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Original article (Orijinal araştırma)

**A study of Ichneumonidae (Hymenoptera) from Northeastern Anatolia
III, with new records and description male of *Temelucha
pseudocaudata* Kolarov, 1982**

Kuzeydoğu Anadolu'dan Ichneumonidlerle ilgili bir çalışma, yeni kayıtlar ve *Temelucha
pseudocaudata* Kolarov, 1982'nin erkeğinin tanımı

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Summary

Forty-four ichneumon species belonging to Anomaloninae, Banchinae, Cremastinae, Ctenopelmatinae, Diplazontinae, Metopiinae, Ophioninae, Pimplinae, Tryphoninae and Xoridinae subfamilies were collected from Northeastern Anatolia, of Turkey in 2015. The newly discovered male of *Temelucha pseudocaudata* Kolarov, 1982 is described. Eight species are recorded for the first time from Turkey: *Ctenochira meridianator* Aubert, 1969, *Diplazon deletus* (Thomson, 1890), *Exochus flavifacies* Kusigemati, 1984, *Homotropus pallipes* (Gravenhorst, 1829), *Odontocolon rufiventris* (Holmgren, 1860), *Sussaba cognata* (Holmgren, 1858), *Triclistus congener* (Holmgren, 1858) and *Tromatobia lineatoria* (Villers, 1789). New data on the distribution of 36 known species is also reported. Additionally, a short zoogeographical characterization is given for each of the species.

Keywords: Fauna, Ichneumonidae, Northeastern Anatolia, zoogeographical notes

Özet

Anomaloninae, Banchinae, Cremastinae, Ctenopelmatinae, Diplazontinae, Metopiinae, Ophioninae, Pimplinae, Tryphoninae ve Xoridinae altfamilyalarına ait 44 ichneumonid türü, 2015 yılında Kuzeydoğu Anadolu'dan toplanmıştır. *Temelucha pseudocaudata* Kolarov, 1982 türünün erkeğinin tanımı ilk defa yapılmıştır. Sekiz tür Türkiye'den yeni kayıt olarak verilmiştir. Bunlar: *Ctenochira meridianator* Aubert, 1969, *Diplazon deletus* (Thomson, 1890), *Exochus flavifacies* Kusigemati, 1984, *Homotropus pallipes* (Gravenhorst, 1829), *Odontocolon rufiventris* (Holmgren, 1860), *Sussaba cognata* (Holmgren, 1858), *Triclistus congener* (Holmgren, 1858) ve *Tromatobia lineatoria* (Villers, 1789)'dir. Bilinen 36 tür için yeni lokasyonlar eklenirken, her bir tür için de zoocoğrafik notlar da verilmiştir.

Anahtar sözcükler: Fauna, Ichneumonidae, Kuzeydoğu Anadolu, zoocoğrafik notlar

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Received (Alınış): 14.11.2016

Accepted (Kabul ediliş): 02.03.2017

Published Online (Çevrimiçi Yayın Tarihi): 25.04.2017

Introduction

Hymenoptera is one of the few mega diverse insect orders. Some 300 thousand to 2.5 million hymenopteran species are estimated to exist worldwide and nearly 115 thousand species have been described (Stork, 1988; La Salle & Gauld, 1992; Gauld & Gaston, 1995; Grissell, 1999).

The order Hymenoptera is an important group in class Insecta as it contains agriculturally, ecologically and economically significant species. They are also ecological indicators. Hymenopteran parasitoids are potential biocontrol agents of agricultural pests (Anbalagan et al., 2015).

Ichneumonidae is a family within the order Hymenoptera and are commonly called ichneumon wasps. As larvae, they parasitize a wide range of hosts, most frequently the larvae and pupae of the larger holometabolous insect orders (Coleoptera, Diptera, other Hymenoptera and Lepidoptera), although a small number also attack the immatures of other holometabolous insects such as Trichoptera (Agriotypinae), Mecoptera (a few Campopleginae), Raphidioptera (a few Campopleginae), and Neuroptera (Brachycyrtinae, some Cryptinae) (Eberhard, 2000).

Despite the abundance, diversity and ecological importance of Ichneumonidae, faunistic studies are not yet adequate in Turkey. The number of species of Ichneumonidae recorded in the Ichneumonidae World catalog Turkey is 1056. An intense series of studies over the last 3 years (Çoruh & Kolarov, 2013, 2016; Çoruh & Özbek, 2013; Çoruh et al., 2013, 2014a, b, c; Kolarov et al., 2014a, b, c, 2015, 2016; Özdan, 2014; Riedel et al., 2014; Yaman, 2014; Yurtcan & Kolarov, 2015; Çoruh & Çalmaşur, 2016) has raised this number to 1220. With eight new records in this paper, the number is now 1228.

The work describe here is recent progress in an ongoing project (Kolarov et al., 2014a, b, 2015, 2016; Çoruh et al., 2014a, b). The objective of this study was to determine the faunal richness in the study area, provide habitat and associated plant data for collected insects, and contribute to the knowledge of Ichneumonidae distribution in Turkey, and also describe the male of *Temelucha pseudocaudata* Kolarov, 1982.

Material and Methods

Sampling and collection method

Adult specimens, 182 in total, were collected from various habitats in Erzincan, Erzurum, Giresun, Gümüşhane, Ordu, Rize and Trabzon Provinces in Northeastern Anatolia, Turkey (Figure 1, Table 1). The ichneumonid specimens were collected on flowering plants by insect net in July 2015. All samples were collected by the first two authors. The identified samples were deposited in the collection of the University of Plovdiv (Bulgaria). Classification, nomenclature, distributional data and associated plants of some species follow Yu et al. (2012). Plant samples were also collected and determined according to Davis (1965-1988) by the third author, and deposited in the herbarium of the Department of Plant Protection of Atatürk University (Erzurum).

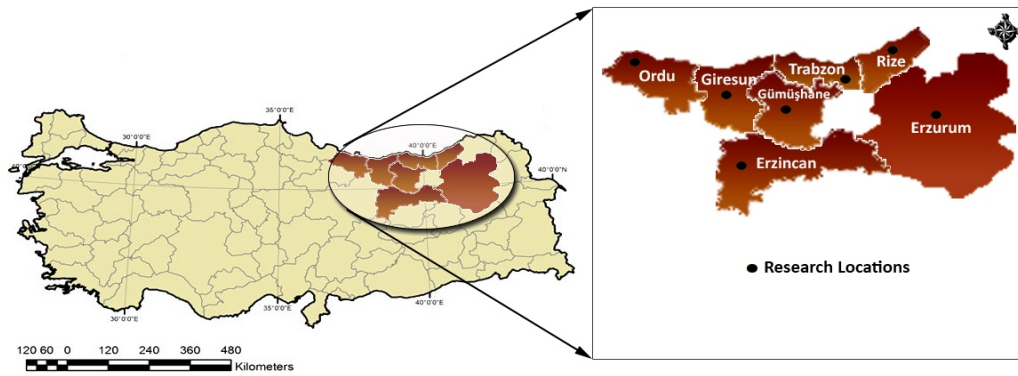


Figure 1. Research locations in Northeastern Anatolia, Turkey.

Study area

Collection areas with geographic coordinates, altitude and habitat details are shown in Table 1 and vegetation data for each location in Table 2.

Table 1. Collection areas in seven provinces in Northeastern Anatolia, Turkey

Collection areas			Coordinates	Altitude (m)	Habitat
Province	District	Locality			
Erzincan	Avcılar		39°36.899' N, 39°49.328' E	1221	Hillside, dominant plant <i>Medicago sativa</i> L.
	Merkez	Ahmetli	39°53.481' N, 39°21.197' E	1988	Roadside, semi-wet mown pasture
		Pöske Mountain	39°51.160' N, 39°21.515' E	1838	Roadside, oak area (red soil)
Erzurum	Aşkale	Kandilli	39°57.420' N, 40°52.550' E	1904	Roadside, meadow area near water channel, dominant plant <i>M. sativa</i>
	Aşkale	Tepebaşı Valley	39°51.741' N, 40°37.454' E	2009	Roadside, dry land, pasture, dominant plant <i>Quercus petraea</i> (Mattuschka) Liebl.
	Merkez	Gelinkaya	40°01.741' N, 40°54.855' E	1803	Roadside, dominant plants <i>Salix triandra</i> L., <i>Populus tremula</i> L. and <i>M. sativa</i>
Giresun	Bulancak		40°55.984' N, 38°13.045' E	16	<i>Corylus avellana</i> L.
	Eynesil	Kekiktepe	41°02.723' N, 39°05.641' E	4	Seaside, hazelnut garden on highest peak
	Keşap	Yolağzı	40°55.998' N, 38°34.578' E	1	Roadside, half shade, semi-wet, hazelnut garden
Gümüşhane	Kelkit	Köycük	40°08.584' N, 39°25.354' E	1393	Roadside, garden areas
Ordu	Turnasuyu	Turnasuyu	40°58.572' N, 37°58.577' E	1	Roadside, mown pasture, shade, hazelnut garden
Rize	İkizdere, Çamlık	İkizdere, Çamlık	40°42.979' N, 40°37.017' E	1099	Mown and unmown pasture, dominant plant <i>Ulmus glabra</i> Huds.
Trabzon	Yomra	Yomra	40°56.365' N, 39°52.131' E	20	Roadside, 20 m above the road, shade, hazelnut garden with cut herbs

Table 2. Vegetation at the collection areas in seven provinces in Northeastern Anatolia, Turkey

Plant species	EA	EmA	EP	EAK	EAT	EmG	GB	GEK	GKY	GKK	OT	RIC	TY
<i>Acantholimon caryophyllaceum</i> Boiss.					x								
<i>Achillea biebersteinii</i> Afan.	x												
<i>Achillea millefolium</i> L.		x				x							
<i>Agrimonia eupatoria</i> L.								x			x		x
<i>Alchemilla caucasica</i> Buser		x											
<i>Alchemilla sintenisii</i> Rotmh.												x	
<i>Allium armenum</i> Boiss. et Kotschy			x										
<i>Allium atrovioleaceum</i> Boiss.										x			
<i>Allium rotundum</i> L.					x								
<i>Alyssum repens</i> Baumq.											x		
<i>Amaranthus retroflexus</i> L.							x	x					
<i>Anchusa leptophylla</i> Roemer et Schulters			x	x									
<i>Anthemis tinctoria</i> L.					x		x				x		
<i>Arctium minus</i> (Hill) Bernh.												x	
<i>Artemisia austriaca</i> Jacq.			x										
<i>Artemisia vulgaris</i> L.							x		x		x		
<i>Asperula orientalis</i> Boiss. et Hohen.	x			x									
<i>Astragalus lagurus</i> Willd.					x								
<i>Astragalus lineatus</i> Lam.						x							
<i>Astragalus microcephalus</i> Willd.					x								
<i>Astrodaucus orientalis</i> (L.) Drude													
<i>Bromus arvensis</i> L.						x							
<i>Bromus japonicus</i> Thunb.			x	x									
<i>Bupleurum falcatum</i> L.			x										
<i>Bupleurum rotundifolium</i> L.				x	x					x			
<i>Calystegia sepium</i> (L.) R. Br.							x	x					x
<i>Campanula rapunculooides</i> L.												x	
<i>Campanula stevenii</i> Bieb.						x							
<i>Carduus nutans</i> L.		x											
<i>Carex panicea</i> L.		x											
<i>Carum carvi</i> L.				x	x								
<i>Centaurea pseudoscabiosa</i> Boiss. et Buhse				x									
<i>Centaurea solstitialis</i> L.	x									x			
<i>Centaurea virgata</i> Lam.			x										

EA: Erzincan, Avclar; EmA: Erzincan, Merkez, Ahmetli; EP: Erzincan, Pöske Mountain; EAK: Erzurum, Aşkale, Kandilli; EAT: Erzurum, Aşkale, Tepebaşı Valley; EmG: Erzurum, Merkez, Gelinkaya; GB: Giresun, Bulancak; GEK: Giresun, Eynesil, Kekiktepe; GK: Giresun, Keşap, Yolağzı; GKK: Gümüşhane, Kelkit, Köycük; OT: Ordu, Turnasuyu; RIÇ: Rize, İkizdere, Çamlık; TY: Trabzon, Yomra.

Table 2. (Continued)

Plant species	EA	EmA	EP	EAK	EAT	EmG	GB	GEK	GKY	GKK	OT	RIÇ	TY
<i>Cephalaria procera</i> Fisch. et Lall.		x			x	x							
<i>Cerastium glomeratum</i> Thuill.											x		
<i>Cerintho minor</i> L.	x	x											
<i>Chenopodium album</i> L.							x	x					
<i>Chenopodium vulvaria</i> L.										x			
<i>Chondrilla juncea</i> L.			x										
<i>Cichorium intybus</i> L.	x	x	x			x		x			x		
<i>Cirsium arvense</i> (L.) Scop.		x		x	x	x				x			
<i>Cirsium echinus</i> (Bieb.) Hand.-Mazz.				x						x			
<i>Commelina communis</i> L.								x					
<i>Conium maculatum</i> L.							x						
<i>Convolvulus arvensis</i> L.		x		x						x			
<i>Convolvulus galaticus</i> Rostan ex Choisy										x			
<i>Conyza canadensis</i> (L.) Cronquist							x	x	x		x		
<i>Coronilla varia</i> L.					x	x				x			
<i>Corylus avellana</i> L.							x	x	x		x		x
<i>Crataegus orientalis</i> Pallas ex Bieb.					x								
<i>Crepis armena</i> DC.		x	x		x								
<i>Crepis vesicaria</i> L.											x		
<i>Cynanchum acutum</i> L.			x										
<i>Cynodon dactylon</i> (L.) Pers.				x									
<i>Dactylis glomerata</i> L.		x		x	x	x				x			
<i>Daucus carota</i> L.	x				x			x		x			
<i>Delphinium cyphoplectrum</i> Boiss.					x								
<i>Descurainia sophia</i> (L.) Webb ex Prantl										x			
<i>Digitalis ferruginea</i> L.												x	
<i>Digitaria sanguinalis</i> (L.) Scop.	x												
<i>Echinops galaticus</i> Freyn			x										
<i>Echinops pungens</i> Trautv.										x			
<i>Echium italicum</i> L.	x												
<i>Echium vulgare</i> L.										x			

EA: Erzincan, Avcılar; EmA: Erzincan, Merkez, Ahmetli; EP: Erzincan, Pöske Mountain; EAK: Erzurum, Aşkale, Kandilli; EAT: Erzurum, Aşkale, Tepebaşı Valley; EmG: Erzurum, Merkez, Gelinkaya; GB: Giresun, Bulancak; GEK: Giresun, Eynesil, Kekiktepe; GK: Giresun, Keşap, Yolağzı; GKK: Gümüşhane, Kelkit, Köycük; OT: Ordu, Turnasuyu; RIÇ: Rize, İkizdere, Çamlık; TY: Trabzon, Yomra.

Table 2. (Continued)

Plant species	EA	EmA	EP	EAK	EAT	EmG	GB	GEK	GKY	GKK	OT	RIÇ	TY
<i>Epilobium angustifolium</i> L.								x	x	x	x		
<i>Equisetum ramosissimum</i> Desf.		x											
<i>Erigeron acer</i> L.									x				x
<i>Eryngium billardieri</i> Delar.	x			x	x					x			
<i>Eryngium campestre</i> L.	x												
<i>Eryngium giganteum</i> Bieb.						x							
<i>Euphorbia palustris</i> L.							x						
<i>Euphorbia peplus</i> L.									x				
<i>Euphorbia petrophila</i> C. A. Meyer		x	x										
<i>Euphorbia stricta</i> L.	x				x					x	x		
<i>Euphorbia virgata</i> Waldst. et Kit.			x			x							
<i>Falcaria vulgaris</i> Bernh.							x			x			
<i>Festuca callieri</i> (Hackel ex St.Yves) F. Markgraf apud Hayek			x										
<i>Festuca ovina</i> L.		x	x	x	x								
<i>Filipendula vulgaris</i> Moench						x							
<i>Galium verum</i> L.		x	x	x	x	x							
<i>Geranium asphodeloides</i> Burm. Fil.											x		
<i>Geranium pyrenaicum</i> Burm. Fil.												x	
<i>Geranium sanguineum</i> L.												x	
<i>Geranium tuberosum</i> L.						x							
<i>Gladiolus atrovioleaceus</i> Boiss.			x										
<i>Globularia trichosantha</i> Fisch. et Mey.					x								
<i>Grammosciadium daucooides</i> DC.		x											
<i>Helichrysum arenarium</i> (L.) Moench			x										
<i>Heracleum pastinacifolium</i> C. Koch						x							
<i>Holcus lanatus</i> L.							x		x		x		x
<i>Hypericum elongatum</i> Ledeb.		x	x	x	x					x			
<i>Hypericum perforatum</i> L.								x	x		x		
<i>Inula oculus-christi</i> L.				x									
<i>Isatis</i> sp.			x	x						x			
<i>Juncus acutus</i> L.											x		

EA: Erzincan, Avçılar; EmA: Erzincan, Merkez, Ahmetli; EP: Erzincan, Pöske Mountain; EAK: Erzurum, Aşkale, Kandilli; EAT: Erzurum, Aşkale, Tepebaşı Valley; EmG: Erzurum, Merkez, Gelinkaya; GB: Giresun, Bulancak; GEK: Giresun, Eynesil, Kekiktepe; GKY: Giresun, Keşap, Yolağzı; GKK: Gümüşhane, Kelkit, Köycük; OT: Ordu, Turnasuyu; RIÇ: Rize, İkizdere, Çamlık; TY: Trabzon, Yomra.

Table 2. (Continued)

Plant species	EA	EmA	EP	EAK	EAT	EmG	GB	GEK	GKY	GKK	OT	RIÇ	TY
<i>Koeleria cristata</i> (L.) Pers.			x										
<i>Lactuca serriola</i> L.	x			x		x	x		x	x			
<i>Lapsana communis</i> L.						x	x		x	x		x	
<i>Lathyrus pratensis</i> L.						x							
<i>Leontodon crispus</i> Vill.				x									
<i>Leontodon hispidus</i> L.								x				x	
<i>Leopoldia comosa</i> (L.) Parl.					x								
<i>Lepidium draba</i> L.		x								x			
<i>Linaria kurdica</i> Boiss. et Hohen.				x									
<i>Lolium perenne</i> L.						x	x				x		
<i>Lotus corniculatus</i> L.						x							
<i>Marrubium parviflorum</i> Fisch. et Mey.		x											
<i>Medicago lupulina</i> L.													x
<i>Medicago sativa</i> L.	x					x							
<i>Melampyrum arvense</i> L.				x	x								
<i>Melilotus alba</i> Desr.	x												
<i>Melilotus officinalis</i> (L.) Desr.				x	x								
<i>Mentha longifolia</i> (L.) Hudson		x		x								x	
<i>Nasturtium officinale</i> R. BR.									x				
<i>Onobrychis altissima</i> Grossh					x								
<i>Ononis spinosa</i> L.		x											
<i>Onopordum acanthium</i> L.										x			
<i>Origanum vulgare</i> L.												x	
<i>Papaver dubium</i> L.		x											
<i>Papaver tauricola</i> Boiss.										x			
<i>Paspalum dilatatum</i> Poir.							x	x					x
<i>Phleum pratense</i> L.		x				x							
<i>Phlomis pungens</i> Willd.				x	x								
<i>Phragmites australis</i> (Cav.) Trin. ex Steudel						x							
<i>Pilosella hoppeana</i> (Schult.) F.W. Schultz & Sch.Bip.												x	
<i>Plantago atrata</i> Hoppe		x											
<i>Plantago lanceolata</i> L.								x		x		x	

EA: Erzincan, Avcılar; EmA: Erzincan, Merkez, Ahmetli; EP: Erzincan, Pöske Mountain; EAK: Erzurum, Aşkale, Kandilli; EAT: Erzurum, Aşkale, Tepebaşı Valley; EmG: Erzurum, Merkez, Gelinkaya; GB: Giresun, Bulancak; GEK: Giresun, Eynesil, Kekiktepe; GKY: Giresun, Keşap, Yolağzı; GKK: Gümüşhane, Kelkit, Köycük; OT: Ordu, Turnasuyu; RIÇ: Rize, İkizdere, Çamlık; TY: Trabzon, Yomra.

Table 2. (Continued)

Plant species	EA	EmA	EP	EAK	EAT	EmG	GB	GEK	GKY	GKK	OT	RIÇ	TY
<i>Plantago major</i> L.							x	x	x		x	x	
<i>Poa bulbosa</i> L.	x	x		x									
<i>Poa longifolia</i> Trin.								x					
<i>Poa nemoralis</i> L.		x											
<i>Poa pratensis</i> L.							x				x	x	x
<i>Polygonum persicaria</i> L.							x		x		x		
<i>Populus tremula</i> L.						x							
<i>Potentilla argentea</i>			x		x								
<i>Prunella vulgaris</i>								x			x	x	
<i>Pteridium aquilinum</i>								x	x			x	x
<i>Quercus petraea</i> (Mattuschka) Liebl.					x								
<i>Ranunculus kotschyi</i> Boiss.												x	
<i>Rhinanthus angustifolius</i> C. C. Gmelin		x		x		x						x	
<i>Rorippa sylvestris</i> (L.) Besser								x			x		
<i>Rubus discolor</i> Weihe et Nees.								x					x
<i>Rubus hirtus</i> Waldst. et Kit.							x		x				
<i>Rumex acetosella</i> L.		x							x				
<i>Rumex crispus</i> L.				x		x	x						
<i>Salix pentandra</i> L.			x										
<i>Salix triandra</i> L.						x							
<i>Salvia candidissima</i> Vahl			x		x								
<i>Salvia sclarea</i> L.			x										
<i>Salvia verticillata</i> L.										x		x	
<i>Sambucus ebulus</i> L.							x				x		
<i>Sanguisorba minor</i> Scop.	x	x	x		x								
<i>Scabiosa argentea</i> L.	x				x								
<i>Scabiosa caucasica</i> Bieb.			x										
<i>Senecio nemorensis</i> L.									x				
<i>Senecio paucilobus</i> DC.		x											
<i>Senecio vernalis</i> Waldst. et Kit.		x											
<i>Senecio vulgaris</i> L.						x	x				x		
<i>Seseli libanotis</i> (L.) W. Koch					x	x							

EA: Erzincan, Avclar; EmA: Erzincan, Merkez, Ahmetli; EP: Erzincan, Pöske Mountain; EAK: Erzurum, Aşkale, Kandilli; EAT: Erzurum, Aşkale, Tepebaşı Valley; EmG: Erzurum, Merkez, Gelinkaya; GB: Giresun, Bulancak; GEK: Giresun, Eynesil, Kekiktepe; GK Y: Giresun, Keşap, Yolağzı; GKK: Gümüşhane, Kelkit, Köycük; OT: Ordu, Turnasuyu; RIÇ: Rize, İkizdere, Çamlık; TY: Trabzon, Yomra.

Table 2. (Continued)

Plant species	EA	EmA	EP	EAK	EAT	EmG	GB	GEK	GKY	GKK	OT	RIÇ	TY
<i>Setaria viridis</i> (L.) P. Beauv.							x						
<i>Silene vulgaris</i> (Moench) Garcke	x	x			x								
<i>Solanum nigrum</i> L.								x					
<i>Sonchus arvensis</i> L.				x									
<i>Sonchus oleraceus</i> L.							x	x				x	
<i>Sorghum halepense</i> (L.) Pers.							x		x				
<i>Stachys sylvatica</i> L.									x				
<i>Stipa pulcherrima</i> C. Koch					x								
<i>Tanacetum balsamita</i> L.		x											
<i>Tanacetum macrophyllum</i> (Waldst. et Kit.) Schultz-Bip.													x
<i>Taraxacum crepidiforme</i> DC.		x										x	
<i>Telekia speciosa</i> (Schreber) Baumg.									x				
<i>Teucrium orientale</i> L.				x									
<i>Tragopogon dubius</i> Scop.						x							
<i>Trifolium montanum</i> L.					x								
<i>Trifolium pratense</i> L.						x		x			x	x	x
<i>Trifolium repens</i> L.												x	x
<i>Turgenia latifolia</i> (L.) Hoffm.				x									
<i>Ulmus glabra</i> Huds.												x	
<i>Urtica dioica</i> L.							x	x			x	x	
<i>Verbascum cheiranthifolium</i> Boiss.			x		x					x			
<i>Verbascum oreophilum</i> C. Koch		x	x										
<i>Verbascum speciosum</i> Schrader				x									
<i>Vicia cracca</i> L.				x		x		x	x				x
<i>Xanthium strumarium</i> L.							x			x	x		
<i>Xeranthemum annuum</i> L.	x		x	x	x					x			

EA: Erzincan, Avcılar; EmA: Erzincan, Merkez, Ahmetli; EP: Erzincan, Pöske Mountain; EAK: Erzurum, Aşkale, Kandilli; EAT: Erzurum, Aşkale, Tepebaşı Valley; EmG: Erzurum, Merkez, Gelinkaya; GB: Giresun, Bulancak; GEK: Giresun, Eynesil, Kekiktepe; GK Y: Giresun, Keşap, Yolağzı; GKK: Gümüşhane, Kelkit, Köycük; OT: Ordu, Turnasuyu; RIÇ: Rize, İkizdere, Çamlık; TY: Trabzon, Yomra.

Results

In the present study, a total of 44 species were identified. These species belong to Anomaloniinae, Banchinae, Cremastinae, Ctenopelmatinae, Diplazontinae, Metopiinae, Ophioninae, Pimplinae, Tryphoninae and Xoridinae subfamilies. Eight species, marked in the text by an asterisk, are new records for Turkish fauna. The newly discovered male of *T. pseudocaudata* is described.

Subfamily Anomaloninae Viereck, 1918

Agrypon gracilipes (Curtis, 1839)

Material examined: Giresun: Keşap, Yolağzı, 25.VII.2015, 2 ♀♀; Ordu: Turnasuyu, 24.VII.2015, ♀.

Distribution in Turkey: Ankara and Bayburt (Özdemir & Kılınçer, 1990; Çoruh et al., 2004, 2014c).

Distribution in World: Palearctic region.

Associated plants: *Foeniculum vulgare* Miller

Anomalon cruentatum (Geoffroy, 1785)

Material examined: Erzincan: Pöske Mt. 23.VII.2015, ♂; Erzurum: Aşkale, Tepebaşı Valley, 23.VII.2015, ♀; Gümüşhane: Kelkit, Köycük, 23.VII.2015, ♀.

Distribution in Turkey: Adana, Adıyaman, Afyon, Antalya, Batman, Bayburt, Bingöl, Diyarbakır, Edirne, Elazığ, Erzincan, Erzurum, Gaziantep, Hatay, Iğdır, İstanbul, Isparta, Kahramanmaraş, Kars, Kırklareli, Malatya, Mardin, Muğla, Tekirdağ and Tunceli (Kolarov et al., 1994, 2002, 2014a; Çoruh et al., 2004; Gürbüz, 2005; Boncukçu, 2008; Kırtay, 2008; Birol, 2010; Gürbüz et al., 2011; Çoruh & Kolarov, 2016; Özdan & Gürbüz, 2016).

Distribution in World: Azerbaijan, Turkey, Kazakhstan and Tajikistan.

Associated plants: *Anthriscus sylvestris* (L.) Hoffm. and *Peucedanum oreoselinum* (L.) Moench.

Subfamily Banchinae Wesmael, 1845

Lissonota (Loxonota) flavovariegata (Lucas, 1849)

Material examined: Erzincan: Ahmetli, 23.VII.2015, 6 ♂♂, ♀, Avcılar (Figure 2), 23.VII.2015, ♀; Erzurum: Gelinkaya, 26.VII. 2015, 53 ♂♂, 9 ♀♀, Kandilli, 6 km from Aşkale, 22.VII.2015, 6 ♂♂, ♀; Giresun: Eynesil, Kekiktepe, 25.VII. 2015, ♂, ♀, Keşap, Yolağzı, 25.VII.2015, ♀; Gümüşhane: Kelkit, Köycük, 23.VII.2015, 6 ♂♂, 4 ♀♀; Ordu: Turnasuyu, 24.VII.2015, ♀.

Distribution in Turkey: Ankara, Bayburt, Bolu, Çankırı, Erzincan, Erzurum, Kars, Kırşehir, Konya, Nevşehir, Trabzon and Yozgat (Özdemir, 1996; Pekel, 1999; Çoruh et al., 2004, 2014c).

Distribution in World: Algeria, Europe, Turkey, Armenia and Iran.

Remark: this species was collected on *M. sativa* while feeding in Kandilli and Avcılar.



Figure 2. Study areas in Avcılar, Erzincan Province, Turkey.

Lissonota (Loxonota) histrio (Fabricius, 1798)

Material examined: Erzurum: Kandilli, 6 km from Aşkale, 22.VII.2015, 2 ♀♀.

Distribution in Turkey: Diyarbakır, Elazığ, Erzurum, Mardin and Ordu (Pekel & Özbek, 2000; Akkaya, 2005, Kolarov et al., 2016; Özdan & Gürbüz, 2016).

Distribution in World: Holarctic region.

Remark: This species was collected on *M. sativa* while feeding.

Lissonota (Loxonota) lineata Gravenhorst, 1829

Material examined: Erzincan: Ahmetli, 23.VII.2015, ♂.

Distribution in Turkey: Diyarbakır, Hatay and Osmaniye (Akkaya, 2005; Gürbüz et al., 2011).

Distribution in World: Europe, Turkey, Iran and Mongolia.

Lissonota (Lissonota) apleuralis Brischke, 1880

Material examined: Erzincan: Avcılar, 23.VII.2015, ♂, ♀; Erzurum: Kandilli, 6 km from Aşkale, 22.VII.2015, ♀; Giresun: Keşap, Yolağzı, 25.VII.2015, ♀.

Distribution in Turkey: Bursa and Çanakkale (Kolarov et al., 1997a, b).

Distribution in World: Europe, Turkey and North China.

Associated plants: *Anethum graveolens* L., *Chaerophyllum bulbosum* L., *Cirsium vulgare* (Savi) Ten., *Daucus carota* subsp. *sativus* L., *Epilobium angustifolium* L., *Fraxinus excelsior* L., *Heracleum sphondylium* L., *Pastinaca graveolens* M. Bieb., *Peucedanum oreoselinum* and *Quercus sessiliflora* (Herb Smith).

Remark: this species was collected on *M. sativa* while feeding near Avcılar.

Lissonota (Lissonota) variabilis Holmgren, 1860

Material examined: Erzurum: Kandilli, 6 km from Aşkale, 22.VII.2015, 2 ♂♂.

Distribution in Turkey: Erzurum and Kars (Pekel, 1999).

Distribution in World: Europe, Georgia and Turkey.

Associated plants: *Angelica sylvestris* L., *Heracleum sphondylium*, *Peucedanum oreoselinum* and *Picea* sp.

Remark: This species was collected on *M. sativa* while feeding.

Subfamily: Cremastinae Forster, 1869*Pristomerus armatus* (Lucas, 1849)

Material examined: Erzurum: Aşkale, Tepebaşı Valley, 23.VII.2015, ♀.

Distribution in Turkey: Elazığ, Erzurum, Malatya and Sivas (Pekel & Özbek, 2000; Kolarov & Yurtcan, 2009).

Distribution in World: Algeria, Morocco, Europe, Georgia, Turkey, Armenia, Iran, Kazakhstan, Turkmenistan, Uzbekistan, Kyrgyzstan and Siberia.

Pristomerus vulnerator (Panzer, 1799)

Material examined: Erzincan: Avcılar, 23.VII.2015, 2 ♂♂, 2 ♀♀.

Distribution in Turkey: Ankara, Black Sea Region, Bursa, Erzurum, Samsun and Tekirdağ (Kolarov, 1995b; Kolarov, 1997; Pekel & Özbek, 2000).

Distribution in World: Holarctic and Oriental regions.

Associated plants: *Alnus glutinosa* (L.), *Angelica sylvestris* and *Peucedanum oreoselinum*.

Temelucha pseudocaudata Kolarov, 1982

This species was described only by female (Kolarov, 1982). In materials investigated we found male specimen and it is described below:

Male. Front wing 4.5 mm long (2.5 as long as hind tibia). Head moderately narrowed behind eyes. Frons concave behind each antennal socket, raised laterally. Ocellus large, its diameter 1.4 times as long as distance between lateral ocellus and eye. Flagellum with 31 (in female with 27-30) segments, first segment 1.5 times as long as fifth, all segments elongated (in female apical segments square to transversal). Inner eye orbita divergent downwards. Clypeus moderately convex with arched apical ridge.

Lower lateral ridge of pronotum produced as distinct sharp tooth. Sternaulus distinct in front half. Nervulus almost interstitial, nervellus not intercepted, discoidella as unclear depigmented trace. Areolation of propodeum complete. Areola shorter than in female, closed behind. Legs slender, hind femur 4.4 as long as wide. Correlation between hind tarsal segments as 44:18:14:9:12. Tarsal claws simple.

Black. Frontal and upper half of face orbits. Predominant part of outer orbits yellow; front and middle legs without black except coxae; apical 0.25 of second tergum, third and fourth terga red colored.

In other as in female.

Material examined: Erzurum, Kandilli, 6 km from Aşkale, 22.VII.2015, ♂, ♀.

Distribution: Bulgaria, Ukraine (Narolsky, in litt.) and Turkey.

Temelucha turcata Kolarov & Beyarslan, 1999

Material examined: Erzurum: Kandilli, 6 km from Aşkale, 22.VII.2015, 6 ♀♀.

Distribution in Turkey: Ankara, Elazığ, Eskişehir, Kayseri, Malatya and Sivas (Kolarov & Yurtcan, 2009).

Distribution in World: Turkey.

Subfamily: Ctenopelmatinae Forster, 1869

Absyrtus vicinator (Thunberg, 1822)

Material examined: Rize: İkizdere, Çamlık, 26.VII.2015, ♀.

Distribution in Turkey: Konya (Özbek et al., 2000).

Distribution in World: Palearctic region.

Associated plants: *Rubus* spp.

Subfamily: Diplazontinae Viereck, 1918

**Diplazon deletus* (Thomson, 1890)

Material examined: Erzincan: Ahmetli (39°53.481' N, 39°21.197 E), 1988 m, 23.VII.2015, ♂.

Distribution in World: Holarctic region.

Associated plants: *Phragmites communis* (Cav.).

Diplazon laetatorius (Fabricius, 1781)

Material examined: Giresun: Eynesil, Kekiktepe, 25.VII.2015, ♀.

Distribution in Turkey: Adana, Adıyaman, Afyon, Ankara, Antalya, Artvin, Aydın, Bolu, Burdur, Denizli, Erzincan, Erzurum, Eskişehir, Hatay, Isparta, Izmir, Kahramanmaraş, Nevşehir, Kırklareli, Konya, Osmaniye, Sinop, Trabzon, Şanlıurfa and Zonguldak (Özdemir, 2001; Çoruh, 2011; Gürbüz et al., 2011; Kolarov, 2015).

Distribution in World: Cosmopolitan species.

Associated plants: *Baccharis pilularis* DC., *Citrullus lanatus* (Thunb.) Matsum. & Nakai, *Citrus sinensis* (L.) Osbeck, *Cynara cardunculus* subsp. *flavescens* L. *Heracleum sphondylium*, *Malus domestica* Borkh., *Neottia ovata* (L.), *Oryza sativa* L., *Peucedanum oreoselinum*, *Picea excelsa* Lam., *Poa pratensis* L. and *Vicia faba* L.

Diplazon scutatorius Teunissen, 1943

Material examined: Rize: İkizdere, Çamlık, 26.VII.2015, 2 ♂♂, ♀.

Distribution in Turkey: Rize (Çoruh, 2009; Çoruh et al., 2014c).

Distribution in World: Palearctic region.

Homotropus nigratarsus (Gravenhorst, 1829)

Material examined: Erzurum: Gelinkaya, 26.VII.2015, ♂.

Distribution in Turkey: Afyon, Antalya, Ardahan, Denizli, Erzurum, Gümüşhane, Isparta and Izmir (Kolarov, 2015).

Distribution in World: Holarctic region and Mexico.

Associated plants: *Betula nana*.

Remark: This species was collected on *M. sativa* while feeding.

**Homotropus pallipes* (Gravenhorst, 1829)

Material examined: Rize: İkizdere, Çamlık, 26.VII.2015, 4 ♀♀.

Distribution in World: Holarctic region and Mexico.

Associated plants: *Poa pratensis* and *Tsuga heterophylla* (Raf.) Sarg.

**Sussaba cognata* (Holmgren, 1858)

Material examined: Rize: İkizdere, Çamlık, 26.VII.2015, 3 ♀♀.

Distribution in World: Holarctic and Oriental regions.

Associated plants: *Angelica sylvestris* and *Oryza sativa*.

Sussaba pulchella (Holmgren, 1858)

Material examined: Rize: İkizdere, Çamlık, 26.VII.2015, ♂.

Distribution in Turkey: Ankara and Çankırı (Özdemir, 2001).

Distribution in World: Holarctic and Oriental regions.

Associated plants: *Angelica* sp., *Oryza sativa*, *Peucedanum oreoselinum*, *Picea abies* (L.) H. Karst., *P. excelsa*, *Polygonum* sp. and *Prunus padus* L.

Syrphophilus bizonarius (Gravenhorst, 1829)

Material examined: Giresun: Dereli, Çamağzı, 23.VII.2015, ♀; Rize: İkizdere, Çamlık, 26.VII.2015, ♂.

Distribution in Turkey: Adana, Ankara, Burdur, Çankırı, Eskişehir, Hatay, Isparta, Konya, Niğde and Sinop (Özdemir, 2001; Gürbüz et al., 2011; Kolarov, 2015).

Distribution in World: Holarctic and Oriental regions.

Associated plants: *Angelica* sp., *Betula nana* L., *Epilobium angustifolium*, *Oryza sativa*, *Poa pratensis* L. and *Veronica anagallis-aquatica* L.

Subfamily: Metopiinae Förster, 1869

**Exochus flavifacies* Kusigemati, 1984

Material examined: Ordu: Turnasuyu, 24.VII.2015, 2 ♀♀; Rize: İkizdere, Çamlık, 26.VII.2015, 2 ♂♂.

Distribution in World: Mongolia.

Exochus mitratus Gravenhorst, 1829

Material examined: Erzincan: Avcılar, 23.VII.2015, ♀.

Distribution in Turkey: Antalya, Denizli, Erzurum, Giresun, Isparta, Kars, Kırklareli and Rize (Kolarov et al., 2009; Çoruh et al., 2014a)

Distribution in World: Holarctic region.

Associated plants: *Peucedanum oreoselinum* and *Quercus lusitanica* Lam.

Remark: this species was collected on *M. sativa* while feeding.

Exochus prosopius Gravenhorst, 1829

Material examined: Giresun: Eynesil, Kekiktepe, 25.VII.2015, 2 ♂♂; Trabzon: Yomra, 25.VII.2015, ♂.

Distribution in Turkey: Giresun, Izmir and Rize (Kolarov et al., 2009; Çoruh et al., 2014a)

Distribution in World: Palearctic region.

Exochus suborbitalis Schmiedeknecht, 1924

Material examined: Erzurum: Gelinkaya, 26.VII.2015, ♀.

Distribution in Turkey: Antalya, Erzurum, Giresun, Isparta, Kahramanmaraş, Ordu and Osmaniye (Kolarov et al., 2009, 2016).

Distribution in World: Palearctic region.

Associated plants: *Peucedanum oreoselinum*.

Exochus thomsoni Schmiedeknecht, 1924

Material examined: Erzurum: Gelinkaya, 26.VII.2015, ♂; Rize: İkizdere, Çamlık, 26.VII.2015, 2 ♂♂.

Distribution in Turkey: Erzurum and Isparta (Çoruh & Kolarov, 2012; Özdan, 2014).

Distribution in World: Palearctic region.

Exochus vafer Holmgren, 1873

Material examined: Erzurum: Gelinkaya, 26.VII.2015, ♀.

Distribution in Turkey: Isparta, Erzincan and Izmir (Kolarov et al., 2009; Çoruh & Kolarov, 2012; Özdan, 2014).

Distribution in World: Europe, Turkey and Siberia.

**Triclistus congener* (Holmgren, 1858)

Material examined: Erzurum: Aşkale, Tepebaşı Valley (Figure 3), 23.VII.2015, ♀.

Distribution in World: Europe and USA.

Subfamily: Ophioninae Scuckard, 1840

Enicospilus ramidulus (Linnaeus, 1758)

Material examined: Erzurum: Aşkale, Tepebaşı Valley, 23.VII.2015, 2 ♀♀; Rize: İkizdere, Çamlık, 26.VII.2015, ♀; Trabzon: Yomra, 25.VII.2015, ♀.

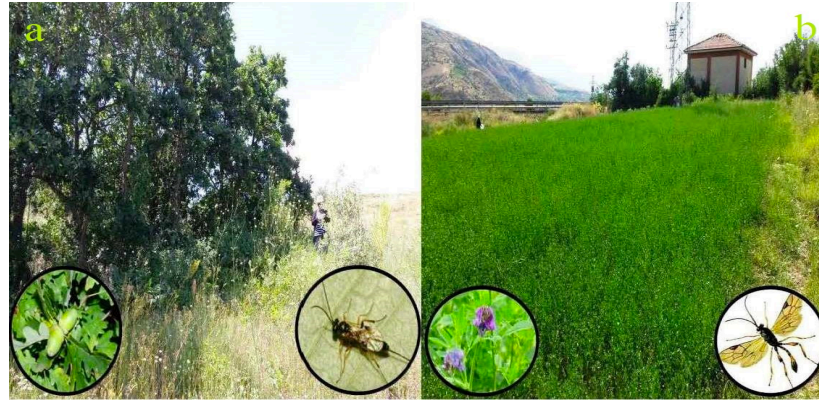


Figure 3. Study areas in Tepebaşı Valley, Aşkale, Erzurum Province (left) and Avcılar, Erzincan Province (right), Turkey.

Distribution in Turkey: Ankara, Erzincan, Erzurum, Isparta, Karabük, Kastamonu, Malatya, Nevşehir, Konya, Rize, Sinop and Tekirdağ (Kolarov, 1995a; Kolarov et al., 2000; Akkaya, 2005; Kolarov & Gürbüz, 2006; Okyar & Yurtcan, 2007; Çoruh & Çalmaşur, 2016).

Distribution in World: Palearctic, Oriental and Afrotropical regions.

Associated plants: *Alnus glutinosa*, *Carum carvi* L., *Oryza sativa*, *Salvia glutinosa* L. and *Seseli libanotis* (L.) W. Koch.

Remark: this species was collected on *S. libanotis* (L.) W. Koch while feeding in Tepebaşı Valley.

Enicospilus tournieri (Vollenhoven, 1879)

Material examined: Erzurum: Gelinkaya, 26.VII.2015, ♂, 8 ♀♀.

Distribution in Turkey: Ankara, Erzurum and Hatay (Kolarov et al., 2000; Çoruh et al., 2014a)

Distribution in World: Palearctic region.

Remark: this species was collected on *M. sativa* while feeding in Gelinkaya.

Subfamily: Pimplinae Wesmael, 1845

Endromopoda detrita (Holmgren, 1860)

Material examined: Erzincan: Ahmetli, 23.VII.2015, ♂.

Distribution in Turkey: Afyon, Bayburt, Burdur, Bursa, Çanakkale, Denizli, Edirne, Erzincan, Erzurum, Gümüşhane, Iğdır, Isparta, İstanbul, İzmir, Kars, Kırklareli, Rize, Tekirdağ and Tunceli (Kolarov, 1987, 1995a; Özdemir & Kılınçer, 1990; Öncüer, 1991; Kolarov & Beyarslan, 1994; Kolarov et al., 1997a, b, 1999, 2002, 2014c; Kolarov & Gürbüz, 2004; Çoruh, 2005; Yurtcan, 2007; Çoruh & Kolarov, 2010).

Distribution in World: Holarctic and Oriental regions.

Associated plants: *Adonis vernalis* L., *Angelica sylvestris*, *Chaerophyllum aromaticum* L., *Cirsium palustre* (L.) Scop., *Daucus carota* L., *Daucus carota* subsp. *sativus*, *Foeniculum vulgare*, *Heracleum* sp., *Juniperus communis* L. and *Peucedanum oreoselinum*.

Exeristes roborator (Fabricius, 1793)

Material examined: Erzincan: Pöske Mt., 23.VII.2015, ♂.

Distribution in Turkey: Ankara, Ardahan, Artvin, Balıkesir, Bayburt, Bilecik, Bingöl, Bitlis, Burdur; Bursa, Çanakkale, Denizli, Edirne, Erzurum, Erzincan, Gümüşhane, Hakkari, Isparta, İstanbul, İçel, Kars, Kırklareli, Muğla, Muş, Rize, Tekirdağ and Tunceli (Özdemir & Kılınçer, 1990; Öncüer, 1991; Kolarov & Beyarslan, 1994; Kolarov, 1995a; Kolarov et al., 1997a, b, 1999, 2002, 2014c; Kasparyan & Gültekin,

2002; Gürbüz, 2004, 2005; Kolarov & Gürbüz, 2004; Çoruh, 2005; Yurtcan, 2007; Çoruh & Kolarov, 2010; Tozlu & Çoruh 2011; Özbek & Çoruh, 2012).

Distribution in World: Palearctic, Oriental and Afrotropical regions, introduced into North America (including Mexico).

Associated plants: *Anethum graveolens*, *Chaerophyllum bulbosum*, *Daucus carota*, *Euphorbia virgata* L., *Fraxinus excelsior*, *Heracleum sphondylium*, *Pastinaca graveolens*, *Peucedanum oreoselinum*, *Quercus sessiliflora*, *Salvia sylvestris* Linne, *Seseli tortuosum* Sibth. & Sm., *Tamarix* spp.

Itoplectis maculator (Fabricius, 1775)

Material examined: Erzurum: Aşkale, Tepebaşı Valley, 23.VII.2015, 4 ♀♀.

Distribution in Turkey: Adana, Ankara, Afyon, Artvin, Balıkesir, Bitlis, Bolu, Çanakkale, Çorum, Denizli, Edirne, Eskişehir, Erzurum, Gümüşhane, Isparta, İçel, İzmir, Kars, Kastamonu, Kırklareli, Kırşehir, Konya, Nevşehir, Niğde, Muğla, Rize, Sinop, Tekirdağ, Van, Yozgat and Zonguldak (Kolarov, 1987, 1995a; Özdemir & Kılınçer, 1990; Öncüer, 1991; Kolarov & Beyarslan, 1994; Erol & Yaşar, 1996; Kolarov et al., 1997a, 1999, 2002; Özdemir & Özdemir, 2002, Gürbüz, 2004; Kolarov & Gürbüz, 2004; Çoruh, 2005; Gürbüz, 2005; Yurtcan & Beyarslan, 2005; Çoruh et al., 2007, 2014a; Okyar & Yurtcan, 2007; Gürbüz et al., 2009; Çoruh & Kolarov, 2010; Birol, 2010; Eroğlu et al., 2011).

Distribution in World: Palearcticregion, introduced into USA.

Associated plants: *Adonis vernalis*, *Alnus glutinosa*, *Chaerophyllum bulbosum*, *Cirsium palustre*, *Daucus carota*, *Epilobium angustifolium*, *Euphorbia nicaeensis* All., *Fraxinus excelsior*, *Heracleum sphondylium*, *Peucedanum oreoselinum*, *Picea abies*, *P. excelsa*, *Pinus sylvestris* L., *Quercus ilex* L., *Quercus sessiliflora*, *Rubus* spp. and *Taxus baccata* L.

Liotryphon caudatus (Ratzeburg, 1848)

Material examined: Erzincan: Avcılar, 23.VII.2015, ♀.

Distribution in Turkey: Anatolia and Isparta (Öncüer, 1991; Kolarov, 1995a; Kolarov & Gürbüz, 2004; Gürbüz, 2005; Çoruh, 2016).

Distribution in World: Palearctic region and New Zealand.

Associated plants: *Acer pseudoplatanus* L.

Scambus nigricans (Thomson, 1877)

Material examined: Erzurum: Gelinkaya, 26.VII. 2015, ♀; Rize: İkizdere, Çamlık, 26.VII.2015, ♀.

Distribution in Turkey: Afyon, Artvin, Balıkesir, Bayburt, Burdur, Bursa, Çanakkale, Denizli, Edirne, Erzincan, Erzurum, Isparta, İstanbul, İzmir, Kahramanmaraş, Kars, Kırklareli and Tekirdağ (Kolarov & Beyarslan, 1994; Kolarov et al., 1997a, 1999, 2002; Kolarov & Gürbüz, 2004; Çoruh, 2005; Yurtcan, 2007; Çoruh et al., 2007; Çoruh & Kolarov, 2010; Kolarov & Çalmaşur, 2011).

Distribution in World: Palearctic region.

Associated plants: *Anethum graveolens*, *Chaerophyllum bulbosum*, *Daucus carota*, *Euphorbia nicaeensis*, *Heracleum sphondylium*, *Peucedanum oreoselinum*.

**Tromatobia lineatoria* (Villers, 1789)

Material examined: Erzincan, Pöske Mt., 23.VII.2015, ♀.

Distribution in World: Palearctic region.

Zatypota bohemani (Holmgren, 1860)

Material examined: Gümüşhane: Kelkit, Köycük, 23.VII.2015, ♂.

Distribution in Turkey: Adana, Edirne, Elazığ, Erzurum, Hatay, İçel, Isparta, İstanbul, Kars and Osmaniye (Kolarov, 1987; Kolarov & Beyarslan, 1994; Kolarov & Gürbüz, 2004; Çoruh, 2005; Yurtcan & Beyarslan, 2005; Çoruh & Kolarov, 2010; Gürbüz et al., 2011).

Distribution in World: Holarctic region.

Associated plants: *Anethum graveolens*, *Peucedanum oreoselinum*, *Rubus* spp.

Subfamily: Tryphoninae Shuckard, 1840

Acrotomus lucidulus (Gravenhorst, 1829)

Material examined: Ordu: Turnasuyu, 24.VII.2015, ♂.

Distribution in Turkey: Afyon, Denizli, Edirne, Isparta, Malatya, Muğla and Rize (Yurtcan & Beyarslan, 2002; Çoruh et al., 2005, 2014b; Gürbüz & Kolarov, 2006; Yurtcan et al., 2006; Yaman 2014).

Distribution in World: Palearctic region.

Associated plants: *Heracleum sphondylium* and *Peucedanum oreoselinum*.

**Ctenochira meridionator* Aubert, 1969

Material examined: Ordu: Turnasuyu, 24.VII.2015, ♀.

Distribution in World: Palearctic region.

Netelia (Netelia) silantjewi (Kokujev, 1899)

Material examined: Erzurum: Gelinkaya, 26.VII.2015, ♂.

Distribution in Turkey: Afyon, Balıkesir, Bursa, Kırklareli, Muğla and Uşak (Kolarov et al., 1997b; Yurtcan & Beyarslan, 2002; Yurtcan et al., 2006; Yaman, 2014).

Distribution in World: Palearctic and Oriental regions.

Associated plants: *Quercus robur*.

Remark: this species was collected on *M. sativa* while feeding in Gelinkaya.

Oedemopsis scabricula (Gravenhorst, 1829)

Material examined: Giresun: Bulancak, 24.VII.2015, ♀.

Distribution in Turkey: Erzurum, Giresun, Malatya, Ordu, Rize and Tekirdağ (Çoruh et al., 2005; Beyarslan et al., 2006; Çoruh et al., 2014a, b; Yaman, 2014).

Distribution: Holarctic and Oriental region.

Thymaris tener (Gravenhorst, 1829)

Material examined: Giresun: Keşap, Yolağzı, 25.VII.2015, ♀.

Distribution in Turkey: Çanakkale (Yaman, 2014).

Distribution in World: Palearctic region.

Associated plants: *Picea* spp.

Subfamily: Xoridinae Shuckard, 1840

**Odontocolon rufiventris* (Holmgren, 1860)

Material examined: Rize: İkizdere, Çamlık, 26.VII.2015, ♀.

Distribution in World: Europe.

Xorides gracilicornis (Gravenhorst, 1829)

Material examined: Ordu: Turnasuyu, 24.VII.2015, ♂.

Distribution in Turkey: Isparta and Osmaniye (Kolarov & Gürbüz, 2006; Gürbüz et al., 2011).

Distribution in World: Palearctic region.

Zoogeographical characterization

The zoogeographical characterization follows mainly the chorotype classification of the Near East fauna, proposed by Taglianti et al. (1999). After investigation of the recent geographic distribution of the species, listed above, they can be divided into the following groups:

1. Cosmopolitan distribution; *Diplazon laetatorius*.
2. Multiregional ranges; *Enicospilus ramidulus* and *Exeristes roborator*, distributed in three zoogeographical regions; Palearctic, Oriental and Afrotropical.
3. Species with ranges in two zoogeographical regions; *Anomalon cruentatum*, *Pristomerus vulnerator*, *Sussaba cognata*, *S. pulchella*, *Syrphophilus bizonarius*, *Endromopoda detrita*, *Netelia (Netelia) silantjewi*, *Oedemopsis scabricula*, distributed in Palearctic/Holarctic and Oriental regions; *Homotropus nigratarsus* and *H. pallipes*, distributed into Holarctic and Neotropic (Mexico) regions; *Liotryphon caudatus*, distributed in Palearctic and Oceanic (New Zealand) regions.
4. Holarctic ranges; *Lissonota (Loxonota) histrio*, *Diplazon deletus*, *Exochus mitratus*, *Triclistus congener* and *Zatypota bohemani*.
5. Most numerous species with Palearctic ranges; *Agrypon gracilipes*, *Absyrtus vicinator*, *Diplazon scutatorius*, *Exochus prosopius*, *Exochus suborbitalis*, *Exochus thomsoni*, *Enicospilus tournieri*, *Itoplectis maculator*, *Scambus nigricans*, *Tromatobia lineatoria*, *Acrotomus lucidulus*, *Ctenochira meridionator*, *Thymaris tener* and *Xorides gracilicornis*.
6. Sibero-European distributions; *Pristomerus armatus* and *Exochus vafer*.
7. Centralasiatic range; *Exochus flavifacies*.
8. Turano-European-Mediterranean distribution; *Lissonota (Loxonota) flavovariegata*.
9. Turanian distribution; *Barylypa torquata*.
10. European ranges; *Lissonota (Loxonota) lineata*, *Lissonota (Lissonota) pleuralis*, *Lissonota (Lissonota) variabilis* and *Odontocolon rufiventris*.
11. Ponto-Caucasian sub endemic; *Temelucha pseudocaudata*.
12. Anatolian endemic; *Temelucha turcata*.

Acknowledgements

The preparation of this article was supported by Project Nr. BAP–2012/234 (Atatürk University) and we are very thankful for this support. We are also indebted to Halil Çoruh for collecting some specimens.

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Original article (Orijinal araştırma)

Bioecological characteristics of *Planococcus citri* Risso, 1813 (Hemiptera: Pseudococcidae) under constant and alternating temperatures¹

Planococcus citri Risso, 1813 (Hemiptera: Pseudococcidae)'nin sabit ve değişken sıcaklıklarda bazı biyoeolojik özellikleri

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Summary

Planococcus citri Risso, 1813 (Hemiptera: Pseudococcidae), is one of the major pest of citrus and many other orchards crops, and ornamental plants in subtropical and tropical regions of the world. The influence of temperature on *P. citri* development and fecundity has a critical role in integrated pest management strategies to reduce the population to below the economic threshold by biological or chemical control methods. The study investigated some bioecological characteristics, such as, development time, duration of biological stages, sex ratio, daily and total fecundity per female, and longevity of *P. citri*, under different temperature regimes during 2015-2016 in Citrus Pest Laboratory at Çukurova University. The shortest egg stage development for females and males were determined as 2.7 and 2.7 d with alternating temperatures of 25/30°C (12:12 h), respectively. The first nymph stage lasted 7.86 d for females, and 8.1 d for males at 25°C. The longest duration for the second nymph stage was obtained at 15°C with 25.7 and 22.5 d for females and males, respectively. The third nymph stage for *P. citri* females completed in 7.0 d at 25°C, and the pupal stage for *P. citri* males lasted 7.8 d at 25°C. The development thresholds of females and males were calculated as 8.5 and 9.5°C, respectively. Also, thermal constants of females and males were 666.67 and 500.00 degree-days. The optimum development temperature was determined as 25/30°C.

Keywords: Citrus mealybug, development time, life table, *Planococcus citri*, thermal constant

Özet

Planococcus citri Risso, 1813 (Hemiptera: Pseudococcidae) dünyanın subtropikal ve tropikal bölgelerinde bulunan başta turunçgil olmak üzere pek çok bahçe ve süs bitkileri üzerindeki ana zararlılardan birisidir. *P. citri*'nin gelişme süresi ve üremesi üzerinde sıcaklığın etkisi zararlının popülasyon seviyesini ekonomik zarar eşliğinin altına düşürmek için uygulanacak bir biyolojik veya kimyasal mücadele programı için kritik bir role sahiptir. Çukurova Üniversitesi Turunçgil Zararlıları Laboratuvarı'nda 2015-2016 yılları arasında, *P. citri*'nin toplam gelişme süresi, cinsiyet oranı, günlük ve toplam yavru sayıları ve ergin ömrü gibi bazı biyoeolojik özellikleri farklı sıcaklıklar altında çalışılmıştır. En kısa yumurta gelişme süresi sırasıyla dişi ve erkekler için 2.7 ve 2.7 gün olarak 25/30°C'de (12:12 h) tespit edilmiştir. Birinci dönem nimflerin gelişme süresi ise 25°C'de dişiler için 7.9 gün sürerken erkekler için ise 8.1 gün olmuştur. En uzun ikinci nimf dönemi gelişme süresi ise 15°C'de dişiler için 25.7, erkekler için ise 22.5 gün olarak hesap edilmiştir. Ergin olma öncesi son gelişme dönemi olarak dişilerin üçüncü dönem nimfler gelişimini 25°C'de 7.0 günde tamamlarken, erkek bireylerin pupa dönemleri ise 7.8 gün sürmüştür. Dişi ve erkek bireylerin gelişme eşikleri ise sırasıyla 8.5 ve 9.5°C olarak hesaplanmıştır. Yumurtadan ergin olması için gerekli sıcaklıklar toplamı olan thermal konstant ise dişiler için 666.67, erkekler için ise 500.00 gün-derecedir. Turunçgil unlubütünün optimum gelişme sıcaklığı ise 25/30°C olarak belirlenmiştir.

Anahtar sözcükler: Turunçgil unlubiti, gelişme süresi, yaşam tablosu, *Planococcus citri*, termal konstant

¹ This article is part of the PhD thesis of the first author.

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Received (Alınış): 14.11.2016 Accepted (Kabul ediliş): 02.03.2017 Published Online (Çevrimiçi Yayın Tarihi): 25.04.2017

Introduction

Citrus mealybug, *Planococcus citri* Risso, 1813 (Hemiptera: Pseudococcidae), is one of the major pest of citrus and many other orchards crops, and ornamental plants in subtropical and tropical region of the world (Williams & Watson, 1988; Blumberg et al., 1995). This pest is also recognized as a major pest of citrus in Turkey (Bodenheimer, 1953; McKenzie, 1967; Düzgüneş, 1982; Lodos, 1986; Williams & Watson, 1988; Ben-Dov, 1994; Uygun, 2001; Franco et al., 2004; Uygun & Satar 2008). It feeds on fruits and twigs of citrus by sucking the sap, and therefore, the plants often become stunted, distorted, or yellowed and show reduced vigor. A dark-colored sooty mold, known as fumagine, grows on the honeydew secreted during its feeding on the host plant (Uygun, 2001; Polat et al., 2007; Uygun & Satar, 2008). Moreover, fruits attacked by *P. citri* often drop in early maturity (August-September) because of injury to the calyx of the fruit. The population density of this pest can reach a level that may cause serious damage in citrus orchards if not controlled. The damage of this pest causes a decrease in the market value of citrus fruit and thus, the pest affects adversely the citrus exports. Several studies determined that there is a significant number of natural enemies of this mealybug in the East Mediterranean Region of Turkey (Alkan, 1953; Soylu & Ürel, 1977; Kansu & Uygun, 1980; Uygun, 1981; Lodos, 1986; Yayla & Satar, 2012; Satar et al., 2013; Kütük et al., 2014). These natural enemies are important in integrated pest management of *P. citri* in Turkey. However, chemical control is often the preferred method to control the pest because it is easy use and requires less knowledge and labor (Yiğit et al., 1994; Satar et al., 2013; Birgücü et al., 2014, 2015).

Cryptolaemus montrouzieri Mulsant, 1850 (Coleoptera: Coccinellidae) and *Leptomastix dactylopii* Howard, 1885 (Hymenoptera: Encyrtidae), which are mass reared and released, are the most commonly used natural enemies of *P. citri* (Yiğit et al., 1994; Erkılıç & Demirbaş, 2007). However, the increasing chemical applications to control *P. citri* in citrus orchards leads to a decline in natural enemy populations and has conversely resulted in a rise in *P. citri* populations in last two decades (Karacaoğlu & Satar, 2010; Satar et al., 2011). Successful application of pesticide also depends on appropriate timing relative to the pest population. So, the influence of temperature on *P. citri* development and fecundity has a critical role of integrated pest management (IPM) strategies to reduce populations to below the economic threshold by both biological and chemical control methods. The temperature conditions also provide information about population dynamics of insects (Keena, 2006), and are important in their bioecological characteristics. Therefore, the effects of temperature on insects in citrus orchards need to be investigated.

To this end, the study investigated some bioecological characteristics, such as, development time, sex ratio, duration of biological stages, daily and total fecundity per female, and longevity of *P. citri*, under different temperature regimes. This knowledge is key for further understanding of the arthropod's biology, especially its adaptation to different temperatures and should provide the basis for better pest management strategies. Furthermore, it can be used to estimate the potential damage level (Sánchez-Ramos & Castañera, 2005) and thus to improve pest management (Satar et al., 2005; Ebrahimi et al., 2009). Also, the model of best fit for the effect of temperate on the development rate of the pest was assessed by linear regression.

Material and Methods

Breeding of the plant and pest

Grapefruit seedlings (*Citrus paradise* Macfad.) cv. Star Ruby obtained from the Subtropical Fruits Research and Application Center of Çukurova University in Adana, Turkey were planted in 20 l plastic pots containing a mixture of soil and peat (1:1 v/v) and then kept in climate room at 26±1°C and 60±10% RH with a 12:12 h L:D photoperiod.

The leaves, twigs and fruits of grapefruit trees infected with *P. citri* were collected from Alata Horticultural Research Institute in Mersin, Turkey and brought to laboratory within culture plates. Then, citrus mealybug individuals were gently transferred to the grapefruit seedling using the fine paintbrush, under a binocular microscope. Afterwards, the infected grapefruit seedlings were transferred to cages (100x85x67 cm) with the upper and lateral sides of which was covered by net, in a climate room at $26\pm 1^\circ\text{C}$ and $60\pm 10\%$ RH with a 16:8 h L:D photoperiod. These were the stock cultures of *P. citri* used in the experiments described below. Maintenance and control for the plant and pest were done daily and irrigation was applied as necessary.

Experimental establishment

A total of 10 individual *P. citri* in the third nymph stage from stock culture were randomly selected and transferred using a fine paintbrush to dissected grapefruit (cv. Star Ruby) leaf discs in 5-cm diameter petri dishes containing 1% water agar. Each of egg mass deposited by females in the petri dish were transferred using a fine paintbrush to new petri dishes with fresh leaf discs containing 1% water agar and monitored for hatching. Each neonate crawler (first-instar nymph) was separately transferred using a fine paintbrush to new petri dishes containing 1% water agar and observed every day to record biological parameters, such as the development time, sex ratio.

Then an experiment was designed to provide mating opportunity, so that each petri contained two males and one female. All replicates in which the nymphs died within 24 h after transfer were omitted from the experiment. The durations of biological stages, daily and total fecundity per female and longevity of adults were observed. Also, new egg masses deposited by females were removed from the petri dishes after recorded and the grapefruit leaf disc in the Petri dishes was renewed every 3 to 5 d, if necessary. The experiment was conducted under five constant (15, 20, 25, 30 and $35\pm 1^\circ\text{C}$) and one alternating ($25/30\pm 1^\circ\text{C}$) temperature regimes under constant relative humidity and photoperiod conditions ($65\pm 10\%$ RH with 16:8 h L:D photoperiod for constant temperatures and 12:12 h L:D photoperiod for alternating temperatures at 8-10 klux light intensity). At least 20 replicates were included for each temperature regime. The experiment continued until the death of all individuals. All the experiments were conducted in Citrus pest laboratory at Department of Plant Protection, Faculty of Agriculture Çukurova University, Adana Turkey during 2015-2016.

Life table and statistical analyses

Life table parameters were calculated using the following formulas:

The age-specific survival rate (l_x) and fecundity (m_x , female/female) was computed by multiplying the mean number of offspring by the sex ratio (Birch, 1948),

Net reproductive rate ($R_0 = \sum l_x \cdot m_x$) (female/female/offspring), i.e. the mean number of offspring which are laid by a female in her lifetime (Birch, 1948) were assed individually for each replication in each temperature then, all cohort in each temperature were used in bootstrap (Efron & Tibshirani, 1993) techniques to estimate the means, variances, and standard errors of the population net reproductive rate.

Intrinsic rate of increase (r_m , female/female/day) by taking advantage from Euler-Lotka equation ($\sum e^{(-r_m \cdot x)} l_x \cdot m_x = 1$) (Birch, 1948),

Mean generation time (day), $T_o = \frac{\ln R_0}{r_m}$ (Birch, 1948),

Gross reproduction rate, $GRR = \sum m_x$ (Birch, 1948),

Where x is female age in days, e is Euler's number which is a mathematical constant (approx. 2.71828).

Pseudo- r_{mj} values of the intrinsic rate of increase (r_m) values were calculated according to the jackknife resampling method (Meyer et al., 1986) for the comparison test, and then, Tukey multiple comparison test (Tukey, 1949) was applied after one-way ANOVA for these pseudo- r_{mj} values of the intrinsic rates. Statistical analyses were performed by using IBM® SPSS® Statistics (Version 20.0) (SPSS 2011).

Also, development rates of the individuals bred under the different temperature regimes were determined by linear regression ($y = a \pm bx$). The mean of the alternating temperatures (27.5°C for 25/30°C) was used in the regression analysis. Afterwards, the development threshold ($-a/b$) and thermal constant (the total effective temperature required to complete a generation, $1/b$) of *P. citri* was calculated according to the linear regression equation (Campbell et al., 1974).

Results and Discussion

The individuals from both sexes of *P. citri* reached adult stage at temperatures between 15 and 30°C; however, eggs of *P. citri* showed no development at 35°C. When neonate crawlers that hatched at 30°C were kept at 35°C, only 5% of them progressed to the second nymph stage. However, these individuals did not progress to further biological stages, and they all died within a month. The development durations of different biological stages of *P. citri* females on grapefruit leaves under different temperature regimes are given in Table 1. Those of *P. citri* males are also given in Table 2.

Table 1. Development durations (d) of immature biological stages of *Planococcus citri* females on grapefruit leaves under different temperature regimes (mean ± SE)*

Temperature (°C)	n	Egg stage	First nymph stage	Second nymph stage	Third nymph stage	Total development
15	16	8.00±0.22 c	14.9±0.81 c	25.7±2.34 c	20.2±1.19 c	68.8±3.00 d
20	15	7.73±1.22 c	13.4±0.77 c	13.0±1.24 b	14.7±0.78 b	48.9±1.55 c
25	23	4.34±0.14 b	7.9±0.29 a	6.4±0.74 a	7.0±0.29 a	25.6±0.95 a
25/30	30	2.73±0.82 a	7.9±0.26 a	7.4±0.50 a	5.8±0.26 a	23.9±0.60 a
30	11	3.70±0.19 a	11.1±0.99 b	9.5±1.07 ab	12.2±0.92 b	36.5±0.86 b
35	50	No development				

* Means (± SE) sharing same letter within same column are not significantly different from each other (Tukey's HSD multiple range test at $P < 0.05$) ($F_{\text{egg}} = 181.467$, $df = 4,90$, $P = 0.000$; $F_{\text{Nymph}_1} = 36.260$, $df = 4,90$, $P = 0.000$; $F_{\text{Nymph}_2} = 43.241$, $df = 4,90$, $P = 0.000$; $F_{\text{Nymph}_3} = 91.780$, $df = 4,90$, $P = 0.000$; $F_{\text{Total_development}} = 167.759$, $df = 4,90$, $P = 0.000$).

Table 2. Development durations (d) of different biological stages of *Planococcus citri* males on grapefruit leaves under different temperature regimes (mean ± SE)*

Temperature (°C)	n	Egg stage	First nymph stage	Second nymph stage	Pupal stage	Total development	Longevity of males
15	30	7.8±0.14 d	14.2±0.65 b	22.5±1.93 b	26.5±1.17 c	71.0±1.97 c	8.3±0.47 c
20	13	8.1±0.26 d	13.4±0.78 b	11.9±0.85 a	11.9±0.94 b	45.3±1.74 b	4.5±0.63 b
25	7	4.7±0.28 c	8.1±0.14 a	5.6±0.20 a	7.8±0.34 ab	26.3±0.28 a	4.7±0.47 b
25/30	20	2.7±0.10 a	7.8±0.41 a	6.3±0.28 a	7.8±0.28 ab	24.6±0.44 a	1.8±0.18 a
30	10	3.6±0.22 b	9.7±1.01 a	9.2±2.20 a	6.4±0.42 a	28.9±3.20 a	2.7±0.30 ab
35	50	No development					

* Means (± SE) sharing same letter within same column are not significantly different from each other (Tukey's HSD multiple range test at $P < 0.05$) ($F_{\text{egg}} = 197.920$, $df = 4,80$, $P = 0.000$; $F_{\text{Nymph}_1} = 19.651$, $df = 4,80$, $P = 0.000$; $F_{\text{Nymph}_2} = 20.619$, $df = 4,80$, $P = 0.000$; $F_{\text{Pupa}} = 85.577$, $df = 4,80$, $P = 0.000$; $F_{\text{Total_development}} = 132.743$, $df = 4,80$, $P = 0.000$; $F_{\text{Longevity}} = 37.897$, $df = 4,80$, $P = 0.000$).

The shortest egg stage was calculated as 2.7 d at 25/30°C, followed by 30, 25, 20 and 15°C, respectively (Table 1). For males, the shortest egg stage was observed at 25/30°C with 2.7 d, and the longest egg stage was 8.1 d, obtained at 20°C (Table 2). Arai (1996) suggested that hatching time of *P. citri* males in egg stage were 4 d at 25°C, 3.2 d at 27°C and 4 d at 27.5°C. According to the results of this study, the hatching time initially decreased with increasing temperature, and then increased again. These results were similar to the results in this present study (Table 2).

The first nymph stage lasted 7.9 d at 25°C for females, and 8.1 d for males. The duration of this stage decreased with increasing temperature up to 25°C, and then increased again at 30°C (Tables 1 & 2). The longest duration for the second nymph stage was obtained at 15°C with 25.7 and 22.5d for females and males, respectively, and the shortest duration for this stage was seen at 25°C with 6.4 and 5.6 d for females and males, respectively (Tables 1 & 2). For last period of immature stages, the third nymph stage for *P. citri* females completed in 7.0 d at 25°C, and the pupal stage for *P. citri* males lasted 7.8 d at 25°C (Table 1 & 2). Similarly, research in Brazil (Cecilia et al., 2009) determined that the development time of the first instar nymphs of *P. citri* females fed on coffee plant was 7.8 d at 25°C and 70% RH. Also, that study found that the duration of the third nymph stage of *P. citri* females fed on coffee plant under the same conditions was 7.02 d (Cecilia et al., 2009).

Polat et al. (2007) studied on the development of *P. citri* fed on four different ornamental plants *Schefflera arboricola* (Hayata) Kanehira, *Kalanchoe blossfeldiana* Poelln., *Nerium oleander* L. and *Syngonium podophyllum* Schott, and found that the first nymph stage of *P. citri* females fed on those ornamental plants were 7.90, 6.74, 6.66 and 5.61 d at 28°C, respectively. Also, Polat et al. (2007) reported that the durations of the first instar nymphs of *P. citri* males fed on those ornamental plants were 7.55, 6.78, 6.60 and 5.55 d at 28°C, respectively. The results of the present study showed similarities with the results obtained for the first instar nymphs of *P. citri* females fed on *S. arboricola* by Polat et al. (2007). However, the results obtained by Polat et al. (2007) on the development of *P. citri* fed on other ornamental plants were different from the results of this present study. Based on this, it is reasonable to conclude that the development of this pest depends on host plant species and temperature.

The durations of preoviposition, oviposition, and postoviposition periods decreased with increasing temperature especially between 15 and 25°C (Table 3). The longest longevity for *P. citri* females was determined as 80.4 d at 15°C, and the shortest longevity was 29.5 d at 30°C (Table 3) and the longest longevity for *P. citri* male was the same as for females, but shortest longevity for male was at 25/30°C instead of 30°C (Table 2). However, the results for total fecundity followed a different trend. The highest total fecundity was observed as 149.7 eggs/female at 25/30°C, followed by 144.9 eggs/female at 20°C, 112.7 eggs/female at 25°C, 104.1 eggs/female at 15°C and 60.72 eggs/female at 30°C. A study of Francis et al. (2012) on the biology of the passion vine mealybug, *Planococcus minor* (Maskell, 1897) (Hemiptera: Pseudococcidae) demonstrated that females laid no eggs at 15°C but laid 269.8 eggs/female at 20°C, 205.6 eggs/female at 25°C, and 187.9 eggs/female at 29°C, under 60±10% RH and 24 h light.

A study conducted on host plants, geographical distribution, natural enemies and the biology of *P. citri* in Egypt by Ahmed and Abd-Rabou (2010) demonstrated that the mean daily fecundity of this pest on citrus was 136.1 eggs/female/day at 18°C, and 65-75% RH. Also, a study conducted by Kim et al. (2008) on the effect of temperature on the development and fecundity of *Pseudococcus cryptus* Hempel, 1918 on zucchini determined the mean daily fecundity of *P. cryptus* as 111 eggs/female/day at 25°C. In addition, Kim et al. (2008) stated that the preoviposition period and longevity of *P. cryptus* females on zucchini at 32°C were 12.5 and 31.3 d, respectively. The results have a similarity with the results obtained on the preoviposition period at 30°C in the present study. In addition, the longevity of *P. cryptus* was determined as 35.3, 27.3, 20.7 and 17.0 d at 16, 20, 24 and 28°C, respectively by Kim et al. (2008), unlike the results of this present study. This was thought to be due to host plant and species differences. Also, it is reasonable to conclude that fecundity and longevity of *P. citri* females were affected by temperature (Table 3).

Table 3. Development durations (d) of mature biological stages and the total fecundity (Eggs/female/day) of *Planococcus citri* females on grapefruit leaves under different temperature regimes (mean ± SE)*

Temperature (°C)	n	Preoviposition period	Oviposition period	Postoviposition period	Longevity of females	Total fecundity
15	16	32.0±1.87 b	40.9±3.92 c	7.5±1.82 b	80.4±4.84 c	104.1±23.89 a
20	15	17.5±2.26 a	21.7±1.85 b	3.7±1.35 ab	41.4±2.53 b	144.9±28.39 a
25	23	13.3±0.59 a	15.4±1.09 ab	2.8±0.45 a	30.7±1.11 a	112.7±15.81 a
25/30	30	13.1±0.91 a	15.7±0.81 ab	3.1±0.43 a	31.9±1.16 ab	149.7±15.45 a
30	11	13.3±1.87 a	13.4±1.06 a	3.1±0.62 a	29.5±2.31 a	60.7±17.47 a
35	50	No development				

* Means (± SE) sharing same letter within same column are not significantly different from each other (Tukey's HSD multiple range test at P < 0.05) ($F_{\text{Preoviposition}} = 31.569$, df = 4,90, P = 0.000; $F_{\text{Oviposition}} = 33.650$, df = 4,90, P = 0.000; $F_{\text{Postoviposition}} = 4.073$, df = 4,90, P = 0.005; $F_{\text{Longevity}} = 74.832$, df = 4,90, P = 0.000; $F_{\text{Number of eggs}} = 2.646$, df = 4,90, P = 0.039).

As shown in Table 4, the highest hatching rate of *P. citri* eggs on grapefruit leaves was obtained at 25/30°C with 100%, and the lowest one was 76.3% at 20°C. However, no hatching was observed at 35°C.

Mortality in immature stage ranged from 8.8 to 42.2% under different temperature regimes and while the most death was in the third nymph stage among immature stages, the least death ratio was calculated in pupal stage. Also, the mean amount of death in immature stage was higher than that in the preoviposition period. The highest mortality was at 30°C with 42.2% in immature stage and at 15°C with 27.3% in the preoviposition period (Table 4). A study on the effect of temperature on biological parameters of *P. citri* by Goldasteh et al. (2009) found that the lowest mortality on *Solenostemon scutellarioides* (L.) R.Br. was in the first and second nymph stages at 25°C.

Table 4. Hatching rate of eggs and mortality rates in different biological stages of *Planococcus citri* on grapefruit leaves under different temperature regimes

Temperature (°C)	Hatching rate		Mortality rate (%)						Sex ratio	
	n	n	First nymph stage	Second nymph stage	Third nymph stage	Pupal stage	Mortality in immature stage	Mortality in preoviposition period	♂:♀	
15	104	85.9	63	3.2	8.2	11.11	6.2	17.5	27.3	1:0.5
20	118	76.3	42	2.4	14.6	15.80	0.0	23.8	15.8	1:1.2
25	105	99.1	41	2.4	2.5	12.50	0.0	14.6	17.9	1:3.0
25/30	100	100.0	57	0.0	1.8	8.60	4.8	8.8	6.2	1: 1.5
30	106	92.5	45	20.0	13.9	19.20	0.0	42.2	26.7	1: 1.1
35	600	No development and hatching								

Life table parameters of *P. citri* on grapefruit leaves under different temperature regimes are given in Table 5. While the highest intrinsic rate was obtained at 25/30°C with 0.108 females/female/day, the least one at 15°C with 0.036 females/female/day (Table 5).

Table 5. Life table parameters of *Planococcus citri* on grapefruit leaves under different temperature regimes

Temperature (°C)	n	Intrinsic rate of increase, r_m^1	Net reproductive rate, R_o^2	Mean generation time, T_o	Gross reproduction rate, GRR	Theoretical population-doubling time, T_2	Finite rate of increase, λ
15	24	0.036±0.002 c	52.17±2.185	115.2	75.7	22.4	1.031
20	21	0.059±0.004 b	67.09±3.193	74.5	117.9	12.4	1.057
25	31	0.100±0.005 a	80.69±2.359	45.6	132.9	6.9	1.105
25/30	35	0.108±0.003 a	82.28±1.935	42.7	133.0	5.0	1.123
30	20	0.064±0.008 b	22.69±1.696	53.8	44.8	13.7	1.052
35	50	No development					

¹ Means (± SE) of the intrinsic rate of increase sharing same letters within same row do not differ significantly from each other (Tukey's HSD multiple range test at $P < 0.05$; $F_{\text{Intrinsic_rate}} = 44.583$, $df = 4, 126$, $P = 0.000$). ²The net productive rate and SE were assed bootstrap technique (5000 times).

The net reproductive rate ranged from 22.69 to 82.28 females/female/offspring and the highest one was seen at 25/30°C. The longest mean generation time was observed at 15°C with 115.210 d (Table 5). Moreover, the age-specific survival rates (l_x) and fecundities (m_x) of *P. citri* females on grapefruit leaves under different temperature regimes are shown Figure 1.

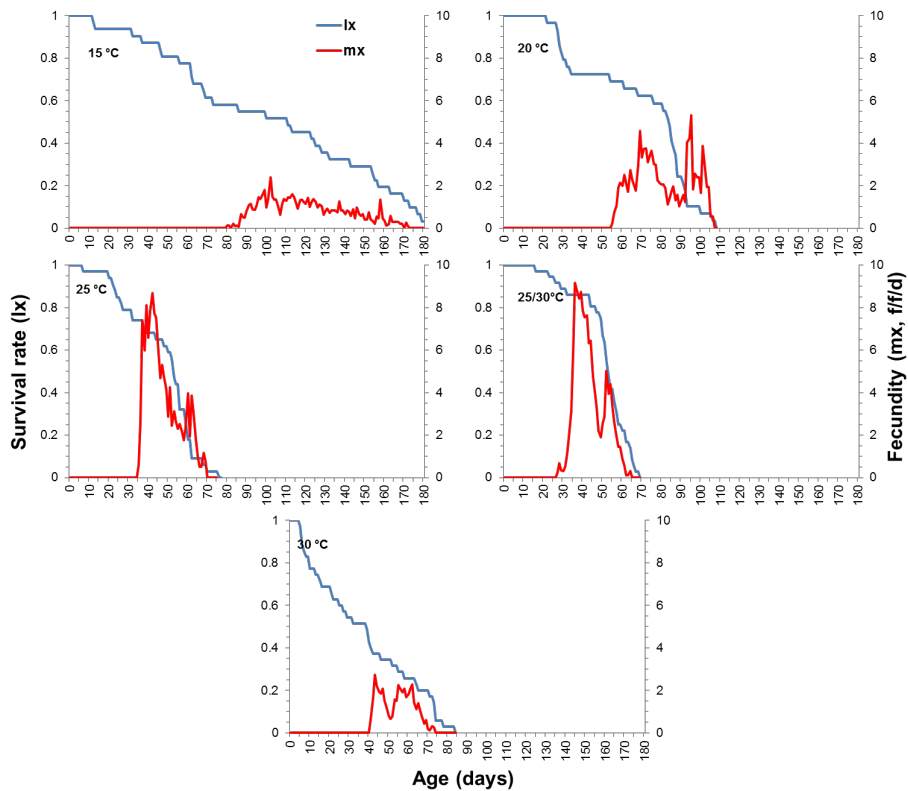


Figure 1. Age-specific survival rates (l_x) and fecundities (m_x) of *Planococcus citri* females on grapefruit leaves under different temperature regimes.

A linear regression analysis was applied to the developmental points within the 15 to 27.5°C range. Development at 30°C was outside the linear segment of the growth curve and therefore excluded from the linear regression. Within the chosen temperature range the developmental rates of *P. citri* increased linearly with increasing temperature. Development rates of *P. citri* at the different temperature regimes were modeled by linear regression line ($y = a \pm bx$) (Figure 2). According to the results of the regression model fitted separately for the data obtained from males and females of *P. citri*, the development rate equation of females was found as Develop. rate = 0.0024*Temp.-0.0231 ($R^2 = 0.95$; $P \leq 0.05$) for egg to adult stage and Develop. rate = 0.0015*Temp.-0.0128 ($R^2 = 0.97$; $P \leq 0.05$) for egg to egg stage. For male *P. citri*, the formula of the development rate equation was Develop. rate = 0.002*Temp.-0.0191 ($R^2 = 0.99$; $P \leq 0.05$) in egg to adult stage. Based on these equations, the development thresholds and thermal constants were calculated as shown in Table 6.

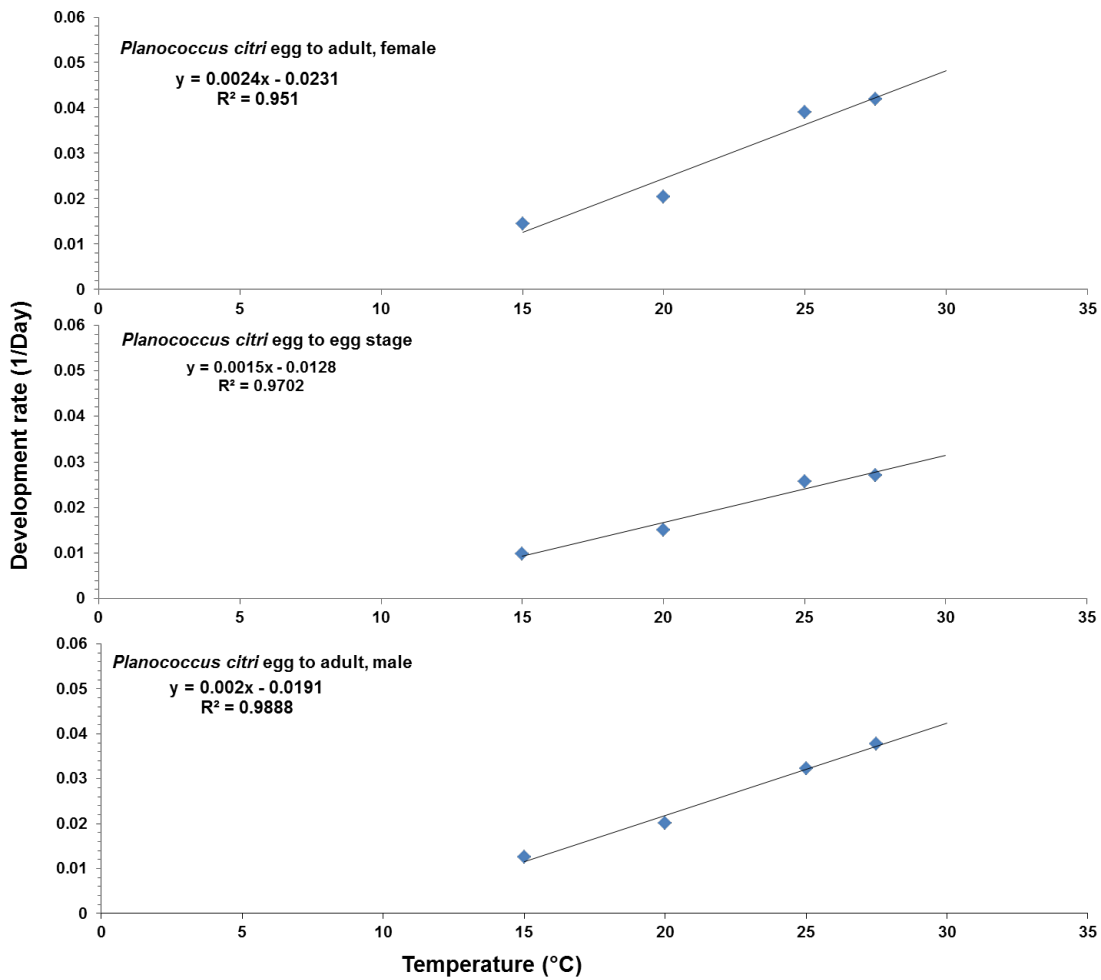


Figure 2. Temperature-dependent development rate of *Planococcus citri* at three constant and one alternating temperature (27.5°C for 25/30°C) on grapefruit leaves. Line is the linear regression analysis of developmental rate and temperature within the range of 15 to 27.5°C

Table 6. Regression equations and parameters of development rates of *Planococcus citri* on grapefruit leaves under different temperature regimes

Equations and parameters	Female from egg to adult stage	Female from egg to egg stage	Male from egg to adult stage
Equation	$y = 0.0024x - 0.0231$	$y = 0.0015x - 0.0128$	$y=0.002x - 0.0191$
Development threshold (-a/b) (°C)	9.63	8.53	9.55
Thermal constants (1/b) (°C.Day)	416.67	666.67	500.00
R ²	0.95	0.97	0.99

Asiedu et al. (2014) determined that total developmental time, female longevity and mean egg number laid by female of *P. citri* were 24.4 to 37.0 d, 32.9 to 38.1 d and 257 to 497 eggs/female, respectively. Francis et al. (2012) studied on the biological characteristics of the passion vine mealybug, *P. minor* at five constant temperatures (15, 20, 25, 29 and 35 °C) and 60±10% RH under constant light, and found that there was no development at 15 and 35°C. Also, male of *P. minor* completed their development from egg to adult in 51.5, 32.8 and 27.5 d under 20, 25 and 29°C, respectively. For females, the development from egg to adult lasted 48.8, 30.8 and 26.9 d at 20, 25 and 29°C, respectively.

The data analysis conducted at the end of the study showed that the optimum development temperature for *P. citri* was 25/30°C and the results showed that the pest can complete the most successfully its life cycle at 25/30°C. The last point should be noted as this temperature has a significant role on planning of control programs against this pest. When viewed from this aspect, the period in which the average temperature in the microclimate of the tree, especially grapefruit that has fruit settle inside the tree canopy, is 25/30°C in Adana Province is between July and September, and the most effective period for this insect in terms of reproductive and abundance is September. However, insecticide application conducted against this pest in this period may create significant residue problems for fruit in marketing and consumption. Herewith, the application of insecticides with low side effects against overwintering individuals on citrus trees, and before neonate crawlers settled under calyx of citrus fruit in end of spring will reduce the reproductive capacity and enable an increase in the success of biological control. In addition, such an approach will be a more environmentally-friendly in terms of IPM.

Acknowledgments

We would like to thank Scientific Research Projects Management Department of Çukurova University for their financial support for this study (project number ZF2014D2). Also, the study was supported by General Directorate of Agricultural Research and Policies and Biological Control Research Institute.

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Original article (Orijinal araştırma)

Nematode biodiversity in cereal growing areas of Bolu, Turkey

Bolu ili buğday alanlarında nematod biyoçeşitliliği

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Summary

Agricultural fields usually contain both plant parasitic and beneficial free-living nematodes. Plant parasitic nematodes have a negative impact on plant productivity and quality traits, whereas free-living nematodes can have beneficial effects on the agricultural soils health. This study was conducted to investigate the diversity of both plant parasitic nematodes and beneficial free-living nematode. Soil samples were collected in 2015 from wheat growing areas of Bolu Province to investigate soil nematode diversity. Bolu Province was divided into two sub-areas according to elevational. Forty-three nematode taxa were found in the samples; 13 taxa plant parasites, 12 bacterivores, 4 fungivores, 10 omnivores and 4 predators. Relative distribution of nematode trophic groups indicated a bacterivore dominated community, followed by plant parasites, fungivores and omnivores; predators represented only a small proportion. Free-living nematodes, especially bacterivores of basal fauna members and large bodied omnivore members were in good condition in abundance and diversity. General community and maturity indices were calculated for each sample and for the two sub-areas. They produced narrow range values with no significant differences. The study revealed that soil food web in wheat growing areas of the province was in fair to good condition based on nematode diversity.

Keywords: Bioindicator, diversity, nematodes, trophic groups, wheat

Özet

Tarım arazileri hem bitki paraziti nematodlar hem de serbest yaşayan yararlı nematodları birlikte barındırmaktadır. Bitki paraziti nematodlar bitki kalite parametreleri üzerinde olumsuz etkiler doğururken, serbest yaşayan nematodlar ise yaşamsal faaliyetleri sonucu olumlu etkilere sahiptir. Dolayısıyla, çalışma hem bitki paraziti nematodların çeşitlilik yapısı hem de yararlı nematodların çeşitlilik yapısını incelemek üzere yürütülmüştür. Bu amaçla, 2015 yılında Bolu ili buğday alanlarından toprak örnekleri toplanmıştır. Bolu ili rakım farklılıkları göz önüne alınarak iki farklı alt-bölgeye ayrılmıştır. Çalışma alanında, 13 bitki paraziti, 12 bakterivor, 4 fungivor, 4 predatör ve 10 omnivor gruba ait olmak üzere, toplam 43 nematod taksonu tespit edilmiştir. Nematod trofik gruplarının oransal dağılımlarına göre, bakterivorların baskın olduğu bir kommunité yapısına sahip olduğunu, bunu bitki parazitleri, fungivorlar, omnivorlar izlemiş, predatörler ise oldukça düşük bir orana sahip olduğu saptanmıştır. Serbest yaşayan nematodlardan, özellikle temel fauna bileşeni olan bakterivor ve iri cüsseli omnivore nematodların hem çeşitlilik hem de yoğunluk bakımından iyi durumda olduğu görülmüştür. Genel kommunité ve nematode maturity indisleri her iki alt-bölgeden alınan toprak örnekleri için hesaplanmış, indis değerleri arasında ise istatistikî olarak bir farklılığın görülmediği, dar bir aralıkta dağıldığı saptanmıştır. Çalışma sonuçlarına göre, nematode biyoçeşitlilik yapısına dayanılarak elde edilen bilgiler ışığında, Bolu ili buğday alanlarındaki toprak besin ağı orta ve iyi dereceler arasında yer aldığı sonucuna varılmıştır.

Anahtar sözcükler: Biyoindikatör, çeşitlilik, nematodlar, trofik gruplar, buğday

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Received (Alınış): 14.02.2017

Accepted (Kabul ediliş): 14.04.2017

Published Online (Çevrimiçi Yayın Tarihi): 08.06.2017

Introduction

Soil contains a relatively large number of different groups of organisms that maintain ecological services through their physical and metabolic activities. Some of the ecological functions of soil organisms are decomposition of organic matter from plant and animals, cycling of minerals and nutrients, redistribution of minerals and nutrients, reservoirs of minerals and nutrients in their bodies, sequestration of carbon, detoxification of pollutants (mostly by micro fauna), regulation of soil structure, community self-regulation and biological regulation or suppression of pest species (Ferris, 2016). The diversity, abundance and functions of soil biota in the soil food web determine the quality of the services or so called soil health.

Nematodes are one of the most abundant and diverse microscopic animals in the soil environment and they are represented at different soil food-web levels interacting with soil biota in multiple ways (Freckman & Ettema, 1993; Ritz & Trudgill, 1999; Ferris et al., 2001; De Deyn et al., 2004; Viketoff et al., 2011). Nematodes are known to be among the most taxa rich organismal group on earth, having with an estimated 500 000 to 1 000 000 species. Only about 20 000 species have been described, and the systematic literature is widely dispersed (Hodda, 2007).

To date, the most extensively studied group of nematodes have been the agricultural pest nematodes that cause economic losses by damaging plants. However, a vast range of ecologically beneficial free-living nematodes are also present in the same soil environment that is inhabited by plant parasitic nematodes. Nematodes are usually divided into five trophic groups including plant parasitic (herbivores), bacterial feeding (bacterivores), fungal feeding (fungivores), nematodes that feed on other nematodes (predators) and nematodes that can feed on plants, fungi, bacteria and other nematodes (omnivores). These groups are associated with their food by examining the morphology of their feeding apertures (Yeates et al., 1993; Bongers & Bongers, 1998; Neher, 2001).

Nematodes live in water-filled soil pores exposed to physical, chemical and biological changes in soil conditions that affect them and change their composition. Their dynamic community composition, long life and their ease of extraction, examination and allocations to trophic groups have made them one of the most preferred indicators of soil biodiversity around the world over the last two decades (Bongers, 1990; Yeates, 2003; Mulder et al., 2005). It is accepted that a healthy soil should contain an abundant and a diverse community of free-living nematodes but less abundant and less diverse composition of plant parasitic nematodes.

The health of a soil ecosystem can be estimated by examining the diversity and abundance of nematodes. As proposed by Bongers (1990), nematode communities have been examined by using a specifically developed maturity index (MI) and the generalist community diversity indices.

Little information is available on nematodes in agricultural soils of Bolu Province. The most recent and the extensive study on the subject was that of Imren et al. (2015), which determined the occurrence of the plant parasitic nematodes associated with cereal-production areas of Bolu Province. However, there has been no comprehensive study of nematode communities including free-living species in the region. Since the nematode assemblage of every region tend to change with the variations in ecosystem components, geographical differences and climate, locally focused studies are valuable to assess nematode biodiversity and its consequences.

Through this study, it is expected that the baseline data will be provided for the future nematological studies in this area. The objective of this study was to investigate the status of the nematode community composition in relation to soil conditions in wheat growing areas of Bolu Province, Turkey.

Material and Methods

Study site and sampling

Bolu Province, covering about 1% of the total land area of Turkey, is located in the Western Black Sea Region with an 8.276 km² land area, of which, 55% is forests and 18% cultivated agricultural land. The climate in the area is characterized by wet cold winters and hot dry summers. Average annual temperature and annual precipitation is estimated as 10.9°C and 573 mm, respectively. Due to the

geographical and climatic features, agricultural production in the province is limited to a few main crops including wheat, potato and sugar beet. Wheat is the most commonly grown crop of the province because of its best suited to the region's precipitation (Anonymous, 2016).

In order to investigate nematode community, structure a total of 26 soil samples were collected in wheat growing areas of Bolu Province from June to August 2015. Sampling sites were categorized into two sub-areas to compare community variables. Area 1 was around Bolu City and Area 2 included Gerede, Dörtdivan and Yeniçağa Districts. The districts in Area 2 were at least 7-10 km apart from each other and, and have similar elevation, soil type and topographical features. The two study areas were about 60 km apart separated by mountains, therefore they had significant differences in elevation (726 and 1475 m for Area 1 and 2, respectively).

Each soil sample was collected from a visually determined section (0.5 ha) of a field from 10-20 cm depth using a 25-mm soil auger. Each sample consisted of 10-15 soil cores totaling approximately 1-1.5 kg in weight. Samples were placed in labeled plastic bags and transported to the laboratory in insulated boxes.

Nematode extraction

Nematodes were extracted from individual soil samples by using a modified Baermen funnel technique (Whitehead & Hemming, 1965). Subsamples (100 g) of homogenized soil were placed in 15-cm plastic Petri dishes and water added until the sample was covered. After 48 h, nematodes that had migrated into the water in the lower part of dish, which was then transferred to 100 ml measuring cylindrical and the nematodes allowed settle for 8 h. The volume was then reduced to 15 ml by discarding excessive water from the Petri dish. Finally, nematode containing suspension were rinsed into 15 ml and transferred to plastic Falcon tubes for storage at 4°C until assessed.

Identification and grouping of nematodes

The water was removed from the tubes by pastor pipette concentrating the nematodes in 1 ml. For counting, the nematode suspension was mixed with a micropipette and 100 µl transferred to a glass slide for examination under light microscope at 100X magnification. Nematodes were identified mostly to genera; however, four taxa were only identified to family, and they were also allocated to trophic group based on morphological structure of mouth parts specialization for feeding habit and diet.

Nematode community analyses

Nematode community structure was analyzed in relation to the site differences especially the distance between the sampling sites and two locations in wheat fields (alpha and beta diversity). Diversity indices, the summarized numerical expression of many taxa have been calculated to assess the diversity of nematode communities in soil. Generalist diversity indices such as the number of taxa (SR), the Shannon diversity index (H'), the Shannon evenness index (E), Hills N1 and N2, Simpsons D; MI family and trophic diversity [bacterivore to fungivore ratio, Ba/(Ba+Fu)] indices developed specifically for nematode biodiversity and trophic diversity indices were computed as described in (Neher & Darby, 2009). Colonizers-persisters (C-P) groups, based on the r-K reproductive strategy, were placed on a 1-5 scale based on their reproductive capacities (Bongers, 1990).

Statistical analysis

Diversity indices were calculated and log transformation applied to all nematode abundance data to examine any significance between the locations prior to performing ANOVA and T-test in SPSS at $P \leq 0.05$ significance level.

Results and Discussion

Nematode faunal structure

Forty-three nematode taxa were found in the samples; 13 plant taxa plant parasites, 12 bacterivores, 4 fungivores, 10 omnivores and 4 predators. The abundance of nematodes at taxon level ranged from 3 to 390 (individuals/100 g soil) and abundance among the samples ranged from 531 to 2145 (individuals/100 g soil). The list of nematode genera, their abundance and frequencies are given in in Table 1.

Table 1. Nematode abundance (average number of individuals/100 g of soil), frequency (percent occurrence in samples) and associated colonizers-persisters (C-P) scale values of the nematode genera

Nematodes	Area 1		Area 2		C-P value
	Abundance	Occurrence (%)	Abundance	Occurrence (%)	
Plant Parasitic					
<i>Filenchus</i> Andrassy, 1954	74.4±10.6	68.8	60.4±12.5	51.5	2
<i>Helicotylenchus</i> Steiner, 1945	6.9±2.5	43.8	13.2±3.8	30.9	3
<i>Heterodera</i> Schmidt, 1871	0.6±0.6	6.3	1.2±0.9	10.3	3
<i>Merlinius</i> Siddiqi, 1970	38.8±7.3	68.8	21.7±5.6	61.8	3
<i>Pratylenchoides</i> Winslovv, 1958	26.3±5.5	50	5.2±1.2	20.6	3
<i>Paratylenchus</i> Micoletzky, 1922	18.8±4.7	43.8	3.1±2.0	20.6	2
<i>Paratrophurus</i> Arias, 1970	0.6±0.6	6.3	0±0.0	0	3
<i>Pratylenchus</i> Filipjev, 1936	20.6±5.5	62.5	6.5±2.9	41.2	3
<i>Psilenchus</i> De MAN, 1921	0.3±0.3	6.3	0.5±0.5	10.3	2
<i>Rotylenchus</i> Linford &Oliveira, 1940	0.3±0.3	6.3	0±0.0	0	3
<i>Trophurus</i> Loof, 1956	0,1±0.0	6.3	0±0.0	0	3
<i>Tylenchorhynchus</i> Cobb, 1913	48.8±5.8	68.8	39.3±7.2	61.8	3
<i>Tylenchus</i> Bastian, 1865	22.5±5.1	31.3	46.4±5.8	61.8	2
Bacterivores					
<i>Rhabditis</i> Dujardin, 1845	28.1±7.9	68.8	31.1±9.2	92.7	1
Monhysteridae	77.5±13	93.8	83.3±11.4	100	2
<i>Cephalobus</i> Bastian, 1865	105.6±9.4	100	76.7±11.3	92.7	2
<i>Eucephalobus</i> Steiner 1936	91.3±7.3	93.8	110.2±11.2	100	2
<i>Acrobeloides</i> Thorne, 1937	105.6±16.2	100	51.8±17.6	92.7	2
<i>Achromadora</i> Cobb, 1913	0±0.0	0	4.3±2.5	20.6	3
<i>Cervidellus</i> Thorne, 1937	2.5±1.4	18.8	0±0.0	0	2
<i>Alaimidae</i>	1.3±1.2	6.3	0±0.0	0	4
<i>Alaimus</i> de Man, 1880	5.0±2.0	37.5	3.5±1.4	30.9	4
<i>Wilsonema</i> Cobb, 1913	3.8±1.7	25	1.1±0.9	10.3	2
<i>Plectus</i> Bastian, 1865	68.8±8.8	87.5	95.6±12.5	100	2
<i>Prismatolaimus</i> de Man, 1880	0.6±0.6	6.3	1.0±0.9	10.3	3

Table 1. (Continued)

Nematodes	Area 1		Area 2		C-P value
	Abundance	Occurrence (%)	Abundance	Occurrence (%)	
Fungivores					
<i>Aphelenchoides</i> Fischer, 1894	107.5±7.3	100	79.6±7.4	100	2
<i>Aphelenchus</i> Bastian, 1865	63.8±9.3	93.8	37.2±10.7	72.1	
<i>Ditylenchus</i> Filipjev, 1936	68.8±11.7	93.8	37.3±7.3	72.1	2
<i>Tylencholaimus</i> de Man, 1876	3.1±2.1	12.5	0±0.0	0	4
Predators					
<i>Mononchus</i> Bastian, 1865	6.3±3.4	25	10.2±2.4	72.1	4
<i>Clarkus</i> Jairajpuri, 1970	0±0.0	0	1.1±0.9	10.3	4
<i>Seinura</i> Fuchs, 1931	0±0.0	0	1.3±0.6	20.6	4
<i>Tripyla</i> Bastian, 1865	0.7±0.0	7.2	0±0.0	0	3
Omnivores					
Dorylaimidae	2.5±2.4	6.3	0±0.0	0	4
<i>Actinolaimidae</i>	7.5±3.6	25	1.8±0.9	10.3	5
<i>Dorylaimus</i> Dujardin, 1845	1.3±1.2	6.3	0±0.0	0	4
<i>Campydora</i> Cobb, 1920	1.3±1.2	6.3	0±0.0	0	4
<i>Mesodorylaimus</i> Andrassy, 1959	0.3±0.3	6.3	0.5±0.5	10.3	4
<i>Prodorylaimus</i> Andrassy, 1959	11.9±4.0	50	13.6±2.8	82.4	4
<i>Aporcelaimus</i> Thorne & Swanger, 1936	4.4±4.2	6.3	11.3±4.3	20.6	5
<i>Aporcelaimellus</i> Heyns, 1965	38.8±5.3	87.5	46.0±6.0	82.4	5
<i>Eudorylaimus</i> Andrassy, 1959	5.6±5.4	6.3	0±0.0	0	4
<i>Belondira</i> Thorne, 1939	21.3±8.5	56.3	38.3±9.9	61.8	5

Although the study area was divided into sub-areas based on the altitude differences, the results revealed no significant differences on nematode community parameters between the sub-areas, such as SR, abundance and occurrence in the samples. However, previous studies from various parts of the world have indicated that altitude was an important parameter in shaping soil nematode communities, regarding characteristics like distribution, species richness and abundance (Hoschitz & Kaufmann, 2004; Háněl & Čerevková, 2010; Zhang et al., 2012; Tsiafouli et al., 2017).

The maximum plant parasitic nematode abundance in the study was 210 individuals/100 g soil. Fourteen genera of plant parasitic nematodes were identified. Most abundant and common genera were *Filenchus*, *Tylenchorhynchus*, *Merlinius*, *Pratylenchus* and *Tylenchus*, in that order (Table 2). There was no significant difference between the two areas in terms of the abundances of plant parasitic nematodes.

Abundances of plant parasitic nematodes in our study were not as high as those of the study by Bao & Neher (2011), in which the abundance of plant parasitic nematodes ranged 719 to 3,578 individuals/100 cm³ of dry soil from Vermont, USA. Bulluck et al. (2002) also reported that plant parasitic nematodes were the most abundant group and ranged from 83 to 88% of the total community. However, Akyazi et al., 2014 reported lower abundance values of plant parasitic nematodes from Ordu Province in Central Black Sea Region of Turkey. Low plant parasitic nematode densities in agricultural soils are considered a good indicator of soil health indicating relative minimal damage and consequent less economic losses on crops.

Table 2. Nematode maturity indices and general community indices (Neher & Darby, 2009) for two wheat growing areas in Bolu Province, Turkey

Indices	Area 1	Area 2
SR	17.13±05	16.4±0.1
PPI	2.51±0.0	2.38±0.1
FI (PPI/MI)	1.07±0.0	1.01±0.1
ΣMI	1.74±0.1	1.84±0.1
MI	2.37±0.1	2.41±0.1
ΣMI 2-5	2.69±0.1	2.82±0.1
MI 2-5	2.40±0.1	2.47±0.1
Ba/(Ba+Fu)	0.66±0.0	0.73±0.0
H' (Shannon)	2.52±0.0	2.47±0.1
E (H/Hmax)	0.8±0.0	0.8±0.0

Bacterivores consisted of 11 genera in one family and were the dominant nematode fauna in both areas, with no significant difference in abundance between the areas. The genera *Cephalobus*, *Acrobelloides*, *Eucephalobus* and family Monhysteridae, in that order, were the most abundant and the widespread bacterivores (Table 2). These findings are consistent with those of Ettema & Bongers (1993), Neher (1999) and Yeates (2003), who reported that Cephalobidae and Rhabditidae were the most abundant families in agricultural soils. These nematodes are the basal faunal members that are stress tolerant and long-lived groups in the C-P 2 group (Bongers & Bongers, 1998). Therefore, they are fairly common in various agroecosystems throughout the seasons even if other free-living nematode groups are missing.

In both study areas, Rhabditids were found to be widespread (with 28 and 31 individuals/100 g soil in Area 1 and 2, respectively) but their abundance was not significantly different. Their presence is considered to be an indication of the presence of decomposing organic matter or recent chemical fertilization. Given their opportunistic nature, rapidly increasing or decreasing in number with changes in nutrient availability (especially nitrogen), and their short life cycle, they placed in the C-P 1 group (Ferris, 2001). Ferris et al. (2004) reported that Rhabditid nematodes tended to have low population numbers when soil organic matter was inadequate. Thus, their presence and wide distribution in the study areas suggest that wheat growing soils of Bolu Province contained decomposing organic matter when the samples were taken. However, this was not the case in wheat growing areas of Central and South Eastern Anatolian where Rhabditidae were relatively scarce (Şahin et al., 2009; Yıldız, 2012; Yıldız & Elekçioğlu, 2012). Relative low annual precipitation can reduce organic matter decomposition as a result of the slow activity of microfauna in soil (Laakso et al., 2000).

However, there have been various studies that report findings consistent with our results, supporting the view that bacterivore nematodes are the most abundant nematode trophic group in agricultural soils (Porazinska et al. 1999; Carmen & Zaborski, 2014; Tsiafouli et al. 2017).

Fungivores were represented by *Aphelenchoides*, *Ditylenchus*, *Aphelenchus* and *Tylencholaimus*, and their abundance listed in Table 2. The abundance and the distribution of *Aphelenchoides* was not significantly different between the study areas, however, *Aphelenchus* and *Ditylenchus* had a slightly lower abundance in Area 2 ($P \leq 0.05$). The fungivore trophic group was the third most abundant (Figure 1).

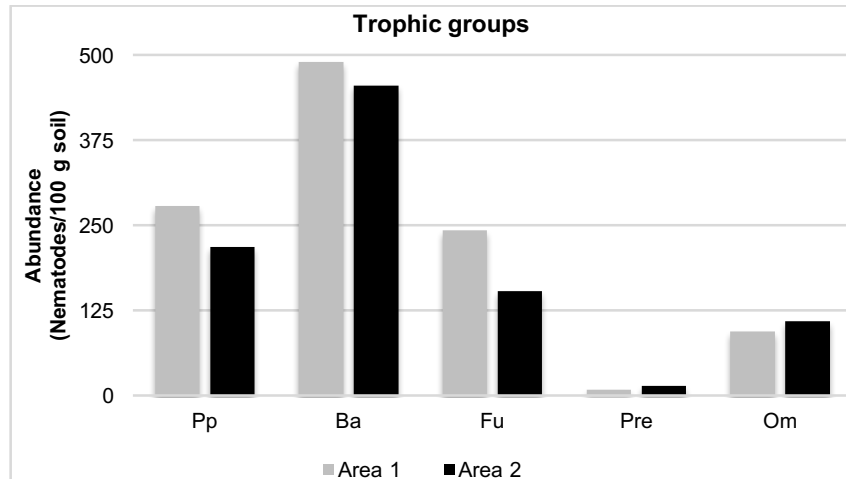


Figure 1. Trophic structure of nematode fauna in two wheat growing areas in Bolu Province, Turkey, based on abundance.

Fungivore nematodes feed on soil fungi that are important in the decomposition of high C:N ratio organic matter (Liang et al., 2009), such as dry wood or straw, thus the diversity and abundance of these nematodes is influenced by the soil organic matter quantity, sufficient soil humidity and soil temperature.

Predators were the least abundant and distributed trophic group in the study (Table 2, Figure 1). Since the predator species in a community occupy the narrow top portion of the trophic pyramid, it is expected that they will be detected in low numbers in a community. Predatory nematodes of C-P 5 have larger body, longer life cycle and extreme sensitivity to soil disturbances, such as heavy tillage in agricultural areas. Predatory nematodes are more likely to be abundant and diverse in undisturbed natural ecosystems, such as grasslands and pastures (Bongers & Bongers, 1998; Georgieva et al., 2002).

The omnivore group consisted of eight genera in two families in Area 1 and five genera and one family in Area 2. *Aporcelaimellus* and *Belondira* were the most abundant and the common nematodes in both areas. Other genera were present with in lower numbers (Table 2). Omnivorous nematodes have similar behavioral and structural patterns to those of predators in agroecosystem and undisturbed natural ecosystems due to their similar features and also being in the C-P 5 group (Bongers & Bongers, 1998). However, they differ from predatory nematodes in their broader diet. Due to their permeable cuticle, large bodies, long life and low fecundity, omnivores are considered to be the soil nematode group most sensitive to stress conditions (Shao et al., 2008).

Relative distribution of nematode trophic groups of the study areas indicated a bacterivore dominated community, followed by plant parasitic nematodes, then fungivores and omnivores, with predators the least abundant group (Figure 1).

Nematode diversity

C-P groups displayed a very similar distribution pattern in both areas (Figure 2). The results revealed that the differences between the areas, especially the altitude, did not have any significant influence on the abundance of the C-P groups. C-P groups of nematodes are based on the life history similarities, referred to the *r-K* strategist principles, to be able to condense information and to facilitate interpretations about these complex species rich nematode communities. Also, functional groups of nematodes can be considered as groups of species that have similar functions in ecosystem services even if they belong to distant taxonomic groups (Bongers & Bongers, 1998; Schratzberger et al., 2007).

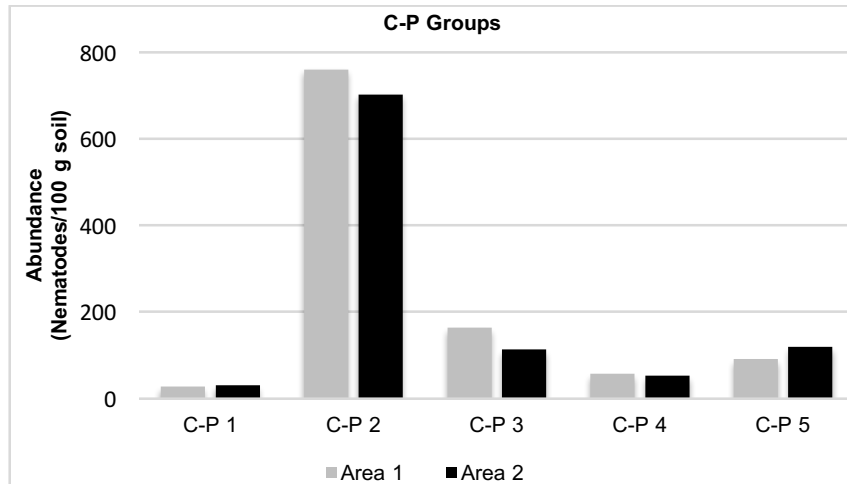


Figure 2. C-P group abundances of nematode communities in two wheat growing areas in Bolu Province, Turkey.

The findings of the study indicate that there was no significant difference in nematode community and maturity indices among the samples (alfa diversity) or between the areas (beta diversity). The average values of indices in both areas were: SR, 17 and 16; H', 2.52 and 2.47; E, 1.56 and 1.54; Ba/(Ba+Fu), 0.66 and 0.73; PPI, 2.51 and 2.38; MI, 2.38 and 2.41; MI 2-5 2.40 and 2.47 (Table 2).

The stability of the index values across the study area, either among the samples or between the sub-areas, suggest that nematode community structure also share common characteristics in these two areas and there was no obvious influencing factor detected over the nematodes in any of the sampled sites. This can be attributed to the commonalities in the use and history of the fields sampled. These fields had primarily non-irrigated, low intensive agricultural regimes that limit causative factors on land properties and ultimately nematode community structure.

To measure nematode community and soil environment interactions, general community indices and specifically developed maturity indices for nematode faunal analysis have been extensively used to obtain information and monitor soil condition from the seminal work of Bongers (1990) through to the latest study by Tsiafouli et al. (2017). Community indices are referred to assess SR, the relative abundances of the species and the evenness in a community H', Simpsons index and Hills index. Whereas, MI family has been the most reliable tool to assess the degree of stress in soil due to its greater sensitivity (Li et al, 2005). Trophic group ratio indices such as bacterivore to fungivore ratios are preferred to assess impact of soil environment over the nematode community depending on the nematode community changes in the diversity and numbers (Neher & Darby, 2009).

In conclusion, a well-established mature nematode community is also considered to be good indicator of the wider soil biota and, ultimately, soil health. In Bolu Province, nematode fauna is dominated by bacterial-feeding, plant parasitic, fungal-feeding and omnivorous nematodes. Its nematode community is well balanced and diverse structure; consequently, indicating a healthy environment for soil biota and agricultural production.

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Original article (Orijinal araştırma)

Neonicotinoid resistance in *Bemisia tabaci* (Genn., 1889) (Hemiptera: Aleyrodidae) populations from Antalya, Turkey¹

Bemisia tabaci (Genn., 1889) (Hemiptera: Aleyrodidae)'nin Antalya, Türkiye popülasyonlarında neonicotinoid direnci

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Summary

Bemisia tabaci (Genn., 1889) (Hemiptera: Aleyrodidae) is one of most important pests of many crops worldwide. Furthermore, it has the ability to rapidly develop resistance to diverse group of insecticides, hence controlling this pest is problematic. The aim of the current study was to investigate neonicotinoid resistance status of different whitefly populations collected from Antalya, Turkey during summer 2011. *Bemisia tabaci* populations from five different locations in Antalya were all biotype B. Resistance to acetamiprid was between 4.4 and 30.4 times the LC₅₀ of a susceptible population. Similarly, resistance to thiamethoxam was between 8.6 and 31.8 times compared to susceptible population. With the exception of one population, there was high correlation ($r^2 = 0.92$) between LC₅₀ for acetamiprid and thiamethoxam. The LC₅₀ confidence limits for all field populations overlapped for both neonicotinoids. Furthermore, LC₉₀ values of all field populations for both neonicotinoids were higher than the recommended doses. These findings indicate development of uniform neonicotinoid resistant for whitefly in the region. The resistance homogeneity and cross resistance in the region sampled is discussed.

Keywords: *Bemisia tabaci*, neonicotinoid, biotype, whitefly, Antalya

Özet

Bemisia tabaci (Genn., 1889) (Hemiptera: Aleyrodidae) tüm dünyada birçok kültür bitkisinde görülen çok önemli zararlılardan biridir. Ayrıca, zararlı birçok gruptan insektisite karşı hızla direnç geliştirme kabiliyetine sahip olması nedeniyle mücadelesi problemlidir. Bu çalışmanın amacı, 2011 yılı yaz döneminde Antalya (Türkiye)'den toplanan farklı beyazsinek popülasyonlarının neonicotinoid grubu insektisitlere karşı direnç durumunu araştırmaktır. Antalya'nın beş farklı bölgesinden toplanan beyazsinek popülasyonlarının hepsinin B biyotipi olduğu belirlenmiştir. Acetamiprid için hassas bir popülasyona göre LC₅₀ dayanıklılık katsayılarının 4,4 ve 30,4 kat arasında olduğu görülmüştür. Benzer şekilde, hassas popülasyonla karşılaştırıldığında thiamethoxam için dayanıklılığın 8,6 ve 31,8 kat arasında değiştiği belirlenmiştir. Bir popülasyon dışında, acetamiprid ve thiamethoxam LC₅₀ değerleri arasında yüksek bir korelasyon ($r^2 = 0.92$) olduğu tespit edilmiştir. Her iki neonicotinoid içinde, araziden toplanan tüm popülasyonlara ait LC₅₀ değerlerinin güven aralıklarının iç içe geçtiği belirlenmiştir. Ayrıca, her iki neonicotinoid içinde, LC₉₀ değerlerinin arazi popülasyonlarında tavsiye dozunun üzerinde olduğu görülmektedir. Bu bulgular bölgede neonicotinoidler için homojen bir dayanıklılığın gelişmekte olabileceğine işaret etmektedir. Beyazsinek popülasyonlarında görülen dayanıklılığın homojenliği ve çapraz direnç konuları test edilen neonicotinoidler için tartışılmıştır.

Anahtar sözcükler: *Bemisia tabaci*, neonicotinoid, biyotip, beyazsinek, Antalya

¹ The study was published as an abstract poster presentation at International Plant Protection Congress held between 24-27 August 2015 in Berlin, Germany.

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Received (Alınış): 25.11.2016

Accepted (Kabul ediliş): 20.04.2017

Published Online (Çevrimiçi Yayın Tarihi): 29.05.2017

Introduction

Bemisia tabaci (Genn., 1889) (Hemiptera: Aleyrodidae) is a cosmopolitan pest causing substantial economic loss in many crops (Menn, 1996) The main damage is caused by direct feeding on plant tissues, excretion of large amounts of honeydew and transmission of several important plant viruses (Bedford et al., 1994; Brown et al., 1995; Denholm et al., 1998; Horowitz et al., 2003; Jones, 2003). Insecticide application has been one the main strategies to minimize the damage potential of pest in many cropping system. However, heavy insecticide use creates substantial selection pressure on the pest, which is known to have 34 biotypes (Tay et al., 2012). As a result, the pest has become resistant to several groups of insecticides shortly after their common use in the field. Consequently, *B. tabaci* resistance was reported from more than 20 countries for 35 active ingredients (Roidakis et al., 2005). Neonicotinoids, a relatively a new class of insecticides, are no exception for this pest and have lost their initial efficacies for certain *B. tabaci* populations around the world (Elbert & Nauen, 2000; Rauch & Nauen, 2003; Wang et al., 2010; Vassiliou et al., 2011). Furthermore, neonicotinoid resistance was reported to be associated with *B. tabaci* biotypes replacing each other completely in areas with heavy neonicotinoid usage (Simon et al., 1999; Pan et al., 2010; Sun et al., 2013). The present study was initiated to determine the neonicotinoid resistance status of whitefly populations from Antalya, Turkey where extensive vegetable growing and pesticide applications take place.

Material and Methods

Insects

Bemisia tabaci populations were initially collected from five different locations in Antalya Province, Turkey during summer 2011 (Table 1). The adults were reared on eggplant in cages maintained at 25-28°C with a 16L:8D h photoperiod for two generations until they reached adequate numbers and homogeneity. A population from southwestern Turkey (Koçarlı, Aydın Province), where pesticide applications have been less than average, was collected in 2009 and maintained at the Entomology Laboratory of Akdeniz University. This population was used as the susceptible reference in bioassays. Also, a lab population originally collected in 2009 from Konaklı, Antalya, where pesticide applications were frequent, was included in the study.

Table 1. Sources of *Bemisia tabaci* populations used in neonicotinoid bioassays

Collection Location	Collection Date	Host	Code
Serik	31 May 2011	Squash	Ser
Aksu	28 July 2011	Sesame	Aks
Tosmur	31 May 2011	Tomato	Tos
Konaklı	31 May 2011	Tomato	Kon
Kumluca	03 June 2011	Tomato	Kum
Koçarlı (susceptible reference population)	2009 and lab maintained since collection	Cotton	Lab1
Konaklı (additional reference population)	2009 and lab maintained since collection	Tomato	Lab2

Insecticides and leaf-dip bioassay

The study used two formulated neonicotinoid insecticides, acetamiprid 200 g L⁻¹ (Mospilan SL, Nippon Soda Co, Tokyo, Japan) and thiamethoxam 240 g L⁻¹ (Actara SC, Syngenta, Basel, Switzerland) for adult whitefly bioassays. Dose-response bioassays were based on Insecticide Resistance Action Committee suggested procedure. Briefly, cotton leaf discs (39 mm in diameter) were immersed in aqueous solution of insecticide containing 0.02% Triton X-100 for 5 s. The leaf discs were allowed to dry and placed upside down on Petri dish containing a thin layer of sterile agar (2%) layer. Between 18 to 22 *B. tabaci* females were placed on each leaf disc after a brief anesthetization using carbon dioxide. The dishes were inverted for the insects to orientate on abaxial side of the leaf disc and placed in a growth chamber (26±1°C, 16L:8D h). Four concentrations of each insecticide were tested to ensure mortality between 0 and 100%, along with a control concentration containing only 0.02% Triton X-100. The mortality data was corrected for each insecticide using Abbott's formula (Abbott, 1925). Each pesticide concentration was replicated three times and final mortality was assessed 48 h after insecticide exposure.

Biotype determination

Mitochondrial cytochrome oxidase I (*mtCOI*) gene was used to determine the biotypes of the pest. Adult whitefly DNA was extracted individually based on the method of Doyle & Doyle (1987). The primers C1-J-2195 and L2-N-3014 were used for *mtCOI* amplification in a thermal cycler according to Frohlich et al. (1999). Amplicons were directly sequenced in both directions using DTCS kit according to manufacturer's instructions (Beckman Coulter 8000 Genetic Analysis System, Brea, CA, USA). Five different individuals from each population were sequenced for *mtCOI* region and the sequences were compared to known biotypes from representative GenBank accessions and previously reported biotypes from the region sampled (Ikten et al., 2007). A divergence criteria <3.5 was used for inclusion in a biotype (Dinsdale et al., 2010).

Statistical analysis

PoloPlus program was used for probit analyses of the concentration-dependent mortality data (LeOra Software, 1987). Resistance ratios were calculated by dividing LC₅₀ and LC₉₀ values of field collected whitefly populations by the values obtained for the susceptible reference population (Lab1).

Results

Amplification of COI region from all whitefly samples produced 850bp DNA fragments. Consensus sequences of each whitefly samples were compared to known biotypes with a divergence threshold of 3.5%. All COI sequences from the whiteflies was aligned using ClustalW and produced an identical haplotype. Comparison of single haplotype to known biotype sequences resulted in 100% identity to biotype B, hence all whitefly samples from different populations were assigned to biotype B.

Bioassay results showed thiamethoxam had varying degrees of toxicities for whitefly populations. The highest LC₅₀ of 115.8 mg/L was obtained from Tos population, whereas Lab1 had an LC₅₀ of 3.7 mg/L for thiamethoxam. Furthermore, Lab2 had an even lower LC₅₀ of 1.2 mg/L (Table 2). The resistance ratios ranged from 8.6 to 31.8 for LC₅₀ and 9.9 to 40.8 for LC₉₀ based on Lab1 (Table 2). Using Lab2, the resistance ratios were 3-7 times higher. Moreover, all field collected whitefly populations had LC₉₀ values of higher than the recommended field dose of thiamethoxam. Whereas, LC₉₀ values of susceptible Lab1 and Lab2 populations were much lower (10% or lower) than recommended field dose (240 µg ai ml⁻¹).

The highest LC₅₀ for acetamiprid (97.5 mg/L) was found in the Aks population. Lab1 population had an LC₅₀ of 3.2 mg/L, giving a 30 times resistance ratio for Aks (Table 3). As in the thiamethoxam assays, Lab2 had an even lower LC₅₀ (0.3 mg/L) than that of Lab1. The range for resistance ratios among the field collected populations was 4.4 to 30.4 for LC₅₀ based on Lab1 (Table 3). These values increased by up to 10 times when based on Lab2. Also, LC₉₀ values of all field collected whitefly populations were more than two times higher than the recommended field dose (60 µg ai ml⁻¹). The LC₉₀ values of Lab1 and Lab2 populations were only 5 and 0.5% of the recommended field dose for acetamiprid.

Table 2. Toxicity values of *Bemisia tabaci* populations from Antalya for thiamethoxam

Population	n	Slope±SE	LC ₅₀ (µg ai ml ⁻¹) (95% CL)	LC ₉₀ (µg ai ml ⁻¹) (95% CL)	RF ₅₀	RF ₉₀
Ser	261	1.0±0.2	31.4 (10.7-115.0)	812.5 (185.2-66149.2)	8.6	24.6
Aks	182	1.0±0.2	73.8 (27.2-443.9)	1349.6 (275.4-144033.2)	20.3	40.8
Kum	191	1.4±0.3	45.7 (22.0-90.6)	370.1 (163.9-2068.4)	12.5	11.2
Tos	237	2.9±1.0	115.8 (24.7-166.5)	325.9 (238.0-927.1)	31.8	9.9
Kon	293	1.2±0.2	38.3 (12.8-96.9)	457.4 (155.5-12570.7)	10.5	13.8
Lab1	237	1.3±0.1	3.7 (1.5-10.8)	33.1 (11.1-319.2)	1.0	1.0
Lab2	180	2.2±0.3	1.2 (0.8-2.2)	4.7 (2.6-15.4)	0.3	0.1

n: Number of whiteflies tested; SE: Standard Error; LC: Lethal Concentration; CL: Confidence Limits; RF: Resistance Factor calculated as (LC₅₀ of field population) / (LC₅₀ of Lab1 population)

Table 3. Toxicity values of *Bemisia tabaci* populations from Antalya for acetamiprid

Population	n	Slope±SE	LC ₅₀ (µg ai ml ⁻¹) (95%CL)	LC ₉₀ (µg ai ml ⁻¹) (95% CL)	RF ₅₀	RF ₉₀
Ser	276	1.4±0.2	14.2 (3.8-117.1)	118.5 (28.3-32091.5)	4.4	11.6
Aks	224	1.3±0.28	97.5 (41.9-1261.0)	920.5 (211.2-986546.3)	30.4	90.4
Kum	128	0.9±0.2	57.0 (16.1-970.8)	1704.2 (234.3-41302279.2)	17.8	167.3
Tos	122	1.1±0.2	25.2 (9.0-138.4)	400.9 (86.7-18284.0)	7.9	39.4
Kon	149	1.0±0.2	21.2 (7.1-111.1)	362.1 (78.8-24972.2)	6.6	35.6
Lab1	157	2.6±0.4	3.2 (2.1-4.7)	10.2 (6.6-20.5)	1.0	1.0
Lab2	150	3.0±0.4	0.3 (0.2-0.4)	0.8 (0.5-1.4)	0.1	0.1

n: Number of whiteflies tested; SE: Standard Error; LC: Lethal Concentration; CL: Confidence Limits; RF: Resistance Factor calculated as (LC₅₀ of field population) / (LC₅₀ of Lab1 population)

Discussion

Neonicotinoid resistance has been documented in different populations of whitefly all around the world (Byrne et al., 2003; Nauen & Denholm, 2005; Wang et al., 2009; Schuster et al., 2010; Vassiliou et al., 2011; Ünal Bahşi et al., 2012). Furthermore, the resistance in biotype Q is reported in several studies to be higher than in biotype B (Horowitz et al., 2005; Luo et al., 2010; Rao et al., 2012). In the current study, the only biotype found was biotype B. However, previous studies have found sympatric presence of biotypes B and Q, and the sole presence of biotype Q in several areas of Antalya Province (Ikten et al., 2007; unpublished data). There are several studies reporting biotype B replacement by biotype Q under neonicotinoid pressure (Simon et al., 1999; Pan et al., 2010; Sun et al., 2013). However, the neonicotinoid resistant populations from the field and lack of biotype Q findings in current study contradict these reports as the region sampled was known to have biotype Q extensively (Ikten et al., 2007). The discrepancy may indicate that either a factor other than neonicotinoid pressure has decisive role in whitefly fitness or biotype B has a genetic pool as wide as that of biotype Q in the region sampled. However, due to the presence of both acetamiprid and thiamethoxam resistance in all whitefly populations collected in this study, it seems biotype B has as wide genetic plasticity than biotype Q to overcome neonicotinoid exposure.

For all populations, except Tos, a high correlation ($r^2=0.92$) was found between LC_{50} for acetamiprid and thiamethoxam indicating cross resistance develops in the field for neonicotinoids. Cross resistance among neonicotinoids in whitefly was previously shown in different regions of the world (Rauch & Nauen, 2003; Nauen et al., 2002; Wang et al., 2009; Schuster et al., 2010; Yuan et al., 2012). However, lack of neonicotinoid cross resistance was also indicated in some studies (Horowitz et al., 2004; Prabhaker et al., 2005; Vassiliou et al., 2011). Hence, it seems there may be more than one mechanism for neonicotinoid resistance and *B. tabaci* populations may gain neonicotinoid resistance through different genetic combinations.

The results of the neonicotinoid bioassays indicate moderate to high resistance levels in field collected *B. tabaci* populations. Compared to current study, lower LC_{50} values were reported for acetamiprid, whereas thiamethoxam values were higher in *B. tabaci* populations in Israel (Horowitz et al., 2004). Similarly, thiamethoxam toxicity was higher, and acetamiprid resistance was comparable for *B. tabaci* populations collected in Cyprus (Vassiliou et al., 2011). However, lower acetamiprid and thiamethoxam toxicity were reported for populations from West Africa (Houndete et al., 2010). With the exception of one population, lower acetamiprid toxicities were reported for populations collected in 2009-2010 from similar localities to those sampled in the current study (Ünal Bahşi et al., 2012). The difference may be an indication of subsequent resistance development in the region.

Resistance ratios were up to 30 times for both neonicotinoids based on Lab1, and 300 times higher based on Lab2. Furthermore, LC_{50} values of the populations collected from field showed overlapping confidence limits (95% CL) for both neonicotinoids. These two findings indicate homogenous resistant development over the region sampled. Similarly, uniform responses for neonicotinoid resistance were reported for different *B. tabaci* populations from Cyprus (Vassiliou et al., 2011). However, a heterogeneous response to acetamiprid was indicated by populations collected in Antalya between 2007 and 2009 (Ünal Bahşi et al., 2012). This indicates that neonicotinoid use on *B. tabaci* populations between 2007 and 2011 exerts contiguous pressure, hence resistance appears all over the region. Furthermore, LC_{90} values higher than recommended field doses for both neonicotinoids warrant careful consideration of other management options.

Acknowledgments

We are grateful to Akdeniz University Scientific Research Projects Coordination Unit (project number 2012.02.0121.026) for their financial aid.

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Original article (Orijinal araştırma)

Differences in succession of Coleoptera species attracted to pig carcasses in rural and urban habitats in Eskişehir Province, Turkey¹

Eskişehir ilinde kırsal ve kentsel habitatlardaki domuz leşlerine çekilen Coleoptera türlerinin süksesyon farklılıkları

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Summary

The aim of this study was to determine Coleoptera succession in pig carcasses in Eskişehir Province, Turkey, and to evaluate differences in successional patterns between rural and urban habitats. A total of 24 pig carcasses were placed in rural and urban areas in Eskişehir Province for one-year period between June 2012 and May 2013. A total of 80 species belonging to the families Staphylinidae, Histeridae, Dermestidae, Silphidae and Cleridae (Coleoptera) were collected either directly from carcasses or sifted for the specimens hiding in the soil. The months and duration of all species on the different stages of carcass decomposition were recorded. In addition, new records were added to the Coleoptera fauna of carcass for Turkey. The study revealed that, both species number and activity periods were different even in areas very close to each other. It is suggested that this type of long-term succession study should be performed across all provinces of Turkey. Acquired data could potentially be used for estimating the minimum post-mortem interval in forensic cases in Turkey.

Keywords: Coleoptera succession, forensic entomology, pig, rural, Turkey, urban

Özet

Bu çalışmanın amacı, Eskişehir ilinde domuz leşi üzerindeki Coleoptera süksesyonunun belirlenmesi ve kırsal ve kentsel habitatlardaki süksesyonel düzen farklılıklarının değerlendirilmesidir. Eskişehir ilinde bulunan bir kırsal bir de kentsel alana toplam 24 domuz leşi, Haziran 2012 ve Mayıs 2013 arasında bir yıllık süre boyunca yerleştirilmiştir. Coleoptera takımı içerisinde Staphylinidae, Histeridae, Dermestidae, Silphidae ve Cleridae familyalarına ait toplam 80 tür, ya direkt olarak leş üzerinden ya da toprakta bulunan türler için elemeye toplanmıştır. Tüm türlerin ayları ve farklı çürüme evrelerinde buldukları süreler belirlenmiştir. Buna ek olarak Türkiye'nin leş Coleoptera faunasına yeni kayıtlar eklenmiştir. Yapılan çalışmada, çok yakın alanlarda bile hem tür sayısının hem de aktivite periyodlarının belirgin şekilde farklılık gösterdiği açığa çıkmaktadır. Bu şekildeki uzun-süreli süksesyon çalışmalarının Türkiye'nin tüm illerinde yapılması gerekliliği ortaya konulmuştur. Elde edilen veriler Türkiye'de adli olaylarda minimum ölüm zaman aralığı tahmininde potansiyel olarak kullanılabilir.

Anahtar sözcükler: Coleoptera süksesyonu, adli entomoloji, domuz, kırsal, Türkiye, kentsel

¹ This study is supported with 1204F072 numbered project by Anadolu University Scientific Researches Unit. Some parts of the manuscript were previously presented as a poster in 22nd National Biology Congress (23-27 June 2014) in Eskişehir, Turkey.

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Received (Alınış): 15.12.2016 Accepted (Kabul ediliş): 10.05.2017 Published Online (Çevrimiçi Yayın Tarihi): 19.06.2017

Introduction

Decomposition is a natural and necessary process responsible for the cycling organic material, such as dead plant or animal matter, through the ecosystem. Carrion represents a temporary and changing food source for a varied and distinct community of organisms (Putman, 1983). Insects are generally the first organisms which come to carcasses and they colonize in a predictable sequence. This sequence depends on nutritional changes in the carcass and variables like geographical region, habitat, season, and climatological and microclimatic conditions, but this sequence is predictable within these parameters (Anderson, 2001). In forensic entomology, Coleoptera seems to be neglected because Diptera locate corpses faster, thus they are more useful in minimum post-mortem interval (PMI_{min}) estimation (Midgley et al., 2010). However, some Coleoptera species can locate a carcass within 24 h of death which could potentially be employed in PMI_{min} estimation (Midgley & Villet, 2009). Moreover, because they cover most of the succession period, Coleoptera could be used in making more accurate PMI_{min} estimations as they colonize in a predictable sequence. In addition, since they exist longer on corpses, their larvae could be used in toxicological analysis for certain drugs (Midgley et al., 2010).

There are many studies regarding carcass fauna in the world. For example, Reed (1958) studied dog carcasses, Payne (1965), Anderson & Van Laerhoven (1996), Wolff et al. (2001), Grassberger & Frank (2004), Carvalho et al. (2004), Matuszewski et al. (2008, 2010a, b, 2011), Bonacci et al. (2010), Anton et al. (2011) and Prado e Castro et al. (2013) studied pig carcasses, Tantawi et al. (1996) studied rabbit carrion and Kočárek (2003) studied rat carrion. In Turkey, succession is rather a new field of study. There are only a few studies which cover the concept in a broad way. Özdemir & Sert (2009) published their study concerning the determination of Coleoptera species attracted to pig carcasses and their succession in Ankara and they detected 40 species. This was the first study which observed the succession of 12 pig carcasses over a one-year period. After this, Sert et al. (2012) published a study of dog carcass also conducted in Ankara. They detected 14 Coleoptera species. In the same year in Edirne, Bana & Beyarslan (2012) studied three pig carcasses and two placements of bovine offal and they also found 14 Coleoptera species.

The aim of the study was to determine the Coleoptera attracted to pig carcass in Eskişehir Province, Turkey, to evaluate successional differences between rural and urban habitats, and to compare the results with previous studies.

Material and Methods

This study was conducted between 21 June 2012 and 31 May 2013 in two different areas of Eskişehir Province. Temperature and precipitation data was recorded daily with Oregon Weather Station WMR89A data logger. The first part of the study was conducted in the Japanese Garden (39°46'10.39" N, 30°28'25.06" E, 796 m.), Yunus Emre Campus, Anadolu University, Eskişehir located 3 km from the city center and represents an urban area because of the constant human activity within 20 m. The flora of Yunus Emre Campus consists of 363 species belonging to 241 genera and 74 families. While most of them are angiosperms, there are 29 gymnosperm species. There are five major angiosperm families especially in the Japanese Garden; Asteraceae (Compositae) 36 taxa (%9,9), Fabaceae (Leguminosae) 29 taxa (%7,9), Poaceae (Graminae) 17 taxa (%4,6), Brassicaceae (Curciferaceae) 18 taxa (%4,9) and Lamiaceae (Labiatae) 22 taxa (%6). Gymnosperms in the Japanese Garden are mainly *Abies*, *Cedrus*, *Pinus* and *Picea* (Pinaceae); *Taxus baccata* L. (Taxaceae); *Biota*, *Chamaecyparis*, *Cupressocyparis*, *Cupressus*, *Juniperus* and *Thuja* (Cupressaceae). The second location (39°56'07.12" N, 30°29'34.11" E, 1143 m) was in a highland rural area surrounded by *Quercus* forests with almost no human activity nearby. It was located within the Central District of Eskişehir Province with the nearest village, Tekeçiler, located approximately 12 km from the study area.

Field studies

The 24 pigs (*Sus scrofa* L., 1758) that were left in the field weighed between 60 and 200 kg each. After obtaining permission to study with pig carcasses from the Experimental Animals Ethics Committee of Anadolu University (permit no. 2012/0001), pigs were supplied from the Production and Research Center of Gazi University, Ankara, the Education-Research and Application Farm of Ankara University and Ankara and Antalya Tropical Animal Farms. They were euthanized at the Experimental Animals Research Center of Anadolu University by means of sodium pentobarbital injection. Each month, two pigs were killed and a carcass placed as soon as possible after death at each of the sites, about 50 m from the nearest carcass. They were placed in iron cages (1 x 1.5 x 1 m), so that they could not be eaten by vertebrate scavengers. The carcasses were checked daily over the one-year period. Adult specimens were collected using tweezers and aspirators directly from carcasses. The carcasses were rolled over to sample from their underside then put back in the original position. The soil around and under the carcasses was sifted to collect specimens which were hiding or had crawled into the soil when disturbed. The collected specimens were preserved in ethyl alcohol (70%) and acetic acid solution (10%) in the field.

Climatic data for Eskişehir are given in Figure 1.

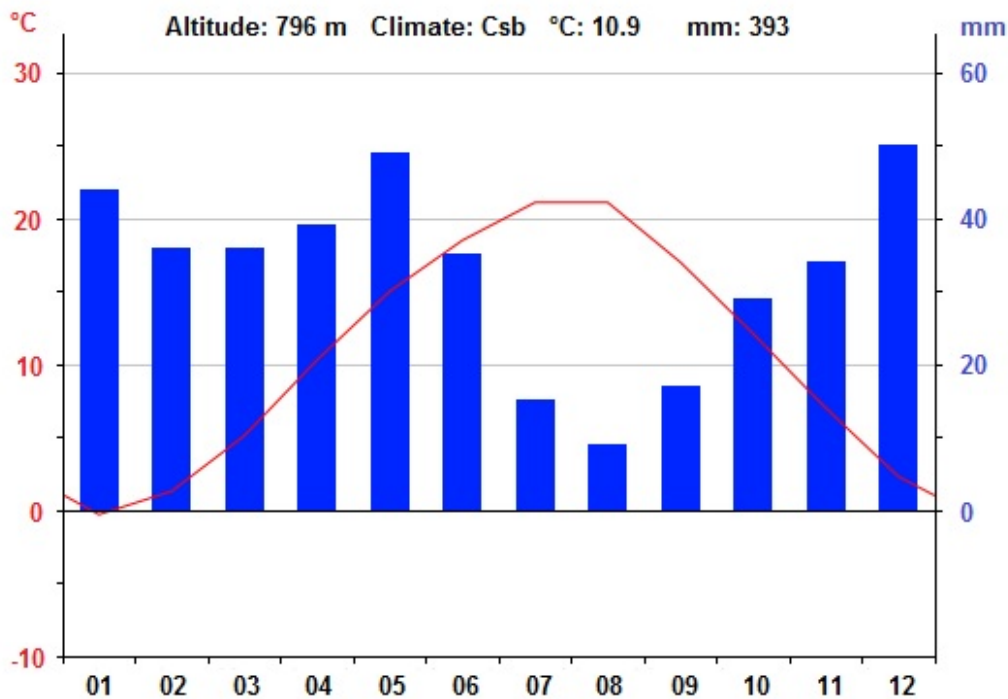


Figure 1. Mean temperature and precipitation in Eskişehir Province, Turkey from 21 June 2012 to 31 May 2013 (recorded with Oregon Weather Station WMR89A data logger).

Laboratory studies

The specimens collected from field were deposited in the collection in the Anadolu University Zoology Museum (AZUM), and then identified using Leica MZ 7.5 and Leica ZOOM 2000 stereomicroscopes. The identification of the specimens was done by authors and in the process identification keys of various researchers were used (Andres, 1925; Pfeffer, 1927; Lesne, 1930; Kalík, 1951; Halstead, 1963; Strand & Vik, 1964, 1968; Osuji, 1975; Kryzhanovskii & Reikhardt, 1976; Miller & Peck, 1979; Zanetti, 1987; Welch, 1997; Eversham, 1999; Ferreira de Almeida & Pires do Prado, 1999; Sikes & Peck, 2000; Secchi, 2002; Assing, 2006a, 2006b, 2007a, b; Lott, 2008; Özdemir & Sert, 2008; Tronquet, 2009; Dekeirsschieter et al., 2011).

Results and Discussion

In this study, 80 species belonging to five coleopteran families (Staphylinidae, Histeridae, Dermestidae, Silphidae, Cleridae) were detected. The highest number of species attracted to carcasses in the study belonged to the family Staphylinidae (39 species). This is followed by the Histeridae (23 species), Silphidae (9 species), Dermestidae (6 species) and Cleridae (3 species). Species distribution in rural and urban habitats over the study period is shown in Figures 2 to 6.

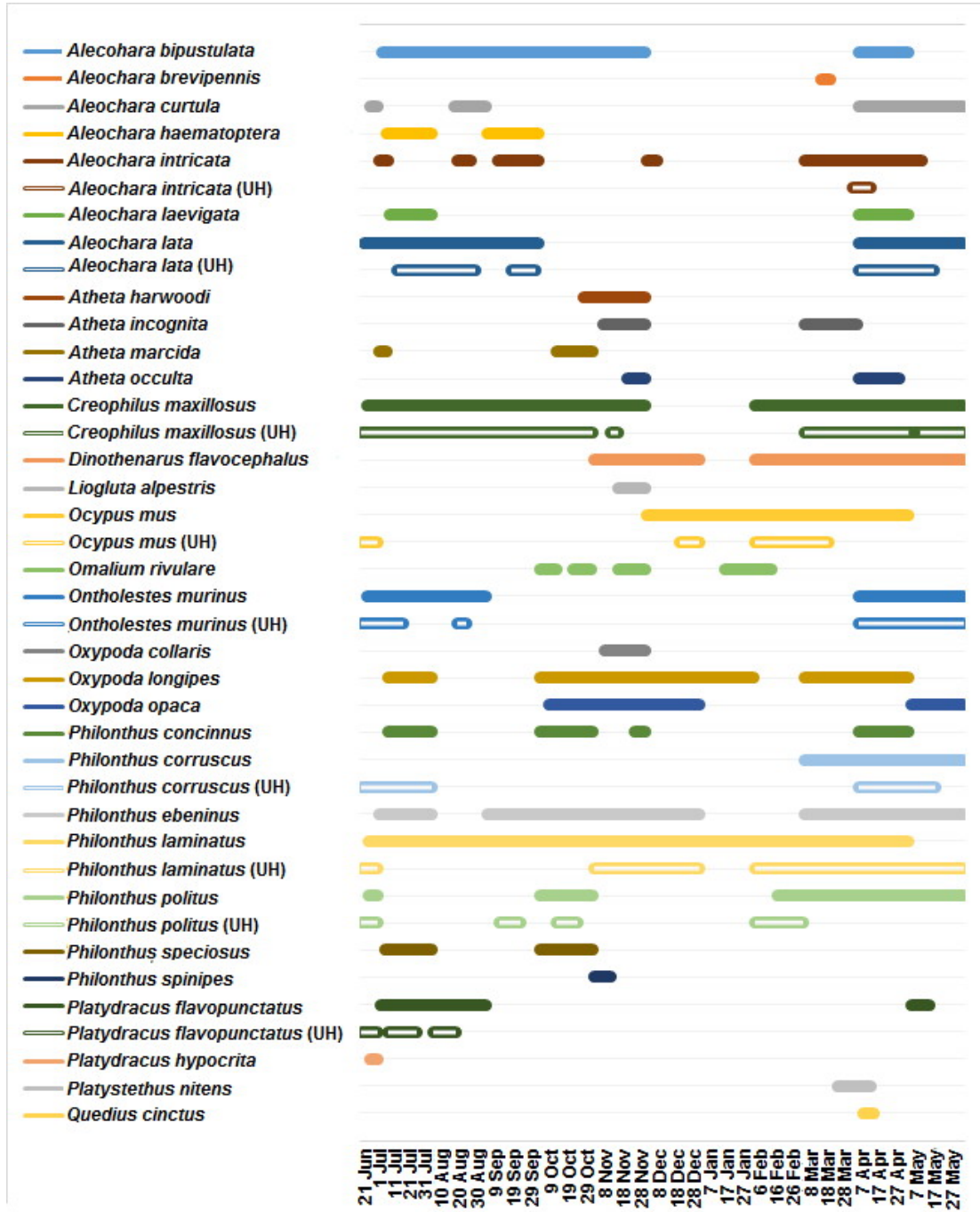


Figure 2. Distribution of Staphylinidae species in both rural and urban habitats in Eskişehir Province, Turkey over a one-year period; the same species are the same colored with solid bars representing the rural habitat, and open bars and the code UH representing the urban habitat.



Figure 3. Distribution of Histeridae species in both rural and urban habitats in Eskişehir Province, Turkey over a one-year period; the same species are the same colored with solid bars representing the rural habitat, and open bars and the code UH representing the urban habitat.

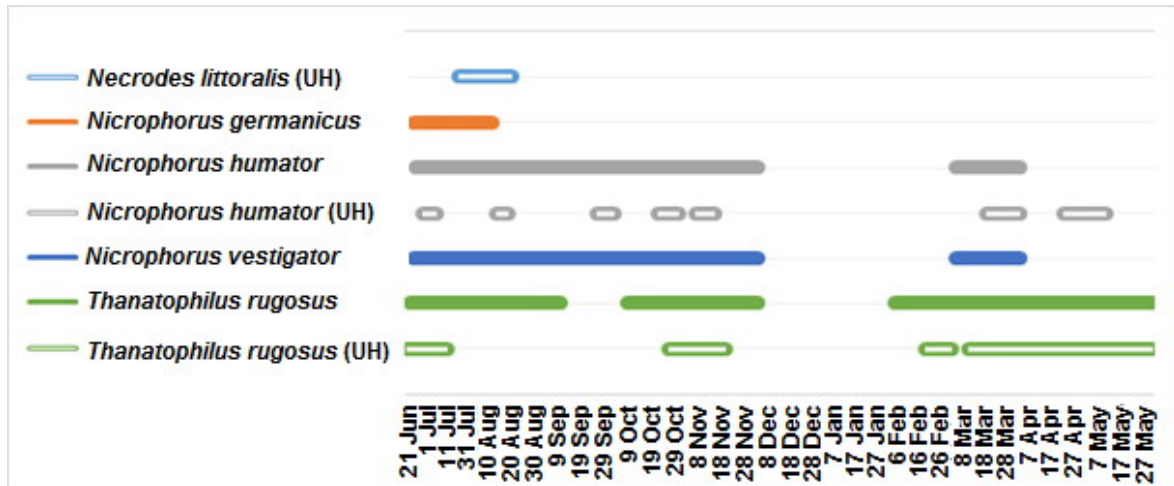


Figure 4. Distribution of Silphidae species in both rural and urban habitats in Eskişehir Province, Turkey over a one-year period; the same species are the same colored with solid bars representing the rural habitat, and open bars and the code UH representing the urban habitat.

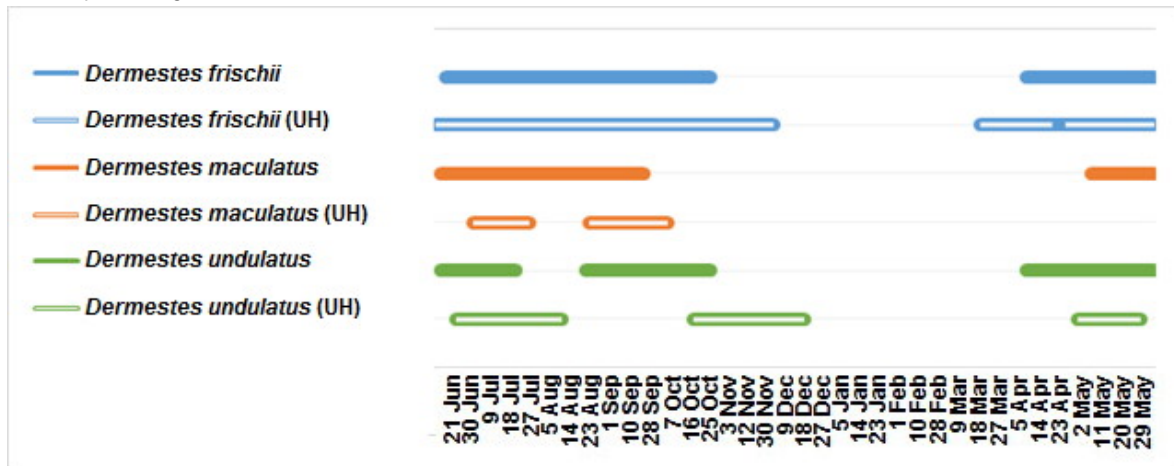


Figure 5. Distribution of Dermestidae species in both rural and urban habitats in Eskişehir Province, Turkey over a one-year period; the same species are the same colored with solid bars representing the rural habitat, and open bars and the code UH representing the urban habitat.

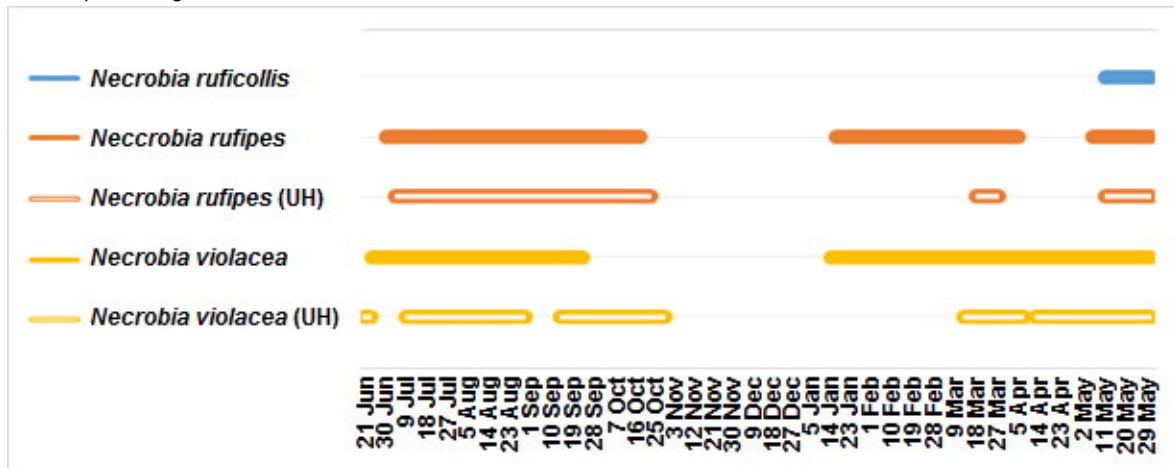


Figure 6. Distribution of Cleridae species in both rural and urban habitats in Eskişehir Province, Turkey over a one-year period; the same species are the same colored with solid bars representing the rural habitat, and open bars and the code UH representing the urban habitat.

Species according to family and their presence in rural and urban habitats are given in Table 1. Eighteen species which were detected only once (only one specimen on one occasion), were not regarded as important to succession. These species are indicated in the table. A total of 51 species were collected from pig carcasses for the first time in Turkey and these are also indicated in Table 1. Regarding habitat differences, except for *Emus hirtus* (L., 1758) and *Necrodes littoralis* (L., 1758), which were collected only from urban habitat, all other species were collected from rural habitat. Except for *Ocypus sericeicollis* (Ménétriés, 1832) and *Margarinotus merdarius* (Hoffmann, 1803) which were collected once during the study, 10 staphylinids, 12 histerids, 2 silphids, 3 dermestids and 2 clerids species were collected in both habitats.

Table 1. Species collected from pig carcasses according to family and their presence in rural and urban habitats in Eskişehir Province (The asterisk indicates species collected as a single specimen and only so are regarded as unimportant. Species which were collected from pig carcasses for the first time in Turkey are underlined)

Family	Species	Rural	Urban	Species	Rural	Urban
Staphylinidae	<i>Aleochara bipustulata</i> (L., 1761)	+	-	* <i>Ocypus sericeicollis</i> (Ménétriés, 1832)	+	+
	<i>Aleochara brevipennis</i> Gravenhorst, 1806	+	-	<i>Omalium rivulare</i> (Paykull, 1789)	+	-
	<i>Aleochara curtula</i> (Goeze, 1777)	+	-	<i>Ontholestes murinus</i> (L., 1758)	+	+
	<i>Aleochara haematoptera</i> Kraatz, 1856	+	-	<i>Oxypoda collaris</i> Saulcy, 1865	+	-
	<i>Aleochara intricata</i> Mannerheim, 1830	+	+	* <i>Oxypoda cristata</i> Assing, 2006	+	-
	<i>Aleochara laevigata</i> Gyllenhal, 1810	+	-	<i>Oxypoda longipes</i> Mulsant & Rey, 1861	+	-
	<i>Aleochara lata</i> Gravenhorst, 1802	+	+	<i>Oxypoda opaca</i> (Gravenhorst, 1802)	+	-
	* <i>Aleochara maculata</i> Brisout de Barneville, 1863	+	-	<i>Philonthus concinnus</i> (Gravenhorst, 1802)	+	-
	* <i>Aleochara spadicea</i> (Erichson, 1837)	+	-	<i>Philonthus corruscus</i> (Gravenhorst, 1802)	+	+
	<i>Atheta harwoodi</i> D.S. Williams, 1930	+	-	<i>Philonthus ebeninus</i> (Gravenhorst, 1802)	+	-
	<i>Atheta incognita</i> (Sharp, 1869)	+	-	<i>Philonthus laminatus</i> (Creutzer, 1799)	+	+
	<i>Atheta marcida</i> (Erichson, 1837)	+	-	<i>Philonthus politus</i> (L., 1758)	+	+
	<i>Atheta occulta</i> (Erichson, 1837)	+	-	<i>Philonthus speciosus</i> Cameron, 1926	+	-
	<i>Creophilus maxillosus</i> (L., 1758)	+	+	<i>Philonthus spinipes</i> Sharp, 1874	+	-
<i>Dinothenarus flavocephalus</i> (Goeze, 1777)	+	-	* <i>Philonthus varians</i> (Paykull, 1789)	+	-	
Staphylinidae	* <i>Emus hirtus</i> (L., 1758)	-	+	<i>Platydracus flavopunctatus</i> (Latreille 1804)	+	+
	* <i>Heterothops dissimilis</i> (Gravenhorst, 1802)	+	-	<i>Platydracus hypocrita</i> (Müller, 1925)	+	-
	* <i>Hypnogyra angularis</i> (Ganglbauer, 1895)	+	-	<i>Platystethus nitens</i> (C.R. Sahlberg, 1932)	+	-
	<i>Liogluta alpestris</i> (Heer, 1839)	+	-	<i>Quedius cinctus</i> (Paykull, 1790)	+	-
	<i>Ocypus mus</i> (Brullé, 1832)	+	+			

Table 1. (Continued)

Family	Species	Rural	Urban	Species	Rural	Urban
Histeridae	<i>Atholus duodecimstriatus</i> (Schrank, 1781)	+	-	<i>Saprinus externus</i> (Fischer de Waldheim, 1823)	+	-
	<i>Carcinops pumilio</i> (Erichson, 1834)	+	-	<i>Saprinus georgicus</i> Marseul, 1862	+	+
	* <i>Gnathoncus nannetensis</i> (Marseul, 1862)	+	-	<i>Saprinus godet</i> Brullé, 1832	+	-
	* <i>Gnathoncus rotundatus</i> (Kugelann, 1792)	+	-	<i>Saprinus maculatus</i> (Rossi, 1792)	+	+
	<i>Hister illigeri illigeri</i> Duftschmid, 1805	+	+	<i>Saprinus planiusculus</i> Motschulsky, 1849	+	+
	<i>Hister quadrinotatus</i> Scriba, 1790	+	-	<i>Saprinus prasinus</i> Erichson, 1834	+	+
	<i>Margarinotus carbonarius</i> (Hoffmann, 1803)	+	-	<i>Saprinus semistriatus</i> (Scriba, 1790)	+	+
	<i>Margarinotus brunneus</i> (Fabricius, 1775)	+	+	<i>Saprinus steppensis</i> Marseul, 1862	+	+
	* <i>Margarinotus merdarius</i> (Hoffmann, 1803)	+	+	<i>Saprinus subnitescens</i> Bickhardt, 1909	+	+
	<i>Margarinotus ruficornis</i> (Grimm, 1852)	+	-	<i>Saprinus tenuistrius</i> Solsky, 1876	+	+
<i>Saprinus caeruleus</i> (Hoffmann, 1803)	+	+	<i>Saprinus vermiculatus</i> Reichardt, 1923	+	+	
<i>Saprinus calatravensis</i> Fuente, 1899	+	-				
Silphidae	<i>Nicrodes littoralis</i> (L., 1758)	-	+	<i>Nicrophorus vestigator</i> Herschel, 1807	+	-
	* <i>Nicrophorus antennatus</i> Reitter, 1885	+	-	* <i>Thanatophilus ferrugatus</i> (Solsky, 1874)	+	-
	<i>Nicrophorus germanicus</i> (L., 1758)	+	-	<i>Thanatophilus rugosus</i> (L., 1758)	+	+
	<i>Nicrophorus humator</i> (Gleditsch, 1767)	+	+	* <i>Thanatophilus sinuatus</i> Fabricius, 1775	+	-
	* <i>Nicrophorus investigator</i> Zetterstedt, 1824	+	-			
Dermestidae	* <i>Dermestes dimidiatus</i> Boeber, 1802	+	-	* <i>Dermestes leopardinus</i> Mulsant & Godart, 1855	+	-
	<i>Dermestes frischi</i> Kugelann, 1792	+	+	<i>Dermestes maculatus</i> DeGeer, 1774	+	+
	* <i>Dermestes lardarius</i> L., 1758	+	-	<i>Dermestes undulatus</i> Brahm, 1790	+	+
Cleridae	<i>Necrobia ruficollis</i> (Fabricius, 1775)	+	-	<i>Necrobia violacea</i> (L., 1758)	+	+
	<i>Necrobia rufipes</i> (De Geer, 1775)	+	+			

The coleopteran species collected are compared with previous studies in Table 2. In the present study 78 species were collected in rural habitat. Among the researchers who performed their studies in rural, forest and/or semi-rural habitats, Anderson & Van Laerhoven (1996) collected 13 species, Kočárek (2003) collected 60 species, Watson & Carlton (2005) collected 34 species, Matuszewski et al. (2008) collected 72 species, Özdemir & Sert (2009) collected 37 species, Matuszewski et al. (2010b) collected 19 species, Bonacci et al. (2010) collected 12 species, Anton et al. (2011) collected 35 species, and Prado e Castro et al. (2013) collected 55 species belonging to coleopteran families collected in the present study. In contrast, while 32 species were collected in urban habitat in this study, Tantawi et al.

(1996) collected 18 species, Wolff et al. (2001) collected 11 species and Grassberger & Frank (2004) collected 10 species from urban habitat. Therefore, greater number of species have been collected from rural, forest and/or semi-rural habitats than from urban habitats when previous research is included (Table 2), and species are found for longer periods on and around carcass in rural habitats (Figures 2-6).

Table 2. Comparison of the number of identified species and total number of Coleoptera collected in the present study and previous studies

References	Location	Carcass Type	Habitat	Staphylinidae	Histeridae	Silphidae	Dermestidae	Cleridae	Other Coleoptera	TOTAL
Anderson & Van Laerhoven (1996)	British Columbia, Canada	Pig	Rural	6	1	2	1	3	9	22
Tantawi et al. (1996)	Alexandria, Egypt	Rabbit	Urban	9	5	-	2	2	8	26
Wolff et al. (2001)	Medellin, Colombia	Pig	Urban	7	1	1	1	1	4	15
Kočárek (2003)	Opava, Czech Republic	Rat	Rural (Forest)	44	4	9	2	1	85	145
Grassberger & Frank (2004)	Vienna, Austria	Pig	Urban	2	1	3	2	2	-	10
Watson & Carlton (2005)	Baton Rouge, Louisiana, USA	Bear, deer, alligator, pig	Rural (Forest)	21	6	4	1	2	26	60
Matuszewski et al. (2008)	Western Poland	Pig	Rural (Forest)	50	10	7	2	3	20	92
Özdemir & Sert (2009)	Ankara, Turkey	Pig	Rural	16	11	4	4	2	3	40
Matuszewski et al. (2010b)	Western Poland	Pig	Rural (Forest)	1	7	7	2	2	9	28
Bonacci et al. (2010)	Rende, Italy	Pig	Semi-Rural	5	1	4	1	1	3	15
Anton et al. (2011)	Thuringia, Jena, Germany	Pig	Rural	18	2	8	4	3	16	51
Prado e Castro et al. (2013)	Coimbra, Portugal	Pig	Rural (Forest)	35	10	3	4	3	25	80
Present Study	Eskişehir, Turkey	Pig	Rural/Urban	39	23	9	6	3	-	80

Özdemir & Sert (2009) conducted their study in Ankara Province, which is adjacent to Eskişehir Province. Despite these provinces being close to each other, both species number and activity periods were substantially different. To explain, Beytepe Campus, Ankara (Özdemir & Sert, 2009) can be considered as an urban habitat similar to Yunus Emre Campus, Eskişehir. Nevertheless, 10 different species collected in Ankara were not collected in Eskişehir, and 11 species collected in Eskişehir were not collected in Ankara.

In the present study, *Aleochara maculata* Brisout de Barneville, 1863, *Aleochara spadicea* (Erichson, 1837), *E. hirtus*, *Heterothops dissimilis* (Gravenhorst, 1802), *Hypnogyra angularis* (Ganglbauer, 1895), *Ocypus sericeicollis*, *Oxypoda cristata* Assing, 2006 and *Philonthus varians* (Paykull, 1789) from Staphylinidae, *Gnathoncus nannetensis* (Marseul, 1862), *Gnathoncus rotundatus* (Kugelann,

1792) and *Margarinotus merdarius* (Hoffmann, 1803) from Histeridae, *Thanatophilus ferrugatus* (Solsky, 1874), *Thanatophilus sinuatus* Fabricius, 1775, *Nicrophorus antennatus* Reitter, 1885 and *N. investigator* Zetterstedt, 1824 from Silphidae, and *Dermestes dimidiatus* Boeber, 1802, *Dermestes lardarius* L., 1758 and *Dermestes leopardinus* Mulsant & Godart, 1855 from Dermestidae were collected only once during the study period (Table 1). *Philonthus varians* and genera such as *Thanatophilus*, *Nicrophorus* and *Dermestes* are known to be associated with particular stages of decomposition (Reed, 1958; Anderson & Van Laerhoven, 1996; Kočárek, 2003; Grassberger & Frank, 2004; Matuszewski et al., 2008, 2010b), but it is possible that because of the dominance of their congeners they were found only once. Other staphylinids are commonly found in dung, decaying organic material, and mammalian and ant nests. From the Histeridae, while *G. nannetensis* and *G. rotundatus* are known to be associated with carcasses, *M. merdarius* is normally found in forest litter, or in the moist substrate of hollow trunks, and it is less likely to be collected from carrion (Lackner & Mazur, 2015), as is in present study.

Aleochara brevipennis Gravenhorst, 1806, *Liogluta alpestris* (Heer, 1839) and *Oxypoda collaris* Saulcy, 1865 from the subfamily Aleocharinae (Staphylinidae) are often observed in forest habitats (Kočárek, 2003) and they were observed in the present study for only a relatively short period and only in the rural habitat. In the current study *Platystethus nitens* (C. R. Sahlberg, 1932) was observed on a carcass for a short time period only; it was reported by Lü & Zhou (2015) that *Platystethus* (Oxytelinae) species are most commonly found in dung and other decaying plant materials. *Philonthus spinipes* Sharp, 1874, *Platydracus hypocrita* (Müller, 1925) and *Quedius cinctus* (Paykull, 1790) from the subfamily Staphylininae can be found on decaying organic material and dung in rural habitats because they are predators on other insects, especially fly larvae. *Platydracus hypocrita* and *Quedius cinctus* were also collected from carcasses in previous studies (Özdemir & Sert, 2009; Fernández et al., 2010; Dekeirsschieter et al., 2013; Mađra et al., 2014). The fact that these species were collected only once in the present study suggests that they are relatively uncommon or carrion is not their primary habitat. *Hister quadrinotatus* Scriba, 1790, *Margarinotus carbonarius* (Hoffmann, 1803), *Saprinus calatravensis* Fuente, 1899, *Saprinus externus* (Fischer de Waldheim, 1823), *Saprinus godet* Brullé, 1832, *Nicrophorus germanicus* (L., 1758) and *Necrobia ruficollis* (Fabricius, 1775) are also known to be associated with carrion at various stages, but they were probably collected in low numbers because congeners were common. *Necrodes littoralis* from the family Silphidae were observed only in the urban area, which was different from the other species detected for a short period also and in the studies of Watson & Carlton (2005) and Dekeirsschieter et al. (2011). It is possible that this species could be found in urban areas if the vegetation is suitable and food is available.

The remaining species, those not mentioned above, were collected from carcass at different times of the year. While some of them were found only in the rural habitat, others were collected from both rural and urban habitats. For example, *Philonthus laminatus* (Creutzer, 1799), *Margarinotus brunneus* (Fabricius, 1775), *Thanatophilus rugosus* (L., 1758) and *Necrobia rufipes* (De Geer, 1775) were observed almost throughout the whole year and could be found in any season without habitat preference. Although, the number of insect species decreased with the reduction of insect activity in winter.

Despite the fact that they were not observed in winter, *Aleochara lata* Gravenhorst, 1802, *Saprinus planiusculus* Motschulsky, 1849, *Saprinus semistriatus* (Scriba, 1790), *Saprinus subnitescens* Bickhardt, 1909, *Nicrophorus humator* (Gleditsch, 1767) and *Dermestes frischi* Kugelann, 1792 were the most commonly observed species on carcass without habitat preference. In addition, *Creophilus maxillosus* (L., 1758), *Saprinus caerulescens* (Hoffmann, 1803), *Necrobia violacea* (L., 1758) and *Dermestes undulatus* Brahm, 1790 were also determined as common species, but while first three were not observed in winter in the urban habitat, *D. undulatus* was not observed in winter in the rural habitat. In contrast, as a

common species, *Philonthus politus* (L., 1758) was observed in winter in both habitats but not observed partly in summer and autumn. *Ocypus mus* (Brullé, 1832) was mostly found in winter and spring in both habitats and for a short period in summer only in the urban habitat. *Aleochara intricata* Mannerheim, 1830 was observed in every season in the rural habitat and only in spring in the urban habitat. *Dermestes maculatus* DeGeer, 1774 was not observed in winter and only in the urban habitat in spring. Among the species which were collected in both habitats, *Ontholestes murinus* (L., 1758), *Philonthus corruscus* (Gravenhorst, 1802), *Saprinus steppensis* Marseul, 1862 and *S. tenuistrius* Solsky, 1876 were observed through spring and summer. Another species, *Platydracus flavopunctatus* (Latreille 1804) was not found in spring in the urban habitat. While *Saprinus georgicus* Marseul, 1862 was collected in summer and autumn, *Saprinus prasinus* Erichson, 1834 and *Saprinus vermiculatus* Reichardt, 1923 were observed only in spring, and *Saprinus maculatus* (Rossi, 1792) and *Hister illigeri illigeri* Duftschmid, 1805 were collected only in summer.

Although the most of the species were distributed in both habitats, species such as *Dinothenarus flavocephalus* (Goeze, 1777), *Philonthus concinnus* (Gravenhorst, 1802), *P. ebeninus* (Gravenhorst, 1802), *P. speciosus* Cameron, 1926, *Aleochara bipustulata* (L., 1761), *A. curtula* (Goeze, 1777), *A. haematoptera* Kraatz, 1856, *A. laevigata* Gyllenhal, 1810, *Atheta harwoodi* D.S. Williams, 1930, *A. incognita* (Sharp, 1869), *A. marcida* (Erichson, 1837), *A. occulta* (Erichson, 1837), *Oxygoda longipes* Mulsant & Rey, 1861, *O. opaca* (Gravenhorst, 1802) and *Omalius rivulare* (Paykull, 1789) from Staphylinidae, *Atholus duodecimstriatus* (Schrank, 1781), *Carcinops pumilio* (Erichson, 1834) and *Margarinotus ruficornis* (Grimm, 1852) from Histeridae, and *Nicrophorus vestigator* Herschel, 1807 from Silphidae were only collected from carcass in the rural habitat. *Dinothenarus flavocephalus*, *O. longipes*, *P. concinnus* and *P. ebeninus* were observed through the whole year, whereas the other species were collected in various combinations of seasons.

In this study, the decomposition process was classified in five stages, namely fresh, bloated, active, advanced and dry, after the classification system of Carvalho et al. (2004). Seasonal changes in the decomposition stages of pig carrion in rural and urban habitats and the Coleoptera species collected are given in Figures 7 to 12. Also, the time of arrival and the period of presence of species are shown in these figures. No Coleoptera species were collected in fresh stage. The decomposition process advanced rapidly in the spring and summer months, with increased temperature and insect activity, thus carcasses in both habitats reached the dry stage (Figures 7a, b; 8a, b) and generally the succession proceeded quickly (Figures 7a, b, 8a, b and 12a, b). The number of species collected decreased with the progress of the seasons through to winter (Figures 9a, b). No Coleoptera were collected from the sixth carcasses in both habitats, even though the decomposition process had continued in winter. Limited insect activity could be detected on the carcasses placed in winter, and activity was mostly delayed until the end of winter (Figures 10a, b; 11a, b). Only one species (*Thanatophilus rugosus*) was attracted to the seventh carcass at dry stage in May after being placed in December in the urban habitat. The remaining carcasses did not reach the dry stage before the end of the experiment.

In conclusion, the Coleoptera fauna of animal carcasses placed outside in Eskişehir Province, Turkey and their successional and seasonal distributions over a one-year period was determined with this study. Overall, 80 species were detected, 51 of those were collected from carcasses for the first time in Turkey. These data could potentially be used for estimating the PMI_{min} in forensic cases in Turkey. The succession of these species should be taken into consideration in forensic cases. In addition, different environmental variables, such as elevation, soil type and vegetation, should also be investigated in further studies. As a result, it is suggested that this type of study should be conducted in rural and urban areas across Turkey.

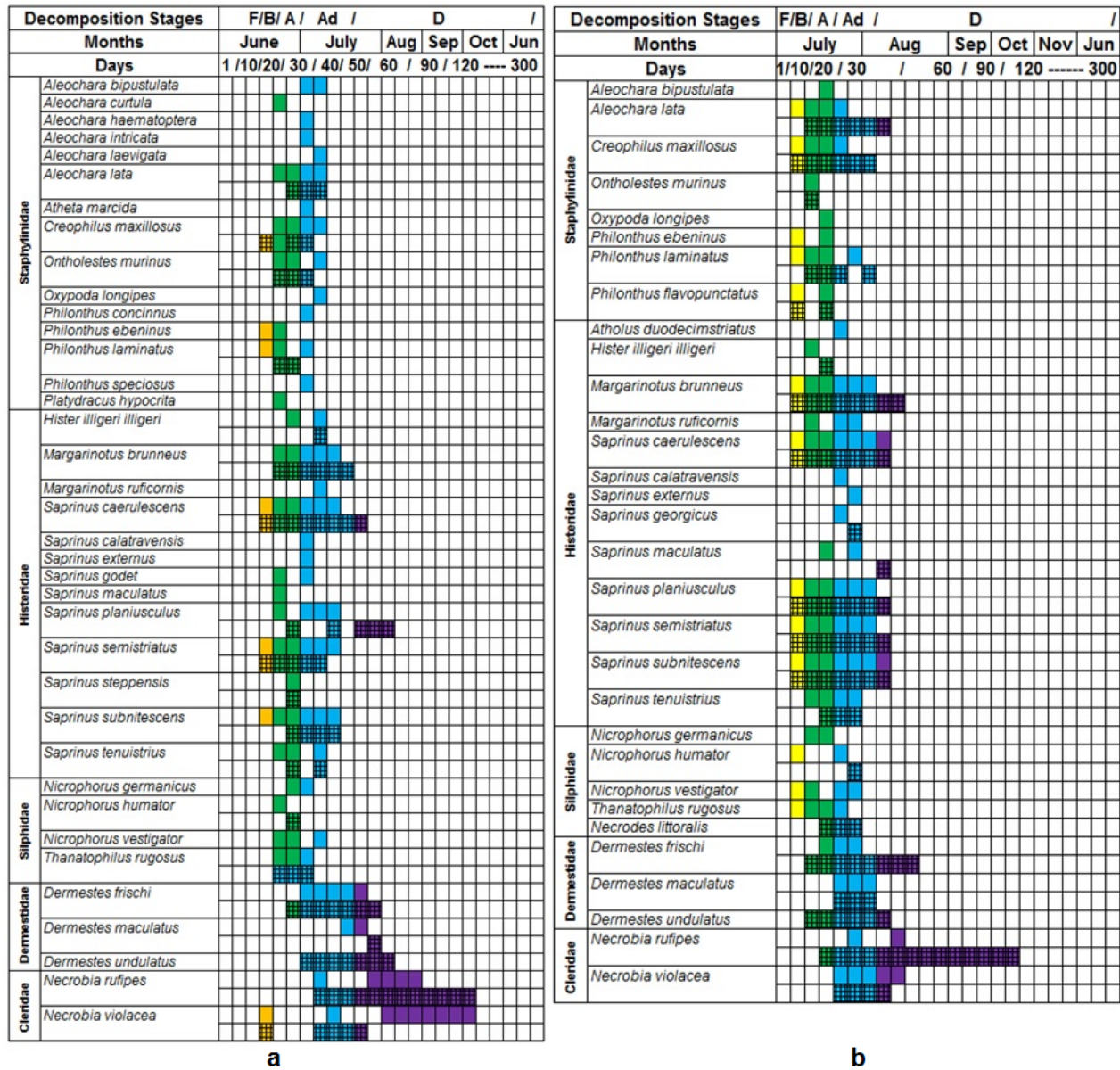


Figure 7. Succession through decomposition stages of carcasses in rural and urban habitats: a) first carcass; b) second carcass (solid, rural; crosshatched, urban). Decomposition stages: F, fresh (not shown in color because none of the insects were attracted); B, bloated (yellow); A, active decay (green); Ad, advanced decay (blue); and D, dry (purple).

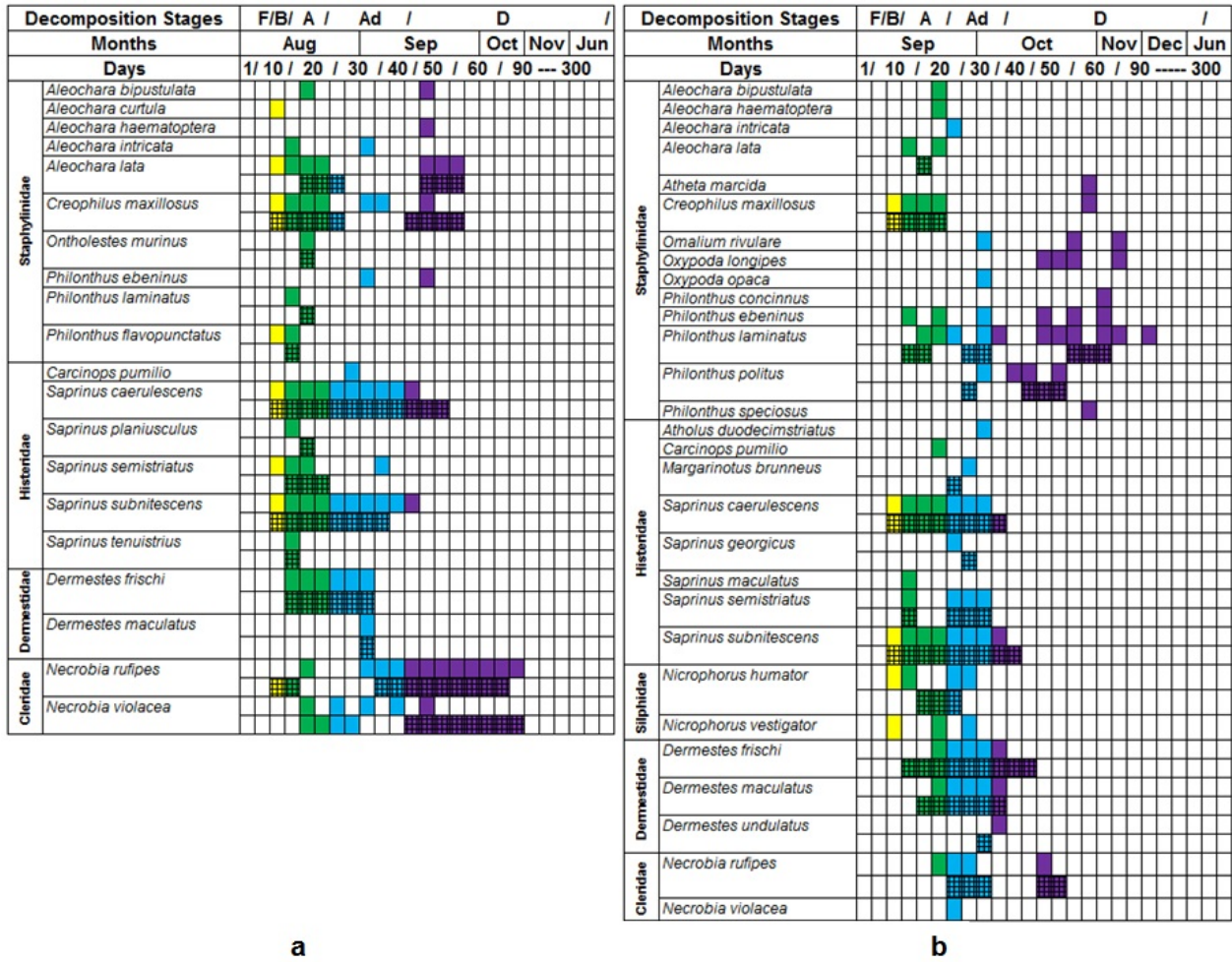


Figure 8. Succession through decomposition stages of carcasses in rural and urban habitats: a) third carcass; b) fourth carcass (solid, rural; crosshatched, urban). Decomposition stages: F, fresh (not shown in color because none of the insects were attracted); B, bloated (yellow); A, active decay (green); Ad, advanced decay (blue); and D, dry (purple).

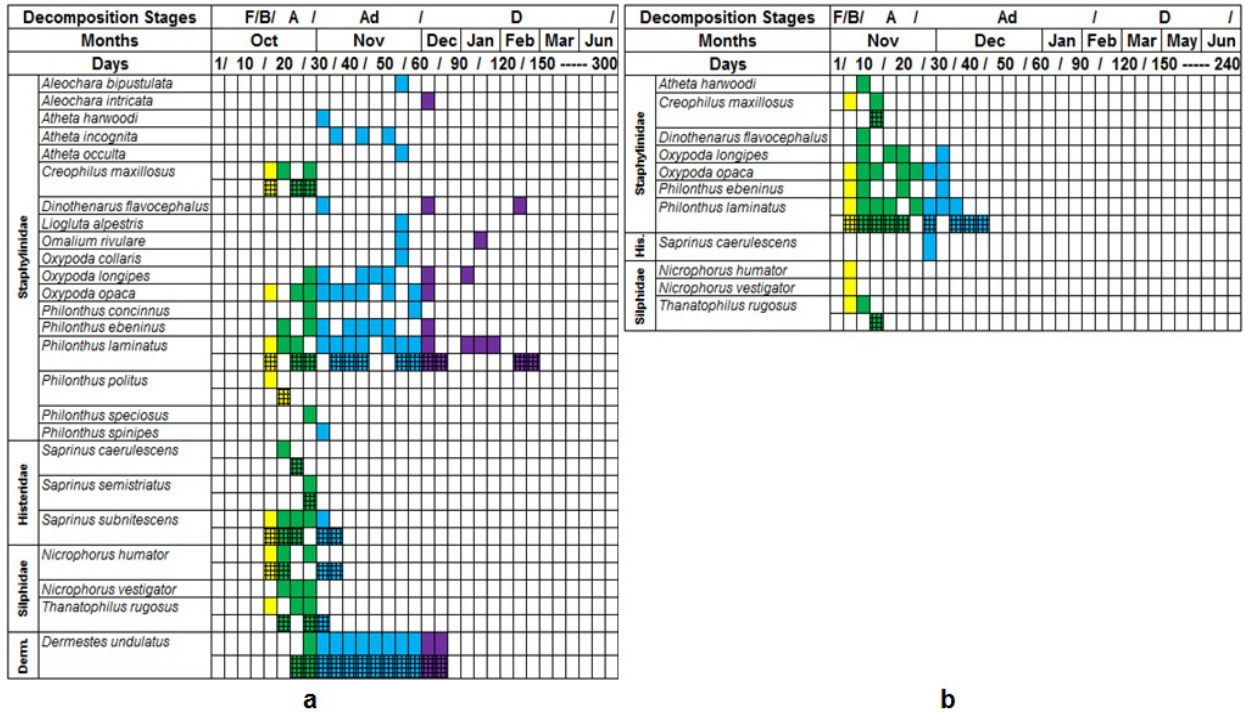


Figure 9. Succession through decomposition stages of carcasses in rural and urban habitats: a) fifth carcass; b) sixth carcass (solid, rural; crosshatched, urban). Decomposition stages: F, fresh (not shown in color because none of the insects were attracted); B, bloated (yellow); A, active decay (green); Ad, advanced decay (blue); and D, dry (purple).

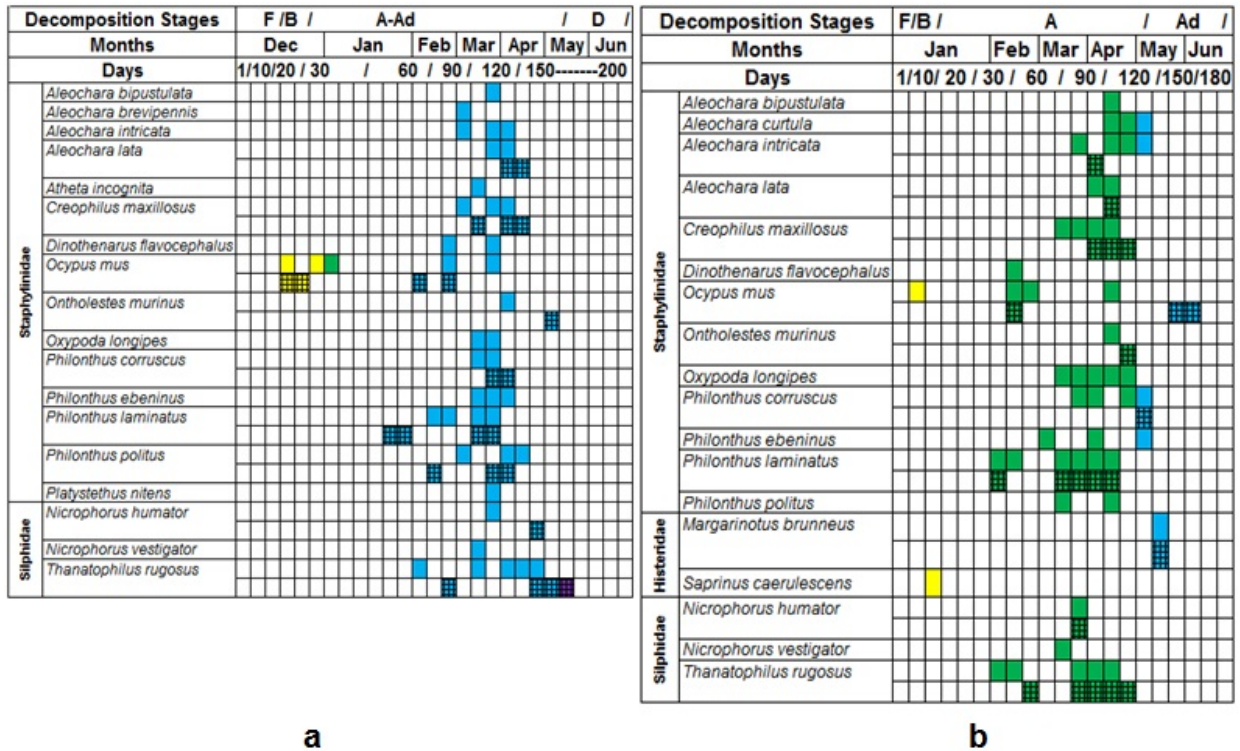


Figure 10. Succession through decomposition stages of carcasses in rural and urban habitats: a) seventh carcass; b) eighth carcass (solid, rural; crosshatched, urban). Decomposition stages: F, fresh (not shown in color because none of the insects were attracted); B, bloated (yellow); A, active decay (green); Ad, advanced decay (blue); and D, dry (purple).

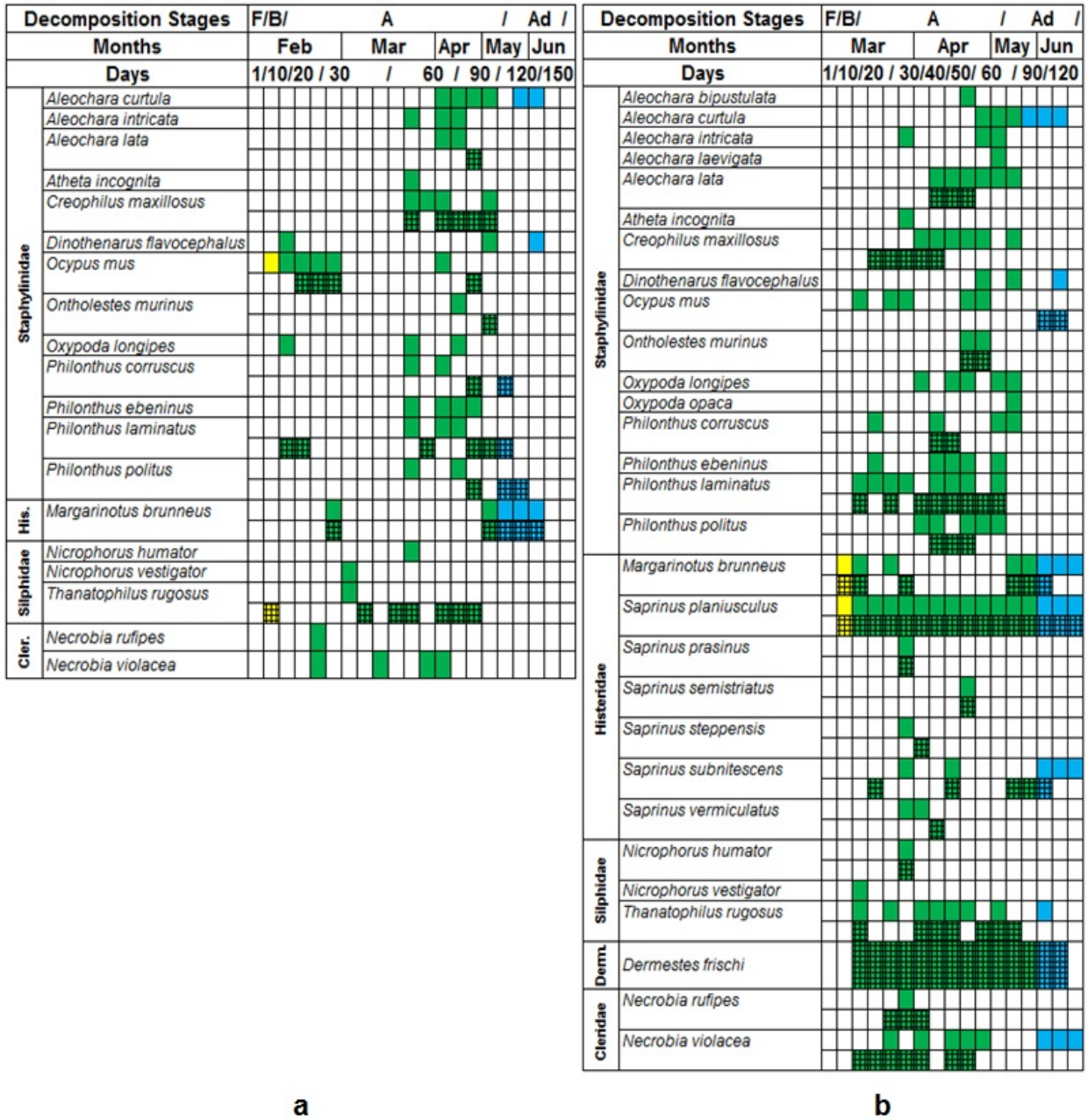


Figure 11. Succession through decomposition stages of carcasses in rural and urban habitats: a) ninth carcass; b) tenth carcass (solid, rural; crosshatched, urban). Decomposition stages: F, fresh (not shown in color because none of the insects were attracted); B, bloated (yellow); A, active decay (green); Ad, advanced decay (blue); and D, dry (purple).

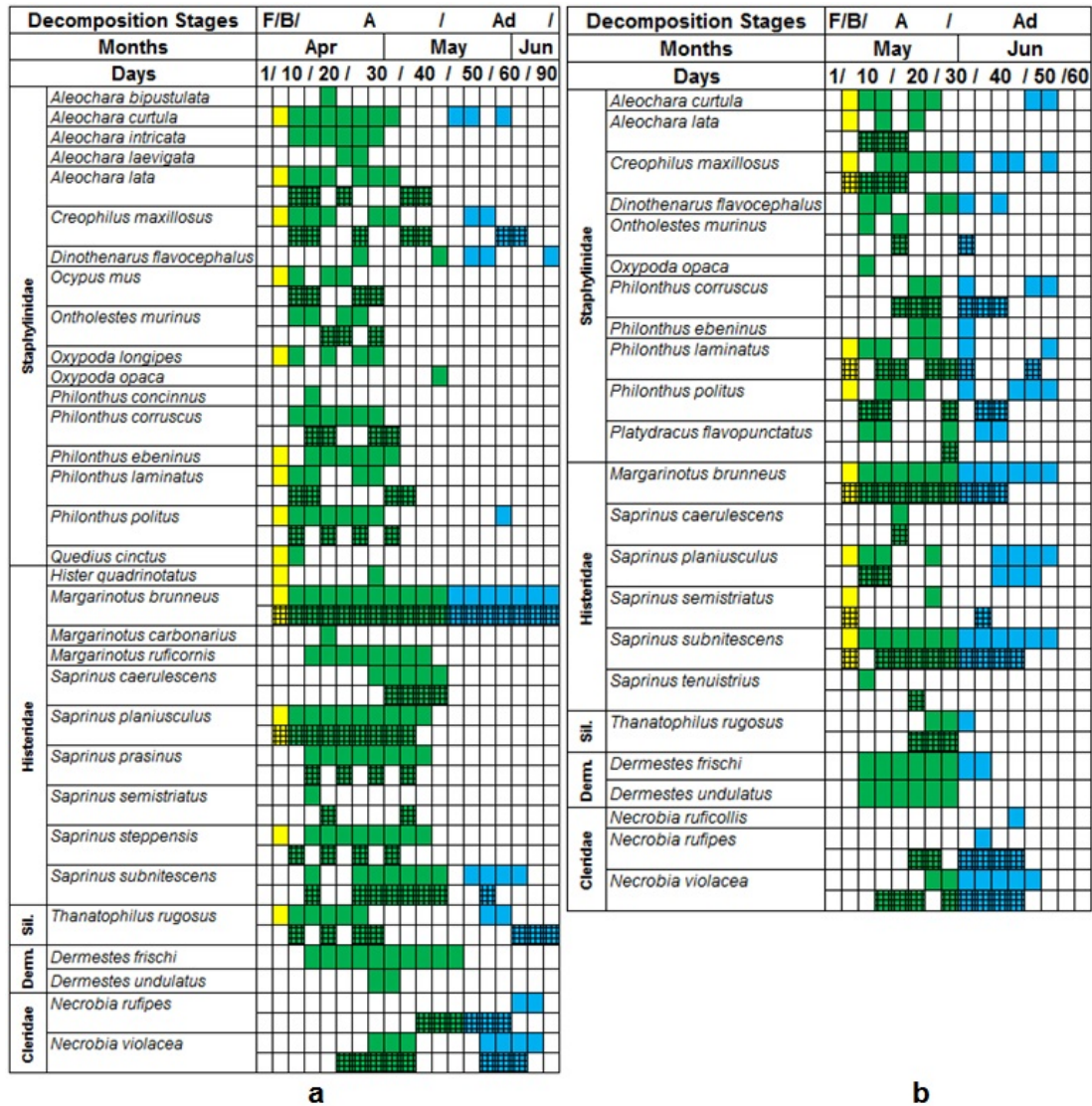


Figure 12. Succession through decomposition stages of carcasses in rural and urban habitats: a) eleventh carcass; b) twelfth carcass (solid, rural; crosshatched, urban). Decomposition stages: F, fresh (not shown in color because none of the insects were attracted); B, bloated (yellow); A, active decay (green); Ad, advanced decay (blue); and D, dry (purple).

Acknowledgments

The pigs used in the study were euthanized by Erdem ERKUŞ, a veterinarian working at the Experimental Animals Research Center, Anadolu University, Eskişehir.

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Original article (Orijinal araştırma)

**Notes on the genus *Rugilus* Leach, 1819 in the Palearctic Region
(Coleoptera: Staphylinidae: Paederinae)¹**

Paleartik Bölge'deki *Rugilus* Leach, 1819 cinsine bağlı türler üzerinde notlar
(Coleoptera: Staphylinidae: Paederinae)

Sinan ANLAŞ^{2*}

Summary

The genus *Rugilus* Leach, 1819 represented by 95 species in the Palearctic Region. New and additional records are provided for 16 species of the genus *Rugilus* from different countries of the Region. The studied material has been collected between 1907-2016 and contained types and additional specimens in the European museums and private collections. Among them eight species are new country records: Azerbaijan (1), Bulgaria (2), Czech Republic (1), Iran (2), Iraq (1) Kazakhstan (1) and Syria (1). Besides, the doubtful species *Rugilus couloni* (Drugmand, 1989) is redescribed and illustrated. A distributional checklist is presented for 10 Turkish *Rugilus* species.

Keywords: Coleoptera, fauna, new records, Paederinae, Palearctic Region, *Rugilus*, Staphylinidae

Özet

Rugilus Leach, 1819 cinsi, Paleartik Bölgede 95 türle temsil edilmektedir. Bu bölgedeki farklı ülkelerden *Rugilus* Leach, 1819 cinsine bağlı 16 türe ait yeni ve ek kayıtlar verilmiştir. İncelenen materyal 1907-2016 yılları arasında toplanmış olup, Avrupa müzelerindeki ve kişisel koleksiyonlardaki tip ve diğer örnekleri içermektedir. Bunlardan sekiz tanesi ilk ülke kaydı niteliğindedir: Azerbaycan (1), Bulgaristan (2), Çek Cumhuriyeti (1), İran (2), Irak (1), Kazakistan (1) ve Suriye (1). Ayrıca, şüpheli bir tür olan *Rugilus couloni* (Drugmand, 1989) yeniden tanımlanmış ve şekillendirilmiştir. Ek olarak, Türkiye'deki 10 *Rugilus* türüne ait bir yayılışsal kontrol listesi sunulmuştur.

Anahtar sözcükler: Coleoptera, fauna, yeni kayıtlar, Paederinae, Paleartik Bölge, *Rugilus*, Staphylinidae

¹ This study was in part supported by the Scientific and Technological Research Council of Turkey (TÜBİTAK, projects 112T907 and 215Z080).

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Received (Alınış): 13.12.2016

Accepted (Kabul edilmiş): 16.05.2017

Published Online (Çevrimiçi Yayın Tarihi): 14.06.2017

Introduction

The genus *Rugilus* Leach, 1819 contains more than 200 species (Newton et al., 2001; Assing, 2012). According to the recent catalogue of Schülke & Smetana (2015), and recent revisions by Assing (2012, 2013b), this genus is represented by 95 species in the Palearctic Region. The genus is divided into two subgenera: the nominate subgenus (76 species); *Eurystilicus* Fagel, 1953 (16 species). Moreover, three species are placed in incertae sedis.

In the present study, 16 species are reported from different countries of the Palearctic Region. Eight *Rugilus* species are reported as new country records. In addition, the doubtful species *Rugilus couloni* (Drugmand, 1989) is redescribed and illustrated.

Material and Methods

The descriptions of the primary and secondary sexual characters of the species redescribed here use the terminology of Coiffait (1984) and Assing (2012). The morphological studies were conducted using a Stemi 2000-C microscope (Zeiss, Germany). Photographs were taken with a digital camera (Zeiss Axiocam ERC5s).

Abbreviations used for measurements (in mm) are: AL, length of antenna; AW, maximal width of abdomen; EL, length of elytra from apex of scutellum to posterior margin; EW, combined width of elytra; HL, head length from anterior margin of clypeus to posterior margin of head; HW, head width (including eyes); ML, length of aedeagus from apex of ventral process to base; PL, length of pronotum along median line; PW, maximal width of pronotum; TaL, length of metatarsus; TiL, length of metatibia; and TL, total body length.

The *Rugilus* material referred to in this study is preserved in the collections: AZMM, Alaşehir Zoological Museum, Manisa, Turkey (S. Anlaş); HNHM, Hungarian Natural History Museum, Budapest, Hungary (G. Makranczy, O. Merkl); IRSNB, Institut royal des Sciences naturelles de Belgique, Bruxelles, Belgium (W. Dekoninck); MHNG, Muséum d'Histoire Naturelle, Genève, Switzerland (G. Cuccodoro); NMNHS, National Museum of Natural History, Sofia, Bulgaria (R. Bekchiev); NMPC, National Museum, Praha, Czech Republic (M. Fikáček); and cKha, private collection of Eduard A. Khachikov, Rostov, Russia.

Results

Subtribe Stilicina Casey, 1905

Genus *Rugilus* Leach, 1819

Rugilus angustatus (Geoffroy, 1785)

Material: IRAN: 2 exs., 13.VIII.1970, northern Iran, western Elbruz, Kalardash, Rudbarak, 1850-2400 m (NMPC). GEORGIA: 1 ex., 17-19.V.1985, Batumi, leg. Dvořák (NMPC). RUSSIA: 1 ex., 10.VII.1999, Rostov, Sholochovsky district, Staroe Lake, leg. Khachikov (cKha). 1 ex., 22.VII.1999, Rostov, Veshenskaya village, leg. Khachikov (AZMM). TURKEY: 2 exs., 02.V.2015, Afyonkarahisar, Ahır Mountain, Büyükhacet Hill, 38°39'42" N, 30°06'05" E, 1925 m, leg. Yağmur & Örgel (AZMM). 3 exs., 22.III.2015, Aydın, Dilek Peninsula National Park, 37°39'49" N, 27°12'57" E, 969 m, leg. Yağmur & Örgel (AZMM). 1 ex., 18.IV.2015, Denizli, Çameli, Değirmentaşı Hill, 37°07'21" N, 29°20'35" E, 1497 m, leg. Anlaş, Yağmur, Örgel & Altın (AZMM). 1 ex., 04.V.2015, Manisa, garden of Alaşehir Vocational School, leg. Örgel (AZMM). 1 ex., 13.IV.2015, Kütahya, Simav, Akdağ, 39°14'58" N, 28°49'41" E, 1670 m, leg. Anlaş & Örgel. (AZMM). 1 ex., 04.IV.2013, Muğla, Datça, Emecik 2 km SW, 36°46'01" N, 27°48'39" E, 107 m, leg. Yağmur & Örgel (AZMM). 1 ex., 20.V.2016, Konya, Beyşehir, Erenler Mountain, near a lake, 37°34'12" N, 32°02'53" E, 1768 m, leg. Örgel & Yaman (AZMM). LOCALITY UNKNOWN: 2 exs., 01.VII.1928, Karpaty, Kuzy (NMPC).

Distribution: *Rugilus angustatus* is widespread in Europe, Cyprus, Turkey (Table 1) and western Siberia from Palearctic Region (Schülke & Smetana, 2015). The above specimens from Iran represent the first record for that country.

***Rugilus arabs* (Saulcy, 1865)**

Material: LEBANON: 1 ex., 02.IV.1975, Les Cédres près de Becharré, 1950-2000 m, leg. Besuchet (MHNG). ISRAEL: 3 exs., 21.IV.1982, Galilée, Mountain, Meron, 900 m, leg. Besuchet & Löbl (MHNG). SYRIA: 1 ex., 04.III.2007, Latakia (AZMM). TURKEY: 2 exs., 31.V.2011, Muş, Varto, leg. Khachikov & Kasatkin (AZMM). 1 ex., 16.XI.2010, Şanlıurfa, Birecik, Kelaynak Valley, leg. Anlaş (AZMM). 3 exs., 11.IV.2014, İzmir, Bozdağlar, 38°24'46" N, 28°08'01" E, 939 m, leg. Anlaş (AZMM). 2 exs., 30.XI.2014, Manisa, Spil Mountain, 38°33'44" N, 27°23'10" E, 1100 m, leg. Yağmur & Örgel (AZMM).

Distribution: This species was known from Israel, Lebanon and Turkey (Table 1) (Schülke & Smetana, 2015). The above specimen in Syria represent the first record for that country.

***Rugilus couloni* (Drugmand, 1989)**

Stilicis couloni Drugmand, 1989: 110

Type examined: Holotype: ♂, Israel, Tel Dan, 25.V.1988, leg. G. Coulon (IRSNB). Paratypes: 3♀♀, same data as holotype (IRSNB).

Redescription: Measurements (in mm) and ratios (range, n = 4): AL 1.72–1.76; HL 0.91–0.95; HW 0.98–1.03; PW 0.79–0.82; PL 0.91–0.95; EL 1.01–1.06; EW 1.15–1.20; AW 0.95–0.98; ML 1.03 (n = 1); TL 6.2–6.4 HL/HW 0.92–0.93; PW/HW 0.80–0.81; PW/PL 0.86–0.87; EL/PL 1.11–1.12; EW/PW 1.46; EL/EW: 1.13–1.14; AW/EW 0.82–0.83.

Habitus and forebody as in Figure 1a-b. Coloration: head and pronotum blackish, elytra dark brown, abdomen blackish, with the narrow posterior margins of the tergites and the apex somewhat paler, antennae reddish brown, with the most antennomeres weakly infuscate, legs reddish brown, with the apices of the femora weakly infuscate. Head transverse, approximately 1.10 times as wide as long (see ratio HL/HW and Figure 1a-b); punctuation coarse, very dense and areolate; interstices reduced to narrow ridges, microsculpture absent; pubescence short and reddish brown, eyes large, longer than postocular region in dorsal view (Figure 1b); antennae moderately slender, antennomere III approximately two times as long as wide; antennomeres IV-VI distinctly oblong, antennomeres VII-X slightly oblong (Figure 1a). Pronotum approximately 1.15 times as long as broad and approximately 0.8 times as wide as head (see ratios PW/PL, PW/HW and Figure 1a-b); punctuation similar to that of head, but denser and partly confluent; along midline partly impunctate, midline impunctate only in posterior half, small relict of impunctate area present also near anterior margin and this area shiny (Figure 1b); microsculpture very shallow; pubescence of similar length as that of head, but less fine and more conspicuous. Elytra large, almost 1.5 times as wide as pronotum and approximately 1.15 times as wide as long, (see ratios EW/PW, EW/EL and Figure 1a-b) and at suture longer than pronotum (see ratio EL/PL and Figure 1a-b); punctuation moderately dense, less finer and sparser than that of head and pronotum; interstices on average approximately as wide as diameter of punctures and glossy, microsculpture available; pubescence yellowish, more distinct than that of head and pronotum; hind wings totally developed. Abdomen narrower than elytra, nearly 0.8 times as wide as elytra (see ratio AW/EW and Figure 1a); punctuation very dense and very fine; interstices with distinct fine microsculpture; pubescence brown to dark brown; posterior margin of tergite VII with palisade fringe.

♂: sternite VI unmodified; posterior margin of sternite VII weakly concave, on either side with cluster of several long black setae; sternite VIII posteriorly with deep emargination and on either side of the excision with several long black setae; aedeagus approximately 1.05 mm long, with ventral process of distinctive shape (Figure 1e-f). The illustration of the aedeagus of *R. couloni* in Drugmand (1989) is misleading.

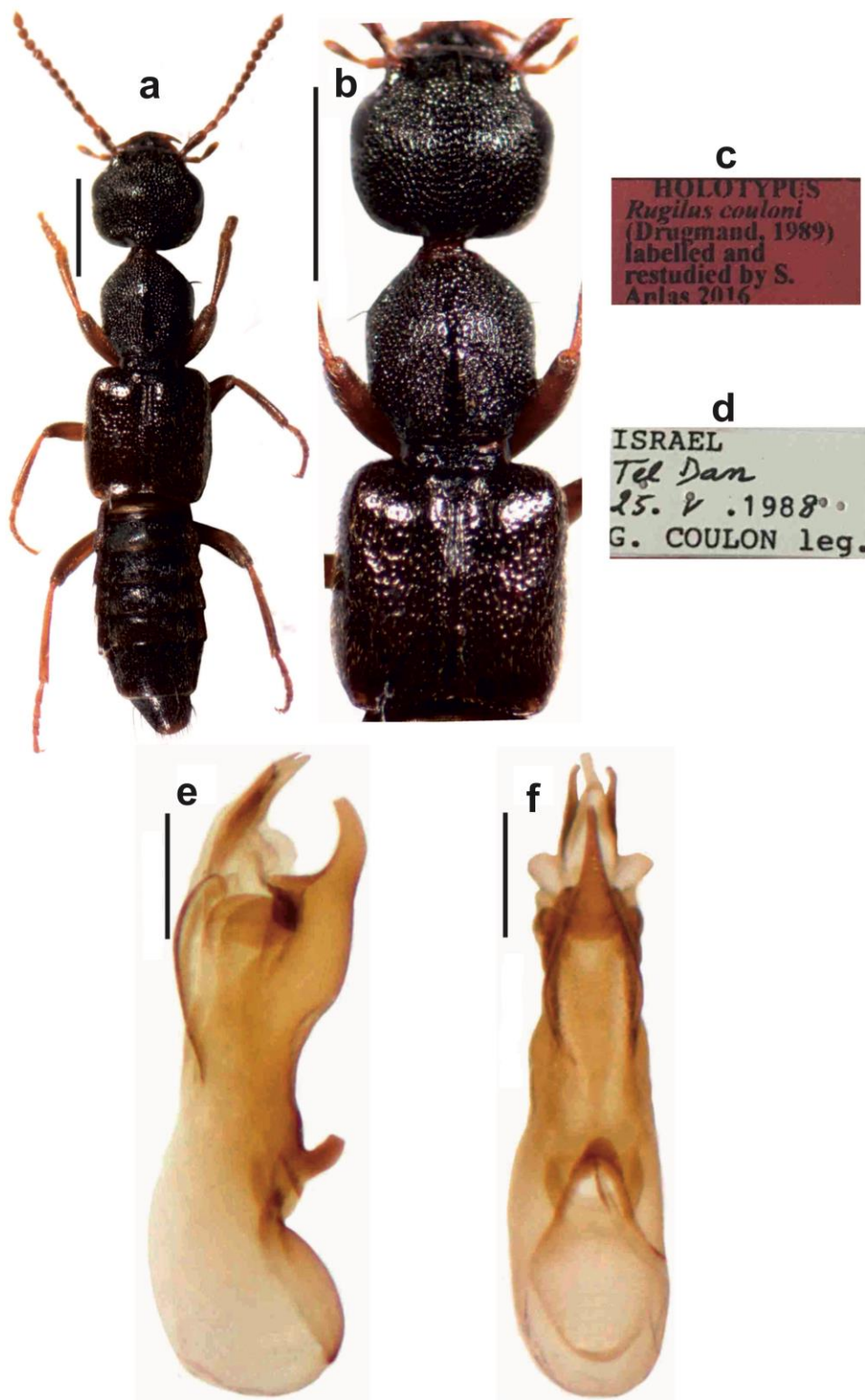


Figure 1. Details of *Rugilus couloni* (Drugmand, 1889): (a) habitus; (b) forebody; (c & d) holotypus labels; (e) aedeagus in lateral view; and (f) aedeagus in ventral view. Scale bars, 1.0 mm (a & b) and 0.2 mm (e & f).

Comparative notes: *Rugilus couloni* is distinguished from all its congeners by the male sexual characters, especially by the morphology of the aedeagus which is of different shape in ventral and lateral view. The species is similar to that of the widespread species *Rugilus orbiculatus* (Paykull, 1789). The species differs from *R. orbiculatus* by larger body (*R. orbiculatus*: body length < 5 mm), by the uniform coloration (*R. orbiculatus*: elytra with yellowish band on the outer apical angle), by the wider elytra than head (*R. orbiculatus*: elytra as wide as head) and by the completely different morphology of the aedeagus.

Remarks: Assing (2012) remarks that the "The Palaearctic fauna includes at least one doubtful species. The type material of *R. couloni*, which was described from Israel, is apparently lost. A clarification of the identity of this name is possible only when material from the vicinity of the type locality and in agreement with the original description becomes available". The Drugmand collection was deposited in the Institut royal des Sciences naturelles de Belgique (IRSNB). The type specimens of *R. couloni* were found in the collections of the IRSNB during a visit in 2016. However, neither the type labels nor identification labels were attached by Drugmand. Thus, the above type specimens were restudied and labeled as a holotype and three paratypes.

Distribution: This species is only known from its type locality in Israel (Drugmand, 1989; Schülke & Smetana, 2015).

***Rugilus erichsonii* (Fauvel, 1867)**

Material: BULGARIA: 1 ex., 09.VIII.2010, Belasitsa Mountain, Kongura hut, 41°34'55" N, 23°18'51" E, leg. Bekchiev (NMNHS). CZECH REPUBLIC: 1 ex., 10.IX.1907, Hluboká (NMPC).

Distribution: *Rugilus erichsonii* is widespread in Europe (Schülke & Smetana, 2015), but it has not been recorded in Bulgaria.

***Rugilus geniculatus* (Erichson, 1839)**

Material: CZECH REPUBLIC: 3 exs., 04.V.1919, Krhanice (NMPC).

Distribution: This species has Atlanto-Mediterranean distribution from northwestern Africa to Ukraine (Assing, 2012). However, it has not been recorded from Czech Republic. Thus, this species is reported here for the first time from Krhanice, Benešov District in the central Bohemian Region of Czech Republic.

***Rugilus korbi* (Fauvel, 1900)**

Material: AZERBAIJAN-IRAN: 1 ex., Talysch, Reitter (NMPC).

Distribution: According to Assing (2012), this species is known from Azerbaijan and northern Iran in Caspian Region.

***Rugilus leibius* Assing, 2005**

Material: TURKEY: 4 exs., 30.V.2014, Denizli, Babadağ, 37°47'43" N, 28°48'47" E, 903 m, leg. Örgel (AZMM). 2 exs., 11.IV.2014, İzmir, Bozdağlar, 38°24'46" N, 28°08'01" E, 939 m, leg. Anlaş (AZMM). 2 exs., 30.XI.2014, Manisa, Spil Mountain, 38°33'44" N, 27°23'10" E, 1100 m, leg. Yağmur & Örgel (AZMM).

Distribution: This species is known from Greece (Lesbos, Samos) and western Anatolia (Table 1) (Assing, 2012, 2013a).

***Rugilus longicollis* (Fauvel, 1900)**

Material: AZERBAIJAN: 1 ex., Caucasus, Leder, Reitter (NMPC). IRAN: 1 ex., 29.VI.1974, Kermanshah, Mâhi Dasht, 34°14'N, 46°42'E, leg. Senglet (det. Rougemont) (MHNG). TURKEY: 2 exs., 28.VI.2016, Yozgat, Aydıncık, Kuşsaray, 40°05'07" N, 35°11'59" E, 1341 m, leg. Örgel & Yaman (AZMM).

Distribution: This species is known from Azerbaijan, Georgia, Iran and central-eastern Turkey (Table 1) (Assing, 2012, 2013b; Sert et al., 2013; Schülke & Smetana, 2015).

***Rugilus maltzevi* Gusarov, 1991**

Material: BULGARIA: 2 exs., 06.V.2010, Ograzhden Mountain, Gega village 42°01'03" N, 27°44'56" E, leg. Bekchiev (AZMM, NMNHS). TURKEY: 1 ex., 07.V.1978, Konya, Beyşehir, 1650 m, leg. Besuchet & Löbl (MHNG). 1 ex., 27.V.2016, Konya, Ereğli, Kartaltepe, 37°22'57" N, 34°00'55" E, 1914 m, leg. Anlaş, Örgel & Yaman (AZMM). 2 exs., 15.X.2013, Denizli, Babadağ, 37°47'55" N, 28°51'26" E, 903 m, leg. Özgen & Örgel (AZMM). 1 ex., 30.XI.2014, Manisa, Spil Mountain, 38°33'44" N, 27°23'10" E, 1100 m, leg. Yağmur & Örgel (AZMM).

Distribution: According to Schülke & Smetana (2015), this species was known from Ukraina and Turkey (Table 1). Thus, it is reported here for the first time from Bulgaria.

***Rugilus orbiculatus* (Paykull, 1789)**

Material: ALGERIA: 1 ex., Algerie (MHNG). BULGARIA: 1 ex., 23.V.2010, Strandzha Mountain, bank of Veleka River, 42°06'10" N, 27°36'53" E, leg. Bekchiev (NMNHS). CZECH REPUBLIC: 1 ex., 12.XI.1939, Závist (NMPC). IRAQ: 3 exs., 4-5.XII.1977, Iraq, Arbil, Eskikalak, near Great Zab River, leg. Topál & Zilahy (HNHM). KAZAKHSTAN: 1 ex., 05.IV.2010, Yuzhno-Kazakhstan Region, Boralday range, satur mts, hole of the Kulan nv., high Krasnye vorota pass, 1000 m, 42°35'13" N, 70°26'53" E, leg. Matalin (AZMM). RUSSIA: 1 ex., 05.V. 1991, Rostov, Rostov-on-Don city, leg. Khachikov (cKha). 1 ex., X.1993, Stavropol Province Pyatigorsk city, Mashuk Mountain, leg. Khachikov (AZMM). 1 ex., 18.VIII.1991, Rostov, Shchepkinskoe Forest, leg. Khachikov (AZMM). SPAIN: 1 ex., 22.V.1991, Espana, Potes Picos de Europa, leg. Podlussány (HNHM). TURKEY: 1 ex., 06.VI.1986, Erczincan (=Erzincan), Tercan, Euphrate, 1400 m, leg. Besuchet, Löbl & Burckhardt (MHNG). 12 exs., 16.IX.2011, Afyonkarahisar, Sandıklı Mountain, 38°27'45" N, 30°21'30" E, 1548 m., by pitfall traps, leg. Yağmur (AZMM). 3 exs., 01.V.2013, Denizli, Çal, Yüğük, 38°00'54" N, 29°20'25" E, 1473 m, leg. Yağmur & Örgel (AZMM). 4 exs., 30.V.2014, Babadağ, 37°47'43" N, 28°48'47" E, 903 m, leg. Anlaş & Örgel (AZMM). 1 ex., 11.IV.2014, İzmir, Bozdağlar, 38°24'46" N, 28°08'01" E, 939 m, leg. Anlaş (AZMM). 1 ex., 06.IV.2015, Bergama, Kozak, Güneşli, 39°21'09" N, 27°07'54" E, 795 m, leg. Yağmur & Örgel (AZMM). 1 ex., 14.VII.1980, Smyrna (=İzmir) (HNHM). 1 ex., 17.V.2013, Kütahya, Simav, Kuyusinir 2 km W, 39°20'21" N, 29°54'08" E, 1310 m, leg. Yağmur & Örgel (AZMM). 2 exs., 30.XI.2014, Manisa, Spil Mountain, 38°33'44" N, 27°23'10" E, 1100 m, leg. Yağmur & Örgel (AZMM). 1 ex., 07.IV.2015, Soma, Yağcılı, 39°20'05" N, 27°40'23" E, 306 m, leg. Yağmur & Örgel (AZMM). 1 ex., 07.IV.2015, Soma, Tabanlar, 39°19'47" N, 27°44'17" E, 682 m, leg. Yağmur & Örgel (AZMM). 1 ex., 28.VI.2012, Konya, Güragaç, Güneysınır, leg. Yağmur (AZMM). 3 exs., 20-21.V.2016, Seydişehir, Erenler Mountain, 37°34' N, 32°00' E, 1500-1900 m, leg. Örgel & Yaman (AZMM). 1 ex., 28.V.2016, Karaman, Ayrancı, Yüğük, 37°00'57" N, 33°46'48" E, 1942 m, leg. Anlaş, Örgel & Yaman (AZMM). 1 ex., 31.V.2016, Niğde, Çiftlik, Gebere Valley, 38°03'03" N, 34°37'19" E, 1798 m, leg. Anlaş, Örgel & Yaman (AZMM).

Distribution: *Rugilus orbiculatus* widespread in western Palearctic, Middle Asia, Nearctic and Australian Regions (Assing, 2012; Schülke & Smetana, 2015). It is reported here for the first time from Iraq.

***Rugilus prolongatus prolongatus* (Solsky, 1874)**

Material: KAZAKHSTAN: 1 ex., 24-30.III.2010, Yuzhno-Kazakhstan Region, Arystandi River, upstream, 7.5 NNE of Shaklak Mountain, 43°15'17" N, 69°26'30" E, 400 m, leg. Matalin (AZMM). UZBEKISTAN: 1 ex., Buchara (=Bukhara) (det. Lokay) (NMPC).

Distribution: This species was known from Kyrgyzstan, Tajikistan and Uzbekistan (Assing, 2012). It is reported here for the first time from Kazakhstan.

***Rugilus rossii* (Zanetti, 1977)**

Material: ITALY: 5 exs., 23.VIII.1990, Calabria, Lamezia, Terme, S. Nicola, leg. Angelini (AZMM).

Distribution: This species is endemic to Italy (Schülke & Smetana, 2015).

***Rugilus rufipes* Germar, 1836**

Material: AZERBAIJAN: 5 exs., Astara, Motlayatag village 2-6.VI.2006, leg. Snegovaya (AZMM). 1 ex., 25-31.V.2008, Lardymly District, near Kiurektshi village, leg. Kasatkin (AZMM). 2 exs., 18.V.2007, Lankaran, Apo, leg. Snegovaya (AZMM). 1 ex., 10.VI.2007, Lankaran, Azfiliag, leg. Snegovaya (AZMM). 3 exs., 14-16.VI.2007, near Peshtatyuk village, leg. Kasatkin (AZMM; cKha). 1 ex., 20.V.2007, Lerik village leg. Snegovaya (AZMM). CROATIA: 1 ex., 03.XI.1916, Istria, Vozice, leg. Fodor (HNHM). CZECH REPUBLIC: 1 ex., 04.VI.1947, Hradec (NMPC). 1 ex., 16.IV.1944, Závist (NMPC). ITALY: 1 ex., 14.V.1989, Basilicata, Accettura Bosco di Montepiano trapp.ossa, 950 m, leg. Angelini (AZMM). 4 exs., 3.VIII.1990, Campania, Cilento, Centaurino, 500 m, leg. Angelini (AZMM). TURKEY: 2 exs., 03.X.2009, Kırklareli, Demirköy, İğneada-Demirköy road 11 km N, under pine forest, 41°51'45" N, 27°53'00" E, leg. Kunt (AZMM). 1 ex., 23.V.2010, Demirköy, İğneada, ca. 20 m, Hamam Lake, 41°49'43" N, 27°57'31" E, leg. Kunt (AZMM). 2 exs., 03.X.2009, İğneada, Siğlioba, leg. Kunt (AZMM). 2 exs., 27.IX.2009, Manisa, Spil Mountain, 1200 m, 38°33'20" N, 27°23'17" E, leg. Anlaş (AZMM). 1 ex., 14.III.2014, Manisa, Selendi-Demirci road, 38°49'35" N, 28°47'55" E, 647 m, leg. Yağmur & Örgel (AZMM). 3 exs., 13.IV.2015, Balıkesir, Sındırgı 20 km W, 39°07'59" N, 28°00'33" E, 408 m, leg. Anlaş & Örgel (AZMM).

Distribution: *Rugilus rufipes* widespread in western Palearctic and western Siberia (Assing, 2012).

***Rugilus similis* (Erichson, 1839)**

Material: AZERBAIJAN: 1 ex., 20.V.2005, Lerik village leg. Snegovaya (AZMM). IRAN: 2 exs., 28-30.V.1973, southern Iran, Korsiah, Exped. Nat. Mus. Praha (NMPC). ITALY: 1 ex., 17.VI.1998, Campania, Matese Lake (CE), 1100 m, leg. Angelini (AZMM). RUSSIA: 1 ex., 30.IX.1990, Rostov, Shchepkinskoe Forest, leg. Khachikov (AZMM). 1 ex., 30.IX.1991, Rostov, Rostov-on-Don city, leg. Arzanov (AZMM). TURKEY: 2 exs., 11.VIII.2010, Afyonkarahisar, Şuhut, Dadak 2 km N, 1320 m, 38°36'18" N, 30°26'59" E, leg. Anlaş (AZMM). 2 exs., 18.III.2015, Denizli, Çameli, Değirmentaşlı Hill, 37°07'21" N, 29°20'35" E, 1497 m, leg. Yağmur & Örgel (AZMM). 2 exs., 14.III.2014, Manisa, Selendi-Demirci road, 38°49'35" N, 28°47'55" E, 647 m, leg. Yağmur & Örgel (AZMM). 2 exs., 20.V.2016, Konya, Beyşehir, Erenler Mountain, 37°34'12" N, 32°02'53" E, 1768 m, leg. Örgel & Yaman (AZMM).

Distribution: *Rugilus similis* was known from Europe, western Siberia, Kazakhstan, Syria and Turkey (Table 1) (Assing, 2012; Schülke & Smetana, 2015). This species is reported here for the first time from Azerbaijan and Iran.

***Rugilus subtilis* (Erichson, 1840)**

Material: BULGARIA: 1 ex., 18.IV.2010, Strandzha Mountain, near Malko Tarnova, 41°58' N, 27°52' E, leg. Bekchiev (NMNHS). GREECE: 1 ex., IX.1984, Olympos, leg. Mahunka (HNHM). SERBIA: 1 ex., 24.III.1918, Arilje, Vrané (NMPC). TURKEY: 1 ex., 27.III.2007, Manisa, Turgutlu, Dağmarmara, Ovacık, leg. Anlaş (AZMM).

Distribution: This species is known from Europe and Turkey (Table 1) (Schülke & Smetana, 2015).

***Rugilus tauricus* (Rougemont, 1988)**

Material: TURKEY: 1 ex., 01.IX.2011, Antalya, Akseki, Çuçur village, 36°52'34" N, 31°40'47" E, 631 m, leg. Örgel (AZMM). 3 exs., 02.VI.2016, Niğde, Ulukışla, Horoz, 37°28'47" N, 34°47'55" E, 1049 m, leg. Anlaş, Örgel & Yaman (AZMM).

Distribution: According to Assing (2012), this species is only known to occur in central southern Turkey (Table 1).

Table 1. Distribution of *Rugilus* species in Turkey

Species	Provinces	References
<i>Rugilus angustatus</i> (Geoffroy, 1785)	Afyonkarahisar, Aksaray, Ankara, Aydın, Çankırı, Denizli, Eskişehir, İzmir, Karaman, Kastamonu, Kayseri, Konya, Kütahya, Manisa, Muğla, Samsun, Yozgat	Rougement (1988), Anlaş (2009), Anlaş & Rose (2009), Sert et al. (2013), Assing (2014), Çiftçi & Hasbenli (2016), Anlaş (present paper)
<i>Rugilus arabs</i> (Saulcy, 1865)	Adana, Amasya, Gaziantep, İzmir, Manisa, Mersin, Muş, Osmaniye, Şanlıurfa, Tokat	Rougement (1988), Anlaş (2009), Assing (2009), Anlaş (present paper)
<i>Rugilus lesbisus</i> Assing, 2005	Aydın, Bolu, Denizli, Eskişehir, İzmir, Manisa, Sakarya	Assing (2013a), Çiftçi & Hasbenli (2016), Anlaş (present paper)
<i>Rugilus longicollis</i> (Fauvel, 1900)	Yozgat	Sert et al. (2013), Anlaş (present paper)
<i>Rugilus maltzevi</i> Gusarov, 1991	Antalya, Denizli, Konya, Manisa	Assing (2009), Anlaş (present paper)
<i>Rugilus orbiculatus</i> (Paykull, 1789)	Afyonkarahisar, Antalya, Denizli, Erzincan, Erzurum, Eskişehir, İstanbul, İzmir, Karaman, Kayseri, Konya, Kütahya, Manisa, Niğde	Rougement (1988), Anlaş (2009), Anlaş & Rose (2009), Kesdek et al. (2009), Sert et al. (2013), Çiftçi & Hasbenli (2016), Anlaş (present paper)
<i>Rugilus rufipes</i> Germar, 1836	Adana, Balıkesir, Bolu, Eskişehir, İstanbul, Kırklareli, Kocaeli, Manisa, Sakarya	Peyron (1858), Rougement (1988), Anlaş (2009), Assing (2011, 2013a), Sert et al. (2013), Anlaş (present paper)
<i>Rugilus similis</i> (Erichson, 1839)	Afyonkarahisar, Aksaray, Çankırı, Denizli, Erzurum, Eskişehir, Karaman, Manisa, Mersin, Muğla, Osmaniye	Rougement (1988), Anlaş (2009), Assing (2013b), Sert et al. (2013), Çiftçi & Hasbenli (2016), Anlaş (present paper)
<i>Rugilus subtilis</i> (Erichson, 1840)	Ankara, Eskişehir, Kastamonu, Manisa	Anlaş (2009), Assing (2011), Çiftçi & Hasbenli (2016), Anlaş (present paper)
<i>Rugilus tauricus</i> (Rougemont, 1988)	Adana, Afyonkarahisar, Antalya, Isparta, Konya, Niğde	Rougement (1988), Anlaş (2009), Anlaş & Rose (2009), Japoshvili & Anlaş (2011), Assing (2013a), Sert et al. (2013), Anlaş (present paper)

Acknowledgments

I am most grateful to my colleagues for making their staphylinid collections available to me. Special thanks to G. Makranczy (Budapest), G. Cuccodoro (Genève), M. Fikáček (Prague), W. Dekoninck (Brussels), R. Bekchiev (Sofia) and E. A. Khachikov (Rostov) for arranging the loan of *Rugilus* material. This study was in part supported by the Scientific and Technological Research Council of Turkey (TÜBİTAK, projects 112T907 and 215Z080).

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Original article (Orijinal araştırma)

Khpra beetle (*Trogoderma granarium* Everts, 1898) in durum wheat (*Triticum durum* Desf): Impacts on some seed characteristics and marketing price

Makarnalık buğdayda (*Triticum durum* Desf) kapra böceği (*Trogoderma granarium* Everts, 1898): Bazı tane özellikleri ve pazarlama fiyatlarına etkileri

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Summary

This study investigated the influence of khpra beetle (*Trogoderma granarium* Everts, 1898, Coleoptera: Dermestidae) on weight, grain quality and marketing price losses in various durum wheat cultivars stored in controlled conditions in laboratory of Faculty of Agriculture at Harran University. Experiment was conducted from 13 January to 4 September 2015. Durum wheat cultivars, Şahinbey, Diyarbakır-81, Zühre, Artuklu, Güney Yıldızı, Fırat-93, Aydın-93, Sarıçanak-98, Eyyübi and Altıntoprak-98, were infested by three different young larval stages. Three samples (80 g) of grain from each cultivar were put into 250-mL glass jars covered with the muslin cloth with the rubber bands and 5, 10 or 15 neonates khpra beetle larvae added. A randomized complete block design with 3 replicates was employed for grain weight losses and a split plot design with 4 replicates (purchasers) was employed for marketing price losses. Grain weight, marketing price and grain quality losses were recorded. The result revealed that geometric mean of weight loss was 4.075% in about 8 months. There were response differences between wheat cultivars against khpra beetle infestation. Except for Zeleny sedimentation, some of quality characteristics such as 1000 kernel weight (g), gluten (%) and gluten index (%) were affected negatively depending on increasing ratio of insect infestations. Geometric means of marketing prices reduced from 418 to 315 USD t⁻¹ in 8 months. Marketing price loss was 103 USD t⁻¹. It was concluded that Fırat-93, Zühre and Altıntoprak-98 were the cultivars least affected by khpra beetle with less weight and marketing price losses.

Keywords: Durum wheat, khpra beetle damage, marketing price, weight and quality loss

Özet

Bu çalışmada laboratuvar koşullarında depolanmış bazı makarnalık buğday çeşitlerinde khpra böceği (*Trogoderma granarium* Everts, 1898, Coleoptera: Dermestidae)'nin yaptığı ağırlık, kalite ve pazarlama fiyatları kayıpları incelenmiştir. Çalışma Harran Üniversitesi, Ziraat Fakültesi, Tarla Bölümü Laboratuvarı'nda 13 Ocak- 4 Eylül 2015 tarihleri arasında yürütülmüştür. Şahinbey, Diyarbakır-81, Zühre, Artuklu, Güney Yıldızı, Fırat-93, Aydın-93, Sarıçanak-98, Eyyübi ve Altıntoprak-98 makarnalık buğday çeşitlerine genç larvalar üç farklı sayıda bulaştırılmıştır. Her bir çeşide ait 3 adet (80 gr) örnek içlerine 5,10 ve 15 adet genç khpra böceği larvası yerleştirilerek 250-mL'lik cam kavanozlara konulmuş ve ağız lastik bantlı tülben bezi ile örtülmüştür. Deneme ağırlık ve kalite kayıpları için tesadüf blokları deneme desenine göre üç tekerrürlü (larva seviyeleri) olarak yürütülmüştür. Pazarlama fiyatları için ise bölünmüş parseller (çeşitler ana parsel, larva bulaşma oranları alt parsel) deneme desenine göre 4 tekerrürlü (borsadaki alıcılar) olarak yürütülmüştür. Dane ağırlığı, pazarlama fiyatı ve dane kalite kayıpları kaydedilmiştir. Elde edilen sonuçlara göre 8 ayda tane ağırlık kayıplarının geometrik ortalaması %4.075 olmuştur. Çeşitler arasında khpra zararına karşı farklı tepki olduğu anlaşılmıştır. Zeleny sedimentasyon değeri dışında, 1000 dane ağırlığı (g), gluten (%) ve gluten indeks (%) değerleri larva bulaşma oranı arttıkça olumsuz yönde etkilenmiştir. 8 ayda khpra'ya bağlı pazarlama fiyatları 418 USD t⁻¹'den 315 USD t⁻¹'a düşmüştür. Pazarlama fiyatı kaybı 103 USD t⁻¹ olmuştur. Fırat-93, Zühre ve Altıntoprak-98 çeşitleri pazarlama fiyatı azalışı ve ağırlık kaybı yönünden khpra zararından en az etkilenen çeşitler olmuşlardır.

Anahtar sözcükler: Makarnalık buğday, khpra böceği zararı, pazarlama fiyatları, ağırlık ve kalite kaybı

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Received (Alınış): 04.02.2017

Accepted (Kabul edilmiş): 24.05.2017

Published Online (Çevrimiçi Yayın Tarihi): 28.06.2017

Introduction

Khapra beetle, *Trogoderma granarium* Everts, 1898 (Coleoptera: Dermestidae) (Munro, 1935), which is one of 115 *Trogoderma* species (Beal, 1982), is one of the most important stored-product pests ranked as one of the 100 worst severe species on earth (Lowe et al., 2000) and considered as an A₂ quarantine level organism by the EPPO (OEPP/EPPO, 1981) (Ahmedani et al., 2007). In Southeastern Anatolia, estimates of storage losses of food grains due to khapra beetle have been reported to vary greatly; 25-35% in 1963 (Kalkan, 1963) and 10% in 2000 (Ekmekçi & Ferizli, 2000). Similarly, in Pakistan khapra beetle damage varied from 4 to 10% (Huque et al., 1969), about 5% (Chaudhry, 1980), 5% (Ahmad, 1984), and from 3.5 to 25.5% (Irshad & Baloch, 1985). Average damage varied between 6 to 33% of stored grain in one season in India (Rahman et al., 1945). The worldwide stored grain loss average was estimated at 10% (Prevett, 1975) and 5% of this was due to insect damage (Esin, 1971). The great importance of this pest relies on its capacity to cause huge loss in stored grain through fast feeding and heating. Mature larvae have potential to withstand starvation for about 3 years. Furthermore, larvae have an ability to live on food with very low moisture content (Ahmedani et al., 2007). Khapra beetle larvae feed on wheat grain and as a result the nutritive quality of the wheat decreases, which lead to lower the marketing price (Ahmedani et al., 2009). Damage occurs in larval stage and adults feed only a little on the grains (Ahmedani et al., 2007) or do not feed at all (Freeman, 1980). Temperature and relative humidity (RH) are the two main physical factors that influence the population of khapra beetle (Cockerel et al., 1971). Larval development is not possible below 12°C but may proceed at very low RH, for example at 25°C and 2% RH. Development is most rapid in warm humid conditions, taking about 18 days at 35°C and 73% RH, and under these conditions the number of larval molts is 4 for males and 5 for females (Hadaway, 1956).

The length of the youngest larva is 1.6-1.8 mm, body width is 0.25-0.30 mm with a tail longer than half of the whole body and tail is made up of quite lot hairs derived from on the last abdominal segment. Mature larva is about 6 mm long and 1.5 mm wide (OEPP/EPPO, 1981). Male pupae are smaller than female ones. The average lengths of males and females are 3.5 and 5 mm, respectively (OEPP/EPPO, 1981). Adults are oblong or oval shape and 1.6-3.0 mm long by 0.9-1.7 mm wide. Males are brown to black in color and the females are lighter in color. Female pupae are larger than male ones. The adults have short life span. The mated and unmated females can survive about 4-7 and 20-30 days respectively and males 7-12 days. They do not fly and can feed very little (Ahmedani et al., 2007). Once-mated females can lay about 60 eggs but more than twice mated females can lay up to 500 eggs.

The youngest larvae are unable to feed on whole grains and can survive eating only damaged grains, older larvae can feed on whole grains. The rate of increase at 33-37°C is about 12.5 times per month (Anonymous, 2005). Khapra beetle has no special preference and can benefit from number of feed products including durum wheat (*Triticum durum* Desf.) (Jha, 2003). Grain quality decreases probably due to abolishment of specific nutrients. It can result in significant decreases in crude fat, total carbohydrates, sugars, protein nitrogen and true protein contents and increases in moisture, crude fiber and total protein at the infestation levels of 75% in wheat, maize and sorghum grains (Jood et al., 1993). Cast skins of khapra beetle may result in dermatitis (Pruthi & Singh, 1950), when the barbed hairs of larvae remain in the grain this may result in a serious hazard, if swallowed (Marison, 1925).

Turkey is one of the most important grain producing countries of world, especially for wheat where it is classified in the top-ten countries globally (FAO Stat, 2009). Southeastern Anatolia is considered to be the durum wheat belt of Turkey. Around the 35% of total durum wheat production is grown in the Southeastern Anatolia (Özberk et al., 2005; 2006). Temperate cereal acreage in the region is about 2 million ha representing 15-17% of total area of Turkey. Total wheat acreage is 1,152,500 ha and annual production is 2,045,990 t. Major growing sites are Şanlıurfa and Diyarbakır Provinces (Özberk et al., 2005). Turkey harbors many species of storage pests due to its suitable climate (Ekmekçi & Ferizli, 2000). *Trogoderma granarium* can reach high infestation rates in wheat samples in Şanlıurfa Province (Işıkber et al., 2014). Grading factors such as the presence of sunn pest (*Eurygaster integriceps* Puton, 1881) damaged kernels in the durum wheat seed lots, presence of red bread wheat kernels, vitreousness

and starchy kernels are major downgrading factors in the region. Some visual characteristics such as 1000 kernel weights and hectoliter weights are also referred by local purchasers (Özberk et al., 2006). The effects of khapra beetle damaged kernels onto marketing prices have not been studied previously. This study investigated the effects of khapra beetle on some seed quality characteristics and the impacts on marketing prices in durum wheat. Cultivar differences were also assessed.

Material and Methods

Widely grown durum wheat cultivars, Şahinbey, Diyarbakır-81, Zühre, Artuklu, Güney Yıldızı, Fırat-93, Aydın-93, Sarıçanak-98, Eyyübi and Altıntoprak-98, were appraised against three different infestation densities (5, 10 and 15 neonate larvae jar⁻¹) of khapra beetle in the laboratory of the Faculty of Agriculture of Harran University, Şanlıurfa, Turkey from February to September of 2015. Some seed characteristics and losses in grain weight and marketing prices were scored periodically. Grain samples in glass jars subjected to khapra beetle infestation in the laboratory were presented to the randomly selected grain purchaser and marketing prices offers were scored. A randomized complete block design with 10 entries and 3 replicates (i.e., 5, 10 and 15 neonate larvae jar⁻¹) was employed for weight loss and some seed characteristics. A split plot design was employed for marketing price losses. Where, the cultivars and three larval infestation densities were assigned to main and subplots respectively. Purchasers in local commodity market were employed as replicates. Grain samples of durum wheat cultivars were received from GAP International Agriculture Research and Training Center in neighboring Diyarbakır Province. Grain samples were treated by high temperature (5 h at 45°C) to abolish the possibility of previous infestation. RH after this treatment was about 10% for all entries. Three samples (80 g) of wheat grain from each cultivar were put into 250 mL glass jars covered with the muslin cloth with the rubber bands and 5, 10 or 15 neonate khapra beetle larvae added. Thousand kernel weights (Uluöz, 1965), Zeleny sedimentation (AACC, 2000; method 56-60), delayed sedimentation (Greenway et al., 1965), gluten (%) and gluten index (%) (AACC, 2000; method 38-12A) were scored initially and at the end of experiment. Khapra larvae were collected from the wheat storage house of the Plant Protection Department of the Provincial Extension Service in Şanlıurfa. The jars were put in an incubator under semi-storage house conditions in summer at 30±2°C, 55±5% RH. The infested grains in each jar were subjected to sieving to isolate the grain dust, exuviate and other residues formed due to the khapra beetle infestation. All live larvae and pupae in jars were put aside and reintroduced to the jar after weighing. Weight losses were scored five times during the period of incubation between 13 January and 4 September 2015. In the same period, marketing price estimates were scored 3 times in local commodity market. JMP-5 statistical software was employed for analysis of variance. A stability analysis called rank (Huehn, 1990) was also performed to detect the less affected cultivars for both weight and market price and losses.

Results and Discussion

Weight losses

Weight losses for all entries under study were scored on 2 and 23 February, 6 April, 16 June and 4 September 2015. Individual analysis of variance indicated the presence of significant cultivar response against khapra beetle infestation ($F = 24.78^{***}$, $P < 0.001$; $F = 12.47^{***}$, $P < 0.001$; $F = 4.94^{**}$, $P < 0.01$; $F = 4.22^{**}$, $P < 0.01$; and $F = 4.91^{**}$, $P < 0.01$ respectively). Geometric grand mean of weight loss was 4.075% in about 8 months. There were no significant differences among replicates (i.e., larval infestation levels) until the last two scoring dates. The effects of larval infestation levels were found to be significant in last two scoring dates ($F = 4.32^*$, $P < 0.05$ and $F = 21.36^{***}$, $P < 0.001$ respectively). Weight loss increased with increasing initial infestation level (Table 1). A rank stability analysis for weight loss occurred after 8 months of artificial infestation by khapra beetle larvae (Figure 1) showed that Fırat-93, Zühre and Altıntoprak-98 were the cultivars least affected, whereas Artuklu and Sarıçanak-98 were the most susceptible cultivars. These results confirmed the previous findings of Ahmad et al. (1986) and Navarro et al. (1978), who reported a high degree of positive correlation between infestation levels and weight loss. Khattak et al. (2000) studied on the effect of khapra beetle infestation employing twelve wheat lines and also found that correlation between progeny development vs. damage and weight loss

was positive and highly significant ($P < 0.01$). Their results matched those of Syed et al. (2006), in which they evaluated the losses caused by khapra beetle to various wheat cultivars. Results of Ahmedani et al. (2011) revealed that increasing infestation levels resulted in significant increase in progeny development, weight loss and weight of frass, the number of broken and insect damaged grains. In general, the insects tend to develop more slowly on khapra beetle resistant wheat cultivars. It is known that there have been several studies about the resistance mechanism of wheat grains against khapra beetle but inheritance of the factors controlling resistance have scarcely been studied (Dobie, 1991).

Table 1. Means and LSD groups of cultivars and larval infestation levels for the weight losses from 80 g of initial sample weight on five consecutive dates

Cultivars Name	Scoring dates				
	02.02.2015	23.02.2015	06.04.2015	16.04.2015	04.09.2015
Şahinbey	78.88 a	78.71 a	78.62 a	77.06 a	68.29 bc
Diyarbakır-81	78.80 b	78.69 a	78.58 a	73.67 b	67.48 bc
Zühre	78.72 c	78.68 a	78.54 a	78.18 a	74.49 a
Artuklu	78.78 b	78.60 ab	78.49 a	77.01 a	71.57 abc
Güney Yıldızı	78.69 cd	78.56 b	78.47 a	77.34 a	67.03 bc
Fırat-93	78.81 b	78.69 a	78.64 a	78.70 a	76.12 a
Aydın-93	78.78 b	78.66 ab	78.52 a	76.23 ab	69.43 bc
Sarıçanak-98	78.65 de	78.38 c	78.23 b	77.81 a	76.01 a
Eyyübi	78.60 e	78.29 c	78.25 b	76.39 ab	66.95 c
Altıntoprak-98	78.89 a	78.65 ab	78.50 a	77.36 a	71.82 ab
Larval intensity (LI)					
5	78.78 a	78.61 a	78.52 a	77.92 a	74.23 a
10	78.75 a	78.60 a	78.49 a	77.17 ab	72.25 a
15	78.75 a	78.56 a	78.43 a	75.83 b	66.27 b
Statistical significance for some sources of variation and some descriptive statistics					
F (Cultivars)	24.78**	12.47**	4.94**	2.22 ^{ns}	4.91**
F (Larval intensity)	2.45 ^{ns}	1.15 ^{ns}	1.74 ^{ns}	4.32*	21.39**
Grand mean	78.76	78.59	78.48	76.97	70.92
Standard deviation (SD)	0.033	0.07	0.11	1.61	2.84
LSD	0.046	0.10	0.15	2.28	4.02
CV%	0.4	0.80	0.13	2.09	4.00

ns, not significant, *significant at $P < 0.05$, **significant at $P < 0.01$, difference between the means with same letter in a column is not significant.

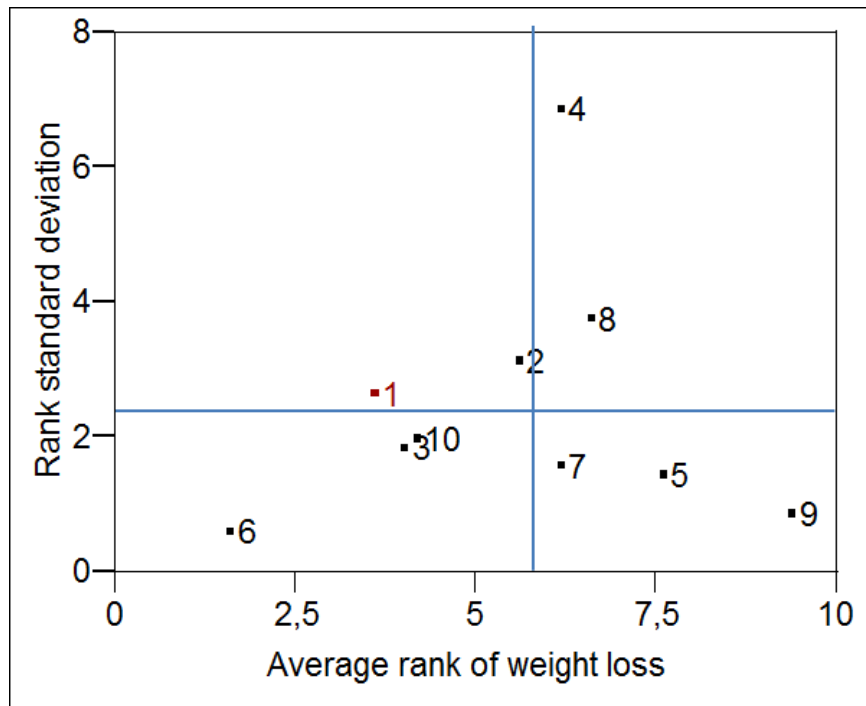


Figure 1. Ranks stability analysis for weight losses of varieties under study (1. Şahinbey, 2. Diyarbakır-81, 3. Zühre, 4. Artuklu, 5. Güney Yıldızı, 6. Fırat-93, 7. Aydın-93, 8. Sarıçanak-98, 9. Eyyubi, 10. Altıntoprak-98).

Grain quality assessment

Thousand kernel weights as one of most affecting grading factor on market price were first assessed on 18 May 2015. Thousand healthy grains and thousand randomly selected grains from glass jars of each entry with different levels of infestation were taken and weighed. An analysis of variance was performed and the means with LSD groups are given in Table 2. The overall mean 1000 kernel weight for healthy grains was 46.19 g whereas for the randomly chosen grains it was 42.21 g. An average of 3.98 g decrease was observed due to khapra beetle larvae damage. Altıntoprak-98, Güney Yıldızı and Şahinbey cultivars exhibited minimum kernel weight loss of 2.90, 2.63 and 3.46 g, respectively. Grain samples of entries with different infestation levels were selected (not adequate for replication) and tested for Zeleny and delayed Zeleny sedimentation tests at the end of study (Table 2). Zeleny sedimentation values ranged between 11 and 19 indicating weakness of durum wheat for this characteristic and the presence of some difference among cultivars. Delayed sedimentation was employed to detected sunn pest damage of grains. Results showed that only Sarıçanak-98 was suffered from sunn pest damage. It also proved to exhibit maximum kernel weight losses. There might be a correlation between susceptibility to sunn pest and khapra beetle damage due to the relatively soft grain structure. Gluten was also tested for all entries (not adequate for replication). Wet gluten ranged from 33 to 48% showing strong nature of durum wheat and the presence of genuine differences among the cultivars. Gluten index values were also scored for all entries under study. There was very high variation among entries with the lowest one of 3.38% and the highest one of 86.95% (Table 2). Eyyübi, Zühre and Altıntoprak-98 gave the highest three ranks with 86.95, 83.86 and 80.90%, respectively. Sarıçanak-98, Diyarbakır-81 and Şahinbey generated the extremely low values with 3.38, 4.60 and 5.10%, respectively. Those low values were attributed to khapra beetle damage totally. Grain quality may downgrade due to reduction of specific nutrients. Significant decreases in crude fat, total carbohydrates, sugars, protein nitrogen and true protein contents and increase in moisture, crude fiber and total protein occurred at the infestation level of 75% by khapra beetle in wheat, maize and sorghum grains (Jood & Kapoor, 1993; Jood et al., 1993; 1996). Starch content decreased at the 50% infestation level (Jood et al., 1993). Severe infestations of grains by khapra beetle may result in unpalatable or unmarketable products for human consumption.

Table 2. Means and LSD groups of 1000 kernel weights and the means of some quality characteristics of cultivars scored on various dates

Cultivars Name	Scoring dates					
	18.06.2015	18.06.2015	04.09.2015	04.09.2015	04.09.2015	04.09.2015
	1000 kernel weight in healthy grains (g)	1000 kernel weight in khapra beetle damaged grains (g)	Zeleny sedimentation in khapra beetle damaged grains (mL)	Zeleny sedimentation (Delay) in khapra beetle damaged grains (mL)	Gluten in khapra beetle damaged grains (%)	Gluten index in khapra beetle damaged grains (%)
Şahinbey	50.56 a	47.10 a	16	22	39.00	5.10
Diyarbakır-81	47.33 b	43.76 bc	16	16	43.00	4.60
Zühre	43.10 cd	39.46 ef	11	20	43.50	83.86
Artuklu	46.53 b	42.46 cd	13	18	40.00	29.95
Güney Yıldızı	42.66 d	39.83 e	19	18	40.35	61.55
Fırat-93	50.53 a	45.53 ab	11	19	41.00	37.85
Aydın-93	43.01 cd	37.96 f	15	23	48.00	6.95
Sarıçanak-98	45.60 bcd	41.26 de	16	12	44.00	3.38
Eyyübi	45.70 bc	40.55 e	14	18	33.00	86.95
Altınoprak-98	47.20 b	44.30 bc	15	21	37.00	80.90
Statistical significance of some source of variation and some descriptive characteristics						
F (Cultivars)	8.11**	22.30**				
F (Larval intensity)	1.26 ^{ns}	0.26 ^{ns}				
Grand Mean	46.21	42.22				
Standard deviation (SD)	1.73	1.07				
LSD	2.44	1.51				
CV%	3.74	2.53				

ns, not significant, *significant at P <0.05, **significant at P <0.01, difference between the means with same letter in a column is not significant.

Market price

Market price estimates of all entries with three infestation levels were received by presenting grain samples in glass jars in local commodity market on 2 February, 16 June and 4 September 2015 respectively. Table 3 shows the statistical significance of various sources of variation and the LSD groups of means of market prices (Table 3). By the end of study, khapra beetle damage was quite high and Fırat-93, Sarıçanak-98 and Altıntoprak-98 received the highest market prices with 0.28, 0.24 and 0.22 USD kg⁻¹ (0.82, 0.72 and 0.67 TL kg⁻¹), respectively. There were genuine differences among the market prices offered by local purchasers. Personal preferences of purchasers also affected market prices significantly. Larval infestation levels also affected market prices where the increasing amount of larval infestations reduced market prices significantly. Cultivar x larval infestation levels interactions were also examined and increasing amount of larval infestation resulted in lower market prices. Fırat-93 and Altıntoprak-98 were found to be the highest market price offered irrespective to larval infestation levels for all scoring dates (Table 4). Sarıçanak-98 seemed to be susceptible to khapra beetle damage initially. However, later on it recovered and was found to be less affected by khapra beetle. A rank stability analysis (Figure 2) indicated that Altıntoprak-98 and Zühre were the highest ranking with lowest SDs. Fırat-93 was also the highest-ranking cultivar with highest SD. Figure 3 shows the overall mean of market price for all entries received above given dates of study. Commodity market ceiling and base market prices for undamaged durum wheat grains are also shown.

It was evident that decreases in market price for all entries were due to khapra beetle damage rather than seasonal price fluctuations. Geometric mean of overall market price was 0.301 USD kg⁻¹ for 8-month period. This resulted in a 117 USD t⁻¹ income loss in the period (i.e., 418 USD initial market price - 301 USD average market price for 8 months). This could be even worse when market prices at the beginning and the end of study were taken into consideration with a 202 USD t⁻¹ loss (i.e., 418 USD initial market price - 206 USD average market price for 8 months). The pests of stored cereal and products are estimated a 10% weight loss annually in Turkey and khapra beetle damage dominates in the Southeastern Anatolia (Yücel, 1988; Işıkber et al., 2004). This damage to durum wheat grains caused by khapra beetle seems to be huge. In December 2016, stock wheat statistics obtained from Turkish Grain Board (TMO, 2016) and the purchasers from local commodity markets and some farmers in Şanlıurfa and neighboring provinces such as Diyarbakır, Mardin, Adıyaman and Gaziantep showed that there was a total stock of 483,000 t of durum wheat. A given amount of stock durum wheat is usually kept from harvest in June until the end of December. When the market prices go up, stored wheat is sold at the end of year. Khapra beetle management by aluminum phosphide fumigation for stored grain is normally practiced when grain is infested. However, it is reported that at least 5% khapra beetle damage always occurs irrespective to khapra beetle management between harvest in June and December. This equates to 24,150 t of wheat. Taking into account for average market price of khapra beetle damage grains in duration of this study (117 USD t⁻¹), the average income loss builds up; 2,825,550 USD (i.e., 24,150 t x 117 USD t⁻¹) for nearly 6-7 months. It could reach a maximum income loss of 4,878,300 USD (i.e., 24,150 t x 202 USD t⁻¹) when the market prices differences at the beginning and end of the study are taken into consideration. Consequently, Zühre, Fırat-93 and Altıntoprak-98 were the cultivars least affected by khapra beetle infestation for weight, market price and grain quality losses. Whereas Şahinbey, Diyarbakır-81, Artuklu, Güney Yıldızı, Aydın-93, Sarıçanak-98 and Eyyübi were moderate or susceptible to khapra beetle infestation for above given characteristics. Resistance mechanism of grains against khapra beetle must become a research focus, but control measures for stored products should not be neglected.

Table 3. Means and LSD groups of marketing prices (Krş kg⁻¹) for all varieties in various days of study

Cultivars Name	Scoring dates					
	02.02.2015		16.06.2015		04.09.2015	
Şahinbey	101.63	bc	86.79	ef	63.75	c
Diyarbakır-81	100.75	ef	87.38	cd	55.79	de
Zühre	101.71	bc	87.75	bc	64.17	c
Artuklu	101.38	bcd	87.75	bc	52.42	e
Güney Yıldızı	101.48	bcd	86.92	de	58.88	d
Fırat-93	101.33	cde	87.96	b	82.33	a
Aydın-93	100.26	g	86.20	f	52.92	e
Sarıçanak-98	100.58	ef	87.30	cde	72.42	b
Eyyübi	102.13	ab	86.83	de	43.33	f
Altıntoprak-98	102.97	a	88.71	a	66.58	c
Larval intensity						
5	101.68	a	87.84	a	62.19	a
10	101.52	a	87.68	a	61.73	a
15	101.07	b	86.58	b	60.46	a
Purchasers						
1	101.49	a	87.30	b	62.82	a
2	101.61	a	87.65	a	62.08	a
3	101.74	a	87.58	ab	59.22	b
4	100.83	b	86.92	c	61.72	a
	242.4 Krş= 1 \$USD		274.3 Krş = 1 \$USD		297.0 Krş = 1 \$USD	
Statistical significance of some sources of variation and some descriptive statistics						
F (Cultivars)	8.59**		14.21**		81.25**	
F (Larval intensity)	4.62*		44.64**		2.10 ^{ns}	
F (Purchasers)	5.71*		7.88**		4.11*	
Grand Mean	101.42		87.36		61.46	
Standard deviation (SD)	0.93		0.65		3.90	
LSD	1.10		0.77		4.60	
CV%	0.92		0.74		6.34	

ns, not significant, *significant at P <0.05, **significant at P <0.01, difference between the means with same letter in a column is not significant.

Table 4. Means (US cents kg⁻¹) and LSD groups of market price of cultivar x larval intensities interactions for all cultivars scored on various dates in local commodity market
Scoring date: 02.02.2015

Larval intensity	Cultivars Name									
	Şahinbey	Diyarbakır-81	Zühre	Artuklu	Güney Yıldızı	Fırat-93	Aydın-93	Sarıçanak-98	Eyyübi	Altıntoprak-98
5	42.02 cde	41.71 defg	42.43 abc	42.12 bcd	41.61 defgh	41.87 def	41.87 def	41.71 defg	41.35 fg	103.50 a
10	41.92 cde	41.71 defg	41.66 defgh	41.81 def	41.94 cde	41.97 cde	41.35 fghi	41.15 hi	42.43 abc	103.80 a
15	41.85 def	41.25 ghi	41.76 defg	41.50 efgh	42.00 cde	41.56 efgh	40.84 i	41.61 defgh	42.60 ab	101.62 cde

F (cultivar x larvae): 4.01**

**significant at P < 0.01, difference between the means with same letter is not significant.

Scoring date: 16.06.2015

Larval intensity	Cultivars Name									
	Şahinbey	Diyarbakır-81	Zühre	Artuklu	Güney Yıldızı	Fırat-93	Aydın-93	Sarıçanak-98	Eyyübi	Altıntoprak-98
5	31.94 cdefg	31.76 efgh	32.12 bcd	32.17 bc	31.94 cdefg	32.17 bc	31.53 hij	31.99 cdef	31.35 j	32.62 a
10	31.62 ghij	31.80 defgh	32.17 bc	32.00 cde	31.71 efghi	32.39 ab	31.80 defgh	31.99 cdef	31.94 cdefg	32.71 a
15	31.35 j	31.99 cdef	31.66 fghij	31.76 efgh	31.39 ij	31.62 ghij	30.98 k	31.48 hij	31.62 fghij	31.66 fghij

F (cultivar x larvae): 4.05**

**significant at P < 0.01, difference between the means with same letter is not significant.

Scoring date: 04.09.2015

Larval intensity	Cultivars Name									
	Şahinbey	Diyarbakır-81	Zühre	Artuklu	Güney Yıldızı	Fırat-93	Aydın-93	Sarıçanak-98	Eyyübi	Altıntoprak-98
5	22.97 g	23.52 fg	27.44 bc	19.69 hi	24.36 efg	28.40 ab	18.85 hij	26.72 bcd	13.97 n	29.46 a
10	25.58 cde	18.68 ij	20.62 h	17.34 jkl	20.62 h	29.46 a	17.92 ijk	27.98 ab	18.01 ijk	18.77 hij
15	15.82 lmn	14.14 mn	16.75 kl	15.90 lm	14.47 mn	25.29 def	18.68 ij	18.43 ijk	11.78 o	19.02 hij

F (cultivar x larvae): 47.42**

**significant at P < 0.01, difference between the means with same letter is not significant.



Figure 2. Rank stability analysis for marketing price losses of varieties under study (1. Şahinbey, 2. Diyarbakır-81, 3. Zühre, 4. Artuklu, 5. Güney Yıldızı, 6. Fırat-93, 7. Aydın-93, 8. Sarıçanak-98, 9. Eyyübi, 10. Altıntoprak-98).

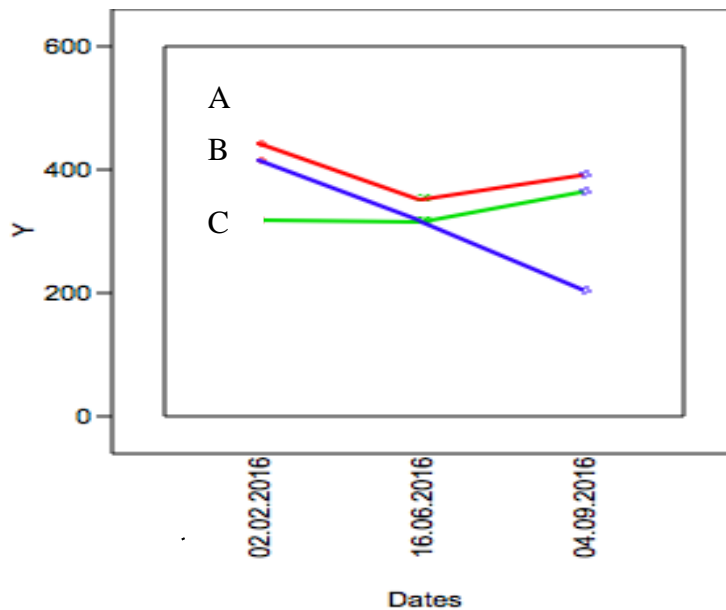


Figure 3. Local commodity market base, ceiling prices and market price of khapra beetle damaging grains.

- A: Ceiling price for undamaged grains in the commodity market (USD t⁻¹)
- B: Base price for undamaged grains in the commodity market (USD t⁻¹)
- C: Average price for khapra beetle damaging grains in the commodity market (USD t⁻¹)

Acknowledgments

Many thanks to the purchasers in local commodity market for their help in market price estimates and laboratory technician, Kübra AYIKGÖZ for laboratory analysis.

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Original article (Orijinal araştırma)

Possibilities for biological control of *Tuta absoluta* (Meyrick, 1917) (Lepidoptera: Gelechiidae) in the western Mediterranean Region of Turkey

Batı Akdeniz Bölgesi'nde *Tuta absoluta* (Meyrick, 1917) (Lepidoptera: Gelechiidae) ile biyolojik mücadele olanaklarının araştırılması

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Ali ÖZTOP²

Summary

After *Tuta absoluta* (Meyrick, 1917) (Lepidoptera: Gelechiidae) was first detected in 2009, it quickly became the dominant pest in tomato-growing areas of Turkey. Both its feeding behavior and resistance to insecticides has forced growers to use alternative control measures. This study examined the effectiveness of individual and combined use of the predatory insect, *Nesidiocoris tenuis* (Reuter, 1895) (Hemiptera: Miridae) and the egg parasitoid, *Trichogramma evanescens* Westwood, 1833 (Hymenoptera: Trichogrammatidae) as biological control measures in the western Mediterranean Region of Turkey. For this purpose, greenhouse trials were conducted in a single-crop tomato cultivation period between 2011-2012 and 2013-2014. A reduction in infested fruit of 95.1 and 94.5% was achieved, respectively, in plots where *N. tenuis* and *T. evanescens* were released together. However, no significant difference was found between the plots with *N. tenuis* alone and the combination of *N. tenuis* and *T. evanescens*.

Keywords: Biological control, *Nesidiocoris tenuis*, tomato, *Trichogramma evanescens*, *Tuta absoluta*

Özet

Domates güvesi, *Tuta absoluta* (Meyrick, 1917) (Lepidoptera: Gelechiidae)'nın Türkiye'de ilk defa 2009 yılında tespit edilmesinden sonra domates yetiştiriciliğinin yapıldığı alanlarda ana zararlı konumuna gelmiştir. Zararlıının gerek beslenme davranışı ve gerekse insektisitlere karşı dayanıklılık oluşturması üreticileri alternatif mücadele yöntemleri kullanmaya zorlamıştır. Bu çalışma ile Batı Akdeniz Bölgesinde zararlı ile biyolojik mücadelede avcı böcek *Nesidiocoris tenuis* (Reuter, 1895) (Hemiptera: Miridae) ve yumurta parazitoidi *Trichogramma evanescens* Westwood, 1833 (Hymenoptera: Trichogrammatidae)'in tek başına ve birlikte etkinliklerinin belirlenmesi hedeflenmiştir. Bu amaçla, 2011-2012 ve 2013-2014 tek ekim örtüaltı domates yetiştirme periyodunda sera denemeleri yapılmıştır. Kontrol parsellerindeki bulaşık meyve oranına göre *N. tenuis* ve *T. evanescens*' in birlikte salındığı parsellerde ilk yıl ve ikinci yıl çalışmasında sırasıyla, %95.1 ve 94.5 oranında azalma sağlamıştır. Bununla birlikte, *N. tenuis* ve *N. tenuis* + *T. evanescens* salınan parsellerde istatistiksel bir farklılık saptanmamıştır.

Anahtar sözcükler: Biyolojik mücadele, *Nesidiocoris tenuis*, domates, *Trichogramma evanescens*, *Tuta absoluta*

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Received (Alınış): 12.12.2016

Accepted (Kabul ediliş): 13.06.2017

Published Online (Çevrimiçi Yayın Tarihi): 04.07.2017

Introduction

Historically, tomato cultivation in Turkey has been seriously impacted by whiteflies, *Bemisia tabaci* (Gennadius, 1889) and *Trialeurodes vaporariorum* (Westwood, 1856) (Hemiptera: Aleyrodidae), vegetable leafminers, *Liriomyza trifolii* (Burgess, 1880) (Diptera: Agromyzidae) and spider mites, *Tetranychus* spp. (Acarina: Tetranychidae) (Tunç & Göçmen, 1994; Bulut & Göçmen, 2000; Yaşarakıncı & Hincal, 1997). Tomato leafminer, *Tuta absoluta* (Meyrick, 1917) (Lepidoptera: Gelechiidae), a native to South America, is one of the most devastating pests in Turkey and worldwide (Desneux et al., 2010, Kılıç, 2010); this species was introduced to Spain in 2006 (Urbaneja et al., 2007). Subsequently, it migrated and was soon discovered in France, Italy, Malta, the Netherlands, England, Hungary and Bulgaria. It was also found in North Africa, including Algeria, Morocco and Tunisia (Desneux et al., 2010). It was first detected in the İzmir Urla, Çanakkale and Balıkesir Provinces of Turkey in August 2009 (Kılıç, 2010) and quickly spread to all other regions in 2010, except for eastern Anatolia. This quick-proliferating pest caused significant product losses in the Mediterranean and Aegean Regions in greenhouse production.

The tomato leafminer is the main pest in open field and greenhouse tomato production. The larvae attack all growth stages of the tomato and feed on everything except for the root. When the larva hatches, it immediately begins to feed and enters the leaves and stems. The feeding process involves opening galleries between two epidermal layers in the leaves. The plant is at risk dehydration from the excess tunneling. Black excrement is left behind in the galleries of the leaf and fruit. The damaged fruit loses its market value and decomposition occurs with infection by secondary microorganisms on the opened fruit (EPPO, 2005). Chemical control of this pest is insufficient on its own and without effective control, the pest can damage up to 80-100% of greenhouse and field tomato crops (Apablaza, 1992; Lopez, 1991).

Controlling *T. absoluta* is very difficult for three reasons; 1) larvae feed in leaf tissue that is hard to access, 2) insecticide resistance has developed in some *T. absoluta* populations, and 3) its rapid proliferate rate. Yalçın et al. (2015) reported that *T. absoluta* populations obtained from the Aydın Province of Turkey had higher resistance; up to 8, 3.79 and 6.4 times for indoxacarb, metaflumizone and spinosad, respectively. Therefore, research on alternative methods for controlling this pest has become a necessity. Mass trapping with pheromone traps and a combination of light and water is one of the alternative control methods that has obtained satisfactory results, especially in low or moderate pest densities (Cocco et al., 2012; Aksoy & Kovanci, 2016). However, the prominent non-chemical control method for *T. absoluta* is biological control. Many studies have been conducted using predators, such as *Nabis pseudoferus* Remane, 1949 (Cabello et al., 2009b), *Nesidiocoris tenuis* (Reuter, 1895) (Calvo et al., 2012), and parasitoids, such as *Trichogramma achaeae* Nagaraja & Nagarkatti, 1969 (Cabello et al., 2009a; Chailleux et al., 2013), *Trichogramma euproctidis* (Girault, 1911) and *T. evanescens* (Chailleux et al., 2013). Most of these studies were conducted over a short-term period, and were laboratory or semi-field studies. However, in the western Mediterranean Region of Turkey, the vegetable-growing period lasts about 9 months from September to June, and pest and beneficial populations are greatly affected by the climatic conditions that include lower temperatures in the winter months. We examined the individual or combined use of *N. tenuis* and *T. evanescens* for biological control of *T. absoluta*. Also, we aimed to determine the performance of other potentially beneficial aspects, such as in the cold period in the winter months.

Material and Methods

Prey, predator and parasitoid rearing

Tuta absoluta rearing

The initial population of the prey, *T. absoluta*, was collected from commercial greenhouses in Antalya. Tomato plants with four to five leaves were placed in 30 x 30 x 50 cm cages with three sides covered in insect net for ventilation. Newly emerged adults were transferred to the cages. Tomato plants with *T. absoluta* eggs were kept in a climate chamber (25±1°C, 65±5% RH and 16L:8D h photoperiod).

***Trichogramma evanescens* rearing**

Trichogramma evanescens was grown on *Ephestia kuehniella* Zell., 1879 eggs in a conditioning chamber (25±1°C, 65±10% RH and 16L:8D h photoperiod). One-day old *E. kuehniella* eggs were kept for 30 min at -20°C. These eggs were then scattered evenly on damp 1-cm wide paper. The egg cards were cut into strips and placed in 16 cm x 1.5 cm glass tubes, where *Trichogramma* individuals were released for 24 h (Bulut & Kılınçer, 1987; Öztemiz, 2001).

***Nesidiocoris tenuis* rearing**

Nesidiocoris tenuis adults were collected from tomato fields in Antalya Province and cultivated on tomato seedlings in cages covered with fine netting. The cages were placed in the conditioning chambers (25±1°C, 65±10% RH and 16L:8D h photoperiod). *Ephestia kuehniella* eggs were placed in cages as prey.

First-year study (2011-2012)

Four treatments were examined; *N. tenuis* released alone, *T. evanescens* released alone, combined release of *N. tenuis* and *T. evanescens*, and control (no beneficial released).

The first-year greenhouse study was conducted over a single tomato-growing period. All plots were surrounded by fine nets before the seedlings were planted. Treatment were replicated three times in the randomized block design. Each plot was 10 m² with 20 plants. *Solanum lycopersicum* L. cv. Bestona was used in the trial and planted on 21 September 2011.

Adult tomato leafminers (1 female + 1 male per plant) taken from the stock culture were released into each cage in all plots on 17 October 2011. A total of 20 mated females were released in each treatment.

Trichogramma evanescens were released by hanging parasitoid cards. Parasitoids on the card were 6-7 days old after parasitization, and considered to be at pupal stage and about to emerge. Each release was 75 parasitoids/m², and this was repeated twice per week for a total of seven releases (Cabello et al., 2009a). Five additional releases were made, starting from 15 March in the spring of 2012 (Table 1).

Nesidiocoris tenuis was also released at 2 adults/m² following the pest release and *E. kuehniella* eggs were used as support food on the plant to establish *N. tenuis*. Beneficial insects were not released in control plots (Table 1), and no pesticides applied against any pest or diseases in all plots.

Second-year study (2013-2014)

The second-year greenhouse study was conducted as above in a single-crop tomato cultivation period. Tomato seedlings (cv. Bestona) were planted on 30 September 2013.

Additional data about releasing the predators and parasitoids for first and second years are given in Table 1.

Sampling of *Tuta absoluta*

Monitoring of the eggs and larvae of *T. absoluta* were performed weekly using five randomly selected plants in each plot. All parts of the plant were examined. The numbers of eggs and larvae of *T. absoluta* were recorded for each plant.

Sampling of the egg parasitoid, *Trichogramma evanescens*

Three leaves (from the upper, middle and bottom side of the plant) of five randomly selected plants were sampled weekly to determine the parasitism levels of *T. absoluta* eggs in each plot. Parasitism was determined by counting blackened eggs (i.e., parasitized) and transparent eggs (i.e., non-parasitized) (Ayvaz et al., 2008).

Sampling of the predator, *Nesidiocoris tenuis*

The nymph and adult stages of *N. tenuis* were separately counted at weekly intervals from five randomly selected plants in each plot. All parts of the plants were examined for the presence of these predators.

Table 1. Biological control of *Tuta absoluta* in the tomato-growing periods 2011-2012 and 2013-2014

Treatment	Release rates (m ²)	Total release (m ²)	Release frequency and dates
2011-2012 growing season			
<i>Trichogramma evanescens</i>	75	900	7 releases in autumn (17, 20, 24, 27 and 31 October, and 3 and 7 November 2011) 5 releases in spring (15,18, 24 and 28 March, and 3 April 2012)
<i>Nesidiocoris tenuis</i>	2	2	1 release (17 October 2011)
<i>Nesidiocoris tenuis</i> + <i>Trichogramma evanescens</i>	2 +75	2 + 900	1 release (17 October 2011) + 7 releases in autumn (17, 20, 24, 27 and 31 October, and 3 and 7 November 2011) 5 releases in spring (15,18, 24 and 28 March and 3 April 2012)
Control			No beneficial released
2013-2014 growing season			
<i>Trichogramma evanescens</i>	75	900	7 releases in autumn (23, 28 and 31 October, and 4, 7,11 and 14 November 2013) 5 releases in spring (11,14, 18, 21 and 25 March 2014)
<i>Nesidiocoris tenuis</i>	2	2	1 release (23 October 2013)
<i>Nesidiocoris tenuis</i> + <i>Trichogramma evanescens</i>	2+ 75	2+ 900	1 release (23 October 2013) + 7 releases in autumn (23, 28 and 31 October, and 4, 7,11 and 14 November 2013) 5 releases in spring (11,14, 18, 21 and 25 March 2014)
Control			No beneficial released

Sampling of damaged fruit

Fifty tomato fruits sampled from randomly selected plants in each plot were inspected to determine whether they were infected at harvest.

Data analyses

The number of *T. absoluta* larvae counted during the season for each plot and the number of damaged fruit were subjected to analysis of variance (ANOVA). The significance threshold for Tukey's HSD test was $P \leq 0.001$. Additionally, biological efficacy was calculated using Abbott's formula (Abbott, 1925).

Results

First-year study

The mean number of tomato leafminer eggs in the control and beneficial released plots were quite low until February, then strongly increased, especially with *T. evanescens* released alone and in the control plots. The control plots yielded the highest tomato leafminer counts at 133.8 eggs/plant on 7 May 2012. Comparable results were obtained for the larval stage of the pest. The larval population reached its highest density in the autumn on 21 November 2011 (8.5 larvae/plant in the control) and in the spring on 14 May (195.4 larvae/plant in the control) (Figures 1 & 2). *Tuta absoluta* larvae in plots with *N. tenuis* released alone, and combined *N. tenuis* and *T. evanescens* release were under 10 larvae/plant. However, in the plots with *T. evanescens* released alone, the pest population was greater than 100 larvae/plant (Figure 2).

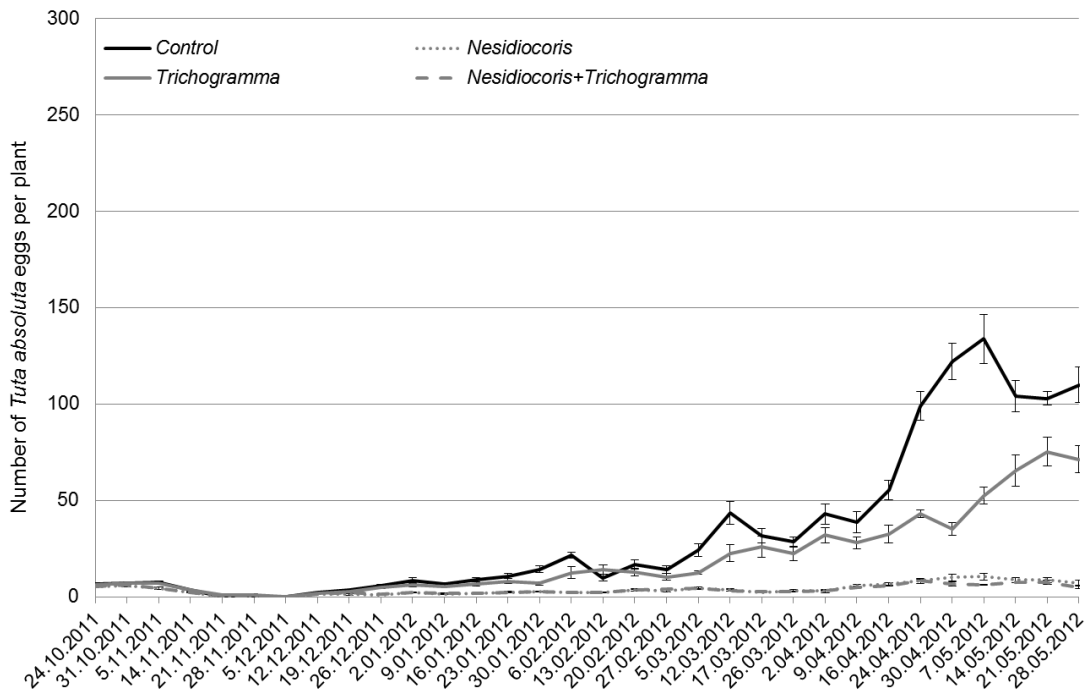


Figure 1. Egg density (mean±SE) of tomato leafminer at all treatments (control, *Nesidiocoris tenuis* and *Trichogramma evanescens* released alone, and in combination) in 2011-2012.

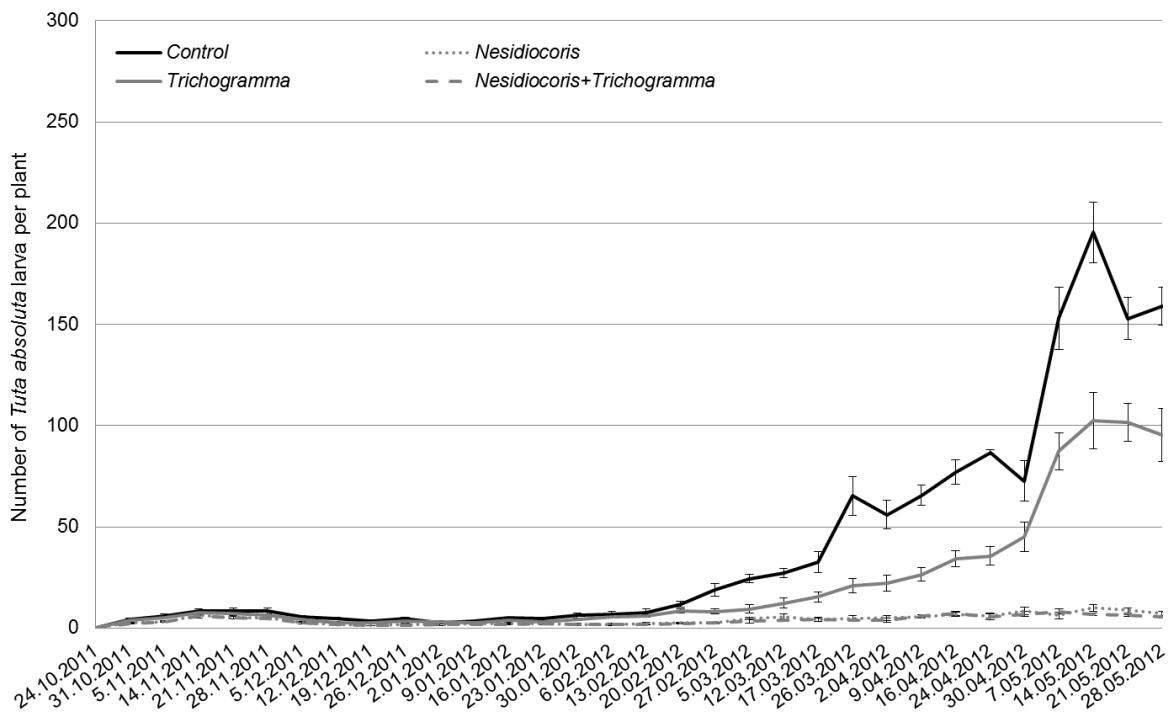


Figure 2. Tomato leafminer larvae (mean ±SE) at all treatments (control, *Nesidiocoris tenuis* and *Trichogramma evanescens* alone, and in combination) in 2011-2012.

Table 2. Total mean numbers of tomato leafminer larvae for all treatments (control, *Nesidiocoris tenuis* and *Trichogramma evanescens* alone, and in combination) in 2011-2012

Treatment	No. of tomato leafminer (mean±SE)
<i>Nesidiocoris tenuis</i>	133.7±21.4 c*
<i>Trichogramma evanescens</i>	696.6±86.0 b
<i>Nesidiocoris tenuis</i> + <i>Trichogramma evanescens</i>	118.7±13.7 c
Control	1288.7±115.8 a

*Means followed by a different letter differ significantly at P < 0.001.

The total number of tomato leafminer larva in the plots with *N. tenuis* (both alone and combined with *T. evanescens*) was significantly different from the control and individual *T. evanescens* released plots ($F_{3,11} = 121.2$; $P < 0.001$). However, the number of *T. absoluta* larvae in plots in which *N. tenuis* and *T. evanescens* were released did not show any significant difference compared to those with *N. tenuis* released alone (Table 2).

The predator, *N. tenuis*, established in plots from the date of release and there was no need for additional releases. The number of *N. tenuis* was higher than 10 adults and nymphs in plots where it was released alone and in combination with *Trichogramma* at 26 March 2012 (Figure 3). The pest population was 4.7 and 3.9 larvae/plant in plots with *N. tenuis* released alone and with *N. tenuis* and *T. evanescens* in combination, respectively, on that day. The numbers of *N. tenuis* alone and combined with *T. evanescens* released plots were almost at the same from the beginning to the end of the experiment (Figure 3).

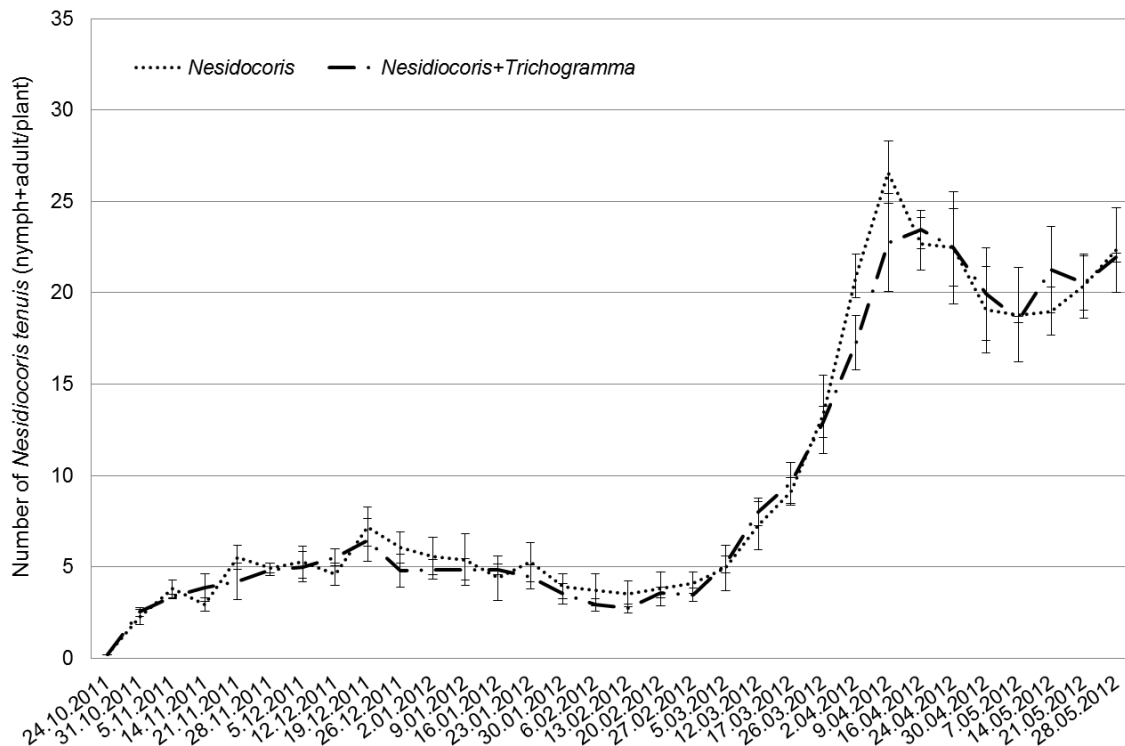


Figure 3. The population of *Nesidiocoris tenuis* (mean±SE) in two biological control treatments (*Nesidiocoris tenuis* released alone, and in combination with *Trichogramma evanescens*) in 2011-2012.

Because no parasitized eggs were detected in the plots where *T. evanescens* released at the beginning of March, five additional releases were performed. The maximum parasitization ratio was observed on 9 April 2012 (45.4%). When evaluating the greenhouse establishment of the beneficial insects after five releases *T. evanescens* in the spring, parasitoids could not be detected in the greenhouse after 7 May 2012 (Figure 4).

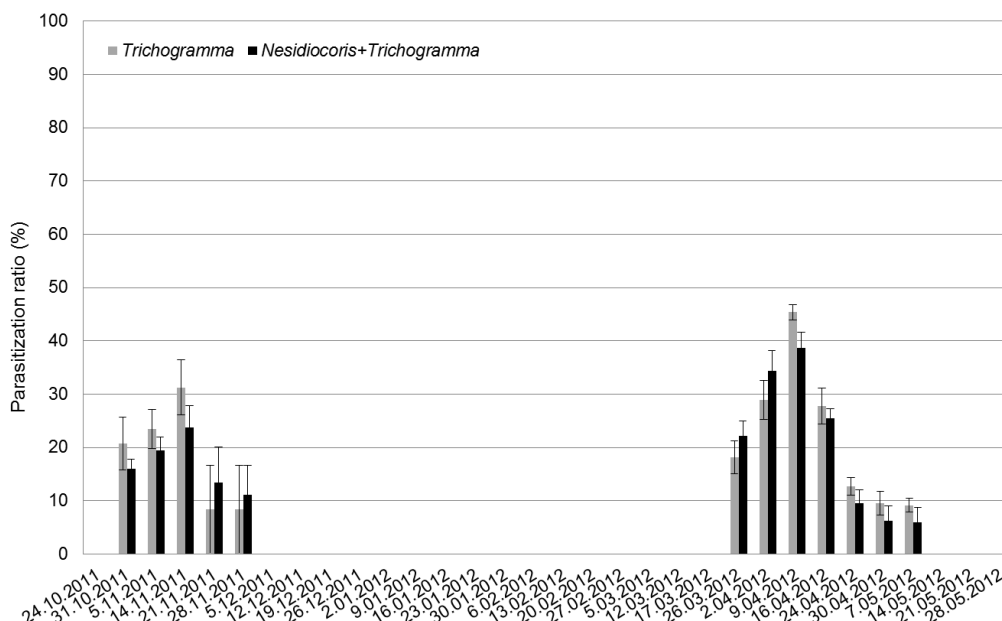


Figure 4. The parasitization ratio (mean \pm SE) of the tomato leafminer eggs in plots with *Trichogramma evanescens* released in 2011-2012.

The mean infested fruit ratios were 5.1, 36.6 and 2.5 in *N. tenuis*, *T. evanescens* and *N. tenuis plus T. evanescens* released plots, respectively and all showed a significant difference compared to the control plots ($F_{3,11} = 317.9$; $P < 0.001$). Compared to the infested fruit ratio in the control, a reduction of 90.0 and 95.1%, respectively, was achieved in the plots with *N. tenuis* released alone and in combination with *T. evanescens*, respectively. In plots with *T. evanescens* was released alone, the efficiency ratio was 28.4% (Table 3).

Table 3. Mean ratio of infested fruit (%) at harvest in the first-year plots and effects of application (corrected with Abbott's formula)

Treatment	Infested fruit ratio (% \pm SE)	Efficiency (%)
<i>Nesidiocoris tenuis</i>	5.1 \pm 0.9 c	90.0
<i>Trichogramma evanescens</i>	36.6 \pm 2.7 b	28.4
<i>Nesidiocoris tenuis</i> + <i>Trichogramma evanescens</i>	2.5 \pm 0.5 c	95.1
Control	51.1 \pm 4.1 a	-

*Means followed by a different letter differ significantly at $P < 0.001$.

Second-year study

The highest number of tomato leafminers in the control plots reached 169.9 eggs/plant on 18 March 2014 (Figure 5). From the beginning of February, the mean number of larvae population rapidly increased in the control plots. The tomato leafminer population reached its highest larval density on 1 April 2014 with 265.1 larvae/plant, again in the spring period and in control plots. On the same date, the pest population in the plots with *T. evanescens* released alone was 208.3 larvae/plant. However, in plots with *N. tenuis* released alone, the maximum pest population reached 24.7 larvae/plant (Figure 6).

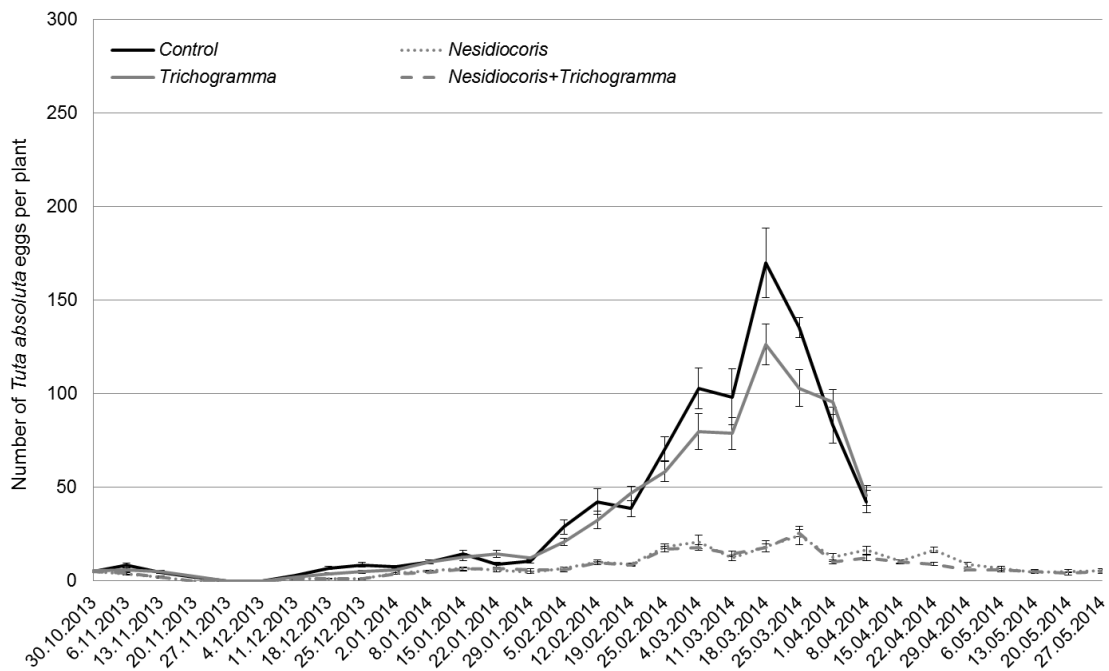


Figure 5. Egg density (mean±SE) of tomato leafminer for all treatments (control, *Nesidiocoris tenuis* and *Trichogramma evanescens* released alone, and in combination) in 2013-2014.

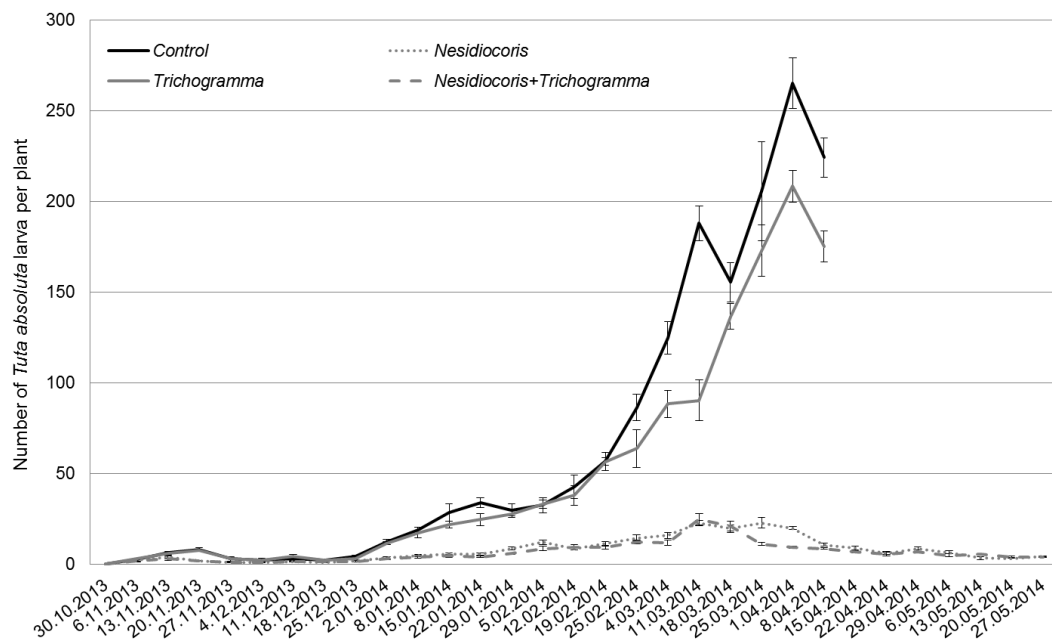


Figure 6. Tomato leafminer larvae (mean±SE) in all treatments (control, *Nesidiocoris tenuis* and *Trichogramma evanescens* released alone, and in combination) in 2013-2014.

Throughout the growing period, a mean of 1536.9 *T. absoluta* larvae per plant were counted in control plots. Although the number of tomato leafminer larvae per plant in plots with *T. evanescens* released alone was statistically different from control plots ($F_{3,11} = 232.9$; $P < 0.001$), it was still quite high and caused serious fruit loss. However, the pest population in the plots with both *N. tenuis* released alone and in combination with *T. evanescens* were 241.1 and 197.9 larvae/plant, respectively (Table 4).

Table 4. Total mean numbers of tomato leafminer larvae for all treatments (control, *Nesidiocoris tenuis* and *Trichogramma evanescens* alone, and in combination) in the second year

Treatment	No. of tomato leafminer (mean±SE)
<i>Nesidiocoris tenuis</i>	241.1±20.1 c*
<i>Trichogramma evanescens</i>	1197.6±88.9 b
<i>Nesidiocoris tenuis</i> + <i>Trichogramma evanescens</i>	197.9±17.3 c
Control	1536.9±94.6 a

*Means followed by a different letter differ significantly at $P < 0.001$.

The *N. tenuis* population was below 10 adults and nymphs until 18 March in plots with *N. tenuis* released alone and in combination with *T. evanescens*. After that date, the *N. tenuis* population remained above that level until the end of the experiment (Figure 7).

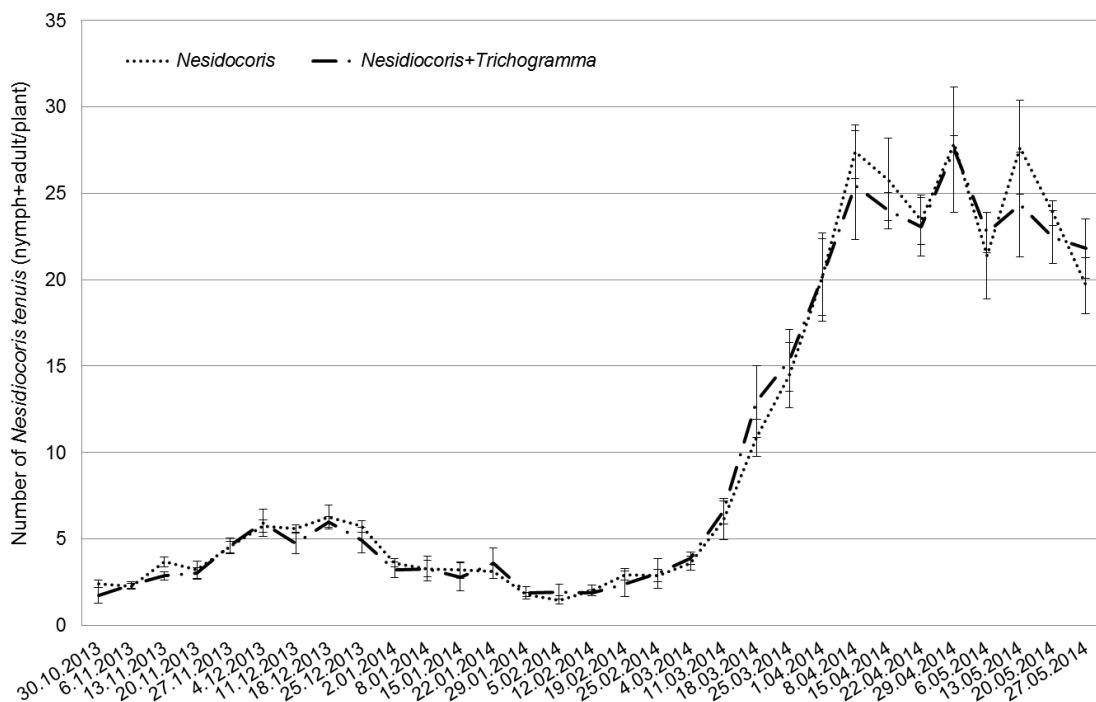


Figure 7. The population of *Nesidiocoris tenuis* (mean±SE) in two biological control treatments (*Nesidiocoris tenuis* alone, and in combination with *Trichogramma evanescens*) in 2013-2014.

In the second-year study, *T. evanescens* releases were made from March onwards. The maximum parasitization ratio was 30.5% on 1 April 2014 in the plot with *T. evanescens* released alone (Figure 8). In the plots with *T. evanescens* released alone, the intensity of tomato leafminers reached 208.3 larvae/plant on 8 April 2014. On that date, in both control plots and plots with *T. evanescens* released alone the damage ratio was 100%.

The damaged tomato fruit number in *N. tenuis* released plots (both alone and combined with *T. evanescens*) showed a significant difference compared to control plots and plots with *T. evanescens* released alone ($F_{3,11} = 179.8$; $P < 0.001$). When *N. tenuis* was released alone, the damage ratio in fruit was 5.1 and 4.3% in the first and second years, respectively. When *N. tenuis* was released in combination with *T. evanescens*, this ratio dropped to 2.5 and 2.9%, respectively. The two beneficial insects together or *N. tenuis* alone did not result in any statistically significant effect on the damage ratio (Tables 3 & 5).

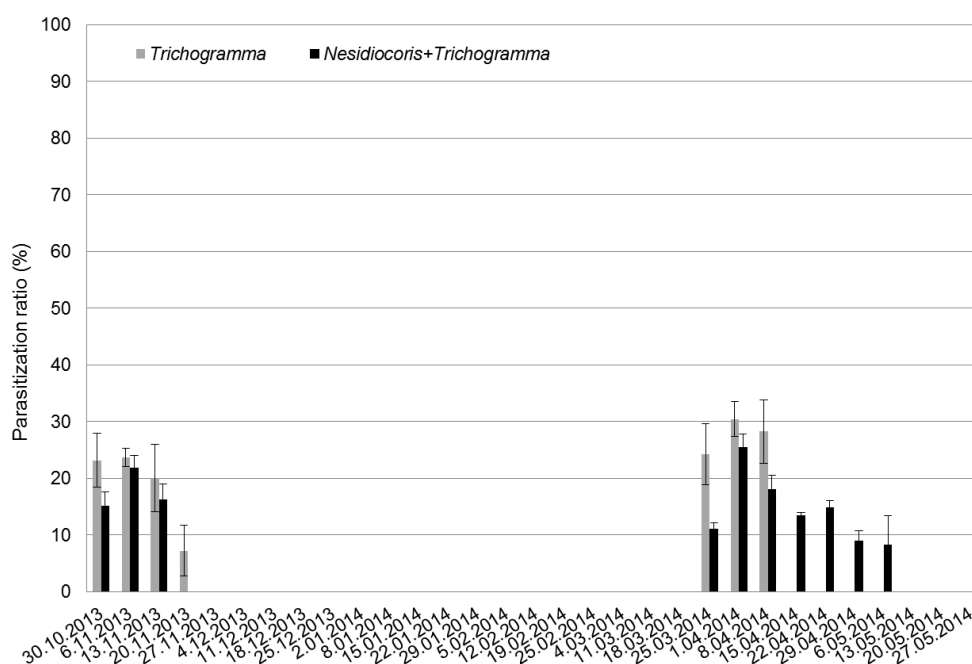


Figure 8. The parasitization ratio (mean±SE) of the tomato leafminer eggs in plots with *Trichogramma evanescens* released in 2013-2014.

Table 5. Mean ratio of infested fruit (%) at harvest in the second-year plots and effects of application (corrected with Abbott's formula)

Treatment	Infested fruit ratio (%±SE)	Efficiency (%)
<i>Nesidiocoris tenuis</i>	4.3±0.4 b*	91.8
<i>Trichogramma evanescens</i>	44.2±2.7 a	16.0
<i>Nesidiocoris tenuis</i> + <i>Trichogramma evanescens</i>	2.9±0.3 b	94.5
Control	52.6±4.7 a	-

*Means followed by a different letter differ significantly at $P < 0.001$.

Discussion

This study aimed to determine the individual and collective effectiveness of the predator insect, *N. tenuis*, and the egg parasitoid, *T. evanescens*, against tomato leafminers, an invasive pest that recently expanded into Turkey.

Given that *T. evanescens* was undetectable in both years at the beginning of March, five additional releases were performed. In the plots with *T. evanescens* released alone, 28.4 and 16.0% reductions in damaged fruit number were obtained compared to the control plots in first and second year study respectively. In the second-year study, the damage ratio in the control plot and the plots testing *T. evanescens* reached 100% in April towards to end of the growing season. Many studies have been conducted to determine the effectiveness of *Trichogramma* species against the tomato leafminer. Domingues et al. (2003) reported a damage ratio in fruit of 13% with *Trichogramma pretiosum* Riley, 1879 release. In a study with another species, *T. achaeae* reduced damage by 92% when used in rate of 75 adult/m² in 3-4 days intervals in August and September (Cabello et al., 2009a). This may explain the differences in egg parasitoid species.

Another key factor for this failure was the climatic conditions. The mean temperatures in the two years were 19.6 and 21.4°C, respectively, during the additional release of *T. evanescens* in the spring. However, these temperatures were not appropriate for *T. evanescens*. The adaptation of *T. evanescens* to the climate, or tomato plant and its cultivars, may be a reason for the failure of its efficiency. Contrary to previous studies, *T. evanescens* was not able to increase its population (Chailleux et al., 2013).

In a study of the predatory insect, *N. tenuis*, the ratio of *T. absoluta* contamination was reduced by 97% in the leaves and 100% in the fruit (Mollá et al., 2009). Similar results were obtained for *Nabis pseudoferus* and the eggs of tomato leafminers were reduced by 92-96% (Cabello et al., 2009b). In the present study, *N. tenuis* reduced fruit damage by more than 90%.

During the study, the mean temperature between December and February was 12.3°C in 2011-2012 and 14.2°C in 2013-2014. The lower development threshold of tomato leafminers is 8°C (Barrientos et al., 1998), while this value is 10.3°C for *N. tenuis* (Sanchez et al., 2009). Furthermore, the weather conditions were more moderate, especially in the second year of the study, which proved advantageous for the tomato leafminers that have a lower development threshold. Remarkable damage on fruit was observed in plots where *N. tenuis* was released in March. Therefore, it is believed that additional releases would be appropriate in the spring, depending on the pest intensity.

Calvo et al. (2012) recommended that applications of *N. tenuis* with *T. achaeae*, and with *N. tenuis*, *T. achaeae* and *Bacillus thuringiensis* have no additional positive effect on *T. absoluta* compared to releasing *N. tenuis* alone. In addition, offspring of *Trichogramma* developed on *T. absoluta* eggs displayed low parasitism and poor biological traits. For instance, some biological characteristics, such as healthy wing occurrence after emergence or adult longevity, were negatively affected compared to the controls reared on the cultured host. Therefore, the parasitoids that developed on *T. absoluta* had little success controlling it (Chailleux et al., 2013). In addition, Chailleux et al. (2013) stated that the combination of *Macrolophus pygmaeus* (Rambur, 1839) and egg parasitoids (*T. achaeae* or *T. euproctidis*) indirectly reduced their effectiveness against *T. absoluta* because the predator insect consumed the parasitoid eggs together with non-parasitoid eggs. However, Oztemiz et al. (2012) reported that the combination of *N. tenuis* and *T. evanescens* has the potential to control *T. absoluta*. This difference can be associated with the effect of host plant cultivar, climatic conditions and time of release.

In plots where *T. evanescens* and *N. tenuis* were released together to control tomato leafminers, no statistically significant benefit was observed with the addition of the egg parasitoid *T. evanescens*. Considering these findings, *N. tenuis* can be effectively used alone to control tomato leafminers.

Acknowledgments

This research was supported by the Turkish Ministry of Food, Agriculture and Livestock, the General Directorate of Agricultural Research and Policy and Bati Akdeniz Agricultural Research Institute. The authors wish to thank Christina HODDLE (University of California, Riverside Department of Entomology, CA, United States) for his critical review of the manuscript.

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Original article (Orijinal araştırma)

**Some biological parameters of *Orius niger* (Wolff, 1811)
(Hemiptera: Anthocoridae) under outdoor conditions in Turkey**

Orius niger (Wolff, 1811) (Hemiptera: Anthocoridae)'in dış koşullarda bazı biyolojik özellikleri

Serkan PEHLİVAN¹

Ekrem ATAKAN^{1*}

Summary

Overwinter biology of *Orius niger* (Wolff, 1811) (Hemiptera: Anthocoridae) was studied during 2014 and 2015, in Adana Province, Turkey. Outdoor experiments were performed at the Department of Plant Protection, Faculty of Agriculture, University of Çukurova on seven different dates between October and April. The predatory bug, *O. niger*, overwinters in Adana Province as adults. Some biological parameters of the *O. niger* were investigated under outdoor conditions at monthly intervals from autumn to spring with cotton seedlings placed in vials with distilled water. Sterilized eggs of the Mediterranean flour moth, *Ephestia kuehniella* Zeller, 1879 (Lepidoptera: Pyralidae), were provided to *Orius* as food. All eggs of *O. niger* hatched even at the low temperatures in winter. Most of the experimental females laid eggs. Total mean numbers of eggs laid by the females was the highest in April (83.55±15.60 eggs/female) and the lowest in December (7.71±1.62 eggs/female). Duration of oviposition of the experimental females was nearly 30 days in October, November and April, but less than 30 days in other winter months. The proportion of non-reproductive females in December-February ranged from 25% to 40%. Longevity of females was nearly a month in winter and they did not survive until spring. While sex ratios (male/female) ranged from 1:2.5 to 1:3 in October-January, the ratio was 1:1 in April. Duration of nymph development was the highest (45 days) in January and the shortest (nearly 18 days) in April. Furthermore, most of the first instars of nymphs died in a short time due to cold winter days in December-February, and nearly 28% total nymphs matured to adults. The experimental results support that *O. niger* can severely decrease the population sizes of pest species in late spring (after April) in countries of the southern zone.

Keywords: Adana, Anthocoridae, biology, *Orius niger*, Turkey

Özet

Orius niger (Wolff, 1811) (Hemiptera: Anthocoridae)'in kışlama biyolojisi Adana'da 2014 ve 2015 yıllarında araştırılmıştır. Dış koşullardaki denemeler Ekim-Nisan periyodunda Çukurova Üniversitesi, Ziraat Fakültesi, Bitki Koruma Bölümü'nde 7 farklı tarihte yürütülmüştür. Avcı böcek *O. niger* Adana'da ergin olarak kışı geçirmektedir. *Orius niger*'in bazı biyolojik özellikleri Sonbahar-İlkbahar periyodunda dış koşullarda incelenmiştir. *O. niger*'e besin olarak sterilize edilmiş Akdeniz un güvesi, *Ephestia kuehniella* Zeller, 1879 (Lepidoptera: Pyralidae) yumurtaları verilmiştir. Kışın çok düşük sıcaklıklarda bile *O. niger*'in yumurtaları açılmıştır. Tüm denemelerdeki dişilerin birçoğu yumurta bırakmıştır. Dişilerin bıraktığı ortalama toplam yumurta sayıları Nisan ayında en yüksek (83.55±15.60/dişi) ve Aralık ayında ise en düşük (7.71±1.62/ dişi) olmuştur. Ekim, Kasım ve Nisan aylarında dişilerin ovipozisyon süresi 30 gün civarında olurken, diğer kış aylarında bu süre 30 günden az olmuştur. Yumurta bırakmayan dişilerin oranı ise Aralık-Şubat döneminde %25-%40 arasındadır. Dişilerin ömür süreleri kışın yaklaşık olarak bir aydır ve dişiler ilkbahar dönemine ulaşamamıştır. Ekim-Ocak ayları arasında cinsiyet oranları (dişi/erkek) 1:2.5'ten 1:3'e kadar değişirken, bu oran Nisan ayında 1:1 olmuştur. Nimf gelişme süreleri Ocak ayında en uzun (45 gün) olurken Nisan ayında en kısa (18 gün) olarak belirlenmiştir. Aralık-Şubat dönemindeki soğuk kış günlerinde birinci dönem nimflerin birçoğu kısa sürede ölürken, yaklaşık %28'i ergin döneme ulaşmıştır. Bu denemenin sonuçları *O. niger*'in güney bölgelerde nisan ayından sonra zararlı türlerin popülasyonlarını azaltabileceğini desteklemektedir.

Anahtar sözcükler: Adana, Anthocoridae, biyoloji, *Orius niger*, Türkiye

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Received (Alınış): 31.01.2017

Accepted (Kabul ediliş): 22.06.2017

Published Online (Çevrimiçi Yayın Tarihi): 03.07.2017

Introduction

The Order Hemiptera is well known for containing important predators of agricultural pests including acari (Acarina) and thrips (Thripidae), as well as other soft bodies pest species (Whitcomb & Bell, 1964). Commonly they are used in integrated pest management programs (IPM) with their high consumption and searching capabilities on their host, their easy mass rearing and quick adaptability to laboratory conditions (Ramakers & van den Meiracker, 1991; Van de Veire & Degheele, 1992; Jacopson, 1993).

The western flower thrips, *Frankliniella occidentalis* Pergande, 1895 (Thysanoptera: Thripidae), is regarded as a very destructive pest of agricultural crops worldwide (Kirk & Terry, 2003). This species was recorded for the first time in Turkey in 1993 (Tunç & Göçmen, 1994). In general, insecticide treatments are not efficient on thrips, because of their cryptic behavior, such as eggs being in plant tissues, and prepupa and pupa mostly in soil. Moreover, resistant population can appear because of high number of chemicals (insecticide) used worldwide (Immaraju et al., 1992; Brødsgaard, 1994; Dağlı & Tunç, 2007). Therefore, there is special attention given to biological control agents belonging to the family Anthocoridae (Hemiptera) to use against pest thrips in agricultural habitats all over the world. *Orius* species are important as biological control agents in controlling of pest thrips species on vegetables grown in tunnels or greenhouses (Schreuder & Ramakers, 1989; Trottin-Caudal et al., 1991; Tavella et al., 1991; Villeveille & Millot, 1991; Van de Veire & Degheele, 1992; Jacopson, 1993). They also feed upon whiteflies (Hemiptera: Aleyrodidae), aphids (Aphididae) and spider mites (Acarina: Tetranychidae) (Riudavets, 1995). These predator species were previously found on different pest species on cotton in the Çukurova Region, Turkey (Ghavami & Özgür, 1992) and they have an important role in suppressing thrips species in cotton fields in Turkey (Atakan, 2006; Atakan & Gencer, 2008) as well as thrips species on vegetables (Atakan, 2007a) and fruit trees (Atakan, 2007b). Also, *Orius niger* (Wolff, 1811) has been commonly found in Turkey (Önder, 1982) and it is widely spread over the Mediterranean basin and is present under natural conditions throughout the year in the Çukurova Region (Atakan & Tunç, 2010). *Orius niger* and *Orius minutus* (L.) have effectively controlled the two-spotted red spider mite and onion thrips in potato fields in the Ardabil region of Iran (Fathi, 2009).

There are several factors that can influence efficiency of *Orius* species in IPM programs. The first factor is their behavior in winter (Ramakers & van den Meiracker, 1991; Gillespie & Quiring, 1993). Temperature and day length are important factors which affect biological control of thrips by *Orius* species, while *F. occidentalis* is well known as a very harmful greenhouse pest thrips, which does not enter reproductive diapause (Kirk & Terry, 2003). It has numerous generations under warm greenhouse conditions, while local or imported strains of some *Orius* species exhibit a diapause, which negatively effects their predation on pest thrips. However, the Antalya strain of *O. niger* (Bahşi & Tunç, 2008) and southern strain of *Orius laevigatus* (Fieber, 1860) from Sicily (Tommasini & van Lanteren, 2003) do not enter reproductive diapause under the short-day-length conditions in the laboratory. However, outdoor conditions are quite variable compared to laboratory conditions. Changeable outdoor conditions (mainly meteorological conditions) would affect winter biology of the predatory bug, i.e., diapause, and would have an impact on its potential predatory capacity in controlling pest insects including thrips in crop and wild plants in open fields and in protected areas.

The purpose of this study was to provide data on overwintering status of *O. niger* under outdoor conditions in Adana, Turkey, with emphasis on its potential usage in IPM programs including thrips pest management in agricultural crops. We investigated how different dates, i.e., months, influence biology of *O. niger*. In addition, we also evaluated whether different day lengths and temperatures at different times from winter to spring affected reproductive behavior of the predator bug. Additionally, investigation of overwintering biology of this predatory bug may help to predict predatory activity of *O. niger* on insect prey in agricultural areas in the following season.

Material and Methods

Host plant source

Cotton (*Gossypium hirsutum* L.) (Malvaceae) cv. SG125 was used in this study. Cotton plants grown in pods in the chambers at 25°C and 60% RH. The cotton seedlings were used for the experiments when they were in two-true leaf stage.

Insect material

Colonies of *Ephestia kuehniella* Zeller, 1879 was supplied by the Biological Control Research Institute of Adana and was reared in the laboratory for two years.

Orius niger was obtained from the cotton flowers in Adana, Turkey. *Orius* individuals were reared in the climatic chamber at 25°C, 60% RH and 16L:8D h photoperiod. Plastic cups (0.5 l capacity) with two ventilation windows covered with muslin were used as rearing units. Fresh bean pods were used as oviposition substrate. *Ephestia kuehniella* eggs were sterilized for 2 h under UV light, then glued on blue cards for *O. niger* feeding. Given of the cannibalism of *O. niger*, adults were put in other rearing cages. The rearing units were used to determine egg hatching and nymphal development. Upper side of the 500-ml plastic containers were cut and covered with fine muslin. Thirty-ml capacity glass vial was fixed onto bottom side and cotton seedling with two cotyledon leaves were immersed in water in the glass vial (7.6 x 2.5 cm).

Experimental design

Experiments were conducted on seven different dates between October and April. The plastic containers with cotton seedlings and insects were randomly placed outdoors. The plastic containers including insects were placed on a platform 3 m above ground outside a south-facing laboratory window. The platform, 90 cm in diameter, was made from concrete.

Meteorological conditions at the experiment site

The experiments were conducted in the Department of Plant Protection, Faculty of Agriculture, University of Çukurova. Experiments were performed under outdoor conditions. The mean, maximum and minimum temperatures and relative humidity were obtained from the Meteorological Station of Department of Agricultural Buildings and Irrigation (about 5 km from the experimental site).

Duration of egg development

Five pairs (10 replicates) of females (in oviposition period) and males of *O. niger* from the laboratory culture were caged in a plastic container and were kept for 24 h in the climatic chamber with the conditions 25°C, 60% RH and 16L:8D h photoperiod. The bugs were released in the cage which contained sterilized eggs of *E. kuehniella* and *Typha* pollen (supplied by the Biological Control Research Institute of Adana, Turkey) on cotyledons of cotton plants. After 24 h, *O. niger* eggs laid into the cotyledons were counted in laboratory and 10 eggs were left in each plant cage. The plastic containers were placed outdoors at each experimental date.

Hatching of eggs inserted into the tissues of caged cotton seedlings was assessed daily under a stereo microscope in the laboratory and recorded. The experiments started on 14 October, 5 November, 4 December, 1 January, 3 February, 3 March and 20 April.

Nymphal survival rate

Newly hatched five nymphs from the egg development experiments under the outdoor conditions, were kept in a plastic container and provided with sterilized eggs of *E. kuehniella* and also *Typha* pollens. Experiment was replicated four times (20 nymphs in total). Nymph development was observed twice a week and emerged adults were sexed. The number of emerged adults in relation to the initial number of eggs was calculated from data of development duration experiments.

The experiments started on 20 October, 17 November, 12 December, 13 January, 9 February, 17 March and 25 April.

Preoviposition, oviposition, postoviposition and fecundity of females

We modified the method of Chyzik et al. (1995) for these studies. Ten pairs of one-day old bugs obtained from the laboratory stock culture were used. One unmated female and a male were taken from the *O. niger* rearing chambers and were kept in a plastic container with cotton seedlings immersed in water in a vial. *Ephestia* sterilized eggs and pollens were added. The plastic containers were placed on the same platform. Cotton seedlings and prey were changed every 2 or 3 days. The numbers of eggs laid into plant tissues by the females were counted under a stereo microscope (40x magnification).

The experiments started on 13 October, 3 November, 3 December, 5 January, 2 February, 9 March and 20 April.

Longevity

Male and female longevity were determined from the individuals used in fecundity test.

Data analysis

Biological data on eggs hatching, preoviposition, oviposition, postoviposition, eggs numbers and nymph development and adult life span according to experimental days were compared by using Tukey's honestly significant difference (HSD) test at $P < 0.05$ significant level. SPSS statistical package program was used (SPSS, 2006).

Results

Duration of egg development

Number of laid eggs ranged between 43 (14 October) and 303 (3 February) eggs/female across the different experiment dates. The duration of first hatching of eggs was similar in experiments in October and November and hatching period lasted 5.70 ± 0.89 and 6.75 ± 0.64 days after first hatching (Table 1). The first hatching occurred 10 days later in December and January experiments. After January, duration of the first hatching was significantly shorter than those in December and January ($F_{(6,43)} = 15.442$, $P < 0.0001$) than other sampling months. Hatching rate was determined as 62.80% in October and 76.78% in November when the eggs hatched within 10 days (Table 1). This rate was 94.01% in experiment of January. Ratio of hatching of eggs within first 5 days in April experiment was 34.31%.

Nymph development period and mortality rate

Total nymphal period was the shortest (17.50 ± 0.18 days) in April ($F_{(6,40)} = 43.447$, $P < 0.0001$) and this was followed by nymphs of September experiment (20.07 ± 1.10 days) (Table 2). Duration of nymph development coming from the experiments of November, December and January were 32.51 ± 0.50 , 37.71 ± 1.74 and 45.00 ± 0.40 days, respectively. Duration of nymph development was the highest at 45.00 ± 0.40 days in January and the shortest at 17.50 ± 0.18 days in April.

Mean temperature ranged from 19 to 23°C and relative humidity was nearly 70% in April (Figure 1). Nymph development time was closely related to temperature and humidity. Mean temperature varied between 5 and 13°C in December-February and dropped to 3°C in the first week of January. Out of 54 nymphs, a total 189 nymphs matured to adults. Mortality rate of nymphs in November-February was over 70%. Sixty percent of nymphs in the October experiment and 44% in the March experiment matured to adults. Numbers of the emerged females were at least two-fold of numbers of the males in October-January (Table 2). After the February experiment, the sex ratio was nearly equal.

Table 1. Duration of *Orius niger* egg development under the outdoor conditions

Experiment date	No of total eggs	^a Mean time±SEM of first hatching of eggs (day)	Hatching rates of eggs (%) according to day intervals		
			0-5 days	6-10 days	>10 days
14 October	43	5.70±0.89 bc (4-10) ^b	4.65	32.55	62.80
5 November	100	6.75±0.64 ab (5-10)	4.05	41.41	54.54
4 December	81	9.66±0.74 a (8-15)	0.00	23.22	76.78
1 January	218	9.87±0.79 a (8-14)	0.00	5.96	94.04
3 February	303	3.75±0.36 bc (2-5)	9.25	64.64	26.07
3 March	150	4.20±0.80 bc (2-7)	10.00	58.00	32.00
20 April	102	2.60±0.76 c (1-5)	34.31	65.68	0.00

^a means with same letter in the same column are not statistically significant according to Tukey's HSD test ($P < 0.05$); ^b parenthesis indicates minimum and maximum values.

Table 2. Total development time of *Orius niger* nymphs under outdoor conditions

Experiment date	n	No of nymphs became adults	No of death nymphs	Mortality of nymphs (%)	^a Mean duration±SEM of total nymph (day)	Sex ratio (Male/female)
15 October	25	13	12	48.00	20.07±1.13c (16-29) ^b	1:3
17 November	25	7	18	72.00	32.51±0.5b (32-33)	1:2.5
12 December	24	2	22	91.67	37.71±1.74a (31-43)	-
13 January	30	4	26	86.67	45.00±0.40a (44-46)	1:3
9 February	30	5	25	83.33	39.00±1.34a (36-42)	1:1.5
17 March	25	9	14	56.00	31.62±2.23b (23-42)	1:1.25
25 April	30	14	16	53.00	17.5±0.18c (17-18)	1:1

^a means with same letter in the same column are not statistically significant according to Tukey's HSD test ($P < 0.05$); ^b parenthesis indicates minimum and maximum values.

Preoviposition, oviposition, postoviposition and productivity of females

Preoviposition period in the October experiment was significantly shorter at 4.33 days than those in the other experiments ($F_{(6,42)} = 11.660, P < 0.0001$) but increased to 9.33 ± 0.76 and 11.71 ± 1.65 days in the November and December experiments, respectively (Table 3). Preoviposition period for the March and April experiments lasted 7.42 ± 1.63 and 8.72 ± 1.46 days, respectively. Oviposition time in the October, November, March and April experiments were significantly greater than those in the winter experiments ($F_{(6,42)} = 6.712, P < 0.0001$) and nearly lasted 30 days. Oviposition was significantly less in the winter experiments ($P < 0.0001$). For example, female oviposited was 11.85 and 18.50 days in the December and January experiments, respectively. Females laid significantly more eggs in October and in April ($F_{(6,42)} = 4.669, P < 0.0001$). Postoviposition in the autumn experiments was significantly longer than in the other experiments, and lasted nearly 10 days.

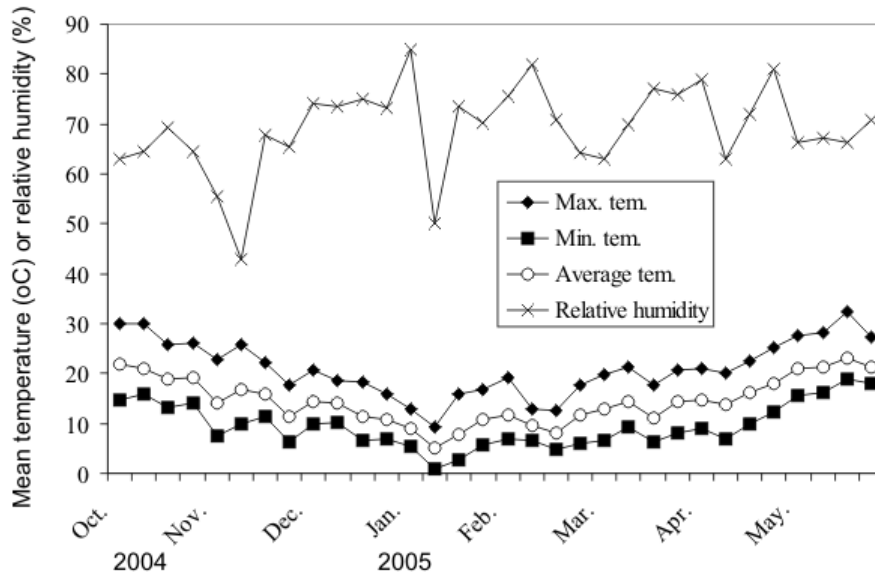


Figure 1. Average, maximum and minimum temperature and relative humidity, October 2014-May 2015 at Balcalı, Adana, Turkey.

Table 3. Duration of preoviposition, oviposition and postoviposition of *Orius niger* females and fecundity under outdoor conditions

Experiment date	n	^a Preoviposition time (day)	Oviposition time (day)	Postoviposition time (day)	Eggs laid (egg/female)
13 October	9	4.33±0.33 c (4-6) ^b	30.33±4.45a (14-48)	7.44±1.66 a (2-18)	58.22±9.14 b (34-109)
3 November	9	9.33±0.76 b (5-12)	28.60±8.31a (4-87)	10.22±4.62 a (2-37)	27.55±9.11 c (8-95)
3 December	9	11.71±1.65 b (7-19)	11.85±3.26b (2-29)	5.28±1.30 b (2-12)	7.71±1.62 e (2-12)
5 January	10	19.25±3.83 a (8-21)	18.50±8.42b (3-35)	4.25±1.03 b (2-14)	18.50±7.18 d (4-36)
2 February	10	13.35±1.21 ab (5-16)	18.50±3.61b (5-32)	4.16±0.74 b (3-6)	15.16±2.41 de (9-26)
9 March	9	7.42±1.63 b (2-14)	25.75±7.56a (3-59)	4.57±0.81 b (2-7)	48.83±12.63 b (22-115)
20 April	7	8.42±1.46 b (4-16)	28.71±4.08a (13-41)	3.42±0.48 b (2-5)	83.85±15.60 a (35-135)

^a means with same letter in the same column are not statistically significant according to Tukey's HSD test ($P < 0.05$); ^b parenthesis indicates minimum and maximum values.

Eggs numbers were significantly greater, with mean of 83.65 ± 15.60 eggs/female, in the April experiment ($F_{(6,42)} = 6.557$, $P < 0.0001$). Mean numbers of eggs laid by females for first 10 days in October and April were similar at 23.3 and 21.30 eggs/female, respectively, but these numbers were significant different from those in the other experiments ($F_{(6,63)} = 9.247$, $P < 0.0001$). Mean numbers of eggs laid by female for first 10 days in the January experiment was 3.10 eggs/female (Table 4). In April, females laid significantly more eggs (50.20 eggs) in 11-30 day period ($F_{(6,63)} = 13.792$, $P < 0.0001$). Mean numbers of eggs laid for more than 31 days were similar, excluding the December experiment, and less even in the October and April experiments. All females laid eggs in first 10 days and 11-30 days in the October and April experiments, respectively. Forty percent of females tested in January and February laid eggs in first 10 days. Only one female oviposited for 31 days (Table 4). Thirty percent of females in December, 40% in January and 25% February did not lay egg. In the other experiments, all females oviposited.

Table 4. Fecundity of *Orius niger* females under outdoor conditions

Experiment date	^a Mean numbers of eggs laid according to day intervals (egg/female)			Rate of ovipositing females (%) according to day intervals		
	0-10	11-30	>31	0-10	11-30	>31
13 October	23.30 \pm 5.18 a	25.0 \pm 4.56 b	6.00 \pm 2.96 a	100	100	30
3 November	11.40 \pm 2.23 b	6.00 \pm 2.04 c	8.60 \pm 5.98 a	90	60	40
3 December	3.90 \pm 1.43 c	1.50 \pm 0.96 c	-	70	30	0
5 January	3.10 \pm 1.36 c	3.20 \pm 2.15 c	1.10 \pm 1.10 b	40	20	10
2 February	4.80 \pm 1.94 c	3.80 \pm 2.04 c	0.50 \pm 0.50 b	40	40	10
9 March	9.10 \pm 2.16 b	14.20 \pm 6.34 b	8.00 \pm 5.17 a	90	60	30
20 April	21.30 \pm 2.78 a	50.20 \pm 9.12 a	9.10 \pm 5.35 a	100	100	40

^a means with same letter in the same column are not statistically significant according to Tukey's HSD test ($P < 0.005$).

Longevity

Female longevity was nearly 45 days in April but was significantly longer at 55 days in November ($F_{(6,54)} = 4.677$, $P < 0.0001$; Table 5). Only one female survived for nearly 100 days in the November experiment. Some females from the November experiment lived for 2 months or slightly longer. Mean longevity of females of December, January and February were less than 30 days. Female life spans in March and April lasted slightly over 30 days (March 34.50 ± 7.38 days; April 44.74 ± 4.21 days). Mean longevity of the males in all experimental months were shorter than those found for females (Table 5). Males assessed in the October and November experiments survived for 30.62 ± 8.82 and 39.44 ± 6.16 days, respectively. Mean longevity of males in December and February were similar, and these males lived for 21 days. Longevity of males approached to nearly 30 days in March and April.

Table 5. Longevity of *Orius niger* adults under outdoor conditions

Experiment date	^a Longevity (day)					
	Female			Male		
	Mean±SEM	Max.	Min.	Mean±SEM	Max.	Min.
13 October	39.77±5.19 b	56	18	30.62±8.82 a	46	13
3 November	55.00±0.10 a	98	16	39.44±6.16 a	67	9
3 December	28.50±4.44 c	56	14	21.80±3.65 a	38	10
5 January	25.20±6.67 c	61	6	21.00±3.55 a	41	6
2 February	20.25±4.62 c	39	3	21.12±5.79 a	51	4
9 March	34.50±7.38 b	57	6	27.00±7.98 a	57	16
20 April	44.74±4.21 b	63	30	29.71±9.52 a	50	4

^a means with same letter in the same column are not statistically significant according to Tukey's HSD test ($P < 0.005$).

Discussion

Winter in Southeastern Mediterranean region of Turkey is mild and rainy. All eggs laid by the females easily hatched during the outdoor experiments. Low rate of nymphs (nearly 20%) survival was recorded in the winter (December-February). Durations of nymph development were nearly 20 days in autumn (October) and spring time but increased up to 30 days or slightly over 30 days in the winter. Mean temperature in October ranged from 19 to 22°C, and 13 to 18°C in April but varied between 5 to 13°C in the winter (Figure 1). Duration of total nymph development of *O. niger*, when temperature increased to over 20°C under the controlled conditions, were less than 20 days. For example, Tommasini & Nicoli (1996) reported that total duration of nymph development of *O. niger* provided with eggs of *E. kuehniella* and *F. occidentalis* as food were 12.9 and 11 days, respectively, at 26°C and 80±5% RH. Duration of total nymphal stage ranged from 16 to 18.8 days when nymphs of *O. niger* were exposed to 26°C and to various day lengths (9-16 h) (Bahşi & Tunç, 2008). However, duration of nymphal instars of *O. niger* provided with nymphs of *Bemisia tabaci* and *Tetranychus cinnabarinus* was 21.5 and 21.8 days, respectively, under the laboratory conditions with 25±1°C and 65±10% RH (Efe & Çakmak, 2013).

Nymphal mortality was particularly higher in the winter and only 28% nymphs of a total 189 nymphs matured to adults. Similarly, Chyzik et al. (1995) found that survival rate of nymphs of *Orius albidipennis* (Reuter, 1884) in the winter in Israel was less than 20%. Females of *O. niger* were found throughout autumn to spring in faba bean (*Vicia faba* L.) fields in the Adana Region, but no *Orius* nymph were detected on faba bean, other arable crops and wild plants until the first week of March (Atakan, 2010). This indicates that mortality rates of the early nymphs were high. Our findings are in accord with results of *O. albidipennis* in Irak (El-Serwi et al., 1985) and *O. laevigatus* in Egypt (Tawfik & Ata, 1973). *Orius* adults survived in the winter in some regions of Israel, but no nymphs were detected until the end of March on mango blossoms (Ben-Dov et al., 1992).

High numbers of eggs were laid in the experiments conducted in October and April. Females oviposited fewer eggs through the winter months. The rate of non-reproductive females was 40% in January and February. In Israel, most adult females laid eggs in the outdoor experiments conducted in October and April, but some females did not lay eggs and died (Chyzik et al., 1995). In our study, females in the October experiment oviposited and laid fewer eggs for two months. Also, one female from the November experiment oviposited for another 2 months. We suggest that the reason why some females did not lay eggs in the winter may be due to their lack of tolerance to low temperatures, and they might have not mated.

In current study, the day length did not induce diapause in *O. niger*. Chyzik et al. (1995) found that both females and males of *O. albidipennis* at the low mean temperature (below 15°C) are active and feed on moth eggs but females do not lay eggs. Bahşi & Tunç (2008) concluded that 9-13 h day length do not induce diapause and did not affect the development of *O. niger*. They suggested that *O. niger* could survive and reproduce under the winter conditions in heated greenhouses. According to data obtained in current study, *O. niger* from Turkey might be useful for controlling *F. occidentalis* under protected conditions in southern Europe and Mediterranean regions. However, It is known that some species of *Orius* show obligatory and facultative diapause, and diapause is induced by low temperature and short day length (Iglinsky & Rainwater, 1950; Gillespie & Quiring, 1993; Ramakers & van den Meiracker, 1991).

In the current study, life spans of females in autumn and winter were less than 30 days and thus they did not survive till spring. Oviposition time of females in winter was less than 20 days. In Israel, mean longevity of *O. albidipennis* from November and December experiments were significantly longer (84.2 and 125.2 days, respectively) (Chyzik et al., 1995), and females started to oviposit again at the end March. Differences between the two studies may be related to different climatic conditions and different species of *Orius*. *Orius niger* does not build up efficient outdoor population in winter and early spring time in the region because of short-lived females and males, low reproductive rates of females and high mortality rates of nymphs. Thus, it will not be possible to control outdoor winter and spring populations of small insects, including *F. occidentalis*, in the region.

Orius niger has effectively controlled flower thrips population in cotton flowers in the region (Atakan & Gencer, 2008) and this predatory bug is more common in the Mediterranean region. Based upon some results of current study, *O. niger* could be employed as a biological control agent in IPM studies on pest thrips. Populations of *F. occidentalis* increase dramatically in spring in greenhouses and outdoors when many crops are flowering, requiring applications of pesticides. However, the present results suggest that densities of *O. niger*, even in the early spring, are still not high. *Orius niger* oviposit again in mid-March, and nymph populations appear on faba bean plants in late March (Atakan, 2010). Nymphs will become adults in mid-April. The predatory effect of the April population will be seen in May.

Finally, mortality rates of *O. niger* nymphs were high in winter to early spring. Additionally, most of the experimental females coming from the autumn and winter months were not able to survive till spring to produce nymphs. Therefore, *O. niger* might not be unable to control pest insects in the early vegetative period under open field conditions. For this reason, further studies in Turkey should focus on efforts to augment winter to early spring populations of this predator emphasizing earlier control of *F. occidentalis*. Diapause does not occur in the *O. niger* under short day length. *Orius niger* should be evaluated for control of *F. occidentalis* in greenhouses in Turkey. However, the reproductive rate of *O. niger* under laboratory conditions is lower than that of *O. laevigatus* (Tommasini et al, 2004), which is commercially used to control pest thrips in greenhouses in Turkey. Further studies are needed on rearing conditions for *O. niger* under laboratory conditions to enhance its mass production for release against pest thrips (mainly *F. occidentalis*).

Acknowledgments

We would like to extend our thanks to Prof. Dr. M. Bora KAYDAN (Vocational School of İmamoğlu, University of Çukurova, Adana, Turkey) and Mr. Tange Denis ACHIRI (MPhil) (Department of Plant Protection, Faculty of Agriculture, University of Çukurova, Adana, Turkey) for reviewing of the manuscript. This study was supported by the Unit of Scientific Research Projects (Project no: FBA-2014-2488), Çukurova University, Adana, Turkey.

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Original article (Orijinal araştırma)

Contact toxicities of some plant extracts in Apiaceae family on different developmental stages of *Tetranychus urticae* Koch, 1836 (Acari: Tetranychidae)

Apiaceae familyasındaki bazı bitki ekstraktlarının *Tetranychus urticae* Koch (Acari: Tetranychidae)'nin farklı gelişme dönemleri üzerine kontakt toksisiteleri

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Summary

The contact toxicities of extracts of anise (*Pimpinella anisum* L.), coriander (*Coriandrum sativum* L.), cumin (*Cuminum cyminum* L.), dill (*Anethum graveolens*) and fennel (*Foeniculum vulgare* Mill.) plants, included in the Apiaceae, prepared with water on different stages of *Tetranychus urticae* Koch, 1836, were studied. The study was conducted in a laboratory and insect breeding rooms between in 2015-2016. Leaf disc - spray tower method was used for the purpose of determining toxic effects of the plant extracts. The effect of plant extracts on adult and protonymphs stages of *T. urticae* were determined. Also, the effects of plant extracts were determined on the hatching of *T. urticae* eggs. Four different concentration of extracts were tested; 1, 3, 6 and 12%. The experiments consisted of one control and four replicates for each concentration. Fifteen individuals were included in each repeat. Dead-live counts were made 1, 3 and 6 days after treatment to determine contact effects of the extracts. It was found that the contact effects of plant extracts on *T. urticae* adult and protonymphs increased with rising concentration, caused 100% death on *T. urticae* adults and nymphs at 12% after 6 days. Also, the greatest effect on *T. urticae* egg hatching was in dill extracts with mortality of over 91%. It is considered that these plant extracts may constitute an alternative acaricides for control *T. urticae*.

Keywords: Apiaceae, plant extracts, *Tetranychus urticae*, toxicity

Özet

Apiaceae familyası içerisinde yer alan anason (*Pimpinella anisum* L.), dere otu (*Anethum graveolens*), kimyon (*Cuminum cyminum* L.), kişniş (*Coriandrum sativum* L.) ve rezene (*Foeniculum vulgare* Mill.) bitkilerinin su ile hazırlanan ekstraktlarının *Tetranychus urticae* Koch, 1836' nin farklı dönemleri üzerinde değme yoluyla toksisiteleri araştırılmıştır. Çalışma 2015-2016 yılları arasında laboratuvar ve böcek üretim odalarında yürütülmüştür. Bitki ekstraktlarının zehir etkilerini belirlemek amacıyla, yaprak disk-ilaçlama kulesi yöntemi kullanılmıştır. Çalışmalarda bitki ekstraktlarının *T. urticae*' nin ergin ve protonimf dönemlerine etkisi belirlenmiştir. Ayrıca bitki ekstraktlarının *T. urticae*' nin yumurta açılımı üzerine olan etkileri de belirlenmiştir. Bitki ekstraktlarının %1, 3, 6, 12 olmak üzere dört farklı konsantrasyonu kullanılmıştır. Denemeler bir kontrol ve her konsantrasyon için dört tekrerrör olarak yürütülmüştür. Her tekrerrörde 15 birey kullanılmıştır. Bitki ekstraktlarının kontakt etkilerini belirlemek amacıyla ölü-canlı sayımları 1, 3 ve 6. günlerde yapılmıştır. Çalışmada bitki ekstraktlarının *T. urticae* ergin ve nimfleri üzerindeki kontakt etkilerinin konsantrasyon miktarı artışına bağlı olarak yükseldiği, altıncı gün sayım sonuçlarına göre bitki ekstraktlarının %12 konsantrasyonunda *T. urticae* ergin ve protonimflerinde %100 ölüme neden olduğu belirlenmiştir. Ayrıca *T. urticae* yumurta açılımı üzerinde en yüksek etki %91 ile dere otu ekstraktında belirlenmiştir. Çalışma sonucunda, kullanılan bitki ekstraktlarının *T. urticae* ile mücadelede sentetik akarisitlere alternatif oluşturabileceği düşünülmektedir.

Anahtar sözcükler: Apiaceae, bitki ekstraktı, *Tetranychus urticae*, toksisite

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Received (Alınış): 07.02.2017

Accepted (Kabul ediliş): 04.07.2017

Published Online (Çevrimiçi Yayın Tarihi): 22.07.2017

Introduction

Tetranychus urticae Koch, 1836 (Acari: Tetranychidae), two-spotted spider mite, causes losses in many agricultural crops of economic importance (Zhang, 2003; Hoy, 2011; Vacante, 2016). *Tetranychus urticae* causes necrotic stain formation as the result of leaf damage, and in high populations it causes drying and leaf fall (Brandenburg & Kennedy, 1987). *Tetranychus urticae* damages more than 1100 plants in 126 families (Migeon & Dorkeld, 2017). To control *T. urticae*, various acaricides and insecticides with extensive effects are used to reduce losses in cultivated plants (Van Leeuwen et al., 2006). *Tetranychus urticae* can develop resistance against insecticides and acaricides as the result of frequent application at short intervals because of its rapid haplodiploid sexual reproduction and short life cycle (Song et al., 1995; Van Leeuwen et al., 2009, 2015).

In recent years, natural pesticides have gained importance as alternatives to synthetic insecticides because of disadvantages of chemical control (Feng & Isman, 1995; Laborda et al., 2013). There have been many studies on the possibility of using compounds found naturally in the plants as an alternative to synthetic pesticides (Shi et al., 2006; Villanueva & Walgenbach, 2006; Cavalcanti et al., 2010; Wei et al., 2011). The studies put emphasis on the compounds obtained from the plants that do not put additional toxic materials into the environment, breakdown in a short span of time, and do not cause soil and water contamination (Liang et al., 2003; Isman & Akhtar, 2007). There have been many studies on the effects of extracts from plants obtained by various methods on *T. urticae* (Choi et al., 2004; Matinez-Villar et al., 2005; Rasikari et al., 2005; Chermenskaya et al., 2010).

An extract of *Satureja hortensis* L. (Lamiaceae) was toxic to *T. urticae* (Aslan et al., 2004). Similarly, extracts of neem (Meliaceae) (Matinez-Villar et al., 2005), some species of Solanaceae (Rasikari et al., 2005), *Capparis aegyptia* Lam. (Capparaceae) (Hussein et al., 2006) and *Nerium oleander* L. (Apocynaceae) (Islam et al., 2008) were found to be effective against *T. urticae*. Kumral et al. (2010) reported that ethanol extracts of *Datura stramonium* L. (Solanaceae) leaves and seeds exhibited acaricidal effects against *T. urticae*. However, there have been no studies in Turkey and only a few elsewhere to determine toxic effects of extracts obtained from the plants in the Apiaceae. It is known that some compounds in plants from the Apiaceae have insecticidal and repellent effects on insects (Regnault-Roger et al., 2005; Isman, 2006). Chermenskaya et al. (2010), determined that extracts of *Angelica tschimganica* (Korov.), *Conium maculatum* L., *Dorema microcarpum* Korov., *Ferula foetidissima* Regel. & Schmalh., *Heracleum dissectum* Ledeb., *Mediasia macrophylla* (Regel. & Schmalh.) plants within the Apiaceae have an average toxic effect on *T. urticae*. However, in this study, the extracts were prepared with an organic solvent. However, we postulate that the water based extracts could have different effects on *T. urticae*. Therefore, in this study, contact toxicities of water-based leaf extracts of five plants within the Apiaceae on different developmental stages of *T. urticae* has been investigated in laboratory conditions to provide an alternative to synthetic insecticide and acaricide.

Material and Methods

Tetranychus urticae culture

The study was conducted in a laboratory and insect breeding rooms at Suleyman Demirel University, Faculty of Agriculture, Plant Protection Department between in 2015-2016. *Tetranychus urticae* susceptible population (German Susceptible Strain, GSS) was brought from Rothamsted Experimental Station (UK) to insect cultivating cabins in Süleyman Demirel University in 2001 and has been cultivated since without exposure to any pesticides. The GSS population was cultured on bean (*Phaseolus vulgaris* L. cv. Barbutia) in climate chambers at 26±2°C, 60±5% RH and 16 h photoperiod.

Plant species investigated

Leaves from five species in the Apiaceae, anise (*Pimpinella anisum* L.), coriander (*Coriandrum sativum* L.), cumin (*Cuminum cyminum* L.), dill (*Anethum graveolens*) and fennel (*Foeniculum vulgare* Mill.) were used to make extracts investigated in this study.

Plant extract preparation

The method of Rezaei & Yarnia (2009) was used to the prepare the plant extracts. Plant material, leaves harvested in flowering phase, were dried for two weeks at room temperature. The dried leaves were pulverized in a blender. Milled material (20 g) from each plant was steeped in 100 mL distilled water for 24 h and then filtered with through cheesecloth. These extracts were transferred to falcon tubes and centrifuged for 5 min at 9,000 rpm then filtered through Whatman filter papers.

Contact toxicity bioassays

The method of Erdogan et al. (2012) was used for testing the contact toxicities of the plant extracts on eggs, protonymphs and adults of *T. urticae*. Concentrations of 1, 3, 6, 12% plant extracts were tested on protonymphs and adults by a leaf disc-spray tower method. In addition, the ovicidal effect of plant extracts on *T. urticae* eggs were evaluated. Fifteen adult females were put on bean leaf discs in 9-cm Petri dishes to obtain eggs, protonymphs and adults at the same stage of development eggs left in the dishes for 24 h. Bioassays with one control (water only) and the four concentrations of each extract were replicated four times. Fifteen individuals were included in each replicate. Triton X 100 at 0.01% was added to the pure water in which extracts were prepared and was also used in the water control as an extender and sticker. The plant extracts are applied as 2 mL on the leaf surface in 1 atm in a spray tower (Kumral et al., 2010). Contact effects of plant extracts were determined by dead-live counts of *T. urticae* nymphs and adults after 1, 3 and 6 days. For the egg hatching experiment, observations continued until all eggs in the control group had hatched.

Statistical analysis

Percentages of dead mites obtained from the contact experiments were calculated according to Abbott formula (Abbott, 1925) and arcsin transformed (Zar, 1999). Contact effects were analyzed by three-way repeated measures ANOVA. Ovicidal effects were analyzed by two-way ANOVA and Tukey's test was used to determine the difference between means ($p < 0.05$).

Results

Contact toxicities of plant extracts on adult period of *T. urticae* are shown in Table 1. The contact effects of the water extracts of all five plants was increased with concentration. The greatest effect occurred after 6 days. The greatest effect on adults after 1 day was with coriander extract at 12% and this was significantly different from other plant extracts ($p < 0.05$). The greatest effect after 3 days was with anise extract and 100% mortality was recorded. On that day, the effects of extracts of fennel, coriander, cumin and dill (in that order) were less than that of the anise extract. After 6 days, all plant extracts cause 100% adult mortality and therefore were not statistically different ($p < 0.05$).

Contact toxicities of plant extracts on *T. urticae* nymphs are shown in Table 2. After 1 day, the greatest effect was with fennel at 12%, which caused 100% mortality, and was significantly different from the other extracts ($p < 0.05$). On that day, the effects of extracts of cumin, coriander, anise and dill (in that order) were less than that of fennel extract. After 3 days, 100% mortality of nymphs occurred with fennel extract at 6 and 12%. After fennel, the extracts of coriander, cumin and dill were also effective. All five extracts cause 100% mortality in nymphs after 6 days. Especially, all concentrations of fennel extract caused 100% mortality in *T. urticae* nymphs. After 6 days, contact effects of all plant extracts were therefore not statistically different ($p < 0.05$).

Table 1. Contact effect of plant extracts at four concentrations on *Tetranychus urticae* adults (mean±SE)*

Time (day)	Concentration (%)	Mortality (%)				
		Anise	Coriander	Cumin	Dill	Fennel
1	1	4.47±0.25bF	10.44±0.16aF	9.09±0.18aF	5.97±0.65bF	5.88±0.35bF
	3	20.89±0.35aE	20.89±0.18aE	19.69±0.25aE	17.91±0.44aE	20.58±0.18aE
	6	32.83±0.27bD	37.31±0.16aD	28.78±0.33bE	31.34±0.27bD	32.35±0.16bE
	12	43.28±0.40bD	50.74±0.56aC	40.90±0.42bD	41.79±0.55bD	45.58±0.32bD
3	1	60.31±0.65aC	24.59±0.42bE	20.00±0.45bE	17.18±0.18cE	25.80±0.36bE
	3	75.01±0.55aB	42.62±0.23bD	35.38±0.75cD	34.37±0.68cD	48.38±0.45bD
	6	88.88±0.28aA	57.37±0.38bC	47.69±0.65cD	53.12±0.65bC	61.29±0.29bC
	12	100.00±0.25aA	80.32±0.45bB	67.69±0.54cC	55.75±0.22cC	83.87±0.47bB
6	1	79.03±0.55aB	57.37±0.55bC	57.37±0.36bC	58.66±0.45bC	63.93±0.32bC
	3	93.54±0.62aA	77.04±0.65bB	88.52±0.46aB	70.00±0.55bB	90.16±0.22aA
	6	100.00±0.25aA	88.52±0.75bA	100.00±0.25aA	85.00±0.38bA	100.00±0.25aA
	12	100.00±0.25aA	100.00±0.65aA	100.00±0.18aA	100.00±0.25aA	100.00±0.35aA

*Different lower case letters in the same line and different uppercase letters on the same column show that the means are significantly different for plant species and application concentration, respectively ($p<0.05$).

Table 2. Contact effect of plant extracts with different concentration on *Tetranychus urticae* protonymphs (mean±SE)*

Time (day)	Concentration (%)	Mortality (%)				
		Anise	Coriander	Cumin	Dill	Fennel
1	1	4.54±0.65cG	16.92±0.75bF	13.63±0.25b	11.11±0.26bG	25.75±0.33aD
	3	16.16±0.35cF	35.38±0.55bE	27.27±0.35b	25.39±0.45bG	50.06±0.45aC
	6	30.30±0.44cE	50.76±0.24bD	40.90±0.45c	36.50±0.55cF	78.78±0.55aB
	12	42.42±0.28cD	64.61±0.28bC	54.54±0.65b	49.20±0.38cE	100.00±0.65aA
3	1	25.80±0.25bE	31.14±0.34bE	24.19±0.23b	19.67±0.45cG	67.21±0.26aB
	3	46.77±0.55bD	49.18±0.54bD	46.77±0.38b	37.70±0.22cF	90.16±0.45aA
	6	62.90±0.16bC	68.85±0.45bC	69.35±0.42b	54.09±0.25cE	100.00±0.25aA
	12	84.64±0.18bB	88.52±0.65bB	88.70±0.65b	77.04±0.45cC	100.00±0.25aA
6	1	60.00±0.22bC	55.73±0.65bD	59.67±0.18b	39.34±0.65cF	100.00±0.25aA
	3	73.33±0.32cB	83.60±0.75bB	85.48±0.16b	65.57±0.75cD	100.00±0.25aA
	6	90.00±0.55bA	96.72±0.65aA	100.00±0.25	88.52±0.45bB	100.00±0.25aA
	12	100.00±0.25aA	100.00±0.25aA	100.00±0.25	100.00±0.25aA	100.00±0.25aA

*Different lower case letters in the same line and different uppercase letters on the same column show that the means are significantly different for plant species and application concentration, respectively ($p<0.05$).

The effects of plant extracts on *T. urticae* egg hatch are shown in Table 3. Inhibition of hatch rose with extract concentration. The greatest inhibition occurred with dill extract at 12% dill extract. The effects fennel, anise, coriander and cumin extracts (in that order) were less than the effect of dill extract.

Table 3. Egg hatching effect of plant extracts with different concentration on *Tetranychus urticae* eggs (mean±SE)*

Concentration (%)	Mortality (%)				
	Anise	Coriander	Cumin	Dill	Fennel
1	20.58±0.65bD	17.64±0.25bD	29.41±0.18aC	14.70±0.55bD	26.47±0.18aD
3	33.82±0.45bC	32.29±0.35bC	41.17±0.65aB	38.23±0.75aC	44.11±0.165aC
6	51.47±0.35bB	47.05±0.44bB	58.82±0.25aA	63.23±0.42aB	60.29±0.25aB
12	69.11±0.26bA	64.70±0.55cA	63.23±0.45cA	91.17±0.25aA	73.52±0.25bA

*Different lower case letters in the same line and different uppercase letters on the same column show that the means are significantly different for plant species and application concentration, respectively ($p < 0.05$).

Discussion

Many volatile oil and plant extracts show acaricide and insecticide effects on economic plant pests, including mites (Prakash & Rao, 1997; Attia et al., 2013). It was determined that when the concentration and the exposure time of five plant extracts used in the study increased the contact effects on *T. urticae* protonymphs and adults. Complete mortality of *T. urticae* protonymphs and adults was observed, especially at 12% plant extracts after 6 days. Other studies have examined effects of plant extracts on *T. urticae*. Coelho et al. (2001) has found that *Petiveria alliacea* L. (Petiveriaceae) extract had acaricide effects. Likewise, Choi et al. (2004) reported that *Rosmarinus officinalis* L. (Lamiaceae) extract was fatal to *T. urticae* adults. Shi et al. (2006) determined that *Kochia scoparia* L. extract caused about 79% mortality of *T. urticae*. Saber (2004) found that *Artemisia monosperma* Delile (Asteraceae) extract has repellent, toxic and prevented egg-laying in *T. urticae*. El-Sharabasy (2010) determined the contact effects of *Artemisia judaica* L. (Asteraceae) leaf extract on *T. urticae* adults and nymphs. Jeon & Lee (2011) found that *Tabebuia impetiginosa* (Mart. ex DC.) (Bignoniaceae) extract had acaricide effects. This literature shows that extracts from many plants cause death in *T. urticae* at high levels.

However, the effects of extracts of *P. anisum*, *C. sativum*, *F. vulgare*, *A. graveolens* and *C. cyminum*, on *T. urticae* have not been previously studied. Chermenskaya et al. (2010) has indicated that the apiaceous species *A. tschimganica*, *C. maculatum*, *D. microcarpum*, *F. foetidissima*, *H. dissectum*, *M. macrophylla* plant extracts caused medium level of mortality in *T. urticae* adults. In the same study, it was found that *Prangos lipskyi* Korov. extracts caused more than 80% mortality in *T. urticae* adults. Similarly, in our study a 12% concentration of five plant extracts caused high rates of mortality of *T. urticae* adult and nymphs. This could indicate that high mortality rates in *T. urticae* are due to common active substance present in plants in the Apiaceae. However, it will be necessary to identify and study the active substances in these extracts in order to determine the validity of this suggestion.

The plant extracts studied also inhibited *T. urticae* egg hatch. There have been some studies in which ovicidal effects of some plant extracts on spider mite eggs were determined. Dimetry et al. (1993) determined that a commercial preparation of neem extracts reduced hatching of *T. urticae* eggs and also had ovicidal effect. Sarmah et al. (2009) reported 87% egg mortality at 10% concentration of aqueous extracts of *Xanthium strumarium* L. (Compositae) against *T. urticae*. Yanar et al. (2011) has reported that *Eucalyptus camaldulensis* (Myrtaceae) extract caused 63% death rate in *T. urticae* eggs. In another study, it was found that *Scaligeria meifolia* Boiss (Apiaceae), *Anisosciadium orientale* DC. (Apiaceae),

Trigonella elliptica Boiss (Leguminosae) and *Dodonaea viscosa* L. (Sapindaceae) extracts had ovicidal activity on *T. urticae* eggs; 46, 41, 40 and 38%, respectively (Ghaderi et al., 2013). Similarly, our results suggest that plant extracts, especially at 12%, strongly inhibit hatching of *T. urticae* eggs. Dill extract, in particular, was highly inhibitory to egg hatch. Therefore, it is recommended that the active substances in plants in Apiaceae family be determined and their activity against pest further investigated.

In conclusion, it is suggested that extracts of these five plants have the potential to be used as an alternative acaricides for control of *T. urticae*. It is an established fact that plant extracts can be effective on many pests and have advantages in respect of human health. However, considering there are many plants in the nature with unknown contents and efficiency, there is a need for laboratory and field studies to investigate the effects of plant extracts on pests. In addition, side effects of some plant extracts on natural enemies must be determined and their safety must be indicated with scientific studies. This study provides some useful new information and it is thought that it can provide a foundation for the future studies.

Acknowledgments

We thank Assoc. Prof. Dr. Nimet KARA, Suleyman Demirel University Faculty of Agriculture of Department of Field Crops for support with the preparation of plant extracts.

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