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Residual Toxicity of Abamectin, Milbemectin and Chlorfenapyr to Different Populations of Two Spotted Spider Mite, Tetranychus Urticae Koch, (Acari: Tetranychidae) on Cucumber in Jordan

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Abstract: Laboratory studies were conducted during the year 2010 to evaluate the duration of residual toxicity of the acaricides; abamectin, milbemectin and chlorfenapyr to six populations of the two spotted spider mite (Tetranychus urticae Koch, (Acari: Tetranychidae) collected from cucumber grown under plastic houses in the six main cultivated areas in Jordan (Ramta, Baqa', Zyzya, Krimeh, Dier Alla and Karamah), compared to a Syrian susceptible strain. Loss of susceptibility in the six populations to the closely related acaricides abamectin and milbemectin were clearly indicated even after one day of application, where the mortality did not exceed 24 and 33%, compared to 96 and 97% for Syrian susceptible strain for the two acaricides, respectively. In contrast, mortality from residual toxicity of chlorfenapyr were considered efficacious after one day of application and reached acceptable level even after 15 days of application. The mortality was ranged between 92 - 98% after one day of application and 48 - 68% after 15 days of application, in four out of six populations. The mite populations collected from Karamah and Krimeh were the least susceptible. Mortality from residual toxicity after one day of application were 75 and 88%, respectively, compared to 98% mortality in the Syrian susceptible strain.

Key words: Residual toxicity · Acaricides · Abamectin · Milbemectin · Chlorfenapyr · Tetranychus urticae · Plastic houses · Cucumber · six population · Jordan · Syrian susceptible strain

INTRODUCTION

The two-spotted spider mite, *Tetranychus urticae* Koch (Acari: Tetranychidae), is a cosmopolitan and extremely polyphagous species with great economic importance for crops in green houses and in the open fields. It can feed on hundreds of plants, including most vegetables and food crops including cucumber, peppers, tomatoes, potatoes, beans, corn, strawberries; and ornamentals such as roses. 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 sever infestations [1]. Greenhouses are ideal areas for *T. urticae* which can complete a generation in one week in suitable areas [2]. In Jordan, the total number of plastic-houses, which were planted with vegetables, was about 78800. More than fifty percent of the total number was planted with cucumber, which is considered as economic important crop, commercially produced in six regions in Jordan [3]. Two spotted spider mite represents one of the most important real threats to cucumber plantation under plastic-houses in Jordan. It is often feed on cell chloroplasts on the underside of the leaf, while the upper surface of the leaf develops whitish or yellowish stippling characteristic, which may join and become brownish as mite feeding continues. As mites move around, their webbing can span leaves and stems. Heavy damage may cause leaves to dry and drop and the plant may be covered with webbing and may die prematurely. It has been reported that two spotted spider mite infestation that causes approximately 14% reduction of total leaf area could result in significant yield loss in cucumber [4].

The Jordanian farmers rely heavily on acaricides to control the two-spotted spider mite. Therefore, they have increased the rate of application, applied a mixture of acaricides and applied acaricides more frequently than they should. They have complained about unsatisfactory results in controlling.

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From field surveillance and screening of various acaricides, it seems that this mite has developed resistance to the most conventional acaricides, but confirmed data are scarce and poorly documented. It is quite possible that *T. urticae* susceptibility to acaricides would differ from one location to another of cucumber cultivation in Jordan. Therefore, it was important to monitor the acaricide susceptibilities of *T. urticae* populations that were collected from cucumber cultivation in Jordan and to determine the duration of acaricide residue toxicity. This study showed the results of laboratory-based tests that determined the response of six field populations collected from the main production regions in Jordan compared to one susceptible strain of *T. urticae* to three used acaricides; abamectin, milbemectin and chlorfenapyr.

MATERIALS AND METHODS

Six populations of *T. urticae* were collected from cucumber plants grown under plastic houses conditions in different regions of Jordan. These geographical regions include Al-Ramtha (100 Km North West of Amman), Baq'a (20 Km North West of Amman), Zyzya (30 Km South of Amman), Krimeh (North of Jordan Valley), Deir-Alla (Central of Jordan Valley) and Karamah (South of Jordan Valley). These regions are considered the main area for cucumber production in the country. A susceptible strain of *T. urticae* was obtained from Lattakia Center for Rearing and Production of Biological Agents (LCRPBA) in Syria. This strain was reared in (LCRPBA) for 5 years without the application of acaricides.

Rearing of the Syrian susceptible strain was done inside special insectaria in University of Jordan, faculty of agriculture. The Syrian strain was reared and maintained on *Phaseolus vulgaris* plants under greenhouse conditions. *P. vulgaris* plants were irrigated and replaced as needed.

Three acaricides used for cucumber protection in Jordan were evaluated for their residual effects towards *T. urticae,* by applying the higher recommended field rate (Table 1).

The residual effects of the three acaricides used on adult females of *T. urticae* were evaluated under room temperature conditions. Cotyledon leaves of bean sowed in polystyrene trays, filled with Peat-moss and Perlite were sprayed by a hand sprayer until run-off. Sprayed cotyledon leaves were removed at 1, 3, 6, 9, 12 and 15 days after application. Removed leaves of each treatment were placed in Petri dishes lined with cotton wool [5]. Twenty five adult female of *T. urticae* were transferred to each cotyledon leaf. The number of dead and alive mites for each treatment was estimated 48 h after contact with the treated cotyledons.

Table 1: Tested acaricides, common and trade names, manufacturer and application rate

Common name	Trade name	Manufacturer	Application rate
Abamectin	Vertimec 1.8% EC	Syngenta AG	7.2 mg/L
Milbemectin	Milbecknock® 1% EC	Sankyo Co. Ltd	12.5 mg/L
Chlorfenapyr	Pirate® 24% SC	BASF AG	120 mg/L

The layout of the residual effect experiment was Complete Randomize Design (CRD) with 4 treatments and 4 replications. Obtained data were subjected to analysis using the SAS programme. Prior to analysis, arcsine-transformation of data was done [6].

RESULTS

The duration of acaricides residue toxicity varied among the compounds tested (Table 2-8). Mortality percentage from residual effect of abamectin and milbemectin in the six populations of *T. urticae* was significantly lower than that for the Syrian susceptible strain. Mortality percentage in the six populations ranges from 9 - 20, 11 - 18, 9 - 15 and 7 - 10 % after 3, 6, 9 and 15 days of abamectin application, while it ranges from 16 - 28, 13 - 23, 9 - 17 and 7 - 10% after 3, 6, 9 and 15 days of milbemctin application. Mortality percentage of the Syrian susceptible strain was 92, 85, 76 and 51% for abamectin; 88, 82, 71, 52% for milbemctin, after 3, 6, 9 and 15 days of application, respectively. Mortality percentage in the control (tap water) ranges between 3 - 5% in all tested populations (Table 2-8).

In contrast, Mortality percentage in the six populations ranges from 78 - 95, 66 - 88, 60 - 86 and 48 - 68% after 3, 6, 9 and 15 days of chlorfenapyr application, except for Karamah population which reach only 63, 51, 46 and 36% after the same durations, respectively; compared to 95, 86, 76 and 63% mortality for the Syrian susceptible strain after the same durations, respectively.

The acaricides abamectin and milbemectin proved to be ineffective in controlling the six populations of the *T. urticae* at the higher recommended field rate even after one day of application. The mortality in the six populations did not exceed 24 and 33%, compared to 96 and 97% in the Syrian susceptible strain, for the two acaricides, respectively. On the other hand, the six populations of the *T. urticae* proved to be moderate to highly susceptible to the acaricide chlorfenapyr. Mortality in the populations collected from Ramtha, Baqa', Zyzya

Table 2: Residual effect of three acaricides against *T. urticae* adult females of Al Ramtha population

		% Mortality after days (d) of application ±SE					
Acaricides	Concentration (mg/L)	1d	3 d	6 d	9 d	12 d	15 d
Abamectin	7.2	15±1.0 b	12±1.6 b	11±1.9 b	10±1.2 b	9±1.0 b	9±1.0 b
Milbemectin	12.5	20±1.6 b	18±2.6 b	14±1.2 b	12±1.6 b	11±1.9 b	7±1.0 bc
Chlorfenapyr	120	95±1.9 a	89±1.0 a	77±2.5 a	73±2.5 a	60±3.3 a	50±2.6 a
Control	Tap Water	5±1.0 c	5±1.0 c	5±1.0 c	4± 0.0 c	5±1.0 b	4± 0.0 c

Means within the same column with the same letter are not significantly different after arcsine transformation using LSD at 0.05.

Table 3: Residual effect of three acaricides against *T. urticae* adult females of Baqa' population

		% Mortality after days (d) of application ± SE					
Acaricides	Concentration (mg/L)	1d	3 d	6 d	9 d	12 d	15 d
Abamectin	7.2	19±1.0 b	9±1.0 c	12±1.6 b	10±1.2 b	8±1.6 bc	7±1.0 b
Milbemectin	12.5	26±1.2 b	20±1.6 b	14±2.6 b	12±1.6 b	11±1.0 b	9±1.0 b
Chlorfenapyr	120	98±1.2 a	95±1.0 a	88±1.6 a	86±1.2 a	70±1.6 a	68±1.6 a
Control	Tap Water	5±1.0 c	4±0.0 d	3±1.0 c	4±0.0 c	5±1.0 c	4±0.0 c

Means within the same column with the same letter are not significantly different after arcsine transformation using LSD at 0.05.

Table 4: Residual effect of three acaricides against *T. urticae* adult females of Zyzya population

		% Mortality after days (d) of application ± SE					
Acaricides	Concentration (mg/L)	1d	3 d	6 d	9 d	12 d	15 d
Abamectin	7.2	24±1.6 b	20±1.6 b	16±1.6 b	14±1.2 b	10±1.2 b	8±1.6 b
Milbemectin	12.5	23±1.0 b	16±1.6 b	13±1.0 b	10±1.2 c	9±1.0 bc	7±1.0 b
Chlorfenapyr	120	92±1.6 a	88±1.6 a	76±1.6 a	69±1,9 a	58±2.6 a	48±1.6 a
Control	Tap Water	5±1.0 c	4±0.0 c	5±1.0 c	5±1.0 d	5±1.0 c	5±1.0 b

Means within the same column with the same letter are not significantly different after arcsine transformation using LSD at 0.05.

Table 5: Residual effect of three acaricides against *T. urticae* adult females of Krimeha population

		% Mortality after days (d) of application ± SE					
Acaricides	Concentration (mg/L)	1d	3 d	6 d	9 d	12 d	15 d
Abamectin	7.2	22±1.2 c	17±1.0 b	14±1.2 b	11±1.0 b	10±1.2 b	8±0.0 b
Milbemectin	12.5	31±1.0 b	25±1.0 b	17±1.0 b	14±1.2 b	12±1.6 b	9±1.0 b
Chlorfenapyr	120	88±1.6 a	78±2.6 a	66±1.2 a	60±1.6 a	54±1.2 a	46±1.2 a
Control	Tap Water	5±1.0 d	3±1.0 c	4±0.0 c	5±1.0 c	5±1.0 c	4±0.0 c

Means within the same column with the same letter are not significantly different after arcsine transformation using LSD at 0.05.

Table 6: Residual effect of three acaricides against *T. urticae* adult females of Dier- Alla population

		% Mortality after days (d) of application ± SE					
Acaricides	Concentration (mg/L)	1d	3 d	6 d	9 d	12 d	15 d
Abamectin	7.2	19±1.0 c	14±1.2 c	12±0.0 c	10±1.2 c	8±1.6 c	7±1.0 c
Milbemectin	12.5	33±1.9 b	28±2.3 b	23±1.9 b	17±1.0 b	13±1.0 b	10±1.2 b
Chlorfenapyr	120	96±1.6 a	92±1.6 a	87±1.9 a	79±3.0 a	72±1.6 a	66±1.2 a
Control	Tap Water	4±1.6 d	4±0.0 d	5±1.0 d	4±0.0 d	5±1.0 c	4±0.0 d

Means within the same column with the same letter are not significantly different after arcsine transformation using LSD at 0.05.

Table 7: Residual effect of three acaricides against *T. urticae* adult females of Karamah population

		% Mortality after days (d) of application ± SE					
Acaricides	Concentration (mg/L)	1d	3 d	6 d	9 d	12 d	15 d
Abamectin	7.2	21±1.0 b	18±1.2 b	17±1.0 b	15±1.0 b	12±1.6 b	10±1.2 b
Milbemectin	12.5	28±1.6 b	20±1.6 b	15±1.0 b	12±1.6 b	10±1.2 b	9±1.0 b
Chlorfenapyr	120	75±1.0 a	63±1,0 a	51±1.0 a	46±2.6 a	41±1.9 a	36±1.6 a
Control	Tap Water	4±1.6 c	4±0.0 c	5±1.0 c	4±0.0 c	5±1.0 c	4±0.0 c

Means within the same column with the same letter are not significantly different after arcsine transformation using LSD at 0.05.

Table 8: Residual effect of three acaricides against *T. urticae* adult females of Syrian susceptible strain

Acaricides	Concentration (mg/L)	% Mortality after days (d) of application ± SE 1d	3 d	6 d	9 d	12 d	15 d
Abamectin	7.2	96±0.0 a	92±1.6 a	85±1.9 a	76±2.8 a	64±2.8 b	51±1.9 b
Milbemectin	12.5	97±1.9 a	88±1.6 b	82±1.2 a	71±2.5 a	60±1.6 b	52±1.6 b
Chlorfenapyr	120	98±1.2 a	95±1.0 a	86±1.2 a	76±1.6 a	70±1.2 a	63±1.9 a
Control	Tap Water	5±1.0 b	5±1.0 c	4±0.0 b	3±1.0 b	5±1.0 c	5±1.0 c

Means within the same column with the same letter are not significantly different after arcsine transformation using LSD at 0.05.

and Dier Alla areas, after one day of application were 95, 98, 92 and 96%, which were close to the mortality in the Syrian susceptible strain that reached 98%, where as mortality in the populations collected from Krimeh and Karamah areas were 88 and 75%, respectively. Satisfactory control results in the six populations from exposure residue of chlorfenapyr even after 12 days of application were recorded, where the mortality reached 70 and 72% in the population collected from Baqa' and Dier Alla areas, compared to 70% for the Syrian susceptible strain and 41 - 60% in the other four populations (Table 2-8).

DISCUSSION

The six populations of *T. urticae* that were collected from plastic houses planted with cucumber in Jordan showed a higher loss of susceptibility against the acaricides abamectin and millbemctin. According to Insecticide Resistance Action Committee - IRAC, which pointed out that the term 'resistance' should be used only when field failure occurs and this situation is confirmed. Acaricides resistance in phytophagous mites is a seriously increasing phenomenon, especially in spider mites which have a remarkable intrinsic potential for rapid evolution of resistance [7].

The acaricide abamectin was introduced into Jordan in 1992 and be considered the most common acaricide for *T. urticae* control by farmers. In the local market, there are about 40 trade names for the acaricide abamectin from different sources with different prices. The total quantities of imported abamectin during 2010 were 48000L, in addition to about 23000 L formulated locally. Where as the acaricide millbemctin was introduced to Jordan in 2007 and it's not well known by all farmers. The total quantity of imported milbemectin in 2010 was only 750 L. It has been reported that repeated application of the same chemical or chemicals with the same mode of action can increase the chances of a pest population developing resistance [8]. However, a positive and significant correlation was observed between the frequencies of milbemectin and abamectin resistance, indicating positive cross-resistance between these two acaricides [9]. In addition to that, it has been conformed that *T. urticae* populations developed 342 fold resistant to abamectin after 5 selection and that, in the resistant population, the cross resistance w as determined against milbemectin 16.3 fold and chlorfenapyr 2.23 fold [10]. On the other hand, it has been indicating that abamectin and melbemectin, which are also broad spectrum insecto - acaricides, are considered safe to beneficial arthropods under field conditions due to their short environmental persistence, rapid uptake into treated plants and fast degradation of surface residues [11].

Chlorfenapyr was commercialized in 1995, but it was registered and introduced to Jordan in 2005. The total quantities of imported and locally formulated of chlorfenapyr in 2010 were only 2700 kg. This acaricide is effective against all stages of spider mite, with long environmental persistence [11, 12].

Differences in mite population's susceptibility to chlorfenapyr were clear for the population collected from Karamah region and to a lesser extent to the population collected from Kraimeh region. These two regions are located in the southern and northern Jordan Valley, with a distant of about 70 km, that are considered from the main regions in egg plant production, which is the a favorite host for the two spotted spider mite, where farmers rely heavily on acaricides to control this mite. This is in agreement with the study conducted in Brazil [13], as mite populations that were collected from different crops (papaya, strawberry, bean, tomato, chrysanthemum, rose), in various counties in the State of São Paulo showed significant differences among populations in their responses to chlorfenapyr, as populations with frequencies of resistance from zero to 65.4% were detected. The highest frequencies of resistance were observed in populations obtained from chrysanthemum in Holambra. In other study in Australia, resistance of *T. urticae* from cotton to chlorfenapyr was first detected in the 2001-2002 season during studying the responses of the mite populations that has been monitored since the

1997-1998 growing season [14]. Low resistant value (RF $\leq$ 3) for mite populations from Korea to the acaricide chorfenapyr has been reported [15, 16].

This acaricide is now out of patentees that mean that the market is expected to be saturated with generics and *T. urticae* may be negatively affected.

Optimal use patterns must be developed to use the acaricides abamectin and milbemectin for controlling the two spotted spider mite in Jordan, by reducing the repeated application during the season and using other acaricides in rotation. Further bioassay and field testing of chlorfenapyr must be done to explore the continued use of this compound in *T. urticae* management.

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