and live larvae were counted on the 14th day of treatment and the data recorded. During the counting, the insects in the plastic jars were touched individually with a fine-tipped brush and observed to see whether they were alive or not. Those that were motionless were considered dead, while those that barely moved were considered alive. The dead insects were kept for 24 hours after the count to see if there was any sign of movement. The same procedure was repeated for the control groups.

Evaluation and analysis of data

As a result of the biological tests on wheat, the mortality of the tested insect species was analyzed according to the Abbott formula (Abbott 1925), and the percentage mortality rates were determined. A one-way analysis of variance

(one-way ANOVA) was applied to the data resulting from the variation of the biological tests. In addition, statistical differences between treatments were compared using Tukey's test at $P \leq 0.05$. All statistical analyses were performed using Minitab software.

RESULTS

The insecticidal activity of the extracts of the plants used in the study at 20% concentration in 3 different solvents was tested 14 days after application against 3rd instar *T. granarium* larvae, and the findings obtained are given in Table 2.

The analysis revealed significant effects of both different plant treatments and different solvents on the mortality rate of *T. granarium* in the 3rd larval instar (for plant: $F_{10,152}=6.48$, $P=0.000$; for solvent: $F_{2,152}=87.61$, $P=0.000$). There was also a statistically significant interaction between the plant and the solvent ($F_{20,132}=13.03$, $P=0.000$). When the mortality rates of the larvae treated with hot water extracts were compared with the control group, significant differences were found between the treatments ($F_{11,48}=13.35$; $P=0.000$). Similar significant differences were found when examining

the mortality rates of the larvae treated with cold water ($F_{11,48}=8.40$; $P=0.000$) and methanol extracts ($F_{11,48}=111.38$; $P=0.000$) compared to the control group.

Examination of the overall mortality rates of the noxious larvae of the hot water extracts of various plants showed that the mortality rates of the plants *Azadirachta indica* A. Juss (Meliaceae), *F. vulgare*, *H. perforatum*, *L. nobilis*, *N. tabacum*, and *O. onites* were statistically in the same group as those of the others. In contrast, the mortality rates of the plants *L. angustifolia* and *N. sativa*, which were in different groups, were statistically significantly lower. The mortality rates of *A. graveolens*, *M. piperita*, and *R. officinalis* were also statistically in the same group, but their mortality rates were statistically significantly lower than those of all other plants. For the larvae of *T. granarium*, the mortality rates of the cold-water extracts of all plants were statistically in the same group. On the other hand, the mortality rates of the methanol extracts of *A. graveolens* and *N. tabacum* were statistically in the same group, which means that the mortality rates of the larvae were statistically significantly higher than the mortality rates of all other extracts (Table 2).

Table 2. Mean percentage mortality rates of 20% concentration of all plant extracts on *Trogoderma granarium* on the 14th day of the application

Plants	Extracts			F Value	P Value
	Hot Water	Cold Water	Methanol		
<i>A. graveolens</i>	28±5.83Cb*	32±3.74Ab	100±0.00Aa	$F_{2,12}=102.33$	$P=0.000$
<i>F. vulgare</i>	34±2.45BCb	32±4.90Ab	62±2.00BCDa	$F_{2,12}=24.82$	$P=0.000$
<i>H. perforatum</i>	34±2.45BCb	36±4.00Ab	52±2.00DEa	$F_{2,12}=11.23$	$P=0.002$
<i>L. nobilis</i>	30±3.16BCa	40±5.48Aa	44±4.00EFa	$F_{2,12}=2.79$	$P=0.101$
<i>L. angustifolia</i>	46±2.45ABb	40±4.47Ab	64±2.45BCa	$F_{2,12}=14.62$	$P=0.001$
<i>M. piperita</i>	26±2.45Cb	30±5.48Aab	44±2.45EFa	$F_{2,12}=6.38$	$P=0.013$
<i>N. tabacum</i>	30±4.47BCb	36±2.45Ab	92±3.74Aa	$F_{2,12}=87.70$	$P=0.000$
<i>N. sativa</i>	56±5.10Ab	44±2.45Ab	70±0.00Ba	$F_{2,12}=15.88$	$P=0.000$
<i>O. onites</i>	38±3.74BCb	26±5.10Ab	58±2.00CDa	$F_{2,12}=17.82$	$P=0.000$
<i>R. officinalis</i>	26±2.45Cb	26±2.45Ab	56±2.45CDa	$F_{2,12}=50.00$	$P=0.000$
Positive Control (<i>A. indica</i>)	36±2.45BCa	36±2.45Aa	36±2.45Fa	$F_{2,12}=0.00$	$P=1.000$
Negative Control (Natural death)	2±2.45Da	0±0.00Ba	2±2.00Ga	$F_{2,12}=0.50$	$P=0.619$
F Value	$F_{11,48}=13.35$	$F_{11,48}=8.40$	$F_{11,48}=111.38$	For plant: $F_{10,152}=6.48$; $P=0.000$ For solvent: $F_{2,152}=87.61$; $P=0.000$ For Plant*Solvent: $F_{20,132}=13.03$; $P=0.000$	
P Value	$P=0.000$	$P=0.000$	$P=0.000$		

*Two-way analysis of variance (ANOVA) was applied to the data and the differences between the averages were determined by Tukey test at 5% significance level. Different capital letters in the same column and different lower case letters in the same row are statistically different from each other.

The mortality rates using hot water as a solvent administered on day 14 of the study were 28% for the extract of *A. graveolens*, 36% for the extract of *A. indica*, 34% for the extract of *F. vulgare*, 34% for the extract of *H. perforatum* extract, 30% for the *L. nobilis* extract, 46% for the *L. angustifolia* extract, 26% for the *M. piperita* extract, 30% for the *N. tabacum* extract, 56% for the *N. sativa* extract, 38% for the *O. onites* extract and 26% for the *R. officinalis* extract. The highest mortality rate (56%) for hot water extracts against *T. granarium* larvae was obtained for the *N. sativa* plant (Table 2).

Using cold water as a solvent administered on day 14 of the study, the mortality rates (%) for *A. graveolens*, *A. indica*, *F. vulgare*, *H. perforatum*, *L. nobilis*, *L. angustifolia*, *M. piperita*, *N. tabacum*, *N. sativa*, *O. onites* and *R. officinalis* were 32, 36, 32, 32, 32, 36, 36, 40, 40, 40, 30, 36, 44, 26 and 26 plants, respectively. *N. sativa* caused the highest mortality rate (44%) among the cold-water extracts on *T. granarium* larvae (Table 2).

When methanol was used as a solvent on day 14 of the study, the percent mortality rates were 100, 36, 62, 52, 44, 64, 44, 92, 70, 58 and 56 for *A. graveolens*, *A. indica*, *F. vulgare*, *H. perforatum*, *L. nobilis*, *L. angustifolia*, *M. piperita*, *N. tabacum*, *N. sativa*, *O. onites* and *R. officinalis*, respectively. As a result of the treatments, it was found that the most effective plant extract against the larvae was the methanol extract of *A. graveolens* and a 20% dose of this extract completely killed the larvae of the pest (Table 2).

DISCUSSION

Research into the properties and effects of plant extracts holds the great promise of obtaining chemical raw materials from our natural resources in a more cost-effective and sustainable way, thereby achieving considerable economic benefits. Research into these properties must be prioritized as part of good agricultural practice, with the aim of developing, producing, advocating, and promoting the widespread use of natural plant extracts. These extracts can serve as viable alternatives to synthetic pesticides, promote healthier food production, and potentially improve international trade in agricultural products. Prioritizing the use of these extracts will not only contribute to a healthier food supply but also improve global agricultural trade.

In contrast to the active ingredients used in chemical control of stored product pests, biopesticides derived from medicinal plant products showed a lower resistance to stored product pests, did not produce toxic residues, persist within the plant, and exhibit lower toxicity to mammals and

the environment. Many researchers had shown in studies on the control of stored product pests that these products were effective in different ways (Isman 2006, Koul 2008).

The results of all biological tests showed that the plant extracts prepared with different solvents exhibited varying degrees of insecticidal activity against the larvae of the 3rd instar of *T. granarium*. A closer look at the study results revealed that these statistical differences in lethal efficacy depended on several factors, such as the specific plant variety and the types of solvents used in the preparation of the extract. This observation is supported by numerous scientific studies that have also emphasized the influence of these variables on the insecticidal efficacy of plant extracts. Sarmamy et al. (2011) reported a mortality rate of 1.54% in *T. granarium* larvae 96 hours after application of a 6% concentration of *N. tabacum* water extract. Zia-ul-Haq et al. (2014) tested the lethal effect of 7 different plant leaf or seed extracts, including *A. indica*, on *T. granarium* and reported that the mortality rate was 24.69% at a concentration of 15%. In agreement with these studies, comparable results were observed in this study. Thus, the use of the cold-water extract of *N. tabacum* at a concentration of 5% resulted in a mortality rate of 2% in larvae four days after application. In contrast, the use of an *A. indica* extract at a concentration of 15% resulted in a 22% mortality rate in the larvae of *T. granarium* on the tenth day after application. Considering these collective results, it was evident that the mortality rate of *T. granarium* larvae was generally relatively low. Eliopoulos (2013) found that the larvae of *T. granarium* have the potential to live in unsuitable environments and can resist many typical insecticides. In addition, Vadivambal et al. (2007) found that the dense hairiness of the larval body forms a protective barrier that prevents direct contact between insecticides and the cuticular layer. These results confirmed the low mortality rate observed in the larvae of *T. granarium* in this study. Their inherent adaptability and physical defenses contributed to their resistance to insecticidal activities, which was consistent with the observed results.

Methanol and distilled water are both polar solvents, but their polarity values are different (Awadh et al. 2008). If the polarity values of solvents are different, the variety and amounts of substances dissolved in the solvents may also vary (Çolak et al. 2020, Navarro del Hierro et al. 2021). Researchers have found that certain secondary metabolites in certain plant organs are extracted with various solvents and that the number of secondary compounds with insecticidal activity decreases when different solvents are used (Çolak et al. 2020, Karakoç and Gökçe 2012, Nawaz et al. 2020). Changes in the polarity of solvents mean that

extracts obtained from the same plants with different solvents have very different insecticidal activities. These different insecticidal activities are attributed to the effective ability of the extracts to form hydrogen bonds and eliminate free radicals. In a study conducted by Dessenbe et al. (2022), it was found that increasing the polarity of the solvents leads to an increase in the number of compounds in the plant. Extracts obtained from the same plant with different solvents had different components, and these extracts showed significant insecticidal activity against *C. maculatus* and *Sitophilus zeamais* (Motsch.) (Coleoptera: Curculionidae). Karakas (2016) reported that leaf extracts of *Anethum graveolens* and *Ocimum basilicum* L. (Lamiaceae) showed a different mortality rate of *S. granarius* beetles depending on the polarity of the solvent. Hiruy and Getu (2018) observed differences in the mortality of *S. zeamais* by the application of solvent extracts from the leaves of *Calpurnia aurea* (Ait.) Benth (Fabaceae) and *Milletia ferruginea* (Hochst) Baker (Fabaceae), depending on the polarity of the solvent. Similarly, Uddin II (2020) found that the mortality of *C. maculatus* when using plant extracts obtained with different polarities from *Trichilia heudelotii* Planch (Meliaceae) varied depending on the polarity of the solvent. As we have seen, the insecticidal activity of plant extracts varies in studies conducted with different methods and solvents with different polarities. There are many supporting studies in the literature on stored pests in this context (Aba-Toumou et al. 2016, Awadh et al. 2008, Gebreslassie and Eyasu 2019, Karakas 2016, Khan et al. 2016, Li et al. 2013, Navarro del Hierro et al. 2021, Rafińska et al. 2019, Suleiman et al. 2018, Uddin II 2020, Wakeel et al. 2019, Zhang et al. 2017, Zhang et al. 2019). In this study, it is hypothesized that the reason for the stronger insecticidal activity of methanol extracts compared to water extracts is related to all this information.

The efficacy of plant extracts as insecticides depends not only on factors such as plant species, age, insect type, and geographical location, but also on the solvents used in the extraction process (Shalan et al. 2005). Most researchers have generally favored solvents such as methanol, ethanol, acetone, and ethyl acetate in their studies on herbal extracts (Truong et al. 2019). The excessive use of these organic solvents poses health and safety risks to researchers and is not suitable for the environment. Therefore, the selection of the appropriate extraction solvent is very important (Dirar et al. 2019). There were limited studies in the literature in which hot and cold water were chosen as extraction solvents. The use of water as solvent was considered the preferred method in the extraction of extracts for human control of stored products. Since it was of great importance

to include extracts from cold and hot water commonly used by humans in scientific research using different solvents, this study was considered to be important.

In this study, the effect of extracts of 10 different plants (*R. officinalis*, *N. sativa*, *L. nobilis*, *A. graveolens*, *O. onites*, *L. angustifolia*, *F. vulgare*, *H. perforatum*, *M. piperita*, and *N. tabacum*) prepared with three different solvents (methanol, hot water, and cold water) on the third instar larvae of *T. granarium* was investigated. The following conclusions were drawn from the results.

The extracts obtained from *A. graveolens*, *N. tabacum*, and *N. sativa* showed remarkably high insecticidal activity against the larvae of *T. granarium*. These particular extracts are promising for effective control of this pest.

The *N. sativa* plant extracts, especially the 20% concentration in the variants with hot water and methanol, showed a mortality rate of more than 50%. In contrast, none of the other plant extracts, whether in hot or cold water, achieved a mortality rate of 50% or more.

The methanol extracts of *N. tabacum*, *A. graveolens*, and *N. sativa* showed mortality rates of 92%, 100%, and 70%, respectively. In contrast, the methanol extracts of the other plants consistently did not exceed a mortality rate of 70%.

The overarching observations indicate that methanol was found to be the most effective solvent for extracting the insecticidal properties of these plants, followed by hot water and cold water in descending order of effectiveness. However, more comprehensive studies should be conducted to determine the applicability of such applications in practice and to establish their applicability on an industrial scale.

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Author's Contributions

Authors declare the contribution of the authors is equal.

Statement of Conflict of Interest

The authors have declared no conflict of interest.

ÖZET

Çalışmanın amacı; Türkiye'nin değişik illerinden toplanan 10 farklı bitkinin [*Rosmarinus officinalis* L. (Lamiaceae), *Nigella sativa* L. (Ranunculaceae), *Laurus nobilis* L. (Lauraceae), *Anethum graveolens* L. (Apiaceae),

Origanum onites L. (Lamiaceae), *Lavandula angustifolia* Mill. (Lamiaceae), *Foeniculum vulgare* Mill. (Apiaceae), *Hypericum perforatum* L. (Clusiaceae), *Mentha piperita* L. (Lamiaceae) ve *Nicotiana tabacum* L. (Solanaceae)] 3'er farklı çözücüde (metanol, sıcak su ve soğuk su) oluşturulan ekstraktlarının *Trogoderma granarium* Everts (Coleoptera: Dermestidae)'un 3. dönem larvalarına karşı toksisitesini belirlemektir. Çalışma sonuçları; bitki türüne ve kullanılan çözücüye göre değişiklik göstermiştir. Yapılan gözlemler sonucunda genellikle etkili çözücü, metanol olarak belirlenmiş ve bunu sırasıyla sıcak su ve soğuk su çözücülerini takip etmiştir. Uygulamanın 14. gününde bitki ekstraktlarının %20 (w/v) konsantrasyonunda çözücü olarak metanol kullanıldığında en yüksek ölüm oranı %100 olarak belirlenirken; bu oran soğuk su kullanıldığında %44 ve sıcak su kullanıldığında ise %56 olarak tespit edilmiştir. Ayrıca; araştırma sonuçlarına göre, *A. graveolens*, *N. tabacum* ve *N. sativa* bitkilerine ait ekstraktların zararlı üzerinde yüksek toksik etki gösterdikleri belirlenerek bu ekstraktların depolanmış ürün zararlıların mücadelesinde oldukça umut verici olduğu düşünülmektedir. Ancak, bu uygulamaların pratikte kullanılabilirliğini kesinleştirmek ve endüstriyel ölçekte uygulanabilirliğini belirlemek için daha kapsamlı çalışmalara ihtiyaç duyulmaktadır.

Anahtar kelimeler: depolanmış ürün zararlısı, bitki ekstraktı, sıcak su, soğuk su, metanol

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5. Bitki Koruma Bülteni'ne gönderilen makaleler, daha önce herhangi bir yayın organında yayınlanmamış veya aynı zamanda başka bir yayın organında değerlendirme aşamasında olmamalıdır.
6. Lisansüstü tezler veya TÜBİTAK, DPT, TAGEM, BAP gibi çeşitli kurumlarca desteklenen projelerin sonuçlarından kısımlar içeren eserler ilgililerinden gerekli izinler alındıktan sonra yayına hazırlanmalı, bu durum teşekkür kısmında mutlaka belirtilmelidir.
7. Bitki Koruma Bülteni'nde yayınlanması istenilen eserler için makale başvurusu DERGİPARK sistemi (<http://dergipark.gov.tr/bitkorb>) üzerinden yapılmalıdır.
8. Sisteme yüklenen makale "Yazarlar için" sekmesinde yer alan "Makale taslağı"na göre hazırlanmalı, sisteme "Makale giriş sayfası" ve tüm yazarlar tarafından doldurulup imzalanan "Bitki Koruma Bülteni Telif Hakkı Devir Formu" ve "Çıkar Çakışması ve Hakem Önerileri Formu" ile birlikte yüklenmelidir.
9. Bitki Koruma Bülteni'nde kör hakemlik değerlendirme süreci izlenmektedir.
10. Değerlendirme sürecine dahil edilen makaleler konu editörü ve belirlenen hakemler tarafından incelenip, onların önerileri doğrultusunda yazarları tarafından düzeltildikten sonra yayınlanır.
11. Bitki Koruma Bülteni'nde yayınlanan makaleler için baskı ücreti alınmamaktadır.

