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Field persistence of some insecticides on okra leaves as determined by cotton leafworm (*Spodoptera littorals)* bioassay

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ABSTRACT

A field experiment was conducted in Moshtohor, Kalubia, Egypt during the summer season of 2004 to study the persistence of spinosad, abamectin, azadirachtin, Dipel 2X and methomyl on okra [(*Abelmoschus esculentus* L.) cv. Balady] leaves. The pesticides toxicity was determined on the 2nd instar larvae of the laboratory strain of *Spodoptera littoralis* (Bosid). The tested insecticides were applied with recommended field rate at 180 gm (25% ai) / feddan, 120 ml (1.8% ai) / feddan, 600 ml (0.15% ai) / feddan, 200 gm (*Bacillus thuringiensis* subsp. *Kurstaki* 32000 IU/mg) / feddan and 300 gm (90% ai) / feddan for spinosad, abamectin, azadirachtin, Dipel 2X and methomyl, respectively. The results revealed that spinosad was highly toxic to larvae compared to the other tested insecticides. The LC_{50} values were 0.4, 1.1, 1.4, 3.1 and 220.8 µg/ml for spinosad, azadirachtin, abamectin, and Dipel 2X,following 72 hr, respectively. In case of methomyl, it was 3.1 mg/ml after 24 hr. The residual toxicity of spinosad, abamectin, azadirachtin and methomyl was sustained up to 10, 7, 5 and 5 days, respectively, while Dipel 2X showed no activity after 3 days of application .The half-life values of the applied insecticides were 1.90, 1.87, 1.40, 0.77, 0.40 day for azadirachtin, abamectin, spinosad , methomyl and Dipel 2X, respectively.

Key words: Insecticides, *Abelmoschus esculentus*, *Spodoptera littoralis*, residual toxicity, half-life

INTRODUCTION

An important consideration in the choice of insecticides for crop protection is the length of time for which the toxic residues will persist on foliage or tissue and on soils. Persistent insecticides might be preferable to use against a continuous, heavy infestation of pests, while those of short persistence might be preferable for the control of sporadic infestations to

allow the survival or rapid reestablishment of natural enemies (Raha *et al.* 1993).

To combat the pests, growers use synthetic organic insecticides, and some biorational insecticides, including products of *Bacillus thuringiensis* (Bt). With the implementation of the Food Quality Protection Act likely to limit the applications of some organic chemical insecticides and the inability of Bt products to provide season-long control of lepidopterous pests, scientists and growers are seeking alternative materials that are effective against the pests and safe to humans and the environment (Liu *et al.* 1999). On the other hand, the main problems with *Bacillus thuringiensis* products for pest control are often narrow activity spectrum and high sensitivity to UV degradation (Sanchis *et al.* 1999).

Okra (*Abelmoschus esculentus* L.) is one of the important vegetable crops in Egypt. It was infested with many insect pests such as cotton leafworm (*Spodoptera littoralis*), spiny bollworm (*Earias* insulana), whitefly (Bemisia tabaci) and aphids (Aphis gossypii).

Persistence of several synthetic insecticides and biopesticides on okra (such as quinalphos, methomyl, alpha-cypermethrin, triazophos, beta-cyfluthrin, deltamethrin, endosulfan and azadirachtin) were investigated by (Samanta *et al.* 1998, Sharma *et al.* 1998 and Dikshit *et al.* 2001).

The objectives of the study were to determine the field stability and toxicity of spinosad, abamectin, azadirachtin, Dipel 2X and methomyl residues on okra foliage through laboratory bioassays.

MATERIALS AND METHODS

Insecticides:
Spinosad (SpinTor 25% WG) produced by Dow AgroSciences, USA. Abamectin, [Vertimec 1.8% EC] produced by Novartis Crop Protection AG, Switzerland. Azadirachtin, [Achook 0.15% EC] produced by Bahar Agrochem and Foods Pvt. Ltd., India. Dipel 2X, [*Bacillus thuringiensis* subsp. *Kurstaki* 32000 IU/mg WP] produced by Valent BioSciences, USA. Methomyl, [Lannate 90% WP] produced by E. I. Du Pont de Nemours, USA.

Rearing technique of *S. littoralis*:

The Egyptian cotton leafworm, *S. littoralis* as a laboratory strain has been reared for several generations according to the method of El-Defrawi *et al.* (1964). The rearing room was kept at 27±2 °C and 65±5 % RH. The larvae of the insect were fed on clean and fresh caster bean leaves, *Ricinus communis* (L.). The adult moth, females and males were supplied with a piece of cotton soaked in 10% sugar solution and 2-3 small branches of Tafla plant (*Nerium oleander* L.), which served as an oviposition site. Deposited egg masses were kept in plastic jars under the mentioned conditions until hatching. The newly hatched larvae were transferred to other plastic jars and provided with fresh caster bean leaves. The second larval instar became available for the bioassay tests.

Filed experiment and sampling:

The field experiment was carried out at the experimental farm of the Faculty of Agric., Moshtohor, Qualyubia Governorate, during the summer season, 2004. An area of ¼ feddan was sown of local okra seeds [(*Abelmoschus esculentus* L.) cv. Balady] on the 4th April 2004. The experiment was designed as follows: plot size, 7 x 4 m; plot to plot distance, 0.7 m; plant to plant distance, 0.25 m and row to row distance, 0.7 m. Treatment plots were arranged in a randomized complete block design with three replications. Irrigation and fertilization were made according to the crop schedule.

The insecticides were applied on the 27th July 2004 using a knapsack sprayer (20 litres), at the recommended field rates, 180 gm/feddan, 120 ml/feddan, 600 ml/feddan, 200 gm/feddan and 300 gm/feddan for spinosad, abamectin, azadirachtin, Dipel 2X and methomyl, respectively. Water was used as controls (or untreated plants) in the field trials and in laboratory bioassay.

Samples of leaves (15–16 leaves with average size of 18–20 cm diameter) were taken randomly from each replicate at intervals of 0 (2h after spray), 1, 3, 5, 7, 10 and 14 days. Leaf samples were collected in polyethylene bags and preserved in a deep freezer in the laboratory at -18 °C (Thompson *et al* 2002).

Bioassay tests:

Bioassay tests were conducted in the laboratory under the same conditions mentioned above. Leaf-dipping technique similar to that

described by Tabashnik and Cushing (1987) was used to determine the toxicity of different insecticides against the 2nd instar larvae of the laboratory strain of *S. littoralis*. The formulated insecticides were diluted with water to obtain at least five progressive concentrations. Four disk (4 cm diameter) of untreated okra leaves were dipped for 5 sec. in each concentration thereafter were left for natural air-dryness and were distributed in four glass jars (250 ml). Ten larvae of the 2nd instar were allowed to feed for 24 h on the treated leaf-disks with the different insecticide concentrations (as four replicates for each concentration). Four replicates of ten larvae each, were fed on water-treated disks of okra leaves for 24 h and served as control. After the exposure period (24 h), the treated and untreated okra leaf disks were replaced daily with fresh caster bean leaves. Mortality was recorded after 1, 2, 3, 5 and 7 days. The larvae were considered dead if no movement was detected when they were touched with a small brush.

Determination of insecticide residual toxicity:

Two leaf disks were used from each plot [2 x 3 (plots) = 6 replicates of each date after spraying]. Leaf disks were individually placed on the bottom of clear glass jar (250 ml), and ten second instar larvae of *S. littoralis* were placed on the leaf disk. Under the laboratory conditions, the exposure period of larvae to treated and untreated okra leaf disks and dates of mortality counts were as the same in the bioassay tests.

Insecticide stability studies under freezing conditions:

Untreated okra leaves were dipped for 5 sec. in LC_{50} of each insecticide separately (four leaves for each insecticide treatment). The okra leaves were left for natural air-dryness and preserved in a deep freezer in the laboratory at –18 °C for 16 d. (the maximum of the experiment freezing time). Four replicates for four leaf disks were used similar to that described by Boisvert and Boisvert (2001) to calculate the insecticide concentration (Table 1).

Data analysis:

Data were analyzed using Probit analysis models (Finney, 1971) using a computer program of Noack and Reichmuth (1978). The lethal concentrations needed to kill 50, 90 and 99% of populations, their confidential limits (95%), slopes and insecticide persistence or residues were computed. The insecticide half-life values of insecticides are calculated according to Welder (1985).

Table (1): Insecticide stability under freezing conditions.

Insecticide	Concentration Before Freezing (LC_{50}) (µg/ml)	Concentration After Freezing (µg/ml) (mean ±SD)
Spinosad	0.4	0.4±0.1
Abamectin	1.4	1.5±0.3
Azadirachtin	1.1	1.0±0.2
Dipel 2X	220.8	189.4±62.9
Methomyl	3.1	3.1±0.0

RESULTS AND DISCUSSION

I- Laboratory toxicity bioassay:

Lethal concentrations values are shown in Table (2) and indicated that spinosad was highly toxic to the second instar larvae of *S. littoralis* compared with the other insecticides. LC_{50} valmes were 0.4, 1.1, 1.4, 3.1 and 220.8 µg/ml for spinosad, azadirachtin, abamectin, methomyl and Dipel 2X, respectively.

Spinosad has relatively broad spectrum and has been effectively used for the control of many species of insect pests in Orders: Lepidoptera, Diptera, Coleoptera and Thysaoptera in various crop systems (Sparks *et al.* 1995, Bret *et al.* 1997, Nolting *et al.* 1997, Peterson *et al.* 1997 and Richardson *et al.* 1998). Also, azadirachtin was highly toxic to *S. littoralis* (Aerts and Mordue 1997). Abamectin was superior to other synthetic insecticides and biopesticides in terms of control efficacy against many insect pests on okra (Ghosh *et al.* 1999).

I I- Field leaf residue bioassay:

Mortality percentages of the second instar larvae of *S. littoralis* showed that spinosad activity was longer than the other tested insecticides and it extended through 10 d., while the activity of abamectin, azadirachtin and methomyl was, 7, 5 and 5 d., respectively. On the other hand, no activity was observed at 3 d. after the field application of Dipel 2X (Table 3 and Figure 1).

Table (2): Lethal concentrations of the tested insecticides against the second instar larvae of *S. littoralis*.

Insecticide	Lethal concentrations (µg/ml) and their 95% Confidence Limits			Slope ±SD	R
	LC_{50}	LC_{90}	LC_{99}		
Spinosad[A]	0.4 (0.1-1.1)	3.0 (1.4-6.4)	15.8 (2.6-96.1)	1.5±0.03	0.983
Abamectin[A]	1.4 (1-1.9)	4.8 (2.7-8.7)	13.3 (5.1-34.6)	2.4±0.36	0.937
Azadirachtin[A]	1.1 (0.6-2.0)	8.2 (1.5-47.5)	67.2 (9.8-490.8)	1.2±0.03	0.974
Dipel 2X[A]	220.8 (80.9-580.2)	3420.0 (850.5-13680)	17100.1 (2440-119700)	1.4±0.03	0.990
Methomyl[B]	3.1 (1.7-5.7)	37.1 (10.3-133)	278.5 (135.4-793)	1.2±0.02	0.922

SD: Standard deviation of mortality regression line; R: Correlation coefficient of regression line

A: Only the evaluated mortality after 3 days was used in the result calculations

B: Only the evaluated mortality after 1 day was used in the result calculations.

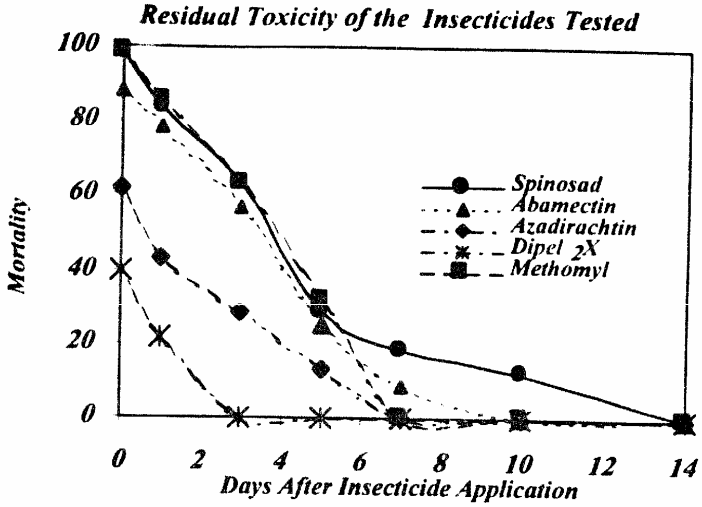

Figure 1. Residual toxicity of the insecticides tested on the second instar larvae of *S. littoralis*.

In the laboratory and other protected environment, leaf residue of spinosad could be toxic to *Trichoplusia ni* larvae longer than 36 d. with relatively slow activity. Because photolysis is the primary route of spinosad degradation, longer residual effect on the foliage in the laboratory compared with that in the field condition is expected (Sanders and Bret 1997). Spinosad applied to field crops generally loses activity after a week (McDonald *et al.* 1998 and Fang *et al.* 2002). Azadirachtin showed low residual toxicity against *Cryptolaemus montrouzieri* (Sundari 1998). On the other hand, the main problems with *Bacillus thuringiensis* products for pest control are often narrow activity spectrum and high sensitivity to UV degradation (Sanchis *et al.* 1999).

III- Persistence of the insecticides tested on the okra leaves:

Azadirachtin and abamectin showed relatively high persistence compared to the other tested insecticides, while spinosad and methomyl were intermediate.The half-life values of the applied insecticides were 1.90, 1.87, 1.40, 0.77 and 0.40 d. for azadirachtin, abamectin, spinosad, methomyl and Dipel 2X, respectively (Table 4 and Figures 2 to 4).

Table (3): Residual toxicity of the tested insecticides on the okra leaves against the second instar larvae of S. littoralis.

Insecticide	Mortality % (mean ±SD)						
	Days after insecticides application						
	Z[C]	1	3	5	7	10	14
Spinosad [A]	98.3±4.1	83.3±5.2	63.3±5.2	28.3±4.1	18.3±4.1	11.7±4.1	0
Abamectin [A]	88.3±4.1	78.3±4.1	56.7±8.2	25.0±5.5	8.3±4.1	0	0
Azadirachtin [A]	61.7±4.1	43.3±5.2	28.3.0±4.1	13.3±5.2	0	0	0
Dipel 2X [A]	40.0±0	21.7±4.1	0	0	0	0	0
Methomyl [B]	98.3±4.1	85.0±5.5	63.3±5.2	31.7±4.1	0	0	0

A: Only the evaluated mortality after 3 days was used in the result calculations
B: Only the evaluated mortality after 1 day was used in the result calculations
C: Two hours after the insecticide application (zero time).

The results also revealed that the Dipel 2X showed the lowest persistence, where *Bacillus thuringiensis* subsp. *kurstaki* spores have a very short persistence in the environment (half-life 10 h.) mainly due to their UV-light sensitivity (Tomlin 2000).

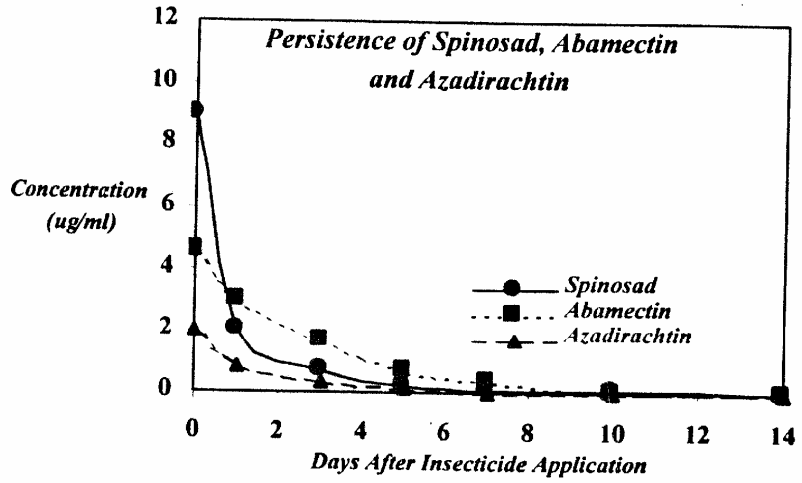

Figure 2. Persistence of spinosad, abamectin and azadirachtin on okral leaves.

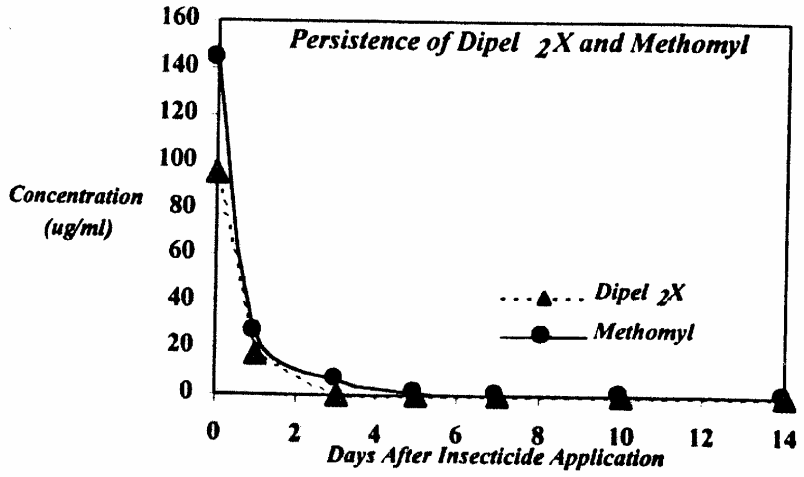

Figure 3. Persistence of Dipel 2X and methomyl on okra leaves.

Table (4): Persistence of the insecticides tested on the okra leaves.

Insecticide	Half-life (day)	Insecticide residues (µg/ml) (mean ±SD)						
		Days after insecticide application						
		Z[C]	1	3	5	7	10	14
Spinosad	1.40	9.0±2.94	2.0±0.77	0.70±0.15	0.18±0.04	0.09±0.02	0.06±0.02	0
Abamectin	1.87	4.52±0.69	2.97±0.33	1.68±0.29	0.70±0.11	0.33±0.16	0	0
Azadirachtin	1.90	2.0±0.49	0.83±0.21	0.37±0.08	0.13±0.06	0	0	0
Dipel 2X	0.40	95.0±0	17.0±1.0	0	0	0	0	0
Methomyl	0.77	143.4±52.05	26.45±11.67	6.27±1.81	1.23±0.33	0	0	0

C: Two hours after the insecticide application (zero time).

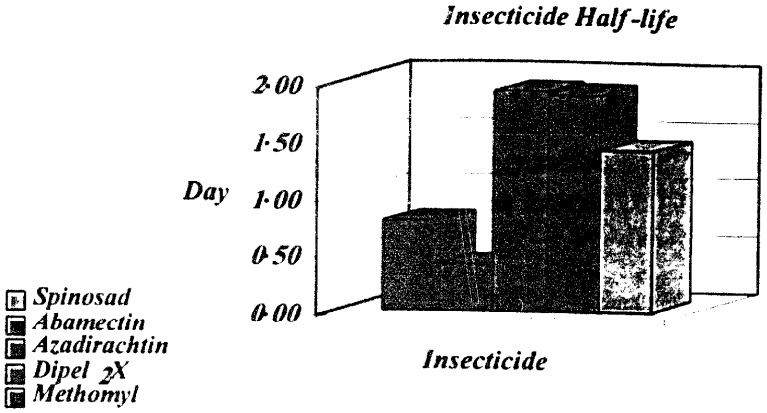

Figure 4. Half-life values of the insecticides tested on okra leaves.

The degradation of spinosad on plant surfaces is mainly by photolysis, its half-life value was ranged from 1.6 - 16 day, methomyl had a half-life in plants c. 3 – 5 day, it rapidly degraded to CO_2 and acetonitrile, with incorporation into natural plant components (Tomlin 2000).

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الثبات الحقلي لبعض المبيدات الحشرية على أوراق نبات الباميا باستخدام يرقات دودة ورق القطن

محمد محمد عزب

قسم وقاية النبات – كلية الزراعة بمشتهر – جامعة بنها

أجرى هذا البحث بهدف دراسة ثبات المبيدات الحشرية إسبينوساد وأبامكتين وأزديراختين ودايبل 2 إكس والميثومايل على أوراق نبات الباميا حقلياً وقد تم إجراء هذه الإختبارات بمزرعة الخضر بكلية الزراعة بمشتهر فى الموسم الصيفى لعام 2004ولقد تم تقدير متبقيات هذه المبيدات بإستخدام التقدير الحيوى بواسطة العمر الثانى لسلالة معملية من دودة ورق القطن وكانت التركيزات الحقلية المستخدمة من كل مبيد كما يلى :
إسبينوساد 180 جم/فدان وأبامكتين 120 مل/فدان وأزديراختين 600 مل/فدان ودايبل 2إكس 200 جم/فدان والميثومايل 300 جم/فدان.

ولقد أظهرت نتائج التقدير الحيوى أن مبيد إسبينوساد كان أشد المبيدات سمية على العمر الثانى ليرقات دودة ورق القطن
بالمقارنة بالمبيدات الأخرى تحت الإختبار وكانت التركيزات القاتلة ل 50 % من الحشرات والمقدرة على أساس نسبة اليرقات الميتة بعد 72 ساعة من المعاملة لكل المبيدات المحتبرة بإستثناء مبيد الميثومايل كانت بعد 24 ساعة هى 0.4 و 1.1 و 1.4 و 3.1 و 200.8 ميكروجرام/مل لكلاً من إسبينوساد وأزديراختين وأبامكتين والميثومايل ودايبل 2إكس على التوالى.

كما أوضحت النتائج إلى إستمرار فاعلية المتبقيات لكلاً من إسبينوساد وأبامكتين وأزديراختين والميثومايل خلال10 و 7 و 5 و 5 أيام من التطبيق حقلياً على التوالى ، بينما لم تظهر فاعلية لمبيد دايبل 2إكس بعد 3 أيام من الرش.

أما فترة نصف العمر للمبيدات تحت الدراسة فكانت 1.90 و 1.87 و 1.40 و 0.77 و 0.4يوم لكلاً من أزديراختين وأبامكتين و إسبينوساد والميثومايل ودايبل 2إكس على التوالى.