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Response to Some Acaricides of the Two-spotted Spider Mite (*Tetranychus urticae* Koch) from Protected Vegetables in Isparta*

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Abstract: The two-spotted spider mite, *Tetranychus urticae* Koch, was tested for susceptibility to various acaricides, propargite (Omite) 570 g l^{-1}, amitraz (Kortraz) 200 g l^{-1} and abamectin (Agrimec) 18 g l^{-1}. Propargite, a selective acaricide, has been used to control *T. urticae* in many crops in Turkey. Amitraz and abamectin have acaricidal and insecticidal properties, and are thus used to control spider mites and some vegetable pests (e.g., whiteflies and *Liriomyza* spp.). Five different *T. urticae* populations collected from vegetable greenhouses in Isparta and their responses to those acaricides were investigated by leaf dip assay and compared with those of a susceptible reference strain. Resistance ratios for the chemicals ranged from <1.00 to 2.5 for propargite, 1.2 to 2.1 for amitraz and <1.0 to 2.9 for abamectin (based on LC_{50}). Since the greenhouses around Isparta province where the two-spotted spider mite populations were collected have recently been constructed, the populations used in this study were not exposed to excessive acaricides and had had no important loss of susceptibility to the acaricides applied in the study. However, it was observed that the growers used pesticides haphazardly.

Key Words: *Tetranychus urticae*, acaricide, bioassay, Propargite, Amitraz, Abamectin

Isparta'da Örtüaltı Sebze Üretim Alanlarında Zararlı Olan *Tetranychus urticae* Koch Populasyonlarının Bazı Akarisitlere Karşı Tepkileri

Özet: İki noktalı kırmızıörümcek, *Tetranychus urticae* Koch'nin bazı akarisitlere, propargite (Omite) 570 g l^{-1}, amitraz (Kortraz) 200 g l^{-1} ve abamectin (Agrimec) 18 g l^{-1}'e karşı duyarlılığı belirlenmiştir. Propargit selektif akarisittir ve Türkiye'de birçok üründe *T. urticae* savaşımında kullanılmaktadır. Amitraz ve abamectin ise akarisit ve insektisit özelliktedir ve bu nedenle kırmızı örümcek ve bazı sebze zararlılarına (mesela, whiteflies, *Liriomyza* spp.). karşı kullanılmaktadır. Isparta da bulunan beş farklı sebze üretim serasından toplanan *T. urticae* populasyonlarının bu akarisitlere karşı duyarlılıkları yaprak daldırma yöntemi ile saptanmıştır ve standart hassas populasyon (GSS) ile karşılaştırılmıştır. Standart hassas (GSS) populasyon ile karşılaştırılarak bulunan direnç oranlarının dağılımı propargite, amitraz, ve abamectin için sırasıyla <1.0 - 2.5, 1.2 - 2.1 ve <1.0 - 2.9 kat düzeylerinde olmuştur (LC_{50}'ye göre). Isparta ve çevresindeki sebze seralarının birçoğu yeni kurulduğu için denemeye alınan populasyonların çalışmada kullanılan akarisitlere karşı önemli düzeyde bir duyarlılık kaybı olmamıştır. Buna karşın üreticilerin bilinçsizce ilaç kullanmaya yöneldikleri gözlenmiştir.

Anahtar Sözcükler: *Tetranychus urticae*, akarisit, bioassay, Propargite, Amitraz, Abamectin

Introduction

The two-spotted spider mite, *Tetranychus urticae* Koch (Acarina: Tetranychidae) is an important and highly polyphagous pest. It is particularly dominant in intensive, high–yield cropping systems, and affects crops by direct feeding, thereby reducing the area of photosynthetic activity and causing leaf abscission in severe infestations (Gorman et al., 2001). Greenhouses are ideal areas for spider mites, which can complete a generation in 1 week in suitable areas (Düzgüneş and Çobanoğlu, 1983). For

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the past 25 years, *T. urticae* control in Turkey has been based almost exclusively on pesticides. Many insecticides and acaricides have been registered to control *T. urticae* in Turkey. A major problem in controlling spider mites is their ability to rapidly develop resistance to acaricides after only a few applications. Their high reproductive potential and short life cycle, combined with 10 or 12 pesticide applications per season in vegetable greenhouses, result in development of resistance in spider mites. In recent years, acaricides have not been performing well against *T. urticae* populations in Turkey, in consideration of selection pressure that might encourage the evolution of pesticide resistance. It was reported that *T. urticae* populations in pear orchards in the USA developed resistance to cyhexatin (Hoyt et al., 1985), and that *T. urticae* populations from roses in Australia developed a 2.6-464.0-fold resistance to fenbutatin-oxide, a 39.0-135.0-fold resistance to propargite and a 11.0-51.0-fold resistance to fluvalinate (Goodwin et al., 1995). There are many claims in the literature that *T. urticae* individuals can develop resistance to insecticides and acaricides (Hoyt et al., 1985; Keena and Granet, 1987; Sawicki and Denholm, 1987; Herron and Rophail, 1998; Gorman et al., 2001). It was suggested that development of resistance can be prevented by developing appropriate strategies (Sawicki and Denholm, 1987). In this study, 5 Isparta populations of *T. urticae*, having different histories of exposure to pesticides, were selected for toxicological studies. In the Isparta region, commercial vegetable farming is newly developing and most enterprises are small scale, being family-owned farms. In the past, growers used fewer chemical applications than they do today. Amitraz, propargite, and abamectin were registered in 1981, 1992 and 1992, respectively, in Turkey. These acaricides are contact and stomach poisons acting on all stages of *T. urticae*. Propargite, a selective acaricide, has been used to control *T. urticae* on many crops in Turkey. Amitraz and abamectin have acaricidal and insecticidal properties, and are thus used to control spider mites and some vegetable pests (e.g., whiteflies and *Liriomyza* spp.).

The objective of this study was to determine the level of resistance developed against those pesticides in field–collected spider mite populations from protected vegetables in Isparta.

Materials and Methods

Spider mite populations

Five populations of *T. urticae* were collected from 5 greenhouses (Table 1) and cultured on bean plants in an insectarium at 27 ± 2 °C, at 60-65% rh and with a photoperiod of 16:8 h (L:D). A susceptible strain (GSS) had been obtained from Rothamsted Experimental Station, Harpenden (England), in 2001 and reared in laboratory conditions since then. The GSS population has been maintained in England as a laboratory culture since 1965 (Dennehy et al., 1993).

Synchronized cultures of *T. urticae* for use in bioassays were produced from each of 5 stock populations and the GSS population. Adult females were transferred from stock populations to bean leaves in small plastic containers.

Chemicals

Chemicals used in the experiments were propargite (Omite® Süper 570 EW) 570 g l^{-1}, amitraz (Kortraz 20 EC) 200 g l^{-1} and abamectin (Agrimec EC) 18 g l^{-1}).

Bioassays

In bioassays, a leaf-dipping method was used to assess the resistance levels (Tian et al., 1992). Bioassays were performed on each *T. urticae* population with a control (water) and different concentrations (5-7) of the tested acaricide. The concentrations of propargite mixed in distilled water were 1.5625, 3.125, 6.25, 12.5, 25.0, 50.0, 100.0, 200.0 and 400.0 µl 100 ml^{-1}, those of amitraz were 4.6875, 9.375, 18.75, 37.5, 75.00, 150.0 and 300.0 µl 100 ml^{-1} and those of abamectin were 0.01562, 0.03125, 0.625, 0.125, 0.25, 0.50 and 1.00 µl 100 ml^{-1}. These concentrations were at X 1/2 intervals. Bean leaf disks 20 mm in diameter were dipped in each pesticide suspension for 5 s, and 3 replicates were used per dose. Leaves were then drained and placed individually on moistened cotton in 90 mm Petri dishes. Twenty-five adult females of the same age were placed on leaf disks and the Petri dishes were covered with a ventilated lid and incubated (27 ± 2 °C, 60-65% rh and 16 L:8 D) for 24 h. At the end of this period, the numbers of dead mites were counted. The criterion for mortality was an inability on the part of mites to walk at least one body length when lightly prodded.

Table 1. Origins of *Tetranychus urticae* populations.

Population	Location of collection	Host	Date of collection
AKSUD	Aksu-Isparta	Tomato, Greenhouse	04.07.2002
AKSUF	Aksu-Isparta	Bean, Greenhouse	04.07.2002
SEN	Keçiborlu-Isparta	Bean, Greenhouse	23.07.2002
ŞAK	Şakikaraağaç-Isparta	Bean, Greenhouse	06.08.2002
KULE	Kuleönü-Isparta	Bean, Field	31.07.2002

Statistical analyses

For each concentration-mortality experiment, data from all replicates were pooled and subjected to probit analysis. LC_{50} and LC_{90} values with a 95% CL and slopes ± SE of the regression were estimated using the POLO computer program (LeOra Software, 1994). The LC_{50} and LC_{90} values of the field populations were compared to those of the susceptible population (GSS).

Results and Discussion

LC_{50} and LC_{90} values of the two-spotted spider mite populations collected from greenhouses in the Isparta region and the resistance level of each population are given in Tables 2-4. The LC_{50} and LC_{90} values and resistance ratios of each population varied. However, based on lethal concentration results, there was no important decrease in the susceptibility levels of any population against propargite, amitraz or abamectin. Resistance factors relative to the response of the GSS range from 1 to 2.9 for all tested chemicals (Tables 2-4). The AKSUF population showed higher LC_{50} values and resistance ratio to propargite and amitraz than did the other populations. Abamectin was very effective against this population. The SEN population showed a higher loss of susceptibility against abamectin than did the other

Table 2. Probit statistics for susceptible population (GSS) and field populations of *T. urticae* tested against propargite (Omite) 570 g l^{-1}.

Population	n*	Slope ± SE	LC_{50} (µl 100 ml^{-1}) (0.95 CL)	LC_{90} (µl 100 ml^{-1}) (0.95 CL)	Resistance factor LC_{50}**	Resistance factor LC_{90}**
AKSUD	555	1.4 ± 0.1	30.6 18.9-47.6	253.1 132.0-911.0	≈ 1	2.3
AKSUF	647	1.7 ± 0.2	80.0 44.8-121.9	467.0 268.1-520.0	2.5	4.3
SEN	490	2.3 ± 0.3	51.9 25.5-7.4	162.01 108.5-414.6	1.6	1.5
ŞAK	590	2.2 ± 0.2	31.6 21.3-43.0	122.1 84.7-218.9	1.0	1.1
KULE	730	1.8 ± 0.1	39.5 22.6-62.1	203.1 118.7-540.8	1.2	1.9
GSS (Susceptible)	689	2.4 ± 0.3	31.5 20.9-44.5	107.4 68.9-0.290.7	-	-

* Sample size refers to number of adult females
**Resistance factor = LC_{50} or LC_{90} of the field-collected / LC_{50} or LC_{90} of the susceptible population (GSS)

Table 3. Probit statistics for susceptible population (GSS) and field populations of *T. urticae* tested against amitraz (Kortraz) 200 g l^{-1}.

Population	n*	Slope ± SE	LC_{50} (µl 100 ml^{-1}) (0.95 CL)	LC_{90} (µl 100ml^{-1}) (0.95 CL)	Resistance factor LC_{50}**	Resistance factor LC_{90}**
AKSUD	564	2.7 ± 0.2	36.7 22.0-53.9	109.8 72.6-225.4	1.2	<1.0
AKSUF	541	1.9 ± 0.2	63.4 51.3-76.6	283.6 218.8-402.1	2.1	2.2
SEN	585	1.9 ± 0.2	36.2 18.7-58.6	171.2 99.2-490.6	1.2	1.3
ŞAK	613	2.2 ± 0.2	40.4 25.2-56.7	155.1 106.5-286.3	1.3	1.2
KULE	525	1.9 ± 0.2	37.3 27.3-48.9	175.6 123.3-297.2	1.2	1.3
GSS (Susceptible)	554	2.1 ± 0.1	30.8 22.9-41.9	129.9 86.9-238.3	-	-

*Sample size refers to number of adult females
**Resistance factor = LC_{50} or LC_{90} of the field-collected / LC_{50} or LC_{90} of the susceptible population (GSS)

Table 4. Probit statistics for susceptible population (GSS) and field populations of *T. urticae* tested against abamectin (Agrimec) 18 g l^{-1}.

Population	n*	Slope ± SE	LC_{50} (µl 100 ml^{-1}) (0.95 CL)	LC_{90} (µl 100 ml^{-1}) (0.95 CL)	Resistance factor LC_{50}**	Resistance factor LC_{90}**
AKSUD	767	2.3 ± 0.2	0.056 0 .033 - 0.081	0.204 0.138-0.393	1.6	2.2
AKSUF	783	2.9 ± 0.2	0.031 0.022 - 0.041	0.082 0.057 - 0.169	<1.0	<1.0
SEN	742	2.3 ± 0.1	0.103 0.075 -0.142	0.371 0.249 - 0.713	2.9	3.9
ŞAK	733	3.3 ± 0.3	0.039 0.029-0.051	0.093 0.068-0.167	1.1	<1.0
KULE	743	3.9 ± 0.4	0.048 0.042 - 0.054	0.102 0.089 - 0.123	1.4	1.1
GSS (Susceptible)	809	2.9 ± 0.3	0.035 0.027-0.042	0.094 0.076-0.130	-	-

*Sample size refers to number of adult females
**Resistance factor = LC_{50} or LC_{90} of the field-collected / LC_{50} or LC_{90} of the susceptible population (GSS)

populations. According to the LC90 values, the AKSUF and SEN populations showed a 4.3-fold and a 3.9-fold increase in resistance to propargite and abamectine, respectively. It is suggested that there is a high possibility of increased resistance developing following the use of propargite and abamectine against the two-spotted spider mite.

Dose-response curves of the *T. urticae* populations are given in Figures 1-3. As mentioned by Hoskins (1960), the slope of the probit dose line varies according to the

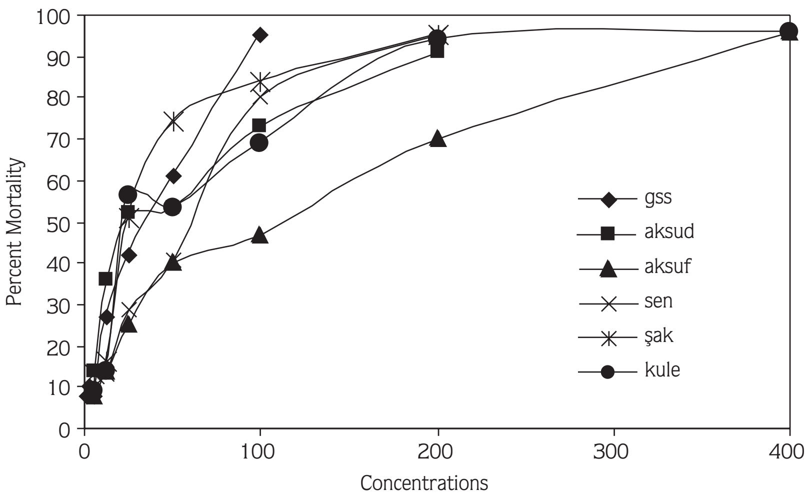

Figure 1. Dose-response for susceptible (GSS) and field-collected populations of *T. urticae* tested against propargite (concentrations; µl 100 ml^{-1}).

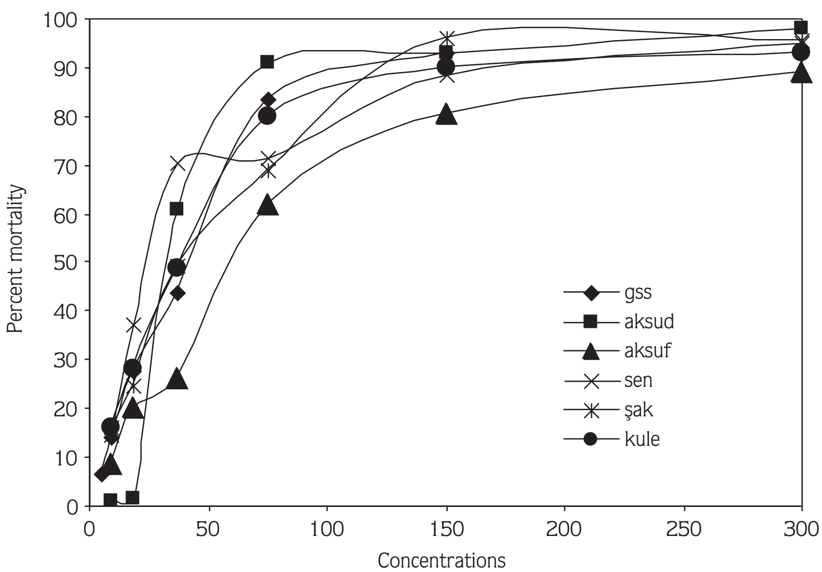

Figure 2. Dose-response for susceptible (GSS) and field-collected populations of *T. urticae* tested against amitraz (concentrations; µl 100 ml^{-1}).

active ingredient. Differences observed in dose-response curves showed variations among individuals within a population. Mortality curves are arranged from left to right, parallel to the susceptibility of the populations (Figures 1-3).

T. urticae resistance to insecticides and acaricides has been widely reported all over the world. Different *T. urticae* populations collected from cotton production areas in Turkey are reported to be resistant against dicofol (1.112-2.497), bromopropylate (<1.0-1.106) and bifenthrin (<1.0-669.120) (according to LC_{50}) (Ay, 2001). It was reported that there was a 38-fold difference between *T. urticae* populations collected from pear orchards in California in terms of resistance to cyhexatin (Tian et al., 1992), and that *T. urticae* populations collected from cotton in Australia could

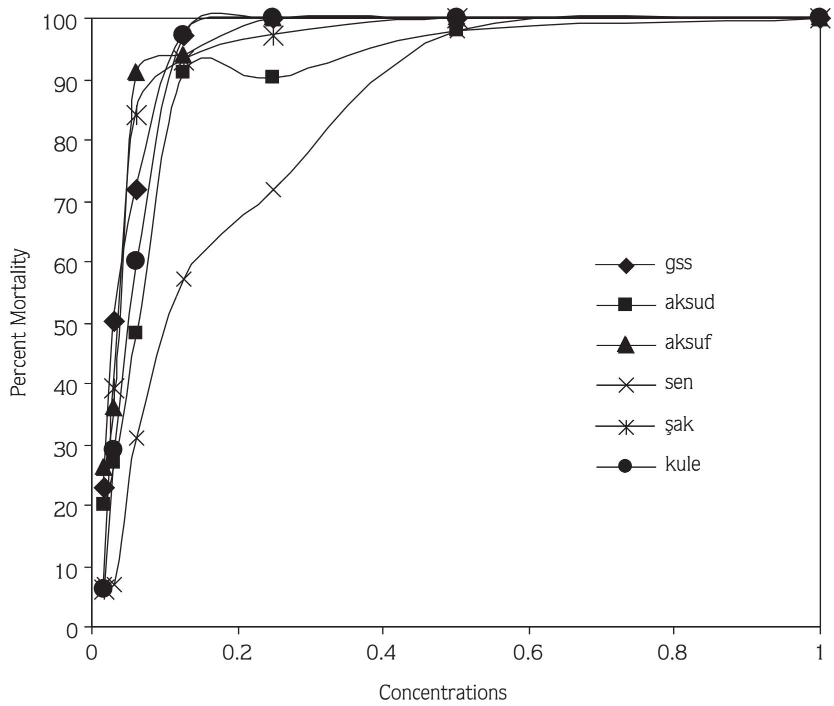

Figure 3. Dose-response for susceptible (GSS) and field-collected populations of *T. urticae* tested against abamectin (concentrations; µl 100 ml^{-1}).

develop resistance against many organophosphate preparations including dimethoate, parathion, demeton-S-methyl, monocrotophos and profenofos (Herron et al., 1998).

Resistance ratios reported in this study are low, but spider mites' high reproductive potential and short life cycle (allowing numerous generations in a growing season), combined with the frequent application of acaricides to suppress mite populations below economic thresholds, facilitate resistance development in a short period (Nauen et al., 2001). Susceptible spider mites had shorter development times and a high percentage of survival and therefore dominated the population in the absence of selection pressure from the chemicals. Moderately resistant populations reverted to susceptibility in 3 to 6 generations. This can be explained by fluctuation in some *T. urticae* populations during a season, depending on recent exposure history to acaricides. The tested populations were collected from fields where growers were unable to control *T. urticae* with those pesticides.

Unless a strong management plan is implemented, the resistance problem is likely to become even more severe. In circumstances where growers have difficulty in controlling *T. urticae*, the problem can be overcome by increasing the application rates of chemicals within sensible limits that will not cause selection pressure.

Resistance rate is related to the number of chemicals applied in a production season, and to the continuous use of preparations having the same effect mechanism (Campos et al., 1996). Among *T. urticae* populations no resistance against abamectin was found in a population that had been subjected to fewer than 6 applications per year (Campos et al., 1995).

Since commercial vegetable growing is a newly developing system around Isparta it is estimated that producers do not use many chemical preparations. From the latest observations, however, it can clearly be seen that producers are heading for a chemical preparation to control all pests. As stressed above, the resistance rate is directly related to the frequency of pesticide applications per season. In the production season, in addition to chemical control, other pest control methods must also be taken into account as much as possible, and in case of necessity chemical applications having different active ingredients should be used in each application.

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