

Evaluation of three pesticides against phytophagous mites and their impact on phytoseiid predators in an eggplant open-field

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ABSTRACT — The present study aimed to evaluate the effects of two insecticides (deltamethrin and acetamiprid), and an acaricide (fenbutatin oxide) on diversity and abundance of mite fauna in replicate experiments carried out in an open-eggplant field in Latakia, Syria. Each pesticide was applied three times with an interval of three weeks between two consecutive applications. Two phytophagous mites were observed in all treatments: *Tetranychus urticae* and *Polyphagotarsonemus latus*. However, the number of predatory mite species (in particular Phytoseiidae) was different according to the pesticide applied and the lowest number observed was in the fenbutatin oxide treatment. This acaricide caused high mortality of *T. urticae*, decreased the abundance of *P. latus*, and negatively affected Phytoseiidae. Acetamiprid and deltamethrin induced the resurgences of *T. urticae* (3.5-fold and 1.5-fold for the former and the latter respectively). The abundance of *P. latus* was not affected by acetamiprid but by deltamethrin. The results clarified also that these insecticides seemed harmless to Phytoseiidae [*i.e.* *Phytoseiulus persimilis*, *Phytoseius finitimus* and *Typhlodromus (Anthoseius) recki*]. Data obtained here are interesting for integrated management programs of the major eggplant phytophagous mites, although the generalization of these results requires some caution and additional experiments are needed.

KEYWORDS — fenbutatin oxide; deltamethrin; acetamiprid; *Tetranychus urticae*; *Polyphagotarsonemus latus*; *Phytoseiulus persimilis*; eggplant; integrated control

INTRODUCTION

Eggplant, *Solanum melongena* L. is the main vegetable crop in open fields in Latakia province, Syria. The crop was cultivated on an area of 641 ha producing about 15.4 thousand tons according to data from Syrian Ministry of Agriculture and Agrarian Reform (2014).

The major pests of this crop include several species of lepidopteran insects, whitefly, thrips, aphid and phytophagous mites, causing severe damages and substantial losses in crop yield (Srinivasan, 2009; Reddy and Miller, 2014). The control

of these pests has been based generally on broad-spectrum pesticides in numerous Latakian fields and the number of sprays performed varied according to eggplant growers. However, the great indiscriminate use of these compounds has negative environmental impacts and can impair the performance of natural enemies, causing consequently the emergence of other population pests as tetranychid mites (Reddy, 2001; Kumral and Kovanci, 2005; Van Driesche *et al.*, 2009; Marčić *et al.*, 2012).

In the recent years, in many cases, the application of pesticides was minimized or stopped essentially due to economic reasons. The results of a

survey carried out in a free pesticide eggplant field in 2015 (Barbar, 2016), revealed the presence of the tetranychid *Tetranychus urticae* Koch and the tarsonemid *Polyphagotarsonemus latus* (Banks), considered as very important pests of eggplant (Bostanian *et al.*, 2003; Maanen *et al.*, 2010; Hoy, 2011). However, these phytophagous mites were accompanied by several predatory mite species essentially belonging to the family Phytoseiidae (Barbar, 2016).

The aim of the present work was to evaluate the effects of two insecticides (deltamethrin and acetamiprid) and an acaricide (fenbutatin oxide) on diversity and abundance of phytophagous mites and their natural enemies (in particular phytoseiid mites) in an eggplant open-field in Latakia province. These pesticides were selected as they were the most common and repeatedly applied on numerous eggplant fields and other crops in this province. This initial study could clarify some points to develop management strategies including biological control of eggplant phytophagous mites.

MATERIALS AND METHODS

Applied pesticides

The pesticides tested were the acaricide Jolly® SC (fenbutatin oxide, with a recommended concentration of 0.5 ml ai.l⁻¹, Slipcam, Italy, applied against *T. urticae*) and the insecticides Dinamethrin® EC (deltamethrin with a recommended concentration of 0.0125 g ai.l⁻¹, Agri Pes, Syria) and Setar® SP (acetamiprid with a recommended concentration of 0.1 g ai.l⁻¹, Agri Pes, Syria) applied against whitefly, thrips and lepidopteran eggplant pests.

Field experiment and mite sampling

The experiment was carried out in an eggplant open-field of approximately 2000 m² located in Latakia province during 2016 season. The crop was grown in the nursery seed bed and then transplanted into the experimental field during mid-April.

The experimental design consisted of four treatments (three pesticides and a water control). Each treatment was replicated four times. The replicates were randomly assigned to plots in a complete

randomized block design. Each plot contained 27 plants and was separated from other plots by 1.2 m to avoid the spray drifts or treatment effects. The plots were left without any pesticide treatments for 1.5 month allowing thus the occurrence of natural infestation by the target pest and their natural enemies. The agronomic practices (mineral fertilizers and irrigation) were kept uniform on all the plots throughout the experiment. Three applications of each pesticide were carried out on 3, 25 June and on 16 July using a ten-liters hand-held sprayer with hollow cone nozzle.

To determine mite population levels, four plants were randomly selected per plot. In each plant, three leaves were randomly selected, one per top, middle and bottom third of the plant (a total of 48 leaves per treatment). Samples were taken one day before the first pesticide applications and 1, 2, 7, 14, 21 days after the first and the second applications. For the third application, samples were taken 1, 2 days after application and then weekly for two consecutive months. All living mites on each leaf were counted using a binocular microscope. Adult stage was mounted on slides in Hoyer's medium and dried in an oven at 40 °C for one week for identification.

Data analysis

Species similarity between the four treatments was explained by Bray-Curtis analysis using the software Biodiversity Professional version 2 (McAleece *et al.*, 1997). For mites abundances and regarding to the results obtained, two ways were performed to evaluate the effect of the pesticides on phytophagous mites and their predatory mites: (1) during experiment, the results revealed that *T. urticae* was approximately the unique mite species on the crop for several weeks after the first application and the reduction of its populations was only observed in plots where fenbutatin oxide was applied. Therefore, mortality ratios by this acaricide were estimated only for this species and calculated according to the equation of Henderson and Tilton (1955). The data were analyzed by one-way analysis of variance (ANOVA) followed by Duncan test ($\alpha = 0.05$) to compare this variable between sampling

TABLE 1: Percentage of phytophagous and predatory mite species observed in different pesticides treatments applied in the eggplant filed studied in Latakia province, Syria.

Mite families	Mite species	% Mite species			
		Control	Fenbutatin oxide	Acetamiprid	Deltamethrin
Tetranychidae	<i>Tetranychus urticae</i>	96.4	91.6	98.2	98.9
Tarsonemidae	<i>Polyphagotarsonemus latus</i>	3.6	8.4	1.8	1.1
	<i>Phytoseiulus persimilis</i>	94.7	84.5	84	94
	<i>Phytoseius finitimus</i>	3.6	11.6	11.6	4
Phytoseiidae	<i>Typhlodromus (Anthoseius) recki</i>	0.4	1.3	4	1
	<i>Typhlodromus (Typhlodromus) athiasae</i>	0.2	1.3	0.1	0
	<i>Amblyseius swirskii</i>	0.2	1.3	0.1	0.3
	<i>Neoseiulus sbaronensis</i>	0.4	0	0	0.4
	<i>Neoseiulus barkeri</i>	0.2	0	0.1	0
Blattisociidae	<i>Lasioseius (Lasioseius) parberlesei</i>	0	0	0.1	0.1
Bdellidae	No identified	0.1	0	0	0.1
Stigmaeidae	<i>Agistemus exsertus</i>	0.1	0	0	0.1
Ascidae	<i>Gamasellodes americanus</i>	0.1	0	0	0

dates (2) some other mite species appeared lately in considerable numbers during season, so the densities of these mites were compared between treatments and between sampling dates for each treatment. Abundances of rarely observed mite species were ignored. A Kruskal-Wallis non-parametric analysis of variance followed by multiple comparisons between ranks were performed as data were not normally distributed. All data were analyzed using SPSS® software version 20 (IBM®, 2011).

RESULTS

Mite diversity

Two phytophagous mite species were collected in all treatments: the two spotted spider mite *T. urticae* being dominant (more than 90 %) and the broad mite *P. latus* (Table 1).

A total of 5299 specimens of predatory mites belonging to five families and 11 species were found in all treatments. The highest numbers of species were found in the control (10 species) and the lowest were found in fenbutatin oxide treatment (5 species) (Table 1). Among these predators, members of the family Phytoseiidae were the most frequent, and the specialist *Phytoseiulus persimilis* Athias-Henriot was the most abundant species (more the 80 % of speci-

mens collected) followed by *Phytoseius finitimus* Ribaga and *Typhlodromus (Anthoseius) recki* Wainstein (Table 1). There was a great similarity percent (more than 80 %) between mite species present in control treatment and those found in deltamethrin and acetamiprid treatments. However, relative low percent (less than 26 %) was observed between control and insecticide treatments and those found in fenbutatin oxide treatment (Figure 1).

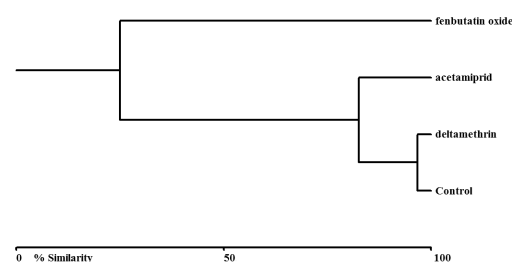


FIGURE 1: Bray-Curtis clustering analysis dendrogram of mite fauna observed in different pesticides treatments applied in the eggplant filed studied in Latakia province, Syria.

Mortality of *Tetranychus urticae* caused by fenbutatin oxide

The ratios of reduction (mortality ratios) of *T. urticae* were significantly different between sampling dates ($F = 2.39$; $df = 19$; $P = 0.006$) (Figure 2). One day af-

ter the first application, fenbutatin oxide produced 42.6 % reduction of *T. urticae*. This ratio increased gradually and reached 79.2 % after 21 days. When the second application was carried out, mortalities were from 92.5 % to 96.0 % during 21 days. The ratio slightly increased to 99.3 % at 21 days after the third application and then, a gradual decrease was observed and reached to 57.0 % at the end of the experiment (Figure 2).

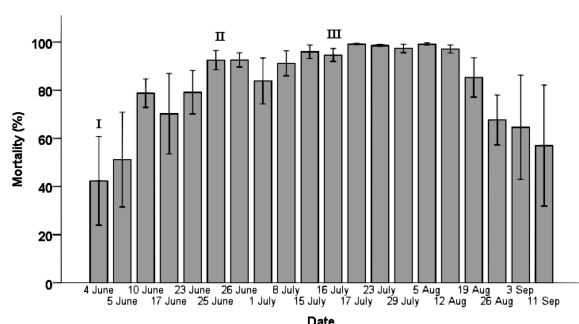


FIGURE 2: Mortality (% \pm S.E.) of *Tetranychus urticae* observed in fenbutatin oxide treatment applied three times (I, II and III) in the eggplant field studied in Latakia province, Syria.

Population of phytophagous and Phytoseiidae mites on eggplant leaves in different pesticide treatments

Tetranychus urticae was the dominant phytophagous mite species on eggplant leaves in all treatments. The lowest densities (general mean \pm SE mites per leaf during season) were observed in fenbutatin oxide treatment (3.44 ± 0.32) compared to other treatments (24.72 ± 0.87 ; 34.96 ± 1.78 and 25.40 ± 1.06 , in control, acetamiprid and deltamethrin, respectively) ($H = 966.14$; $df = 3$; $P < 0.001$) (Figure 3a).

In control treatment, the abundance of this phytophagous species was lower during June, August and September compared to that observed during July (the highest density was 54.65 ± 4.48 in mid-July) ($H = 342.04$; $df = 20$; $P < 0.001$) (Figure 4a). For fenbutatin oxide treatment, the highest abundance was observed before treatment (21.52 ± 3.24 , $H = 206.22$; $df = 20$; $P < 0.001$), decreased to less than two individuals per leaf after two weeks of the 1st application and remained approximately at this

low abundance afterward (Figure 4b). Density variations in time were similar for the two insecticide treatments (acetamiprid and deltamethrin). The population significantly increased two weeks after the first application, reached the highest levels one week after the second application at 1st July (for acetamiprid 138.63 ± 11.18 , $H = 550.46$; $df = 20$; $P < 0.001$; for deltamethrin 72.54 ± 6.43 , $H = 427.58$; $df = 20$; $P < 0.001$) (Figure 4b, c), and then, decreased progressively afterward until the end of sampling.

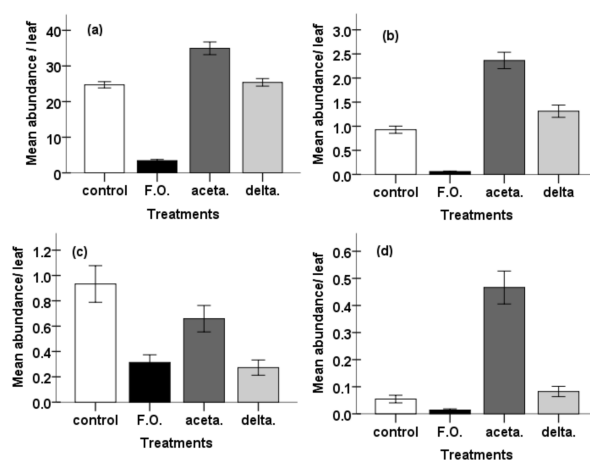


FIGURE 3: General mean abundance per eggplant leaf (\pm SE) of *Tetranychus urticae* (a), *Phytoseilus persimilis* (b), *Polyphagotarsonemus latus* (c) and other phytoseiid species (d) in control, fenbutatin oxide (F.O.), acetamiprid (aceta.) and deltamethrin (delta.) treatments.

During experiments, the specialist predator *P. persimilis* accompanied *T. urticae*. The highest densities of this predator (general mean during season) were observed in acetamiprid treatment (2.36 ± 0.17) followed by those in deltamethrin and control treatments (1.31 ± 0.13 and 0.93 ± 0.07 ; respectively) and the lowest were observed in fenbutatin oxide (0.06 ± 0.01) ($H = 322.17$; $df = 3$; $P < 0.001$) (Figure 3b).

In control treatment, the population started to increase at the beginning of July, reached the highest abundance at 23 July (3.67 ± 0.56), and then decreased until the end of sampling ($H = 366.59$; $df = 20$; $P < 0.001$) (Figure 4a). In fenbutatin oxide treatment, the highest abundance was observed before the 1st application (0.35 ± 0.15 ; $H = 50.01$; df

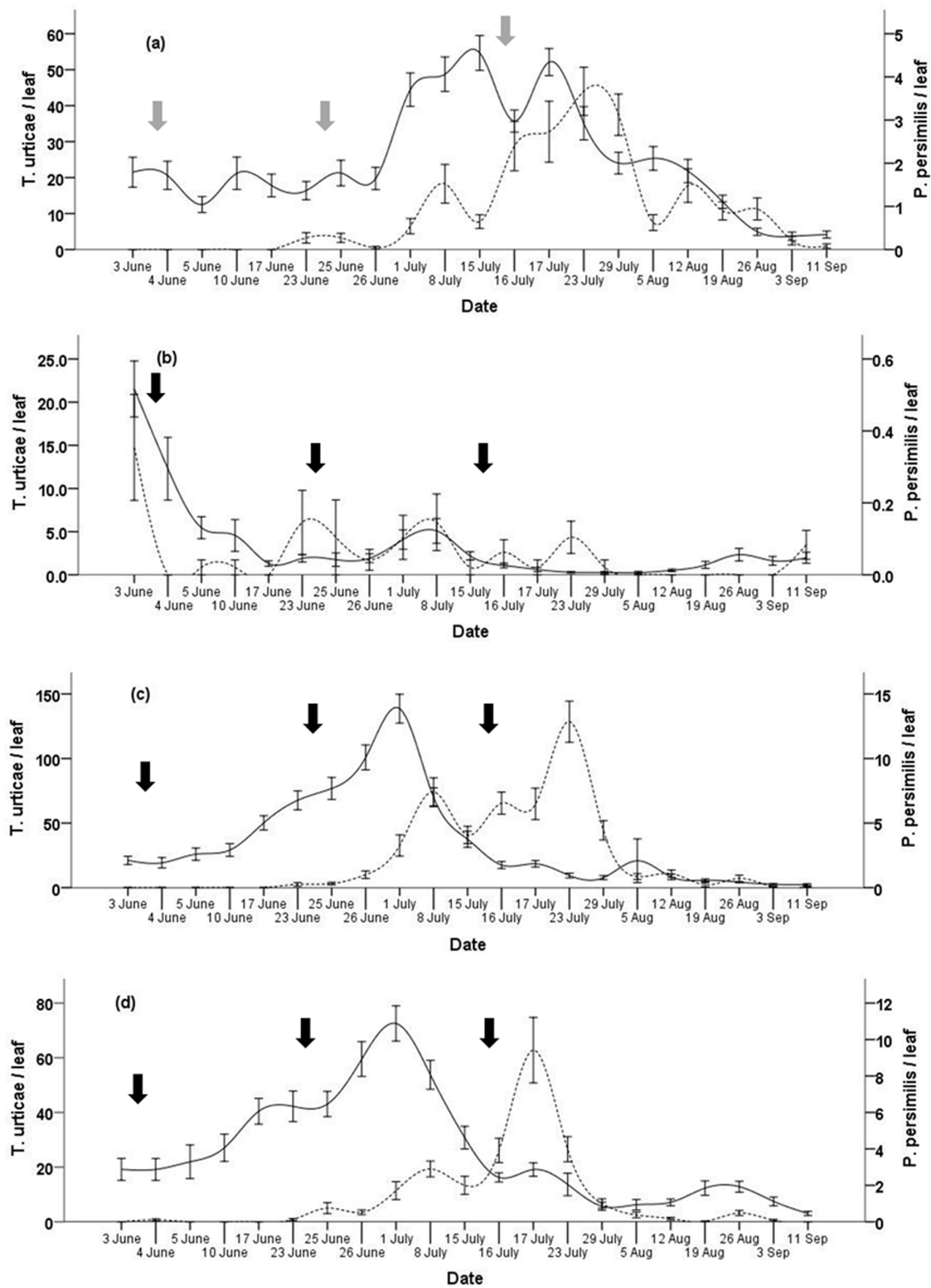


FIGURE 4: Mean abundance per eggplant leaf (\pm SE) of *Tetranychus urticae* (continuous line) and *Phytoseiulus persimilis* (dotted line) observed during experiments in control (a) treated with water (gray arrows), fenbutatin oxide (b), acetamiprid (c) and deltamethrin (d) (black arrows).

= 20; $P < 0.001$) (Figure 4b) and decreased to very low levels afterward. Density variations in time in acetamiprid and deltamethrin treatments were approximately similar. In acetamiprid, the population started to increase at the end of June (one week after the 2nd application), reached the highest abundance at 23 July (12.85 ± 1.59 ; one week after the 3rd application), and decreased until the end of sampling ($H = 475.35$; $df = 20$; $P < 0.001$) (Figure 4c). In deltamethrin treatment, the highest abundance appeared at 17 July (9.42 ± 1.80), ($H = 441.68$; $df = 20$; $P < 0.001$) (Figure 4d).

The tarsonemid mite *P. latus* appeared lately during experiments. The densities were different between treatments. The highest densities (general mean during season) were observed in control and in acetamiprid treatments (0.93 ± 0.14 ; 0.66 ± 0.10 , respectively) and the lowest were observed in fenbutatin oxide and deltamethrin treatments (0.31 ± 0.06 ; 0.27 ± 0.06 ; respectively; $H = 17.01$; $df = 3$; $P = 0.001$) (Figure 3c). The first appearance of this species was observed in control treatment at the first week of July, reached the highest abundance at the end of August (7.83 ± 2.07 ; $H = 235.06$; $df = 20$; $P = 0.001$), and decreased until the end of sampling (Figure 5a). In plots where pesticides were applied, the species was observed in mid-July, reached the highest abundance at the beginning of September, and decreased until the end of sampling (Figure 5b, c and d).

Other phytoseiid species [dominated by *P. finitimus* and *T. (A.) recki*] were also appeared lately during experiments. The abundance (general mean during season) was higher in acetamiprid treatment (0.47 ± 0.06) than in remaining ones (0.06 ± 0.01 in control; 0.014 ± 0.004 in fenbutatin oxide; 0.08 ± 0.02 in deltamethrin; $H = 146.32$; $df = 3$; $P < 0.001$) (Figure 3d). In all treatments (Figure 5a, b, c and d), the abundance was very low from the beginning of sampling until one week after the 3rd application. In acetamiprid treatment, the highest abundance was observed at 12 August (2.46 ± 0.53 ; $H = 337.05$; $df = 20$; $P < 0.001$) and decreased afterward. In control and deltamethrin treatments, the highest abundance was observed in the beginning of September (0.56 ± 0.25 ; $H = 80.37$; $df = 20$; $P < 0.001$ for con-

trol, and 0.81 ± 0.26 ; $H = 160.32$; $df = 20$; $P < 0.001$ for deltamethrin). In fenbutatin oxide treatment the abundance of those predators was the lowest and increased at the end of sampling (eight weeks after the 3rd application) (0.08 ± 0.04 ; $H = 40.81$; $df = 20$; $P = 0.004$).

DISCUSSION

The results showed that the diversity and the abundance of mite fauna were different according to the pesticide applied. **The acaricide fenbutatin oxide** seemed to have an effective performance against *T. urticae* and the application of one spray decreased its densities to very low levels for about one month. Repeated sprays of this compound provided also an extreme suppression of its populations. Effectiveness and long-term effects of fenbutatin oxide against *T. urticae* and other tetranychid species have been previously reported (Knight *et al.*, 1990; Herron *et al.*, 1997; Teodoro *et al.*, 2005). However, field control failure and resistance to this compound could be rapidly occur (Herron *et al.*, 1994; Jacobson *et al.*, 1999; Liburd *et al.*, 2007). Fenbutatin oxicide appeared also effective for management of *P. latus*. Abundance of this species was 3-fold lower and the species was approximately absent until three weeks after the 3rd application compared to untreated control. There were no available data about the efficacy of fenbutatin oxide against this species, so additional studies are needed to confirm this result.

A distinct impact of fenbutatin oxide on Phytoseiidae populations seemed to be present. For *P. persimilis*, the abundance was reduced 15-fold compared to untreated control. However, previous studies (Petitt and Karan, 1991; Kim and Yoo, 2002; Liburd *et al.*, 2007) reported that fenbutatin oxide did not have a significant effect on *P. persimilis*. Indeed, it was no evidence to definitively conclude about the adverse effects of fenbutatin oxide on this predator as this compound reduced the availability of *T. urticae* and this could be detrimental to *P. persimilis*, considered as a highly specific species on *Tetranychus* sp. and attempts to disperse or die when *T. urticae* populations present in low numbers (Liburd *et al.*, 2007; McMurtry *et al.*, 2013).

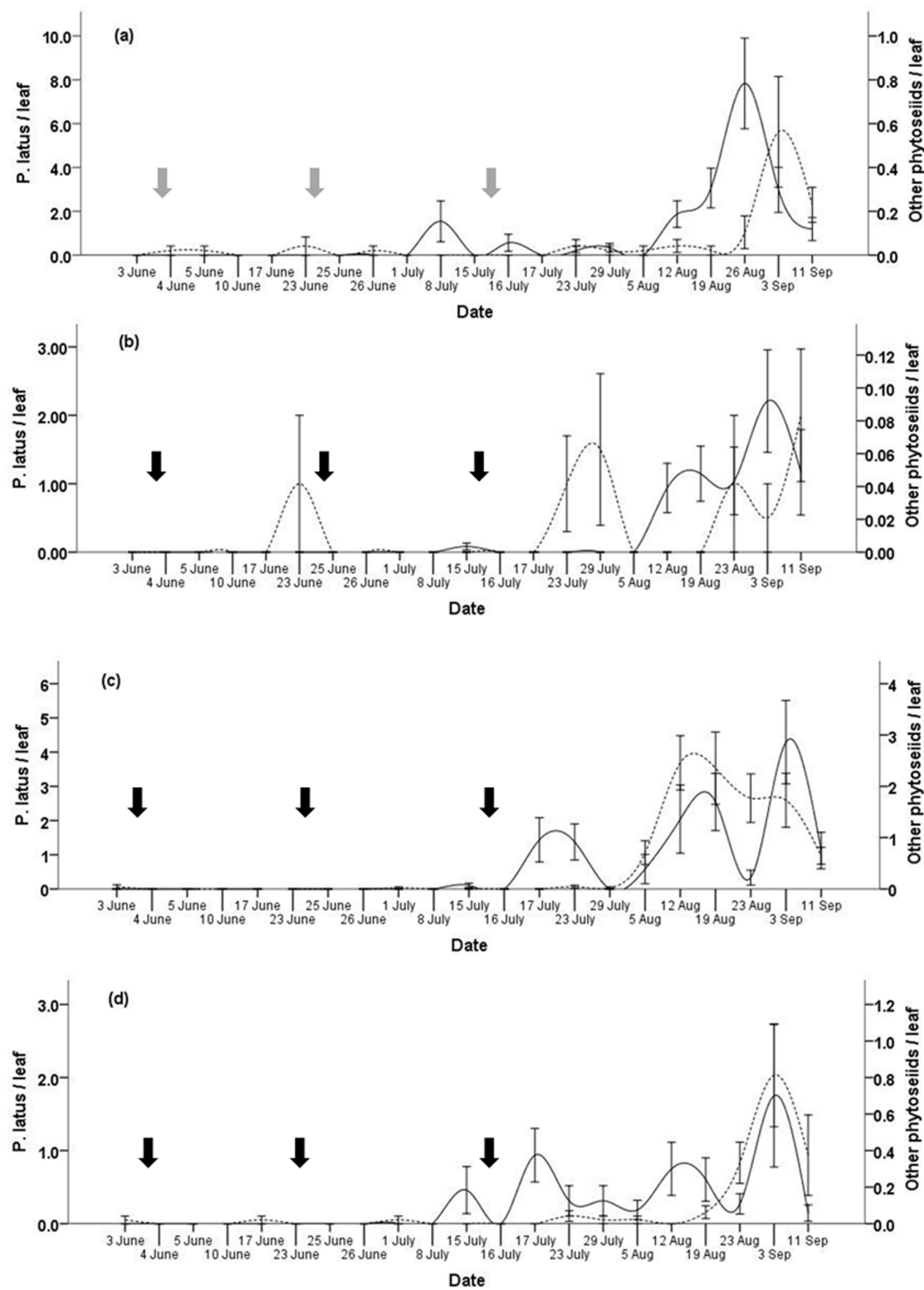


FIGURE 5: Mean abundance per eggplant leaf (\pm SE) of *Polyphagotarsonemus latus* (continuous line) and other phytoseiid species (dotted line) observed during experiments in control (a) treated with water (gray arrows), fenbutatin oxide (b), acetamiprid (c) and deltamethrin (d) (black arrows).

Abundance of other phytoseiid species was 4-fold lower compared to untreated control, so fenbutatin oxide seemed delaying their appearance on eggplant leaves for approximately 1-2 weeks after each application. The results of the previous studies have showed that the impact of fenbutatin oxide is ranked from slightly toxic to moderately toxic depending on the phytoseiid species studied (Overmeer and van Zon 1981; Chen *et al.*, 2003; Teodoro *et al.*, 2009).

The application of acetamiprid and deltamethrin caused a significant and immediate increase of *T. urticae* populations but the increasing was less pronounced in deltamethrin. The mean number of this mite progressively increased after the 1st spray and reached the highest level (3.5-fold and 1.5-fold for acetamiprid and deltamethrin respectively compared to that in untreated control) one week after the 2nd spray. Although the number of *T. urticae* eggs on eggplant leaves did not counted, it is very probably that these insecticides positively affected the fecundity of this mite and resurgences of its populations. This phenomenon has been previously observed for acetamiprid (and other neonicotinoids) and also for several synthetic pyrethroids including deltamethrin and could be linked to several mechanisms such as changes in physiology of the mites or biochemistry of host plants in favour of *T. urticae* (Gerson and Cohen, 1989; James and Price, 2002; Martin *et al.*, 2010; Barati and Hejazi, 2015; Gupta, 2015).

The abundance of the tarsonemid *P. latus* did not negatively affected by the application of acetamiprid but do for deltamethrin (3-fold lower compared to that in untreated control). Opposite results have been reported in the literature: acetamiprid showed effectiveness in reducing populations of *P. latus* (Varghese and Mathew, 2013), whereas deltamethrin was ineffective to control this species (Azandémè-Hounmalon *et al.*, 2015). The results obtained were consequently difficult to interpret and generalized as the species was not observed in all treatments until 1-2 weeks before the 3rd application. Furthermore, these differences in results may be due to differences in strains of *P. latus*, insecticide formulations, number of sprays and

host plant.

Trend in *P. persimilis* abundance in acetamiprid treatment was similar to that observed in deltamethrin but it was less pronounced in the latter. Respectively, mean abundances were 3.5–5-fold and 1–3.5-fold higher after the 2nd and the 3rd acetamiprid and deltamethrin sprays compared to untreated control. Even if the direct effects of acetamiprid and deltamethrin on *P. persimilis* were not determined, these insecticides indirectly increased populations of this predator by increasing the abundance of its "favourable" prey (*T. urticae*). Indeed, the ratios predator : prey were 1: 10 and 1: 25 (for acetamiprid and deltamethrin, respectively) after one week of *T. urticae* peak, and highly increased in favour to the predator and reached to 1: 0.73 and 1: 2 two weeks later. In previous studies, the susceptibility of several phytoseiid species [*Galendromus occidentalis* (Nesbitt), *Neoseiulus californicus* (McGregor), *Phytoseiulus macropilis* (Banks), *Typhlodromus* (*Typhlodromus*) *pyri* Scheuten and *Iphiseius degenerans* (Berlese)] to those insecticides was highly varied from no effect observed to moderately toxic according to phytoseiid species and strains (Gerson and Cohen, 1989; Rodrigues *et al.*, 2002; Poletti *et al.*, 2007; Bonafos *et al.*, 2008; Martinez-Rocha *et al.*, 2008; Beers and Schmidt, 2014; Doker *et al.*, 2015; Poletti and Omoto, 2012). Further laboratory tests should be thus clarify the direct effects of acetamiprid and deltamethrin on *P. persimilis* Syrian populations.

Similarly, the generalist predators [in particular *P. finitimus* and *T. (A.) recki*] appeared simultaneously in insecticide treatments and in untreated control one week after the 3rd application. Their abundances were ≥ 8 -fold in acetamiprid but approximately equal to that in untreated control for deltamethrin, suggesting, thus, that these insecticides are harmless to those predators and agreeing with the previous results (Poletti *et al.*, 2007; Bonafos *et al.*, 2008; Martinez-Rocha *et al.*, 2008). However, these results should be taken with some precautions as the predators appeared lately in season and this late appearance could be related to the availability of *P. latus* as a potential favourable prey to these predators.

Overall, the results of the present study demonstrated the incompatibility of fenbutatin oxide in integrated mite control programs as it reduced diversity and abundance of beneficial mites. On the other hand, repeated applications of acetamiprid and deltamethrin should be avoided as they induced the resurgence of *T. urticae* populations although the species did not have the history of exposure to these insecticides. The results of this case study indicated also that *P. persimilis* naturally intervened when the abundance of *T. urticae* was 40–50 mites/ leaf and decreased this density to low levels during two weeks (Figure 4a). Therefore, this outcome obtained could be economically and ecologically important by reducing the number of pesticides used and the number of sprays applied. However, the generalization of these results, requires some cautions and additional experiments are needed to clarify the threshold tolerance of eggplant crop to phytophagous mites under Syrian environmental conditions and the direct effects of these chemicals (and their combinations) on Phytoseiidae species in order to enhance their efficiency in controlling phytophagous mites on eggplant.

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