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Evaluation of Insecticides in the Management of Whiteflies (*Bemisia tabaci* Gennadius) and their Impacts on Yield of Eggplants

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Abstract The sweet potato whitefly *Bemisia tabaci* (Gennadius) is one of the most destructive widespread polyphagous insects attacking more than 900 plant species. This research aims to compare the effects of pyrifluquinazon and cyantraniliprole insecticides against whiteflies under greenhouse conditions and study their impacts on the yield of eggplants. Two greenhouses divided into three compartments each, were used for this study. Five consecutive treatments were conducted in the two greenhouses at 10-15 day intervals, and 8 pickings were also performed. The first compartment in both greenhouses was treated with pyrifluquinazon, the second with cyantraniliprole, and the third received 5 different conventional insecticides and served as a positive control. Pyrifluquinazon demonstrated statistically high efficacy against eggs and nymphs compared to all other insecticides including cyantraniliprole. The corrected percentage reduction in the whiteflies' egg population was above 99% in both greenhouses and the percentage reduction in the nymph population was above 97% in the compartments treated with pyrifluquinazon alone. Cyantraniliprole showed better results relative to spiromesifen, spirotetramat, tolfenpyrad, sulfoxaflor, and flupyradifurone treated plots. After the fifth application, cyantraniliprole was able to reduce the whiteflies eggs' populations by 23.69% and 42.47% in greenhouses 1 and 2, respectively; whereas whiteflies nymphs' populations were reduced by 76.25% in greenhouse 1. The pyrifluquinazon-treated compartments recorded the highest yield (3.9 Kg/plant) followed by cyantraniliprole-treated compartments (2.2 Kg/plant). The appropriate use of pyrifluquinazon in an integrated pest management program for whiteflies control should be highlighted to prevent a resistance buildup.

Keywords *Bemisia tabaci,* Pyrifluquinazon, Cyantraniliprole, Eggplant

1. Introduction

The sweet potato whitefly or cotton whitefly *Bemisia tabaci* Gennadius (Hemiptera: Aleyrodidae) is one of the most destructive polyphagous sap-feeding insects invading a broad range of vegetables and ornamentals. It is classified as one of the 100 of the World's Worst Invasive Alien Species [1]. According to many reports, *B. tabaci* is distributed worldwide except in cold regions like Antarctica.

It has been reported that *Bemisia tabaci* invades more than 900 host plants [1]. So far, over 300 different plant viruses were recorded to be successfully transmitted by the insect [2]. *Bemisia tabaci* is considered a complex cryptic species. According to mitochondrial cytochrome oxidase subunit I (mtCOI) gene analysis, this complex consists of more than 34 morphologically indistinguishable species.

Moreover, out of 34 species, 11 were stated as highly defined groups based on genetic markers and DNA identification studies [3-8]. Out of the *B. tabaci* cryptic species complex, the Middle East-Asia Minor 1 (MEAM1, known as B biotype) and Mediterranean (MED, known as Q biotype) species, are reported to be the most widespread and polyphagous invaders [9]. It was also demonstrated that MEAM1 and MED species exist in Lebanon [10]. Two factors have helped the extension of these biotypes, the large diversity of host plants and the international plant materials commerce [11-13].

Bemisia tabaci is known for its very fast and effective spread once established on the host crop. Sweet potato whiteflies have two different types of damages, direct and indirect ones. The direct damage is caused by the destruction of the host plant through phloem feeding behavior. The second type is through the excretion of the sugary honeydew on plant parts leading to the growth and development of the saprophyte black fungus, the sooty mold, and thus reducing photosynthesis. A third indirect type of damage is presented through the transmission of plant pathogenic viruses belonging to different genera such as *Begomovirus* (Family: Geminiviridae), the most destructive one, in addition to *Carlavirus*, *Criniviruses*, *Ipomovirus,* and *Torradovirus* [14-18].

Another problematic issue is the evolution of resistance buildup to the commonly used insecticides. The misuse of insecticides due to repeated application of pesticides having the same mode of action and due to the overuse of chemicals has led to the development of resistance in whiteflies *B. tabaci* and decreased their efficacies and their potential to reduce virus transmission. As a result, crop losses have increased [19] and pushed the farmers to follow integrated pest management settings [20-22]. Although insecticide resistance of *B. tabaci* is on the way to aggravation, the use of chemical treatments is still inevitable in many crops [23]. It was demonstrated that *B. tabaci* has developed a resistance to neonicotinoids such as acetamiprid, imidacloprid and thiamethoxamxam [24-27] as well as for flupyradifurone, a butenolide [26]. Resistance for chlorpyrifos an organophosphate (OP) [28], for deltamethrin a pyrethroid [25,29,30] is also reported. Resistance was also recorded for spirotetramat and spiromesifen tetramic acids and spinetoram [31], pyriproxifen an insect growth regulator (IGR) [32], and pymetrozine a Chordotonal organ TRPV channel modulators [33]. Alternatively, biological control of whitefly *B. tabaci* experiments using parasitoids, predators and entomopathogenic showed promising results for the management of whiteflies. One of the most efficient tools was *Beauveria bassiana,* an entomopathogenic fungus [10]. Other biological tools could also be considered effective on homopterans such as *Solanum panduriforme* extracts [34] and the installation of different types of pheromones for monitoring and mass trapping [35].

The Eggplant (*Solanum melanogena* L.) is an important vegetable crop grown throughout the world [36]. Fruits are relatively characterized by the high content of antioxidant substances and they constitute a good source of vitamins and minerals making their total nutritional value comparable with many berry species sand tomatoes [36, 37]. The three most important insect pests on eggplant cultivars are the whitefly, *B. tabaci*, the aphid, *Aphis gossypii* Glover, and the jassid, *Empoasca decipiens* Paoli [38]. In addition, eggplant can be infected by insect pests during the harvesting period. Whitefly feeding can reduce yield directly through sap removal. Honeydew accumulation can also reduce the growth of fruits although light to moderate infestation is well tolerated by the eggplant. However, the fruits become unmarketable in severe cases and the eggplant physiology is negatively affected [39].

The target of this work is to analyze the efficacy of two insecticides pyrifluquinazon and cyantraniliprole compared to that of spiromesifen, spirotetramat, tolfenpyrad, flupyradifurone, and sulfoxaflor on eggs and nymphs of whiteflies *B. tabaci*. The project covers also the examination of the effect of these insecticides on eggplant yield under greenhouse conditions.

2. Materials and Methods

2.1 Experimental Set-up

The field experiment was conducted on an eggplant plantation under greenhouse conditions. The used eggplant variety trade name “Sawad el leil” is produced by SEMINIS and it is characterized by its large size and black color and is designed to be planted in open fields and protected fields. It is also characterized by its high production reaching 4 to 5 kg per plant per season.

Two experimental greenhouses were selected in the coastal area, north of Beirut (34°02’02” N - 35°37’57” E and 34°08’44” N - 35°37’59” E). 34°08’44” N - 35°37’59” E.

The design of both experiments is based on splitting the greenhouse into three different compartments (Table 1).

Table 1. Greenhouse sizes and locations

Greenhouses	Compartment Code	Surface (m^2)	Number of plants
1	A	78	210
	B	78	280
	C	67	210
2	D	50	135
	E	50	180
	F	50	180

Inside each greenhouse, adjacent compartments (plots) were built up with a white insect net avoiding the movement of whiteflies from one compartment to another. The compartments consist of parallel corridors, 3 m wide and 2.5 m high. Walls, ceilings, and doors were all made up with the insect net.

Transplantation took place on June 14, 2019; plot doors were kept open for more than one month to ensure a natural infestation of whiteflies.

2.2. Soil Preparation, Fumigation, and Transplantation

The soil preparation consisted of several plowing followed by a soil covering with a 50-micron plastic film and fumigation using allyl isocyanate (Dominus®). After 14 days, the cover was removed and the soil was ventilated for two days before transplantation.

2.3. Fertilization and Eggplant Plantation

The fertilization program was based on two different but complementary phases, the pre-planting phase, and the fertigation. The first phase took place just after plastic film removal and consisted of granular fertilizer and organic amendment soil application. The fertigation started 10 days after seedling transplantation and consisted of using water-soluble fertilizers based on the developmental stages of eggplant crops.

2.4. Insecticides and their Applications

The used insecticides in the field experiments were as follows; Pyrifluquinazon (Colt® 20WG, Nihon Nohyaku LTD) was used alone in one compartment in each of the two experimental greenhouses. Cyantraniliprole (Benevia® 10OD, FMC) was also used alone. A spray program consisting of 5 insecticides namely, Sulfoxaflor (Closer® 240SC, DAS), Spiromesifen (Oberon® 240SC, BCS), Tolfenpyrad 15EC (Hachi hachi®, Nihon Nohiyaku LTD), Spirotetramat (Movento®, BCS), and Flupyradifurone (Sivanto®, BCS.) used alternatively in one compartment in each of the two experimental greenhouses. Insecticide applications were made at 10-15 day intervals (Table 2). In the first compartment of the two experimental greenhouses, Cyantraniliprole was applied for five consecutive sprays. In the second compartment, plants were sprayed with Pyrifluquinazon for five subsequent applications, and the third compartment of both greenhouses was designed to be a positive control plot since five different insecticides were applied successively at 10-15 day intervals (Table 2).

2.5. Infestation

The induction of the plant infestation in all compartments, and in both greenhouses was made naturally since after the transplantation, the doors of the compartments were left open for five weeks to allow the whiteflies to build their population on eggplant leaves.

2.6. Insecticide Impact on Yield

Eggplants were harvested on the 10, 19, and 24 August 2019, and 5, 10, 14, 18, 25, and 28 September 2019. Eggplant yields were estimated by weight produced by the number of eggplants per plot. Before statistical analysis, the total eggplant yields per plot were calculated.

2.7. Data Collection and Statistics

Five assessments took place just before each one of the five applications of insecticides. In addition, a sixth assessment took place fourteen days after the last application. Each assessment consisted of a random collection of five infested leaves from each compartment in both greenhouses. Collected leaves were brought to the laboratory where eggs and nymphs were counted.

The % reduction in egg or larval population over the positive control was calculated using Henderson & Tilton formula [40].

Total yields were analyzed using an analysis of variance (ANOVA) to assess the effect of both insecticides and positive control. Means were separated using Tukey HSD following a significant effect of insecticide ($P<0.05$).

3. Results

3.1. Efficacy of Insecticides on Whiteflies Eggs and Nymphs

Before any application, both greenhouses presented no significant differences in the level of eggs and nymphs. Moreover, the number of eggs and nymphs of all treatments within the same greenhouse recorded relatively no statistical significance (Table 3).

Table 2. Insecticide application timing and rates of use in eggplant greenhouses

Number of applications	Date of application	Plot codes 1	Plot codes 2	Insecticide treatments	Rate of use (mg A.I. /m^2)
First application	July 24, 2019	A	D	Cyantraniliprole	14.5
		B	E	Pyrifluquinazon	11.7
		C	F	Spiromesifen	12
Second application	August 3, 2019	A	D	Cyantraniliprole	14.5
		B	E	Pyrifluquinazon	11.7
		C	F	Spirotetramat	7.5
Third application	August 18, 2019	A	D	Cyantraniliprole	14.5
		B	E	Pyrifluquinazon	11.7
		C	F	Tolfenpyrad	9.3
Fourth application	August 31, 2019	A	D	Cyantraniliprole	14.5
		B	E	Pyrifluquinazon	11.7
		C	F	Flupyradifurone	17.9
Fifth application	September 14, 2019	A	D	Cyantraniliprole	14.5
		B	E	Pyrifluquinazon	11.7
		C	F	Sulfoxaflor	7.8

Table 3. Number of eggs and nymphs before treatments in greenhouses 1 and 2

Treatments	**Number of eggs per leaf before the first treatment**		**Number of nymphs per leaf before the first treatment**	
	Greenhouse 1	Greenhouse 2	Greenhouse 1	Greenhouse 2
Pyrifluquinazon	558 ±285 a[*]	558 ±215 a	391 ±201 a	481 ±198 a
Cyantraniliprole	741 ±348 a	777 ±339 a	620 ±274 a	387 ±132 a
Positive control	793 ±383 a	1718 ±591 a	314 ±137 a	552 ±229 a

[*] Mans followed by the same letter within the same row are not significantly different ($P>0.05$)

Ten days after the first application, both greenhouses revealed significant differences in the number of eggs ($F = 9.471$; $df = 1$; $P<0.05$) and nymphs ($F = 7.414$; $df = 1$; $P<0.05$). In both greenhouses, the corrected % reduction in egg population was 94.26% and 99.48% in pyrifluquinazon-treated plots (Tables 4 and 5). As for the number of nymphs, just before the second application, pyrifluquinazon showed the lowest nymph infestation. The corrected % reduction in whiteflies nymph population was 98.09% and 98.88% in greenhouses 1 and 2 respectively (Tables 4 and 5). Cyantraniliprole did not succeed to decrease the number of eggs and nymphs two weeks after the first treatments; the corrected percentage reduction in the eggs population was 0 in both greenhouses and the corrected % reduction in the nymph population was 4.56 % and 0% in greenhouse 1 and 2, respectively (Tables 4 and 5).

Two weeks after the second application, the number of eggs ($F= 6.659$; $df = 1$; $P<0.05$) and nymphs ($F = 21.549$; $df = 2$; $P<0.05$) were significantly different in the different treatments in both greenhouses. The corrected % reduction in eggs population in the pyrifluquinazon treated plot was 99.63% and 99.96% in greenhouses 1 and 2, respectively, in comparison to 37.79% and 19.15% in Cyantraniliprole treated plots (Tables 4 and 5). Regarding the number of nymphs, pyrufluquinazon showed the highest control efficiency after the second application where the corrected percentage reduction was 99.59% and 99.98% in greenhouses 1 and 2 respectively, in comparison to a reduction of 36.02% and 44.04% in cyantraniliprole treated plots (Tables 4 and 5).

Thirteen days after the third application, the number of eggs in both locations revealed a significant difference ($F = 5.141$; $df = 1$; $P<0.05$). In addition, the corrected percentage reduction in eggs was 99.67 and 99.98% in pyrifluquinazon-treated greenhouses while the percentage reduction in eggs in cyantraniliprole was 98.03% in greenhouse 1 and 9.43% in greenhouse 2. As for the nymphs, the corrected percentage reduction was 99.77 % and 100% in nymphs' population in the pyrifluquinazon-treated plots of greenhouses 1 and 2. In cyantraniliprole plots the percent reduction was 100% and 40.07% in whiteflies nymphs' population greenhouses 1 and 2, respectively (Tables 4 and 5).

Table 4. Corrected percent reduction in the eggs and nymphs of whiteflies in greenhouse 1

		10 DA first spray	**15 DA 2nd spray**	**13 DA 3rd spray**	**14 DA 4th spray**	**14 DA 5th spray**
Pyrifluquinazon	Eggs	94.26%	99.63%	99.67%	99.94%	99.38%
	Nymphs	98.09%	99.59%	99.77%	100%	97.46%
Cyantraniliprole	Eggs	0%	37.79%	9.43%	10.92%	23.69
	Nymphs	4.56%	36.02%	40.07%	74.01%	76.25

*DA: Days after

Table 5. Corrected percent reduction in the eggs and nymphs of whiteflies in greenhouse 2

		10 DA first spray	**15 DA 2nd spray**	**13 DA 3rd spray**	**14 DA 4th spray**	**14 DA 5th spray**
Pyrifluquinazon	Eggs	99.48%	99.96%	99.99%	99.99%	99.98%
	Nymphs	98.88%	99.98%	100%	100%	97.91%
Cyantraniliprole	Eggs	0%	19.15%	98.03%	73.88%	42.47%
	Nymphs	0%	44.04%	100%	61.18%	0%

*DA: Days after

Two weeks after the fourth application, the corrected percentage reduction in the eggs population was 99.94% and 99.00% in the whitefly eggs population in the pyrifluquinazon-treated plots and 10.92% and 73.88% in the cyantraniliprole-treated plots of greenhouses 1 and 2, respectively (Tables 4 and 5). The populations of the nymphs were reduced by 100% in the pyrifluquinazon-treated plots in both greenhouses whereas in the cyantraniliprole treated plots the corrected percentage reduction in the nymph's population was 74.01% and 61.18%, respectively (Tables 4 and 5).

Fifteen days after the fifth application, a significant difference in the number of nymphs was shown between the two treated plots in both greenhouses. In greenhouse 1, the corrected percentage reduction in the whitefly's eggs population was 99.38% in the pyrifluquinazon-treated plots and 23.69% in the cyantraniliprole-treated plot as compared to the positive control which received 5 applications of different insecticides. The whiteflies nymphs' populations were reduced by 97.46% when pyrifluquinazon was used and by 76.25% in the cyantraniliprole-treated plot in greenhouse 1. In greenhouse 2, the corrected percent reduction in the whiteflies egg population was of 99.98% in the pyrifluquinazon-treated plot and of 42.47% in the cyantraniliprole plot. The nymph populations were reduced by 99.91% in the pyrifluquinazon-treated plot whereas there was no reduction in the cyantraniliprole-treated plot in greenhouse 2 (Tables 4 and 5).

3.2. Impact of Whiteflies and Insecticide Treatment on Yield of Eggplants

In greenhouse 1, there was a significant effect of treatments on the yield of eggplants (F=14.87; df=3; P=0.000063). Maximum yield was recorded in the pyrifluquinazon (3.9Kg/plant) followed by cyantraniliprole (2.2 Kg/plant). Minimum yield was recorded in the positive control where 5 different insecticides were used (0.4 kg/plant).

In greenhouse 2, there was also a significant effect of treatments (F=34.32; df=2; P<0.00001). The maximum yield was recorded in pyrifluquinazon (3.8Kg/plant), followed by cyantraniliprole (2.7kg/plant) and the lowest yield was recorded in the positive control where the yield was only 0.2 kg/plant).

4. Discussion

In this study and based on the field experiments in two different greenhouses, it was statistically demonstrated that the active ingredient pyrifluquinazon exhibits the highest insecticidal potency against whiteflies egg and nymph densities. Moreover, pyrifluquinazon-treated plots revealed the highest yield in comparison to all other treatments.

In the first greenhouse, pyrifluquinazon showed a noteworthy reduction in egg and nymph densities. In pyrifluquinazon plot, the number of eggs decreased from 558 ±285, before the first treatment to 29 ±6, seventy-five days after the first application and after five consecutive treatments at 10-15 day intervals (Table 4). As long as pyrifluquinazon is a TRPV channel modulator with a feeding cessation effect and not an ovicide [41,42], the number of eggs has decreased because of its larvicidal activity on newly hatched crawlers [43] leading to the lowering of whole whiteflies population thus, a sharp reduction in eggs densities. These results confirmed the results obtained by the same authors [44] following four consecutive spraying. Pyrifluquinazon is a contact insecticide with translaminar movement for the control of Hemiptera and some Thysanoptera in vegetables, fruit trees, and citrus [45]. Furthermore, pyrifluquinazon demonstrated a remarkable decrease in nymph density as it was maintained significantly low during all treatments as shown in table 4, this is due to the nymphicidal activity [43]. In Cyantraniliprole treated plot, the number of eggs was remarkably higher than that of pyrifluquinazon as it increased from 741 ± 348 before the first treatment to 3520 ± 275 at the end of the experiment (Table 4). In addition, the nymph density was considerably high during all treatments (Table 4). The low efficacy of cyantraniliprole could be due to the buildup of resistance and / or cross-resistance as demonstrated by Wang et al. [46]. Results after spraying five consecutive different insecticides showed very low efficacy in comparison to pyrifluquinazon. Bielza *et al*. [31] demonstrated the development of resistance buildup for Spiromesifen and spirotetramat in parallel. On the other side, Smith *et al*. [47] revealed high tolerance of whiteflies population to flupyradifurone as an indication of a certain degree of cross-resistance for neonicotinoids. Moreover, the low insecticidal potency of eggs and nymphs of sulfoxaflor is mainly because it is directed to adult whiteflies [48]. Results of all treatments were reflected directly in the production of eggplants since the highest crop yield was obtained in pyrifluquinazon treated plot.

Similar to the first greenhouse, results in the second greenhouse indicated that pyrifluquinazon is an effective tool to be a part of the integrated and resistance pest management control of whiteflies, pyrifluquinazon showed a very high insecticidal efficacy resulting in a sharp decrease of egg and nymph densities (Table 5). Regarding cyantraniliprole-treated plot, results showed a lower efficacy on the nymphs (Table 5). Similar to the first location, the eggplant production, showed a direct relation between the efficacy of insecticides and the decrease of whiteflies *B. tabaci* population. In this case, the highest yield of eggplants was in pyrifluquinazon-treated eggplants as long as whiteflies damage is reduced. Damage caused by a high infestation of whiteflies is an example of how plant growth and production could be negatively affected leading to sharp economic losses [49-53].

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