American-Eurasian J. Agric. & Environ. Sci., 11 (2): 228-236, 2011
ISSN 1818-6769

Bioresidual Effect of Two Insecticides on Melon Aphid *Aphis gossypii* Glover (Homoptera: Aphididae) and its Parasitoid *Aphidius colemani* Verick (Hymenoptera: Brachonidae)

[1]Leena A. Irshaid and [2]Hazem S. Hasan

[1]Department of Biotechnology, Faculty of Agricultural Technology,
Al-Balqa' Applied University, Al-Salt 19117, Jordan
[2]Department of Plant Production and Protection, Faculty of Agricultural Technology,
Al-Balqa' Applied University, Al-Salt 19117, Jordan

Abstract: Bioresidual effects of two insecticides, cypermethrin and chlorpyrifos, were studied using *Aphis gossypii* and its parasitoid *Aphidius colemani* that grown on water melon plants under laboratory conditions. A total of 135 water melon plants were used in different treatments to study the direct and indirect effects of the two insecticides on both aphid adults and parasite as adult and as pupa inside aphid mummies. The application rate of the insecticides was 0.125ml/ 0.25 L for cypermethrin and 0.25ml/0.25 L for chlorpyrifos. 100% mortality of aphid was obtained using chlorpyrifos when aphid introduced to plants at same day of spray, while cypermethrin produced 81.6% aphid mortality. chlorpyrifos revealed longer period of plant protection against aphid infestation for about two weeks more than cypermethrin. For Integrated Pest Management program of *A. gossypii* by using the parasitoid *A. colemani*, cypermethrin showed a high survival percentage for parasitoid when introduced 72 hr after spraying; while chlorpyrifos was severely harmful to the parasitoid adults and parasitoid pupae in aphid mummies at all post-spraying introducing periods.

Key words: *Aphis gossypii* · *Aphidius colemani* · Bioresidual effect · Chlorpyrifos · Cypermethrin · IPM

INTRODUCTION

The melon aphid, *Aphis gossypii* Glover (Homoptera: Aphididae) is one of the most serious insect pests in agriculture causing damages either directly by feeding which resulted in curling and deformation of young leaves and twigs, or indirectly by contaminating thr fruits with honeydew which in turn may case, transmission of plant viruses and the growth of black sooty mould which inhibits photosynthesis and therefore causes substantial yield loss [1-3]. Also, honeydew attracts ants which fend off natural enemies of Homopterans [4].

Chemical treatment remains the main method of control, even though resistance to insecticides has been documented in many *A. gossypii* populations including those in Australia [5], France [6], Hawaii [7], Holland [8], Japan [9], China [10], Sudan [11], Pakistan [12], the USA [13,14] and UK [15-17].

Chemical control and biological control are the two important strategies used in an IPM program, Successful biological control of *A. gossypii is* a pre-requisite for implementation of Integrated Pest Management (IPM) programs. Several aphid parasitoid species (Hymenoptera: Brachonidae: Aphidiinae) have been tested for their usefulness as biological control agent based on population growth rates, searching capacity and host preferences. *Aphidius* species can parasitize many aphids daily during a reproduction period for a few days, whereas *Aphelinus* parasitoids parasitize fewer aphids daily during a reproduction period for many days. *Aphidius colemani* Vrick is a solitary endoparasitoid, which parasitizes many aphid species belonging to Aphididae. *A. colemani* is proved to be most promising parasitoid in *A. gossypii* control [18].

The value of aphid parasitoids in IPM programs that incorporate insecticide, remains limited as the adult stage

Corresponding Author: Leena A. Irshaid, Department of Biotechnology, Faculty of Agricultural Technology, Al-Balqa' Applied University, Al-Salt 19117, Jordan.

of the aphid parasitoid is known to be extremely vulnerable to agrochemicals when compared to other non-target beneficial invertebrates [19]. However, lethal and sub-lethal effects of broad-spectrum and non-selective pesticides are usually considered a high risk to beneficial species [20,21] Misuse of chemical insecticides might be accountable for the outbreaks of the pest because extensive and intensive use of insecticides applied heavy selection pressure on target pests and accelerated resistance development [22,23]. The adult stage of a parasitoid can become exposed to insecticide through direct exposure to spray droplets at the time of spraying, or through residual exposure to insecticide residues on the crop foliage via foraging behavior and through dietary exposure while feeding on contaminated water droplets and aphid honeydew [24]. These routes of exposure are not of equal importance, but each pose different threats to parasitoid survival during different period of time [25].

Desneux *et al.* [26] recently pointed out that the determination of the acute toxicity of pesticides to beneficial arthropods had traditionally and largely relied on the measurement of an acute median lethal dose or concentration and the estimated lethal dose or concentration might only be a partial measure of the deleterious effects. In addition to direct mortality induced by pesticides, their sub-lethal effects on arthropods must be considered for a complete analysis of their impact.

Organophosphate insecticides were reported to be more toxic to developing parasitoids than either pyrethroid or carbamate compounds, if the pesticides' application time not consider parasites appearance and activity, the whole adult parasitoid population can be destroyed by a single insecticide spray to arable crops; however the immature parasitoids are only protected from some pesticides within mummy stage. [24]. Umoru and Powel [27] used a resistant strain of *Myzus persicae* in laboratory experiments to investigate the sub-lethal effects of the Dimethoate and Primicarb on the parasitoid *Diaeretiella rapae.* Both insecticides produced sub-lethal effects on *D. rapae* when the parasitoid attacked and developed in aphid that had been dipped in insecticide solutions.

In this study, two insecticides (Cypermethrin as non-systemic pyrethroid and chlorpyrifos from organo-phosphate group) were evaluated against melon aphid and its parasitoid to assess the feasibility of using pesticides for the reduction of aphid population on host plants without having any adverse effect on the parasitoid in order to improve IPM program for melon aphid.

MATERIALS AND METHODS

Aphid and Parasite Culture: Samples collection and laboratory experiments were conducted at Al-Balqa' Applied University research station (Al-Salt, Jordan) during the period from February 2007 to the end of September 2007. The adults of melon aphid (*Aphis gossypii*) and the parasitic wasps (*Aphidius colemani*) from aphid mummies were collected from an insecticide-free vegetable plants grown in the green house. Water melon plants grown in protected cages were used as an aphid host stock.

To maintain the parasite culture, modified Petri dishes were used. The cover of the Petri dishes were turned upside down and fixed to other half by a Super Glue. A small hole was made in the cover through which the bottom of the Petri dish was filled with distilled water. Water melon leaf infested with the aphid was inserted via its stalk into the hole. Adult parasites were kept on the water melon leaf and Petri dish was covered by muslin for ventilation.

Experimental Plants: Water melon seeds were cultured in small plastic pots (one seedling/pot) containing perlite and kept inside plant growth chamber at 25±1°C; pots were daily irrigated with Hoagland solution [28]. One month old plants were moved into insecticide-free cages to be used for different bioassays work.

Insecticide Preparation and Application: Two insecticides were used, the first was Cypermethrin as non-systemic pyrethroid with 10% (weight / volume) active ingredient and the second one was Chlorpyrifos from organo-phosphate group with 48% (weight / volume) active ingredient. Cypermethrin dose was 0.125ml/ 0.25 L and chlorpyrifos dose was 0.25ml/0.25 L

Third treatment was prepared as control. To study the bioresidual effects of the two insecticides on melon aphid and its parasite, 45 pots of three true leaves water melon plants were sprayed with cypermethrin, 45 pots were sprayed with chlorpyrifos and 45 pots were used as control (sprayed with distilled water and surfactant).

Cypermethrin Treatment: Twenty five of apterous aphids were transmitted using fine hair brush to each of 20 pots that sprayed with either cypermethrin or chlorpyrifous. Aphids were transmitted to pots according to the following program: 5 pots with same day of spray, 5 pots after 1 week of spray, 5 pots after 2 weeks of spray and 5 pots after 3 weeks of spray. The surface of each pot was covered completely with white paper to observe any aphid that may drops from any Plants.

Twenty five of newly emerged adult parasites were introduced to each of 20 sprayed pots which covered with ventilated cylindrical plastic cage (7 cm× 30 cm) to prevent parasites escape. Parasites introduced to the cage using glass vials (1 cm× 5 cm) according to the following program: 5 pots at same day of spray, 5 pots after one day of spray, 5 pots after two days of spray and 5 pots after three days of spray. Parasites were supplied with diluted honey on a piece of cotton as food.

For parasite mummy study, adult parasites were introduced to 5 pots of water melon plants infested with aphid for 24 hours; each pot was covered with ventilated cylindrical plastic cage (7 cm×30cm) to prevent parasites escape. After 7 days of introduction, mummy's appearance where checked and 25 mummies only were left on each pot. Then pots were sprayed with the cypermethrin and were watched for parasite adult's emergence.

All the 45 treated pots were kept in the laboratory at room temperature. Mortality percentage of insects was recorded after 4, 24, 48, 72, 96 hours of treatment.

Data Analysis: SAS program (version 7) based on General Liner Model (GLM) was used for data analysis. Split-Split Plot with Complete Randomize Design (CRD) and variance test were carried out to determine significant difference among treatments in all experiments. For mummy experiment, CRD was used. Significantly different mean values of treatments were then separated using Least Significant Deference test (LSD).

RESULTS

Effect of Cypermethrin on *A. gossypii*: Data of mortality percentage of A. gossypii as affected by cypermethrin and insect introduction time are summarized in Table 1. In general, aphid mortality rate was increased by the treatment and by increasing the exposure time to cypermethrin. Also, the present study examined the effect of different introduction time of the target aphid after the initial insecticide spray. The general trend of the result showed that cypermethrin exerted lesser impact on the mortality percentage of *A. gossypii* when aphid introduction on the sprayed plants was postpond (Table 1).

Effect of Cypermethrin on *A. colemani:* The effect of cypermethrin on the mortality percentage of the parasitoid *A. colemani* summarized in Table 2 indicated that the effect of the insecticide increased by increasing the exposure time and decreased by increasing the introduction time of the parasitoid on the host plant (Table 2). After 72 hr and 96 hr of treatment, mortality percentage were more sever compound to 24 hr and 48 hr. Moreover, introducing the parasitoid at the sane day of treatment resulted in higher mortality compound to the introduction after 2 or 3 days (Table 2).

Effect of Chlorpyrifos on *A. gossypii* and the Parasitoid: The Mortality percentage of *A. gossypii* and its parasitoid *A. colemanii* was further investigated in response to Chlopyrifos insecticide (Table 3 and 4).

Table 1: Mortality percentage of *A. gossypii* introduced to water melon plants sprayed with Cypermethrin at different post-spraying periods

		Aphid mortality percentage			
Treatments	aphid introduction time	24 hr	48hr	72 hr	96 hr
Treated	Same day	46.6fgh	88.7 abc	91.4abc	100 a
Control		13.3lmn	24.4 jklm	24.4 jklm	35.5hij
Treated	After one week	53.3ef	64.7de	82.2 bc	95.6 abc
Control		6.7 n	17.8klmn	24.4 jklm	35.6 hij
Treated	After two weeks	49.3efg	64.7 de	82.2 bc	85 bc
Control		8.9mn	11.1 lmn	26.7 ijkl	33.3hijk
Treated	After three weeks	42.2 fghi	58 ef	80 cd	97.8ab
Control		4.4 n	10.9mn	13.3lmn	17.8klmn

Means followed by the same letter are not significantly different at P=0.05 by LSD=17.0

Table 2: Mortality percentage of *A. colemani* introduced to water melon plants sprayed with Cypermethrin with different post-spraying periods

		A. colemani mortality percentage			
Treatments	*A. colemani* introduction time	24 hr	48hr	72 hr	96 hr
Treated	Same day	73.3 bcde	86.7 abcd	100 a	100 a
Control		13.3ijkl	26.7 ijkl	60 def	82.2 abcd
Treated	After one day	46.7 fghi	90 abc	93.3 ab	100 a
Control		6.7 jklm	10.0 jklm	30.9 ghij	57.8 efg
Treated	After two days	33.3 ghij	60 def	93.3 ab	100 a
Control		8.9 ijkl	11.7 ijkl	14.8 ijkl	20.6 hijkl
Treated	After three days	26.7 ijkl	40 ghij	46.7 ghij	93.3 ab
Control		11.7 klm	13.3klm	15.6klm	20.8 hijkl

Means followed by the same letter are not significantly different at p=0.05 by LSD=17.7

Table 3: Mortality percentage of *A. gossypii* introduced to water melon plants sprayed with Chlorpyrifos at different post-spraying periods

		Aphid mortality percentage			
Treatments	Aphid introduction time	24 hr	48hr	72 hr	96 hr
Treated	Same day	100 a	100 a	100 a	100 a
Control		13.3 ijk	24.4 ghij	24.4ghij	35.5fg
Treated	After one week	77.8bc	89 ab	95.6a	100 a
Control		6.7k	17.8hijk	24.4ghij	35.6fg
Treated	After two weeks	55.6de	62.2 cd	77.8 bc	100 a
Control		8.9 jk	11.1 ijk	26.7 ghi	33.3fgh
Treated	After three weeks	53.3de	62.2 cd	84.4 ab	100 a
Control		4.4 k	10.9 ijk	13.3 ijk	17.8 hijk

Means followed by the same letter are not significantly different at p=0.05 by LSD=20.16

Table 4: Mortality percentage of *A. colemani* introduced to water melon plants sprayed with Chlorpyrifos at different post-spraying periods

		A. colemani mortality percentage			
Treatments	*A. colemani* introduction time	24 hr	48hr	72 hr	96 hr
Treated	Same day	100 a	100 a	100 a	100 a
Control		13.3 jk	26.7 hi	60 def	82.2 bc
Treated	After one day	100 a	100 a	100 a	100 a
Control		6.7 k	10.0 jk	30.9 hi	57.8 efg
Treated	After two days	100 a	100 a	100 a	100 a
Control		8.9 jk	11.7 jk	14.8 jk	20.6 ij
Treated	After three days	100 a	100 a	100 a	100 a
Control		11.7 jk	13.3 jk	15.6 jk	20.8 ij

Means followed by the same letter are not significantly different at p=0.05 byLSD=12.45

Table 5: Effect of pesticides treatment on the *Aphis gossypii* adult mortality when introduced to water melon plants at different post-spraying periods

	Aphid mortality percentage	Time after spraying		
Treatments	5 hr	1 week	2 weeks	3 weeks
Cypermethrin	81.6 b	66.7 c	65.2 c	59.9 c
Chlorpyrifos	100 a	87.4 b	66.8 c	66.7 c
Control	26.4 d	25.2 d	15.5 e	11.8 e

Means followed by the same letter in column and raw are not significantly different at p=0.05 by LSD =7.4

Table 6: Effect of pesticides treatment within time on adult mortality of *Aphidius colemani* when introduced to water melon plants after treated with pesticides

	Aphidius colemani adult s mortality percentage			
	Hours(s)			
Treatments	5	24	48	72
Cypermethrin	90.0 b	80.0 c	62.2 d	37.8 f
Chlorpyrifos	100 a	100 a	100 a	97.8 a
Control	46.7 e	20.0 h	28.9 g	29.6 g

Means followed by the same letter in same column are not significantly different at p=0.05 by LSD =6.81

Table 7: Effect of two Pesticides chlorpyrifos and Cypermethrin on *Aphidius colemani* adult emergence percentage

	Aphidius colemani Adult Emergence Percentage
Cypermethrin	64.00 b
Chlorpyrifos	10.67 c
Control	88.00 a

Means followed by the same letter are not significant different at p=0.05 by LSD=23.22

When introducing the melon aphid at the same day of spraying, treatment with chlorpyrifos resulted in complete mortality irrespective to the exposure time (Table 3). The mortality percentage generally tend to decrease when increasing the aphid introduction time (Table 3).

The insecticide chlorpyrifos induced a severe effect on the parasitoid *A. colemani*. The mortality percentage at different parasitoid introduction time and at different exposure timereached the level of 100% (Table 4).

Direct Effect of Both Insecticides on *A. gossypii* and the Parasitoid *A. colemanii:* The purpose of the present study also extended to investigate the toxicity effects of cypermethrin and chlorpyrifos on the adult of *A. gossypii* (Table 4) and *A. colemanii* (Table 6).

The mortality percentage of melon aphid adults was significantly affected by both insecticides (Table 5). The highest mortality was recorded for chlorpyrifos (100%) after 5 hr of treatment. However, at the same time, cypermethrin resulted in 81.6% adult mortality of melon aphid. Differences in mortality percentage among different treatments were non-significant at 2 and 3 weeks after treatment (Table 5).

All the insecticides were significantly toxic to the adults of *A. colemani* (Table 6).When using cypermethrin the mortality percentage decreased with the time after treatment and reaching its maximum (90%) after 5 hr of treatment. chlorpyrifos resulted in more sever effects with almost complete mortality at all-time points (Table 6).

Effect of Both Insecticides on *A. colemani* Mummies: Results of the effects of cypermethrin and chlorpyrifos on the adult emergence of *A. colemanii* are illustrated in Table 7. Both insecticides exerted significant effects. cypernmethrin resulted in 64% adult emergence, while chlorpyrifos resulted in 10.67% (Table 7).

DISCUSSION

Since *Aphis gossypii* is a polyphagous pest cause different damage levels; directly or indirectly to several plants, [1-4], farmers used to control such pest with insecticides, however, populations of this pest have demonstrated the ability to develop resistance to several insecticides especially for organophosphate and charbamate insecticides [11, 15, 24, 30-32]. Aphid insects examined in this study were susceptible to the examined insecticides and gave high mortality however, an appropriate management strategy for pesticide resistance should be considered. Hymenopteran parasitoids are important in controlling aphid naturally and are used in biological control programs of them in diverse crops [33]. Although biological control is desirable, aphid having high reproductive rate and mobility, so they will be very difficult to control by biological means only and require selective insecticides acting together with natural control [34].

Proper IPM program must be established to control this pest, depending on integration between parasitoid application and a non-persistent pesticide spraying. Suitable insecticide in IPM program is essential to choose effective dose against aphid but still allows the surviving aphid nymphs to be attacked subsequently by adult parasitoids [35].

Aphid mortality with cypermethrin was high and this result is similar to Zhong *et al.* [36], how found that cypermethrin significantly increased the death mortality of *Aphis gossypii.* But *A. gossypii* mortality with cypermethrin differed according to post spray periods, so when aphid introduced to plants in same day treatment, mortality percentage was high since insects have double dose of insecticide at same day treatment (Table 1).

The direct effect of cypermethrin was significantly lower than its indirect effect which is accumulated with time. After three weeks of post spray, both direct and indirect effects were reduced, so cypermethrin reduced aphid population but without eradication of aphid which will be source for parasitoid maintenance in the field.

The sublethal effects of insecticides on insect pests and its parasitoids are of paramount importance when making an insecticide selection cypermethrin cause high mortality percent in parasite *A. colemani* when it was introduced at same day of spray because adults waked over the insecticide deposits and encounter the spray droplets directly. In the current study, *A. colemani* appeared to suffer greater mortality than their aphid hosts after exposure to cypermethrin residues. This difference in susceptibility is likely to be based on mobility. Parasitoids were very mobile and showed an active searching behavior when they were exposed to plant surfaces, thus, they came into contact with higher doses of insecticide residues than the relatively static, phloem-feeding aphids. This is similar to found Langhof *et al.* [37] who recorded that *A. colemani* appeared to suffer greater mortality than their aphid hosts *Myzus persicae* (Hemiptera; Aphididae) after exposure to Lannate® 20 L residues.

cypermethrin affect the parasitoid at same day, one day, two days after spraying but its effect was noticed to be reduced after the third days of spraying (Table 2). At the same time it provide aphid control for about three weeks after spraying. cypermethrin is the most suitable for (IPM) program for *A. gossypii* with using the parasitoid *A. colemani*. but in this case *A. colemani* must be introduced after at least 4 days after spraying to have a high survival percentage for the parasites. So, during the planting season, secondary infestation by *A. gossypii* with the presence of *A. colemani* as mummies can be controlled with cypermethrin spray, it is preferable to spray it in spot treatments for aphid colony and avoiding culture of parasites mummies.

Since adult parasitoids are killed by the spray and also by residual activity, mummies may constitute a reservoir for aphid parasitoids. Mummified aphid can provide high degree of protection for immature parasites against cypermethrin. Therefore, these protected immature parasites may have the ability to serve as effective reservoir of adult parasites, helping to prevent further aphid resurgence in a crop after pesticide treatments.

Other studies have also reported that the pupal stage within a mummified aphid was the least susceptible to insecticide since adult parasitoids are killed by the spray and also by residual activity, mummies may constitute a reservoir for aphid parasitoids [38,39]. Indeed, at the deltamethrin (pyrethroid insecticide) field rate there was no significant effect on emergence of *Aphidius ervi* (active aphid parasite) when exposed at the mummy stage but found high rates of mortality on adults [39]. Krespi *et al.*, [40] and Jansen, [41] also reported an absence of deltamethrin effects on the emergence rate of *Aphidius rhopalosiphi* from mummified aphids. Furthermore Abo El-Ghar and El-Sayed [42] found that cypermethrin is an active insecticide against both the cabbage aphid Brevicoryne brassicae (L.) and the parasitoids Diaeretiella rapae (M'Intosh) within the mummified aphids but it was more active against *B. brassicae* than against the parasitoids.

Another high-risk insecticide group is the organophosphate insecticides. chlorpyrifos, has high acute oral toxicity to *Aphis gossypii.* chlorpyrifos deposits were effective against aphid adults over the first 24 hr, it is effect against aphid declined over 3 weeks but still harmful, same was reported by Ibrahim *et al.* [43] who found that Diafenthiuron (organophosphate insecticides) is able to control *Aphis gossypii* even for long period (three weeks) since it has good post-spray deposit activity.

Chlorpyrifos was more efficient than cypermethrin in controlling aphid even after three weeks of spraying and all aphids were killed even after 96 h of introduction to three weeks post spraying period. With long post spraying periods the direct effect of chlorpyrifos reduced but its indirect effect accumulated with time leading to aphid mortality. Chlorpyrifos gave complete control with all post spraying periods which indicate that it is a long lasting pesticides, this is similar to Desneux *et al.* [44] who found that chlorpyrifos was about 100 times more effective than cypermethrin in affecting *Aphidius ervi* (a relatively generalist parasitoid of aphids).

Chlorpyrifos gave longer period of plant protection from aphid infestation as two weeks more than cypermethrin at least (Table 3) therefore, it can be used as pre-infestation spray before aphid colonizing the plants specially at the beginning of the planting season, which protect the transplants from highly dangerous early infestation, since plants are weak, succulent and affected severely with primary infestation especially if it is associated with virus transmission, which can lead to highly reduction in the plants growth and yield. In a study to determine the response of the melon aphid to certain insecticides in the Jordan Valley, Nazer *et al*. [45] found that pirimiphos-methyl (Organophosphate) and pirimicarb (Charbamate) were more effective compared to cypermethrin (pyrethroid).

Chlorpyrifos prove more toxic to developing parasitoids than cypermethrin, Longley [24] reported that organophosphate compounds prove more toxic to developing parasitoids than pyrethroid and carbamate compounds.

The present experiment proved that chlorpyrifos is harmful to adults and pupae of the parasitoids while cypermethrin is harmless to pupae, same result was obtained by Desneux *et al.* [44] who ranked five different insecticides in order of increasing toxicity as: pirimicarb, triazamate, lambda-cyhalothrin and chlorpyrifos, the latter being about 100 times more toxic than pirimicarb. They found also that 1/68 of the field application for pirimicarb will kill 50% of the parasitoid *Aphidius ervi*, while it was 1/79 for chlorpyrifos.

Wang *et al*. [46] found that chlorpyrifos had the highest contact toxicity to *Anagrus nilaparvatae* (Hymenoptera: Mymanidae) an egg parasitoid of the rice planthopper with 100% mortality 24 h after treatment; other pyrethroid and carbamate insecticides were less effective.

Li and Liu [47] recorded that chlorpyrifos showed 100% mortality 24 h after treatment of pupal stage of the parasite *Cotesia plutellae*(Hymenoptera: Braconidae) In leaf residue experiments, chlorpyrifos had the highest toxicity, with 100% mortality in adult wasps, while cypermethrin, provided 22 to 40% mortality,

The quantity of insecticide taken up via ingestion was positively correlated with time spent feeding, the results suggest that chlorpyrifos showed little or no repellent properties to feeding *A. colemani a*nd could there four represent an important route of exposure to aphid parasitoids in the field (Table 4). The uptake of residual deposits, mainly the tarsal receptors or setae reaching maximum loading and/or subsequent parasitoid grooming behavior removing insecticide particles from the exterior of the body and cause death to the parasitoid [25]. Chlorpyrifos was harmful to parasitoid adult more than aphid, so it is tempting used at early growing season to prevent early infection with aphid, but after release of parasites it will be harmful to parasite adults and pupae in mummies. In fact aphids are not always evenly distributed. Sometimes they are concentrated in certain areas such as field margins or hot spots within the field. If the infested area is large enough, it may require spot treatment before aphids are uniformly spread across the field.

The high percent of aphid mortality at control treatment in same day of spray was due to disturbance of aphid with water droplets which further more encourage aphid fungus infestation, so there survival decline with time. While after one and two weeks of control post treatment, the low survival percentage can be due to aging of aphid adults or miss transmission of aphid to plants because we have highest survival after 3 weeks of spraying (Table.6)

REFERENCES

1. Jacobson, R.J. and P. Croft, 1998. Strategies for the control of *Aphis gossypii* Glover (Hom: Aphididae) with *Aphidius colemani* Viereck (Hym: Brachonidae) in protected cucumbers. Biocontrol Science and Technology, 8(3): 377-387.
2. Capinera, J.L., 2000. Melon Aphid or Cotton Aphid, *Aphis gossypii* Glover (Insecta: Hemiptera: Aphididae). EENY-173 (IN330), one of the Featured Creatures series of the Entomology and Nematology Department, Cooperative Extension Service, Institute of Food and Agricultural Sciences, University of Florida.
3. Andrews, M.C., A. Callaghant, L.M. Field, M.S. Williamson and G.D. Moores, 2004. Identification of mutations conferring insecticide insensitive AchE in the cotton-melon aphid, *Aphis gossypii* Glover. Insect Molecular Biology, 13(5): 555-561.
4. Yokomi, R.K. and Y.Q. Tang, 1995. Host preference and suuitability of two Aphelinid parasitoids (Hym: Aphilinidae) for Aphid (Hom: Aphididae) on Citrus. Journal of Economic Entomology, 88(4): 840-645.
5. Herron, G.A., K. Powis and J. Rophail, 2001. Insecticide resistance in *Aphis gossypii* Glover (Hemiptera: Aphididae), a serious threat to Australian cotton. Australian Journal of Entomology, 40(1): 85-91.
6. Delorme, R., D. Auge, M.T. Bethenod and F. Villatte, 1997. Insecticide resistance in a strain of *Aphis gossypii* from Southern France. Pesticide Science, 49(1): 90-96.

7. Hollingsworth, R.G., B.E. Tabashnik, D.E. Ullman, M.W. Johnson and R. Messing, 1994. Resistance of *Aphis gossypii* (Homoptera, Aphididae) to insecticides in Hawaii-spatial patterns and relation to insecticide use. Journal of Economic Entomology, 87(2): 293-300.
8. Silver, A.R.J., H.F. Van Emden and M. Battersby, 1995. A Biochemical-mechanism of resistance to pirimicarb in 2 glasshouse clones of *Aphis gossypii*. Pesticide Science, 43: 21-29.
9. Satoshi, T., K. Shinkichi, I. Hiroki, N. Ken, K. Shuji, S. Eiichirou and S. Tomoko, 2008. Development of molecular diagnostics of the two point mutations in acetylcholinesterase gene associated with insecticide resistance in the cotton aphid, *Aphis gossypii* Glover (Homoptera: Aphididae) and a survey of genotypic frequency in field populations. Applied Entomology Zoology, 43(1): 127-133.
10. Chuan-Wang, Z.C., G. Jing, Xi-Wu L. Pei and G. Hui-Lin, 2007. Over expression of carboxylesterase gene associated with organophosphorous insecticide resistance in cotton aphid, *Aphis gossypii* (Glover). Pesticide Biochemistry and Physiology, 90(3): 175-180.
11. Grafton-Cardwell, E.E., T.F. Leigh, W.J. Bentley and P.B. Goodell, 1992. Cotton aphids have become resistant to commonly used pesticides. Calif. Agric., 46: 4-7.
12. Amad, M., M. Iqbal and I. Denholm, 2002. High resistance of field populations of the cotton aphid Aphis gossypii Glover (Homoptera: Aphididae) to pyrethroid insecticides in Pakistan. Journal of Economic Entomology, 96(3): 875-878.
13. Grafton-Cardwell, E.E., 1991. Geographical and temporal variation in response to insecticides in various life stages of *Aphis gossypii* (Homoptera, Aphididae) infesting cotton in California. Journal of Economic Entomology, 84(3): 741-749.
14. Kerns, D.L. and M.J. Gaylor, 1992. Insecticide resistance in field populations of the cotton aphid (Homoptera, Aphididae). Journal of Economic Entomology, 85: 1-8.
15. Furk, C., D.F. Powell and S. Heyd, 1980. Pirimicarb resistance in the melon and cotton aphid, *Aphis gossypii* Glover. Plant Pathology, 29(4): 191-196.
16. Furk, C. and S. Vedjhi, 1990. Organophosphorus resistance in *Aphis gossypii* (Hemiptera, Aphididae) on *Chrysanthemum* in the UK. Annals of Applied Biology, 116(3): 557-561.
17. Furk, C. and C.M. Hines, 1993. Aspect of insecticides resistant in the melon and cotton aphid *Aphis gossypii* (Hemiptera: Aphididae). Ann. Applied Biology, 123: 9-17.
18. Van Steenis, M., 1995. Evaluation and application of parasitoids for biological control of *Aphis gossypii* in glasshouse cucumber crops. WAU dissertation no. 1973. Dissertation, internally prepared.
19. Theiling, K.M. and B.A. Croft, 1988. Pesticide side-effects on arthropod natural enemies: a database summary. Agriculture, Ecosystems and Environment, 21(3-4): 191-218.
20. Greathead, D.J., 1995. Natural enemies in combination with pesticides for integrated pest management. In: R. Reuveni, Editor, Novel Approaches to Integrated Pest Management, Lewis Publishers, F.L. Boca Raton, pp: 183-197.
21. Ruberson, J.R., H. Nemoto and Y. Hirose, 1998. Pesticides and conservation of natural enemies in pest management. In: P. Barbosa, Editor, *Conservation Biological Control*, Academic Press, San Diego, CA, USA, pp: 207-220.
22. Kilin, D., N. Nagata and T. Masuda, 1981. Development of carbarmate resistance in the brown planthopper, *Nilaparvata lugens* (Stål), Appl. Entomol. Zool., 16(1): 1-6.
23. Hirai, K., 1993. Recent trends of insecticide susceptibility in the brown planthopper *Nilaparvata lugens* (Stål) (Homoptera: Delphacidae), Appl. Entomol. Zool., 28(3): 339-346.
24. Longley, M., 1999. A review of pesticide effect upon immature aphid parasitoids within mummified hosts. International Journal of Pest Management, 45(2): 139-145.
25. Longley, M. and D. Stark, 1996. Analytical techniques for quantifying direct, residual and oral exposure of an insect parasitoid to an organophosphate insecticide. Bulletin of Environmental Contamination and Toxicology, 57(5): 683-690.
26. Desneux, N., A. Decourtye and J.M. Delpuech, 2007. The sublethal effects of pesticides on beneficial arthropods, Annu. Rev. Entomol., 52: 81-106.
27. Umoru, P.A. and W. Powell, 2002. Sub-lethal Effects of the Insecticides Pirimicarb and Dimethoate on the Aphid Parasitoid *Diaeretiella rapae* (Hymenoptera: Braconidae) When Attacking and Developing in Insecticide-Resistant Hosts. Biocontrol Science and Technology, 12: 605-614.

28. Hoagland, D.R. and D.I. Arnon, 1950. The water-culture method for growing plants without soil. California Agricultural Experiment Station Circular, 347: 1-32.
29. Takada, H. and Y. Murakami, 1988. Esterase variation and insecticide resistance in Japanese *Aphis gossypii*. Entomol. Exp. Appl., 48: 37-41.
30. Gubran, E.E., R. Delorme, D. Auge and J.P. Moreau, 1992. Insecticide resistance in cotton aphid *Aphis gossypii* (Glov) in the Sudan Gezira. Pesticide Science, 35(2): 101-107.
31. Tang, Z., 1992. Insecticide resistance and countermeasures for cotton pests in China. Resistant Pest Manage. Newsl., 42: 9-12.
32. Grafton-Cardwell, E.E., L.D. Godfrey, W.A. Brindley and P.B. Goodell, 1997. Status of Lygus bug and cotton aphidresistance in the San Joaquin Valley, pp: 1072-1074. In Proc. Beltwide Cotton Conf., New Orleans, LA. 7-10 Jan.1997. Natl. Cotton Counc. Am., Memphis, TN.
33. Pungerl, N.B., 1984. Host preferences of *Aphidius* (Hymenoptera: Aphidiidae) populations parasitizing pea and cereal aphids (Hemiptera: Aphididae). Bull. Entomol. Res., 74: 153-161.
34. Stark, J. and T. Rangus, 1994. Lethal and sublethal effects of the neem insecticide formulation, Margosan-O, on the pea aphid. Pestic. Sci., 41: 155-160.
35. Jaime, E. Araya, Manuel Araya and María Angélica Guerrero, 2010. Effects of some insecticides applied in sublethal concentration on the survival and longevity of *Aphidius ervi* (Haliday) (Hymenoptera: Aphidiidae) adults. Chilean Journal of Agricultural Research, 70(2): 221-227.
36. Zhong, Z.Z., P. Zhang and J. Chen, 2005. Inhibition Effects of Three Aphicides on the CMV Transmission by *Aphis gossypii*. Guizhou Agricultural Sciences, 4: 42-43.
37. Langhof, Maren A., Gathmannb Achim, Poehling a Hans-Michael and Meyhöfer A. Rainer, 2003. Impact of insecticide drift on aphids and their parasitoids: residual toxicity, persistence and recolonisation. Agriculture, Ecosystems and Environment, 94(3): 265-274.
38. Starý, P., 1970. Biology of Aphid Parasites (Hymenoptera: Aphidiidae) with Respect to Integrated Control, Junk, The Hague.
39. Desneux, N., R. Denoyelle and L. Kaiser, 2006. A multi-step bioassay to assess the effect of the deltamethrin on the parasitic wasp *Aphidius ervi*, *Chemosphere*, 65: 1697-1706.
40. Krespi, L., Krespi, J.M. Rabasse, C.A. Dedryver and J.P. Nenon, 1991. Effect of three insecticides on the life cycle of *Aphidius uzbekistanicus* Luz. (Hym., Aphidiidae), J. Appl. Ent., 111: 11.
41. Jansen, J.P., 1996. Side effects of insecticides on *Aphidius rhopalosiphi* (Hym. Aphiddidae) in laboratory, *Entomophaga*, 41: 37-43.
42. Abo El-Ghar Gamal, E.S., Abd El-Sayed, 1992. Long-term effects of insecticides on diaeretiella rapae (m'intosh), a parasite of the cabbage aphid. Pesticide Science, 36(2) 109-114.
43. Ibrahim, N.M.M., M.D. Abdallah and M.A. Kandil, 1992. Laboratory tests for assessing the initial and residual toxicity of certain insecticides against the cotton aphid, *Aphis gossypii* (Glover), Bulletin of Entomological Society of Egypt, 19: 19-24.
44. Desneux, N., H. Rafalimanana and L. Kaiser, 2004. Dose-response relationship in lethal and behavioural effects of different insecticides on the parasitic wasp *Aphidius ervi*. Chemosphere, 54(5): 619-627.
45. Nazer, I.K., M.M. Obidat and N.S. Sharaf, 1984. Response of the melon aphid (Aphis gossypii Glov, Homoptera, Aphididae) to certain insecticides in the Jordan Valley. Arab Scientific Conference of Biological Sciences, Amman (Jordan), 3-6 Nov 1984.
46. Wang, H.Y., Y. Yang, J.Y. Su, J.L. Shen, C.F. Gao and Y.C. Zhu, 2008. Assessment of the impact of insecticides on *Anagrus nilaparvatae* (Pang et Wang) (Hymenoptera: Mymanidae), an egg parasitoid of the rice planthopper, *Nilaparvata lugens* (Hemiptera: Delphacidae). Crop Protection, 27: 514-522
47. Li, Z.M. Li and Y.Q. Liu, 2005. Comparative toxicity of several insecticides to *Cotesia plutellae*, *Pesticides* 44(8): 374-376 (in Chinese, with English abstract).