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Research article

Toxicological Effects of Selected Insecticides on *Delphastus catalinae* (Horn) (Coleoptera, Coccinellidae), a Predator of *Bemisia tabaci* Genn (Homoptera, Aleyrodidae)

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Abstract. Toxicological effects of selected insecticides, fenpropathrin+pyriproxifen, acetamiprid, diafenthiuron, pyriproxifen, and chlorfenapyr were tested by direct spray and dry film methods in the laboratory against adult and larvae (L_3-L_4) of *Delphastus catalinae* (Horn) (Col., Coccinellidae), a predator of whiteflies, including cotton whitefly, *Bemisia tabaci* (Genn.). Water was used as untreated control. Fenpropathrin+pyriproxifen, acetamiprid, and diafenthiuron were harmful to larvae and adults of *D. catalinae* in both methods. Pyriproxifen was to be seemed harmless to adults in direct spraying technique; however it was harmful to the adults in dry film method. Eggs laid by a few adults which developed pyriproxifen treated larvae failure to hatch in dry film method. In conclusion, chlorfenapyr was innocuous to larvae and adults of *D. catalinae* in both methods. Fenpropathrin+pyriproxifen, acetamiprid, diafenthiuron and pyriproxifen would be incompatible with biological control of whitefly by *D. catalinae*.

Seçilmiş İnsektisitlerin Beyazsinek, *Bemisia tabaci* Genn (Homoptera, Aleyrodidae)'nin Avcısı, *Delphastus catalinae* (Horn) (Coleoptera, Coccinellidae) Üzerindeki Toksikolojik Etkileri

Anahtar kelimeler:
Delphastus catalinae, insektisitler, toksikolojik etki

Özet. Fenpropathrin+pyriproxifen, acetamiprid, diafenthiurion, pyriproxifen ve chlorfenapyr etkili maddelerini ihtiva eden seçilmiş insektisitlerin toksikolojik etkileri laboratuvar koşullarında Pamuk beyazsineği, *Bemisia tabaci* (Genn.)'nin avcısı *Delphastus catalinae* (Horn) (Col., Coccinellidae)'nın larva (L_3-L_4) dönemlerine karşı kuru film ve püskürtme yöntemleriyle test edilmiştir. Saf su püskürtülmesi şahit kabul edildi. Fenpropathrin+pyriproxifen, acetamiprid ve diafenthiuron'un her iki yöntemde avcı, *D. catalinae*'nın larva ve ergin dönemlerine zararlı olduğu, Pyriproxifen'in ise püskürtme yöntemiyle zararsız ancak kuru film yöntemiyle erginlere zararlı olduğu belirlendi. Pyriproxifen'in kuru film yöntemi uygulamasında canlı kalan az sayıda *D. catalinae* erginlerinin bırakmış oldukları yumurtalar açılmalarına rağmen yumurtadan çıkan larvalar ergin dönemlerine ulaşamadılar. Sonuç olarak chlorfenapyr'in her iki yöntemde *D. catalinae*'nin ergin ve larvalarına zararsız olduğu, buna karşılık fenpropathrin+pyriproxifen, acetamiprid, diafenthiuron ve pyriproxifen etkili maddelerini ihtiva eden insektisitlerin beyazsineklerin *D. catalinae* ile yürütülen biyolojik mücadele programında kullanılamayacağı ortaya konmuştur.

INTRODUCTION

Sweet potato whitefly, *Bemisia tabaci* (Genn.) is still one of the most important cotton pests in the East Mediterranean region of Turkey. Although population levels of *B. tabaci* have been changing year by year especially in irrigated cotton fields, since the major outbreak in 1974, in the East Mediterranean Region of Turkey (Kaygısız 1976; Tunc *et al*., 1983; Anonymous 1994; Şekeroğlu *et al*., 1998). Damage to the plant is caused by reduction in plant vigor and production of honeydew on which sooty molds develop. *B. tabaci* also transmits some plant pathogenic viruses (Lodos 1982). In the East Mediterranean Region of Turkey, estimated crop loss caused by *B. tabaci* in irrigated and rain fed-cotton growing areas are 40% and 15%, respectively (Şekeroğlu *et al*., 1998).

In Turkey, a lot of studies were carried out on the biology and management of *B. tabaci* in cotton fields (Kaygısız 1976; Stam and Tunc 1983; Kişmir 1983; Şekeroğlu *et al*., 1998). Importance of sustainable integrated whitefly management, combining optimally all available tactics to maintain whitefly populations below levels that will cause economic loss, has been increasing (Ellsworth *et al*., 1995). Although a lot of natural enemies of *B. tabaci*, such as *Eretmocerus mundus* Mercet, *Encarsia* sp., *Prospeltella* sp.nr.*aspiticola* M., *Chrysoperla carnea* (Steph), *Nabis pseudoferus* RM, *Geocoris* spp., *Orius* spp. and *Deraecoris* spp. were found in the East Mediterranean Region of Turkey, they are not enough to suppress the whitefly populations (Kismir 1983; Kaygisiz 1976; Mart *et al*., 1995; Sekeroglu *et al*., 1998), because of broad-spectrum insecticide applications. Heinz *et al*. (1994) suggested that releases of *Delphastus catalinae* (Horn) (Col., Coccinellidae) into *B. tabaci* exclusion cages resulted in a 55 % and a 67% decrease in the whitefly densities and has potential to suppress the pest in open cotton fields. All member of the genus *Delphastus* spp. are known as predators of the whitefly species. *D. catalinae* is widely distributed across the central and southern United States (Gordon 1994). Therefore, we introduced *D. catalinae*, as an alternative predator of sweet potato whitefly, *B. tabaci* into the East Mediterranean region of Turkey.

Integration of biological and chemical control requires knowledge of side-effects of insecticides on natural enemies. Hoelmer *et al*. (1994) found that *D. pusillus* adults were not affected with 0.3% azadirachtin for two weeks and females feeding on treated whiteflies for several days continued to lay eggs.

In this study, we evaluated toxicological effects of selected insecticides, advised in IPM programs for cotton pests in Turkey to adult and larvae of *D. catalinae*.

MATERIALS AND METHODS

Rearing of Delphastus catalinae

For culturing the colony, approximately 15-20 individuals of *D. catalinae* were provided by Texas Agricultural Experiment Station, Texas A&M University, USA. The predatory insect was reared on heavily *B. tabaci*-infested cotton, *Gossypium hirsutum* L., by the same method for culturing *Serangium parcesetosum* Sicard, another predator of *B. tabaci* (Yigit 1992). Sweetpotato whitefly, used regular prey supply was reared on cotton. Cottonseeds were sown in soil in plastic pots (18 cm diameter) and maintained in a glasshouse until the seedlings reached a height of 30 cm. They were then transferred to a constant temperature room at 27±2 °C under 16 h illuminations and 70±10% R.H. and placed next to the plants infested with *B. tabaci*. These plants were kept in this room for 2 or 3 weeks to obtain sufficient prey density (25 eggs or larvae + pupa per cm^2 of leaf area). Twelve heavily infested plants (four pots, each with three-four plants) were placed in 50 x 110 x 80 cm growth cages the sides of which were covered with the cheese-cloth and the top with a glass pane. The cages were maintained in a constant temperature room at 25±1 °C under 16 h illumination and 70±10% R.H. Mixed sexes of *D. catalinae* adults were introduced in cage (15-20 adults per cage) containing cotton plants heavily infested with *B. tabaci*.

Insecticides

Commonly used insecticides, taken place in cotton-IPM program for *B. tabaci* control were tested to reveal the toxicological side-effects on the predatory insect. Tap water was used as untreated control (Table 1).

Bioassay

Ten days old adults and larvae (L_3: third and L_4: fourth instars) of *D. catalinae* were used for bioassay to determine the toxicological effects of the pesticides by direct spraying and dry film methods.

Direct spray method (Topical spray method)

Ten predator larvae or adults from the culture were carefully placed using a fine brush on cotton leaf containing 200-300 whitefly larvae and/or pupae lower surface facing up on moistened filter paper in a

glass Petri dish (9 cm x 1.7 cm). The petri dishes were covered with cheesecloth. Application of pesticide solutions at the recommended field concentrations was sprayed to cotton leaf by a hand sprayer until run off, covered as 2 mg/cm^2 leaf surface (Yiğit *et al*., 1992). Each insecticide solution was prepared with tap water at the recommended field dosage that would be applied at the spray volume of 35 liter solution per 0.1 ha (Kismir and Sengonca 1980). Every insecticide was applied by a different hand sprayer.

Dry film method

This method was modified from Brun (1985 and 1988). Inside of Petri dishes was sprayed by the hand sprayer mentioned above as coverage of two ml of pesticide solutions per cm^2 at recommended field dosage. The amount of wet solution received on petri dish was measured by weighing the dish before and immediately after spraying. Petri dishes were allowed to dry for 2 h and then ten adults or larvae were placed inside the Petri dishes with heavily *B. tabaci* infested cotton leaf face down. Water- treated Petri dishes were used as untreated control.

In both methods, Petri dishes were covered with the cheesecloth for ventilation. New cotton leaves, heavily infested with whitefly larvae and/pupae were added in Petri dishes to feed the predator larvae or adults, whenever needed. Ten larvae or adults were used in each treatment with four replications. The mortality data was recorded by 24 h intervals after the exposure of beetles until 120th h of the treatment.

The number of reaching the adult stage of the surviving larvae was recorded and analyzed (ANOVA). The test ended after emergence of the first-laid eggs and the eggs were observed whether hatched or not. Bioassay were conducted in a laboratory at 25±2 ºC, 16:8 (L: D) h photoperiod, and 70±10 % RH.

Treatment effects were analyzed after 120 h using analysis of variance (ANOVA), and means were separated using the Duncan's multiple comparison test following a significant F-test.

Total effect (E) % of the pesticides on the predator was calculated after 120 h, according to Abbott (1925) formula:

$E\% = 100 \times (n_t - n_c) / n_t$

where n_t: is the number of surviving adults or larvae in treated group, n_c: is the number of surviving adults or larvae in untreated group.

The pesticides were classified into four categories depending on the degree of damage (Total effect E %), caused to adults and larvae of *D. catalinae*: 1=harmless (<30%), 2=slightly harmful (30-79%), 3=moderately harmful (80-99%) and 4=harmful (> 99%) (Hassan *et al*., 1994).

RESULTS

Direct Spray Method (Topical spray)

Significant differences were found in mortality of *D. catalinae* adults and larvae 120 h exposure by direct spraying method. In adult treatment, fenpropathrin+pyriproxifen, acetamiprid, and diafenthiuron caused high levels mortality. Surviving adults (females) treated with pyriproxifen and chlorfenapyr laid eggs which hatched like as untreated plots (Table 2).

Fenpropathrin+pyriproxifen, acetamiprid and diafenthiuron were also caused high mortality to larvae of *D. catalinae*. A few larvae treated with diafenthiuron reached to adult stage, but they died before laying eggs. However, at pyriproxifen treated larvae, the mean mortality was low, there was no larvae reaching adult stage. Chlorfenapyr was almost harmless to larvae based on IOBC scale and most of them reached the adult stage which laying eggs. The eggs hatched like laid by adults, developed from the untreated larvae (Table 2).

Table 1. Insecticides tested on *Delphastus catalinae*, a predatory insect of Sweet potato whitefly, *Bemisia tabaci*.
Çizelge 1. Delphastus catalinae'ye denenen bazı pestisitler ile bunların uygulama dozları.

Commercial name	Common name	Formulation and ratio of active ingredient (%)	Application dosage (g, ml 0.1 ha^{-1})	
			Active ingredient	Preparation
Prempt	Fenpropathrin+Pyriproxifen	EC 150+50	15+5	100
Mospilan	Acetamiprid	SL 20	0.6	30
Polo	Diafenthiuron	WP 50	40	80
Admiral	Pyriproxifen	EC 100	5	50
Pirate	Chlorfenapyr	SC 360	28.8	80

Dry Film Method (Residual toxicity)

The data revealed that residual toxicity of insecticides on *D. catalinae* adults and larvae, exposed throughout 120 h was significantly different among the insecticides and untreated control (Table 2). In adult treatment, mortality was very high on fenpropathrin+pyriproxifen, acetamiprid, diafenthiuron and pyriproxifen plots. Chlorfenapyr was the slightly harmful to adults. Surviving adults, treated by chlorfenapyr laid eggs which they hatched like as the untreated adults (Table 3).

In larvae treatment, however, the mortality of diafenthiuron was less than that of fenpropathrin+pyriproxifen and acetamiprid throughout 120 h exposure. Although a few larvae treated with diafenthiuron and fenpropathrin+pyriproxifen reached to adult stage, they died before laying eggs. No larvae reached to adult stage treated with acetamiprid. Pyriproxifen and chlorfenapyr were taken place in the same group based on mean dead larvae throughout 120 exposure to residue, however the number of reaching to adult stage was higher on chlorfenapyr treated larvae than that of pyriproxifen plots. On the other hand, reaching to adults from pyriproxifen treated larvae could not lay eggs, while reaching to adults from chlorfenapyr treated larvae laid eggs, like as untreated plots (Table 3).

DISCUSSION

Fenpropathrin+pyriproxifen, acetamiprid and diafenthiuron were the most detrimental insecticides tested to *D. catalinae*. Similar effects could be expected from many relatively broad-spectrum insecticides. Michaud and Grant (2003) found that the toxicity to coccinellids was generally highest for carbamates (with the exception of methomyl), followed by pyrethroids, including fenpropathrin and organophosphates. Survival induce of Coccinellid species on fenpropathrin treatment at field recommended rate was very low (lower than 1%). Also they suggested that the test on a single species could be sufficient for predicting general insecticide susceptibility within the family Coccinellidae.

Pyriproxifen was almost harmless to adult insects by direct spraying method**;** however it was harmful in dry film method, based on IOBC scale. Also pyriproxifen was harmless or slightly harmful to larvae in both testing methods throughout 120 h exposure. A few larvae reached to adult stage in dry film method, but laid eggs could not hatch, and no adults were developed from direct spraying method. This result explains IGR effects of pyriproxifen on the coccinellid. On the other hand**,** Magagula and Samways (2000) suggested that immediate larval mortality of *Chilocorus nigrita* (Fabricius), a coccinellid predator of California red scale, *Aonidiella aurantii* (Maskell) from pyriproxyfen was not significantly different from untreated plots. They reported that none of the larvae, fed with pyriproxifen-treated *A. aurantii* were pupated. Fecundity of *C. nigrita* was not affected by exposure to IGRs, either in the laboratory or in the field, but all eggs exposed to IGRs failed to hatch. Although larvae developed to the adult stage in the field experiments, the IGRs' ovicidal activity and effects on immature stages still had a detrimental effect on *C. nigritus* populations.

Chlorfenopyr was almost harmless to adults and/or larvae of *D. catalinae* in both methods, with the exception of slightly harmful to adults in dry film method. Pietrantonia and Benedict (1997) also found that chlorfenopyr is slightly harmful to *Orius insidousus* (Say), a general predator in cotton fields.

CONCLUSION

Chlorfenopyr appears to be compatible with biological control of whitefly by *D. catalinae* and could become a useful component of *B. tabaci* control and similar pests where coccinellids taken place as significant contributors to the bio-control practices.

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Table 2. Mean mortality of adults and larvae (L_3-L_4) of *Delphastus catalinae* on cotton leaf throughout 24 h, 48 h, 72 h, 96 h and 120 h exposure to insecticides by direct spraying method under laboratory conditions*.

Çizelge 2. Pamuk tarlasında kullanılan bazı pestisitlerin püskürtme yöntemiyle Delphastus catalinae'nın ergin ve larvalarına laboratuvar koşullarında etkileri.

Insecticides	Adult									Larvae									
	Mean mortality.....h exposure to insecticides					Total effect after 120 h (%)	IOBC	Eggs laid by the surviving adults	Hatch of eggs laid by the surviving adults	Mean mortality.....h exposure to insecticides					Total effect After 120 h (%)	IOBC	Adults recovered from the treated larvae	Eggs laid by the adults	Hatch of eggs laid by the adults
	24 h	48 h	72 h	96 h	120 h					24 h	48 h	72 h	96 h	120 h					
Fenpropathrin+pyriproxifen	10.00	10.00	10.00	10.00	10.00c	100.00	4	-	-	10.00	10.00	10.00	10.00	10.00e	100.00	4	0.00d	-	-
Acetamiprid	10.00	10.00	10.00	10.00	10.00c	100.00	4	-	-	10.00	10.00	10.00	10.00	10.00e	100.00	4	0.00d	-	-
Diafenthiuron	2.00	5.75	9.25	9.25	9.25 c	92.50	3	-	-	0.50	5.50	6.25	8.25	8.50d	84.00	3	1.25c	-	-
Pyriproxifen	0.00	1.25	1.50	2.00	2.00 b	20.00	1	+	+	0.75	2.25	3.25	3.5	4.00c	36.03	2	0.00d	-	-
Chlorfenapyr	1.25	2.75	3.00	3.00	3.00b	30.00	1	+	+	0.00	0.75	2.00	2.00	2.00b	14.70	1	8.00b	+	+
Untreated	0.00	0.00	0.00	0.00	0.00 a			+	+	0.00	0.25	0.62	0.62	0.62a			9.12a	+	+

***** Means in columns followed by different letters indicate significant differences among insecticides tested at p≤ 5 % (ANOVA).

+Means indicate that the surviving adults lay eggs and these eggs hatch.

-Means indicate that the surviving adults could not lay eggs and these eggs could not hatch.

Table 3. Mortality of larvae (L_3-L_4) and adults of *Delphastus catalinae* on cotton leaf 24 h, 48 h, 72 h, 96 h and 120 h after treatment with insecticide by dry film method under laboratory conditions*.

Çizelge 3. Pamuk tarlasında kullanılan bazı pestisitlerin kuru film yöntemiyle Delphastus catalinae'nın ergin ve larvalarına laboratuvar koşullarında etkileri.

Insecticides	Adult									Larvae									
	Mean mortality.....h exposure to insecticides					Total effect after 120 h (%)	IOBC	Eggs laid by the surviving adults	Hatch of eggs laid by the surviving adults	Mean mortality.....h exposure to insecticides					Total effect after 120 h (%)	IOBC	Adults recovered from the treated larvae	Eggs laid by the adults	Hatch of eggs laid by the adults
	24 h	48 h	72 h	96 h	120 h					24 h	48 h	72 h	96 h	120 h					
Fenpropathrin+pyriproxifen	9.25	10.00	10.00	10.00	10.00c	100.00	4	-	-	5.25	9.25	9.25	9.25	9.25c	91.95	3	0.75c	-	-
Acetamiprid	9.50	9.50	9.50	9.50	9.50c	94.39	4	-	-	10.00	10.00	10.00	10.00	10.00c	100.00	4	0.00c	-	-
Diafenthiuron	2.50	5.25	7.25	7.50	9.00c	88.78	3	-	-	1.50	3.25	4.50	4.50	5.00b	42.85	2	0.75c	-	-
Pyriproxifen	4.50	9.25	9.75	10.00	10.00c	100.00	4	-	-	0.25	0.50	2.50	2.50	3.00ab	20.00	1	3.25b	+	-
Chlorfenapyr	1.50	3.00	4.00	4.75	4.75b	41.14	2	+	+	0.00	0.75	1.50	2.25	3.25ab	22.85	1	5.50ab	+	+
Untreated	0.66	0.91	1.08	1.08	1.08a			+	+	0.13	0.37	0.87	1.00	1.125a	-	-	7.125a	+	+

***** Means in columns followed by different letters indicate significant differences among insecticides tested at p≤ 5 % (ANOVA).

+Means indicate that the surviving adults lay eggs and these eggs hatch.

-Means indicate that the surviving adults could not lay eggs and these eggs could not hatch.

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