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Original article (Orijinal araştırma)

Spinosad resistance in a population of *Frankliniella occidentalis* (Pergande, 1895) from Antalya and its cross resistance to acrinathrin and formetanate[1]

Antalya'dan alınan bir *Frankliniella occidentalis* (Pergande, 1895) popülasyonunda spinosad direnci ve bu popülasyonun acrinathrin ve formetanate'a karşı çapraz direnci

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Abstract

Frankliniella occidentalis (Pergande, 1895) (Thysanoptera: Thripidae) is a serious agricultural pest worldwide. Spinosad is used against many major pest species in Turkey, including *F. occidentalis*. However, resistance to spinosad was detected in *F. occidentalis* population collected from a greenhouse-grown peppers in Kumluca, Antalya, Turkey in 2015. This population (Antalya-2015) had been exposed to intensive, long-term spinosad application in this greenhouse. Cross resistance to acrinathrin and formetanate in the Antalya-2015 population was investigated and the stability of its resistance to spinosad was monitored over 1 year. LC values of susceptible and Antalya-2015 populations were determined using a leaf-dip bioassay and resistance ratios calculated. The resistance ratio of the Antalya-2015 population to spinosad was extremely high (up to 235-fold) and showed a 15-fold cross resistance to acrinathrin. Cross resistance to formetanate was not detected. In addition, spinetoram and spinosad were tested on the Antalya-2015 population using the recommended rates; mortalities were 88 and 38%, respectively. In the assay for stability of resistance to spinosad, mortality in Antalya-2015 population did not significantly change even after the population had been maintained in an insecticide-free environment for 1 year. Thus, resistance to spinosad in the Antalya-2015 population was stable.

Keywords: Antalya, cross resistance, *Frankliniella occidentalis*, spinosad resistance, stability

Öz

Frankliniella occidentalis (Pergande, 1895) (Thysanoptera: Thripidae) tarımda dünya çapında yaygın olan önemli bir zararlıdır. Spinosad, *F. occidentalis* türü de dahil olmak üzere Türkiye'de başlıca zararlılara karşı kullanılmaktadır. Fakat 2015 yılında Kumluca, Antalya, Türkiye'den bir biber serasından alınan *F. occidentalis* popülasyonunda spinosad'a direnç belirlenmiştir. Bu popülasyon (Antalya-2015) alındığı serada uzun süre yoğun spinosad uygulamasına maruz kalmıştır. Aynı popülasyonun acrinathrin ve formetanate'a karşı çapraz direnci araştırılmıştır ve bunun spinosad'a karşı direncinin kalıcılığı 12 aydan daha fazla bir süre boyunca izlenmiştir. Yaprak-daldırma biyoassayi ile Hassas ve Antalya-2015 popülasyonlarının LC değerleri tespit edilmiştir ve direnç oranları hesaplanmıştır. Antalya-2015 popülasyonu spinosad'a 235-kat kadar oldukça yüksek düzeyde dirençlidir ve bu popülasyon acrinathrin'e 15-kat çapraz-direnç göstermiştir. Formetanate'a çapraz direnç tespit edilmemiştir. Ek olarak, spinetoram ve spinosad etiket dozlarında Antalya-2015 popülasyonu üzerinde test edilmiştir. Bu aktif maddelerle elde edilen ölüm oranları sırasıyla %88 ve %38'dir. Spinosad direncinin kalıcılık testinde, Antalya-2015 popülasyonu insektisitlere maruz kalmaksızın 1 yıllık bir süre bekletildiğinde bile ölüm oranında 12 ay öncesine göre önemli bir değişiklik bulunmamıştır. Bu yüzden Antalya-2015 popülasyonunda spinosad direnci kalıcı haldedir.

Anahtar sözcükler: Antalya, çapraz-direnç, *Frankliniella occidentalis*, spinosad-direnci, kalıcılık

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Introduction

The western flower thrips, *Frankliniella occidentalis* (Pergande, 1895) (Thysanoptera: Thripidae), is a serious (polyphagous) pest of numerous agricultural crops worldwide, including vegetables, ornamentals, fruits, and industrial crops (Kirk & Terry, 2003; Mouden et al., 2017). *Frankliniella occidentalis* causes significant economic loss to many agricultural crops, both through its feeding and by transmission of several destructive plant viruses, including tomato spotted wilt virus (TSWV) (EPPO, 1999; Kirk & Terry, 2003).

This invasive thrips was first detected in Antalya, Turkey in 1993 (Tunç & Göçmen, 1995). Currently, crops in many regions of Turkey are infested with *F. occidentalis* (Ulubilir & Yabaş, 1996; Bulut & Göçmen, 2000; Atakan, 2003, 2008a, b; Kılıç & Yoldaş, 2004; Özsemerci et al., 2006; Sertkaya et al., 2006; Nas et al., 2007; Doğanlar & Aydın, 2009; Tekşam & Tunç, 2009; Hazır et al., 2011; Yıldırım & Başpınar, 2013). Furthermore, TSWV is still expanding its range across Turkey (Şevik, 2011; Şevik & Arlı-Sökmen, 2012; Fidan, 2016). Greenhouse production amounts to about 3.2 Mt in Antalya, making Antalya the most important center for greenhouse production in Turkey (GTHB, 2017). *Frankliniella occidentalis* is a particularly serious pest in greenhouses across coastal Antalya at almost all times of the year. *Frankliniella occidentalis* is also an important quarantine pest (EPPO, 2018). Much of the produce from greenhouses in Antalya is exported. Therefore, successful management of this pest species is critical for preventing large economic losses for agricultural exports.

Recently, the management of *F. occidentalis* and transmission of TSWV have met several obstacles. First, the number of registered insecticides for use in rotational applications against *F. occidentalis* is limited and some insecticides have become ineffective due to development of resistance in many thrips populations. Second, some crop species that were once resistant to TSWV strains have lost their resistance (Fidan, 2016). Insecticide resistance in *F. occidentalis* has become a common, worldwide problem over the last two decades, including in various parts of the USA, several European countries, Israel, Turkey, and Australia (Immaraju et al., 1992; Brodsgaard, 1994; Jensen, 1998; Kontsedalov et al., 1998; Espinosa et al., 2002; Herron & James 2005; Bielza et al., 2007; Dağlı & Tunç, 2007; Thalavaisundaram et al., 2008; Zhang et al., 2008; Gao et al., 2012). Currently, resistance against 30 active ingredients has been recorded in *F. occidentalis* populations (APRD, 2018).

Spinosad is a critically important bioinsecticide commonly used worldwide against many greenhouse insect pests, including *F. occidentalis*, *Helicoverpa armigera* (Hübner, 1808) (Lepidoptera: Noctuidae), *Spodoptera littoralis* (Boisduval, 1833) (Lepidoptera: Noctuidae) and *Tuta absoluta* (Meyrick, 1917) (Lepidoptera: Gelechiidae). Insecticide resistance management (IRM) is essential for delaying or preventing pest populations from becoming resistant to the active ingredients in insecticides (such as those in spinosad) (Croft, 1990; Soderlund & Bloomquist, 1990). The first step in developing an efficient IRM strategy is to determine the susceptibilities of pest populations to various insecticides. Furthermore, insecticide mode of action, cross-resistance spectrum and resistance mechanisms in insect populations should be investigated (Croft, 1990). Rotational use of insecticides is a major strategy to delay the resistance development in pest populations. However, to inhibit insect resistance to any insecticide, insecticide use should be rotated with other types of insecticides. However, this strategy is most-effectively implemented after determining if there are any cross resistance occurring among insecticides. The most-effective insecticides are those for which the pest population does not exhibit any cross resistance (or multiple resistance).

In a previous study (2007-2009) investigating the susceptibility of *F. occidentalis* populations in Antalya to several insecticides, it was found that only two of 10 *F. occidentalis* populations were resistance to spinosad (Dağlı et al., 2010). However, more recently, growers from many different parts of Antalya have been reporting that spinosad is becoming less effective against *F. occidentalis.* This reported increase in resistance was the impetus for this study.

In the present study, the level of resistance of an *F. occidentalis* population (Antalya-2015) to spinosad was measured. This population, collected from greenhouse-grown peppers in Kumluca (Antalya), Turkey in 2015, had been exposed to intensive, long-term spinosad. Cross resistance of the Antalya-2015 population to acrinathrin and formetanate, two other widely-used insecticides, were investigated. In addition to acrinathrin and formetanate, the efficacy of spinetoram on the Antalya-2015 population was determined at the recommended rate. Furthermore, the stability of resistance to spinosad in the same population was monitored for 1 year at two diagnostic doses. The main objective was to obtain information that could be used to develop rotational insecticide programs to better control *F. occidentalis* outbreaks.

Material and Methods

Frankliniella occidentalis populations

A population of *F. occidentalis* determined to be susceptible to spinosad was obtained in 2014. This population was collected from a bean plant in a home garden in Şuhut, Afyonkarahisar Province, Turkey (38°31'43.79"N, 30°32'42.97"E). The garden was far from fields or greenhouses, and located at an elevation of about 1000 m above sea level. In 2015, a population of *F. occidentalis* (Antalya-2015) from peppers grown in a greenhouse in Kumluca, Antalya Province, Turkey (36°21'51.95"N, 30°14'19.16"E) was collected. This population had been exposed to intensive, long-term spinosad applications. However, other active ingredients (acrinathrin, formetanate and spinetoram) had not been used in this greenhouse before collecting samples.

Insecticides

Active ingredients, commercial names and modes of action of insecticides used in this investigation are detailed in Table 1.

Table1. Active ingredients and commercial names for insecticides and their mode of action

Active ingredient	Commercial name	IRAC mode of action*
Spinosad	Laser, SC 480, Dow Agro Sciences	Nerve action, Nicotinic acetylcholine receptor (nAChR) allosteric modulators, (5).
Acrinathrin	Rufast, 75 EW, AgriNova	Nerve action, Sodium channel modulators, (3 A).
Formetanate	Dicarzol, 50 SP, AMC TARIM	Nerve action, Acetylcholinesterase (AChE) inhibitors, (1A).
Spinetoram	Radiant, 120 SC, Dow Agro Sciences	Nerve action, Nicotinic acetylcholine receptor (nAChR) allosteric modulators, (5).

* (IRAC, 2018).

Rearing of *Frankliniella occidentalis*

Frankliniella occidentalis was reared on green bean pods in a laboratory. Rearing followed the methods used in several other studies (Steiner & Goodwin, 1998; Murai & Loomans, 2001; Espinosa et al., 2002), but with some minor modifications. Transparent plastic cups (18 x 11 cm diameter) were used as thrips rearing cages (Figure 1). The lids of these cups were screened with filter paper to provide ventilation. Several layers of paper towels were placed at the base of the rearing cups to provide shelter for the thrips during their pupal stage. Green bean pods were prepared as follows before releasing thrips into the rearing cages. The pods were (1) soaked in sodium hypochlorite solution (about 6 g/L) for 2-3 min and then thoroughly rinsed with water, (2) immersed in sugar solution (about 5 g/L) for 2-3 min, and (3) dried before placing them into rearing cages. These pretreated pods were put into rearing cages before adult *F. occidentalis* were released into the cages (Figure 1). Generally, the pods in rearing cages were replaced by a fresh pod every 3-4 days. All *F. occidentalis* populations in cages were maintained in a climate-controlled room maintained at 23±1ºC and a 16:8 h L:D photoperiod.

Figure 1. Cages used to rear *Frankliniella occidentalis.*

Bioassay

A leaf-dip method was used for determining population LC values (Zhang et al., 2008). A series of serially-diluted concentrations (5-6) of spinosad, acrinathrin and formetanate were prepared in distilled water with TritonX-100 solution. These concentrations of each insecticide caused 0 to 100% mortality in the tested populations. Additionally, spinetoram was tested, but only at its recommended rate (60 mg a.i./L). Both susceptible and Antalya-2015 populations had been reared in an insecticide-free environment for 1 year. Two diagnostic doses of spinosad, the recommended rate and one tenth of that rate, were used to determine the stability of spinosad resistance in the Antalya-2015 population. Discs (3 cm diam.) were excised from leaves of potted bean plants and dipped (5 s) in one of the various insecticide concentrations or in water (control treatment) mixed with TritonX-100 solution. After drying, the treated leaf discs were placed in Petri dishes that were prepared as follows. (1) Agar (1.5%) was prepared in distilled water and boiled in a microwave, (2) after cooling for 30 min, the agar was poured into 3-cm diam. plastic Petri dishes, and (3) the treated leaf discs were embedded in that agar. Adult female thrips were then collected from rearing cages with a mouth aspirator, anesthetized with CO_2 and transferred to the leaf discs. The Petri dishes were covered with perforated plastic wrap to prevent the thrips from escaping but allow ventilation (Figure 2). Three replicates of each insecticide concentration was prepared. Fifteen to 30 thrips were used in each replicate. Mortality was recorded after 48 h with the aid of a microscope. Thrips were recorded as dead if they did not respond when prodded with a pin or brush.

Figure 2. The insecticide bioassay test cells for *Frankliniella occidentalis* (top view).

Data analysis

Probit analysis was applied to the number of alive and dead thrips (PoloPlus, 2002-2009 LeOra Software, Petaluma, CA, USA) to determine the LC values for the populations. Resistance ratios for spinosad, acrinathrin and formetanate were calculated as LC_{50} values of the Antalya-2015 population divided by the LC_{50} values for the susceptible population. Mortality data obtained from spinetoram bioassays were corrected using the Abbott formula (Abbott, 1925). Mortalities related to stability of spinosad resistance were likewise corrected. Mortalities from the first and the second measurement in resistance stability bioassays were analyzed using Pearson's chi-squared test.

Results

Mortalities of thrips in control treatments were less than 6.5% in all bioassays. LC_{50} values of susceptible and Antalya-2015 populations for spinosad, acrinathrin and formetanate are given in Tables 2, 3 and 4, respectively. Resistance ratios for thrips in the Antalya-2015 population to these insecticides (at LC_{50}) are also presented in these tables. Resistance of the Antalya-2015 population to spinosad, acrinathrin were 235- and 15-fold higher, respectively, than that of the susceptible population. No resistance to formetanate was recorded. LC_{90} values of the Antalya-2015 population for spinosad and acrinathrin were also higher than at the recommended rate for these insecticides. However, LC_{90} values for the Antalya-2015 population for formetanate were lower than at its recommended rate.

Table 2. The resistance ratio (at LC_{50}) to spinosad in susceptible and spinosad-resistant (Antalya-2015) populations of *Frankliniella occidentalis*

Populations	n*	Slope±S.E	LC_{50} mg (a.i.)/L (95% CL)	Resistance Ratio**	LC_{90} mg (a.i.)/L (95% CL)	***Registered rate mg(a.i.)/L
Susceptible	497	1.4±0.2	0.6 0.4-1.0	-	5.4 3.2-10.6	96
Antalya-2015	658	1.4±0.2	141.0 96.0-238.0	235	1216.5 555.9-6503.4	96

* n: number adult female thrips used in bioassay;
** Resistance ratio: LC_{50} of the Antalya-2015 population / LC_{50} of the susceptible population;
*** (GTHB,2018);
Highest mortality in control treatments was 4.8%.

Table 3. The resistance ratio (at LC_{50}) to acrinathrin in susceptible and spinosad-resistant (Antalya-2015) populations of *Frankliniella occidentalis*

Populations	n*	Slope±S.E	LC_{50} mg (a.i.)/L (95% CL)	Resistance Ratio	LC_{90} mg (a.i.)/L (%95% CL)	***Registered rate mg (a.i.)/L
Susceptible	484	1.2±0.1	1.8 0.7-3.5	-	19.5 10.9-38.9	60
Antalya-2015	278	1.5±0.2	26.5 11.6-46.8	14.7	191.5 101.3-614.9	60

* n: number adult female thrips used in bioassay;
** Resistance ratio: LC_{50} of the Antalya-2015 population / LC_{50} of the susceptible population;
*** (GTHB,2018);
Highest mortality in control treatments was 4.7%.

Table 4. The resistance ratio (at LC_{50}) to formetanate in susceptible and spinosad-resistant (Antalya-2015) populations of *Frankliniella occidentalis*

Populations	n*	Slope±S.E	LC_{50} mg (a.i.)/L (95% CL)	Resistance Ratio	LC_{90} mg (a.i.)/L (95% CL)	***Registered rate mg(a.i.)/L
Susceptible	436	2.0±0.3	14.2 6.5-32.7	-	62.0 28.3-612.5	500
Antalya-2015	608	1.3±0.1	16.0 8.5-26.6	1.1	147.3 85.6-305.7	500

* n: number adult female thrips used in bioassay;
** Resistance ratio: LC_{50} of the Antalya-2015 population / LC_{50} of the susceptible population;
*** (GTHB,2018);
Highest mortality in control treatments was 4.2%.

Mortality ratios from with spinetoram and spinosad bioassays are shown in Table 5. The efficacy of spinetoram on the Antalya-2015 population was determined only at its recommended rate. Both insecticides caused 100% mortality to the susceptible population of *F. occidentalis* at their recommended rates. However, mortality in the Antalya-2015 population caused by spinetoram and spinosad were lower: 88 and 38%, respectively (Table 5). Mortality-related stability of spinosad resistance in the Antalya-2015 population is provided in Table 6.

Two diagnostic doses of spinosad, the recommended rate and on tenth of that rate, were used to determine the stability of spinosad resistance in the Antalya-2015 population. In this population, the recommended rate of spinosad caused 30% mortality, whereas the tenth rate caused only 2% mortality. For the Antalya-2015 population that had been reared in an insecticide-free environment for 1 year, mortalities at these two diagnostic doses were 39% (recommended rate) and 5% (one tenth rate), not much different than when the population had not been reared in an insecticide-free environment (Table 6). Therefore, the reversion (i.e., dilution) rate of spinosad resistance in the Antalya-2015 population was quite low (2-9%).

Table 5. Mortality ratios for susceptible and spinosad-resistant (Antalya-2015) populations of *Frankliniella occidentalis*in bioassays with spinosad and spinetoram

Populations	Spinosad recommended rate: 96 mg a.i./L		Spinetoram recommended rate: 60 mg a.i./L	
	n*	Mortality (%)	n*	Mortality (%)
Susceptible	74	100	66	100
Antalya-2015	63	38	69	88

* n: number adult female thrips used in bioassay;
Highest mortality in control treatments was 5.8%.

Table 6. Stability of spinosad resistance in susceptible and spinosad-resistant (Antalya-2015) populations of *Frankliniella occidentalis*

Spinosad rates	Populations	First measurement (first generation)		Second measurement (12 months later)	
		n*	Mortality (%)	n*	Mortality (%)
9.6 mg a.i./L	Susceptible	52	100.0	80	94.4
	Antalya-2015	154	1.6	121	5.1
96 mg a.i./L	Susceptible	55	100.0	109	100.0
	Antalya-2015	138	29.6	214	39.2

* n: number of adult female thrips used in bioassay;
Mortality ratios obtained from second measurements were not significantly different from first measurement, (Pearson's chi-squared test, P>0.05);
Highest mortality in control treatments 6.5%.

Discussion

The susceptible *F. occidentalis* population used in this study was highly susceptible to the insecticides tested. The LC_{90} values of susceptible population for spinosad, acrinathrin and formetanate (5.4, 19.5 and 62 mg a.i./L) were much lower than the recommended rates of those insecticides (96, 60 and 500 mg a.i./L). Furthermore, this susceptible population was also sensitive to spinetoram. The recommended rates of spinetoram (60 mg a.i./L) caused 100% mortality in this susceptible population. The existence of such susceptible populations in nature is needed to provide dilution in resistance and is critically important for monitoring changes in insecticide resistance in greenhouse or field populations.

The *F. occidentalis* population Antalya-2015 was collected from a greenhouse that had been treated with frequent spinosad applications over a long period of time. As expected, a high level of spinosad resistance was detected in this population, which was up to 235 times more resistant to spinosad than the susceptible population. Dağlı et al. (2010) had previously documented resistance of *F. occidentalis* populations in Antalya to spinosad; however, the 235-fold resistance detected in present study was much higher than the previous finding. The LC_{90} value of spinosad for the Antalya-2015 population (1217 mg a.i./L) was much higher than the recommended rate (96 mg a.i./L). The recommended rate of spinosad caused 100% mortality in the susceptible population, whereas mortality was only 30% in the Antalya-2015 population. Thus, spinosad would clearly be ineffective in controlling the Antalya-2015 population. This level of resistance is likely to be the main reason spinosad could not control *F. occidentalis* infestations under field conditions.

Resistance of *F. occidentalis* to spinosad has also been reported in other countries, including Spain, Australia, Japan and China (Herron & James 2005; Bielza et al., 2007; Zhang et al., 2008; Gao et al., 2012). In the case of the population in Murcia, Spain, resistance to spinosad was low in 2001-2002 because it had not been used previously in this region, however, after intensive application of spinosad, resistance had become 3682-fold higher by 2004 (Bielza et al., 2007).

Insecticide resistance in *F. occidentalis* is not limited to spinosad. In fact, Gao et al. (2012) has reported that *F. occidentalis* has significant potential to develop resistance to many other active ingredients used in insecticides. Resistance in *F. occidentalis* has been recorded worldwide against 30 different active ingredients used in insecticides, including a variety of well-known and often-used insecticides, including spinosad, spinetoram, cypermethrin, deltamethrin, acrinathrin, abamectin, chlorpyrifos, malathion, methiocarb and formetanate (Gao et al., 2012; ARDB, 2018).

Spinosad is a valuable bioinsecticide for pest management because it is highly effective in controlling many important pest species and is not very toxic to mammals in low doses. Currently, the most-widely recommended registered insecticides against *F. occidentalis* in Antalya greenhouses are spinosad (spinosyn), acrinathrin (pyrethroid), formetanate (carbamate) and spinetoram (spinosyn). It was shown that the Antalya-2015 population is highly resistant to spinosad, and has moderate cross resistance to acrinathrin (15 times higher than the susceptible population) but has no cross resistance to formetanate. Therefore, acrinathrin would not be a suitable alternative for insect infestations that have already acquired some cross resistance (such as the spinosad-resistant *F. occidentalis* population used in this study). In contrast, formetanate would likely provide an effective alternative for resistant insect populations because in the Antalya-2015 population it had high efficacy and no cross resistance. Probably, the mechanisms of resistance to spinosad and formetanate were not the same in the spinosad-resistant *F. occidentalis* population. In two different studies, resistance to spinosad in *F. occidentalis* population was detected as non-metabolic (Bielza et al., 2007; Zhang et al., 2008). Similarly, Shono & Scott (2003) reported that metabolic detoxification enzymes (monooxygenases, hydrolases or glutathioneS-transferases) were not involved in spinosad resistance in house fly, *Musca domestica* L., 1758 (Diptera: Muscidae). In contrast, metabolic enzymes such as esterase and GST were determined to be a major mechanism of resistance to carbamate and organophosphate insecticides in *F. occidentalis* populations (Jensen, 1998; Jensen, 2000; Maymo et al., 2002). Therefore, the efficacy of formetanate on the spinosad-resistant population possibly had not been affected by the spinosad resistance mechanism. Similarly, spinosad resistance was not associated with cross resistance to formetanate in *F. occidentalis* populations in Spain (Bielza et al., 2007).

In present study, the efficacy of spinetoram on the Antalya-2015 population was evaluated on the basis of its recommended rate. Spinetoram has the same mode of action as spinosad (IRAC, 2018). However, spinosad was registered in 2003 in Turkey, whereas the registration date for spinetoram was 2014 (Tosun & Onan, 2014). In present study, both spinetoram and spinosad caused 100% mortality in the susceptible population of *F. occidentalis*, but spinetoram showed higher efficacy than spinosad in the Antalya-2015 population (88% mortality for spinetoram vs. 38% mortality for spinosad). Considering these differences in mortalities, cross resistance to spinetoram in the spinosad-resistant population was low. However, more detailed data are needed on LC values to determine degree of cross resistance between spinetoram and spinosad in the Antalya-2015 population.

In the assay of the stability of spinosad resistance, mortality rate in Antalya-2015 population did not significantly change after it had been maintained in an insecticide-free environment for 1year. Therefore, its resistance to spinosad was stable (i.e., it is unlikely to revert to a spinosad-susceptible population). Several factors might help maintain stability of spinosad resistance in this population. Initial gene frequencies, mode of resistance inheritance, the fitness cost and immigration of susceptible insect are the major effective factors in resistance stability (Bielza et al., 2008). Based on the results from the present study, the effect of two factors on the resistance stability could be possible. (1) Initial resistant gene frequency in the Antalya-2015 population was quite high with almost the entire population being resistant individuals because a diagnostic dose of spinosad (9.6 mg a.i./L) causes 100% mortality in the susceptible population. However, the same dose caused only 1.6% mortality in spinosad-resistant population. (2) Dilution of resistance might not be possible in Antalya-2015 population because it was maintained in rearing cages. Therefore, immigration of susceptible thrips from the outside was not possible. Additional genetic investigation of the susceptible and resistant populations, such as reciprocal crosses and backcross studies, will be necessary to determine the mode of inheritance and the number of effective genes conferring resistance to these insecticides. Similar results for resistance stability of *F. occidentalis* have been reported from Spain, where resistance remained high and stable for 8 months, until 2 months after susceptible individuals migrated into the resistant population, after which resistance declined (Bielza et al., 2008). This suggests that spinosad resistance may be diluted in greenhouses by *F. occidentalis*-susceptible individuals that migrate from a nearby environment. Therefore, the frequency of insecticide application in any given greenhouse should be reduced to lower the risk of pest populations developing resistance to pesticides. Otherwise, when pesticide resistance develops in a greenhouse population, reduction of resistance will be extremely difficult without purposively introducing susceptible populations to dilute the gene pool of pest species. Therefore, all resistance management strategies should be implemented before resistance in pest populations becomes evident. Otherwise, many active ingredients suitable for integrated pest management, such as those in spinosad, will become useless when pests become resistant to a large variety of pesticides.

Given the high level of resistance to spinosad found in the investigated population of *F. occidentalis*, the prevalence of spinosad resistance in *F. occidentalis* populations should be determined for more populations in greenhouses in Antalya and across Turkey. In addition to spinosad, other registered, commonly used active ingredients should be investigated for pest resistance and insecticide rotational strategies should be updated. However, relying only insecticides may not be adequate to efficiently control *F. occidentalis* and the TSWV it transmits. The frequency of insecticide application of spinosad should be reduced to provide for a more sustainable management of pests using more efficient insecticides. Even more importantly, cultural, biological and other alternatives should be integrated into pest management strategies.

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