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ISSN 2319-3077 Online/Electronic

ISSN 0970-4973 Print

UGC Approved Journal No. 62923

MCI Validated Journal

Index Copernicus International Value

IC Value of Journal 82.43 Poland, Europe (2016)

Journal Impact Factor: 4.275

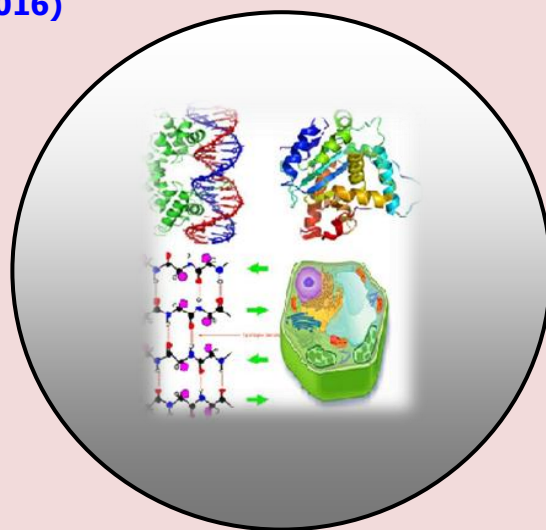
Global Impact factor of Journal: 0.876

Scientific Journals Impact Factor: 3.285

InfoBase Impact Factor: 3.66

J. Biol. Chem. Research

Volume 35 (2) 2018 Pages No. 590-594

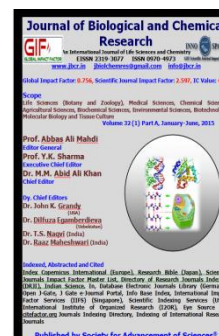


Journal of Biological and Chemical Research

An International Peer Reviewed / Referred Journal of Life Sciences and Chemistry

**Indexed, Abstracted and Cited in various International and
National Scientific Databases**

Published by Society for Advancement of Sciences®

J. Biol. Chem. Research. Vol. 35, No. 2: 590-594, 2018**(An International Peer Reviewed / Refereed Journal of Life Sciences and Chemistry)****Ms 35/02/7010/2018****All rights reserved****ISSN 2319-3077 (Online/Electronic)****ISSN 0970-4973 (Print)**[http:// www.sasjournals.com](http://www.sasjournals.com)[http:// www.jbcr.co.in](http://www.jbcr.co.in)jbicchemres@gmail.com**RESEARCH PAPER**

Received: 17/07/2018

Revised: 28/08/2018

Accepted: 05/09/2018

Resistance Monitoring in Cotton Leaf worm *Spodoptera littoralis* to Certain Bioinsecticides during Ten Cotton Seasons in Eight Governorates on Egypt

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ABSTRACT

Resistance ratio of six bioinsecticides in eight strains of Spodopteralittoralis which collected from bini-swef, fayoum. Sharkia, Dakahlia. Menofia, Gharbia, Kafr- elshakh and Behera during 2008 to 2017 cotton seasons. The results indicate that resistance ratios RR fluctuated from one year to another and from Governorate to another. During ten cotton seasons the tested Bacillus thuringiensis Agerine 6.5% WP, Protecto 9.4 %WP, Dipel 2x 6.4% WP, Dipel Df 5.4 % WG and two other bioinsecticides, Spinosad (Spintor 24 % SC) and Emamectinbenzoate (Radical 0.5 %) against podopteralittoralis that all field strains were very highly susceptible for spintor and Radical followed by Dipel 2X and Dipel Df followed by protect and the last one was Agreïn.

Key words, Spodoptera littoralis, bioinsecticides, and several Governorates.

INTRODUCTION

Cotton leaf worm is one of the most economically important insect pests in cotton in Egypt. Due to repeated application of conventional insecticides over years, resistance in cotton leaf worm was developed against many conventional insecticides. Alternatively, bioinsecticides were used to control such insect through the integrated pest management (IPM) programs. Therefore, the level and prediction of resistance potentially occurred against recommended bioinsecticide is needed to be monitored. In this study, toxicity of different formulations of *Bacillus thuringiensis* was determined against field strains of *S. littoralis* collected from different regions in Egypt. The activity of another two bioinsecticides were also studied namely; spinosad and emamectin benzoate. The objective of this work is to monitor the resistance level in *S. littoralis* to certain bioinsecticides during ten cotton growing seasons in different eight regions in Egypt. *Bacillus thuringiensis* crystal proteins are preferred and widely used as an alternative to chemical pesticides in pest management strategies against insect pests of agriculture crops (Roh *et al.*, 2007). Also, spintor is an insecticide based on a fermentation product of this soil bacterium actinomycete *Saccharopolyspora spinosa*. It exhibits a high degree of selective toxicity towards the insect order Lepidoptera, but less toxic to many beneficial arthropods (Thompson *et al.*, 2000). The purpose of the present study is to investigate the resistance of tested biocides against the cotton leaf worm *Spodoptera littoralis* (Boisd.) collected from different Governorates of Lower Egypt in cotton fields during 2012, 2013 and 2014 seasons.

MATERIAL AND METHODS

A. Eight field strains of the cotton leaf worm were collected from the cotton field in Egypt from several locations (bini-swef, fayoum. Sharkia, Dakahlia. Menofia, Gharbia, Kafr- elshakh and Behera) from 2008 to 2017 cotton seasons. After collection, the egg-masses were kept separately in 400 ml jar, covered with muslin held in position by rubber band until the eggs hatched. The jars were provided with castor oil leaves for larval feeding and to provide the required humidity for hatching. All cotton leaf worm field strains and laboratory strain were reared at 25 ± 2 C and 70 ± 5 % relative humidity. The larvae were then used for bioassay study

B- Bioinsecticides used

The following six bioinsecticides were used: four different formulations of *Bacillus thuringiensis* Agerine 6.5% WP, Protecto 9.4% WP, Dipel 2x 6.4% WP, Dipel Df 5.4% WG and two other bioinsecticides, Spinosad (Spintor 24 % SC) and Emamectin benzoate (Radical 0.5 %).

C. Bioassay tests

Six concentrations for each formulation were prepared as aqueous solutions. Castor – bean leaves were dipped for 15 second in each concentration then left for one hour to dry. Then, the 2nd instar larvae of each strain were fed on treated leaves in kept in plastic for 24 hr in jars and covered with muslin, then the treated leaves were removed and provided with fresh untreated leaves in clean jars for another three days. Three replicates of ten larvae, were tested for each concentration. Mortality percentages were recorded after 5 days from larvae transfer onto untreated leaves and mortality percentage was corrected according to Abbot, 1925. To estimate the LC₅₀ values, the corrected mortality percentages were subjected to probit analysis according to the method of Finney (1971). The level of resistance in the field strains was calculated as resistance ratio (RR) compared with the susceptible strain. Susceptible strain was reared under laboratory condition for more than 15 generations without exposure to any insecticides.

Resistance Ratio (RR) = LC₅₀ for field strain / LC₅₀ for susceptible strain

RESULTS AND DISCUSSION

Resistance ratio of six bioinsecticides in eight strains of *Spodoptera littoralis* which collected from bini-swef, fayoum. Sharkia, Dakahlia. Menofia, Gharbia, Kafr- elshakh and Behera during 2008 to 2017 cotton seasons are shown in (Table 1 and 2). Data are fluctuated from season to other and governorates to another. Data in Table (1) showed resistance ratio (RR) to Agerin, protecto and Dipel 2x against *Spodoptera littoralis* collected from eight governorates during 2008 to 2017 in cotton seasons in Egypt. Data showed in Agerine 6.5 wp (RR) value in was very low in all governorates during in 2008 to 2017 cotton season RR was fluctuated from 0.9 to 8.73 fold in all seasons Beni-swef Governorate but RR increase in Beni-swef during 2012 season RR was 18.70 fold. Infauom Governorate RR fluctuated from 1.20 to 9 fold in all seasons but in Sharkia governorate RR was fluctuated from 1.80 to 5.60 fold in all seasons except in 2014 season RR was 10.70 fold on the other hand in Dakahlia governorate also RR was very low it was fluctuated from 1.80 to 4.60 fold except in 2014 season RR was 10.70. in Menofia Governorate RR don't increase than 5 fold in Gharbia Governorate RR was fluctuated from 1 to 9 fold in all seasons in kafr el shekh RR was fluctuated from 1.8 to 9.40 fold but in 2013-2014 and 2017 seasons RR was very high RR was 11.36, 11.4 and 13.4 fold. Finally in Behera Governorate RR fluctuated from 2 to 9.19 fold in all seasons but in 2013 season RR was 10.47 fold in protectobiocide all of the Governorates RR were less than 10 RR was fluctuated from 0.04 to 8 fold in Beni-swef but in 2012 season RR was very high (31.40) fold, also in Fayoum Governorate RR was very low it was fluctuated from 0.05 to 4.27 fold in Sharkia governorate also RR was very low it was fluctuated from 0.09 to 3.5 fold in all seasons, in Dakahlia Governorate RR was 0.20 to 4.9 in all seasons also in Menofia Governorate RR was fluctuated from 0.07 to 3.3 fold in all seasons in Gharbia Governorate RR was ranged from 0.04 – 7.5 in all seasons but Kafr-elshekh RR was very low RR was ranged from 0.2- 8 fold except in 2012 and 2013 seasons RR were 17.8 and 15.55 fold finally in Behera Governorate RR also very low RR was ranged from 0.2- 8.4 fold but in 2012 season RR was 23.6 fold. In biocide Dipel 2x RR were less than 10 in all Governorates and all seasons except in 2017 in Beni-swef, RR in Beni-swef Governorate was fluctuated from 0.3 to 5.1 fold, in Fayoum RR was ranged between 0.3 and 6.3 fold, in Sharkia RR was fluctuated from 0.4 to 9.05 fold in Dakahlia was 0.5 to 3.07 fold also in Menofia Governorate RR was ranged between 0.2 and 3.2 fold in Gharbia also RR was very low RR ranged between 0.4 and 7.9 in all season except in 2017 season RR was 10.4 fold and in Kafrelshekh data showed low level of resistance RR ranged between 0.3 and 9.5 in all seasons on the other hand in Behera data showed high level of resistance, RR were 10. In 2016 and 2017 seasons RR was 11.14 and 12.15 fold respectively but in all seasons RR was fluctuated from 0.4 and 5.5 fold.

Table 1. Resistance ratio (RR) to Agerin, protecto and Dipel2x against *Spodopteralittoralis* collected from eight governorates during 2008 to 2017 in cotton seasons in Egypt.

Insecticides	season	Bini-swef	fayoum	Sharkia	Dakahlia	Menofia	Gharbia	Kafr-elshakh	Behera
Agerine 6.5 wp <i>Bacillus thuringiensis</i>	2008	0.90	1.20	1.80	1.40	0.60	1.00	1.80	2.00
	2009	2.30	1.40	2.50	2.00	1.70	2.10	1.90	2.60
	2010	4.24	4.20	2.50	4.60	4.00	5.00	3.89	6.10
	2011	0.86	0.50	2.00	1.30	1.40	1.01	3.63	2.10
	2012	18.70	6.40	5.60	4.20	5.20	6.00	9.40	2.80
	2013	8.73	4.62	4.19	2.95	3.66	6.69	11.36	10.47
	2014	8.5	9.79	10.70	10.29	5.10	8.23	11.4	9.13
	2015	8.28	1.37	2.15	1.52	1.27	3.98	7.84	8.48
	2016	3.77	1.37	2.00	4.08	2.11	1.15	2.97	3.84
	2017	5.26	2.01	4.95	1.40	0.79	9.61	13.24	4.42
Protecto 9.4 %Wp <i>Bacillus thuringiensis</i>	2008	0.04	0.05	0.09	0.20	0.07	0.04	0.20	0.20
	2009	0.50	0.40	2.00	0.90	0.90	0.90	1.20	2.00
	2010	0.48	0.50	2.00	1.20	1.40	1.10	1.08	2.10
	2011	0.50	1.21	2.50	2.00	1.20	3.20	1.30	0.90
	2012	31.40	2.80	3.90	4.90	3.30	5.60	17.80	23.60
	2013	8.40	2.25	2.37	3.28	2.40	5.87	15.55	7.14
	2014	4.35	4.27	3.57	3.25	3.23	4.54	2.60	5.46
	2015	3.41	1.61	2.16	1.51	1.48	4.39	2.59	3.71
	2016	2.39	3.10	2.16	1.96	1.88	3.80	3.55	8.41
	2017	8.00	0.94	1.32	1.70	2.33	7.57	8.02	4.01
Dipel 2x 6.4% wp <i>Bacillus thuringiensis</i>	2008	1.00	0.70	1.30	1.80	0.80	0.40	1.50	0.90
	2009	0.10	0.30	0.40	0.60	0.20	0.30	0.60	0.20
	2010	0.30	0.32	0.50	0.50	0.30	0.40	0.35	0.40
	2011	0.90	2.43	7.60	2.70	2.40	7.90	2.90	2.40
	2012	2.40	2.20	1.50	1.40	2.10	1.10	1.50	1.80
	2013	4.46	3.40	2.40	2.82	3.20	4.29	8.42	5.53
	2014	5.1	6.31	4.95	3.07	2.51	3.23	3.90	3.50
	2015	4.34	0.57	1.99	1.32	0.70	3.73	3.27	3.52
	2016	1.71	1.53	9.05	4.86	0.89	1.31	2.31	11.14
	2017	10.86	1.13	1.96	1.97	0.42	10.49	9.59	12.45

In table (2) showed resistance ratio (RR) to Dipel Df, Spintor and redical against *Spodoptera littoralis* collected from eight governorates during 2008 to 2017 in cotton seasons in Egypt. In Dipel Df also all of the Governorates and seasons indicated low level of resistance in Beni-swif RR ranged between 0.3 and 5.3 fold in all seasons except in 2013 season RR was 12.5, in Fayoum RR was fluctuated from 0.6 to 7.0 fold except in 2012 season RR was 13.8 fold on the other hand in Sharkia Governorate RR fluctuated from 0.7 and 9.7 fold in all seasons also in Dakahlia RR was fluctuated from 0.72 and 9.8 fold in all seasons also in Menofia RR fluctuated from 0.9 and 6.77 fold in Gharbia Governorate level of resistance was very low RR was ranged between 0.5 and 3.0 fold in all season except in 2013 season RR was 11.35, in Kafr-Elshak RR was ranged between 0.9 and 4.8 fold in all seasons except in 2013 season RR was 16.96 fold finally Beherea Governorate data showed low level of resistance, RR ranged between 0.8 and 4.5 fold in all seasons except in 2013 season RR was 11.3 fold. This Table also showed spintor and Radical this biocides were not indicated any level of resistance RR less than 5 RR was fluctuated from 0.06 and 4.6 fold in all spintor Biocide for all seasons and Governorate also Radical was ranged between 0.04 and 3.7 in all Governorates and seasons. Finally these two compounds are very important. The results showed that all field strains were very highly susceptible for spintor and Radical followed by Dipel 2X and Dipel Df followed by protecto and the last one was Agerin.

In conclusion, the biocides Radical and Spintor were very effective in the control of *S. littoralis* and they recorded lack and low levels of resistance after three cotton seasons. Therefore, in order to maximize the negative effects of the chemicals on the environment and natural enemies in the management of pests, the natural insecticides could be integrated into IPM programmers. Nehad M. Elbarky *et al* (2008) Radiant SC12% (Spinetoram) is a new generation of spinosyn group. Effect of this bio-insecticide against larvae of *Spodoptera littoralis* (Boisd) was studied to evaluate the susceptibility of 2nd and 4th larval instars in laboratory. The LC₅₀ of radiant was 0.05 and 0.03 ppm. After 24 and 48 hours, respectively for the 2nd larval instars. Where the LC₅₀ were 6.67 and 2.86 ppm after 24 and 48 hours, respectively for the 4th larval instars. In the semi-field experiment, recommended doses of radiant exhibited high mortality 100 & 95.7 % after 0 and 1 days, respectively then decreased gradually to reach 58.1 % after 7 days. Also the field experiment showed high mortality 91.4% after 2 days then reduced gradually to reach 83.1% after 8 days. The effect of different concentrations in laboratory and recommended doses in field showed 100% mortality of entire hatched egg masses. Effect of recommended doses of radiant against predators inhabiting cotton field demonstrated that radiant was safe to natural enemies. The effect of LC 50 of radiant on the major biochemical component of 4th larval instars after 24 hours showed that, this data agree with Adel-Sattar *et al* (2012) studied Pyrethroids and Biocides Resistance in Field Strains of the Cotton Leaf Worm, *Spodoptera littoralis* (Boisd.)

Table 2. Resistance ratio (RR) to Dipel Df, Spintor and redical against *Spodoptera littoralis* collected from eigh governorates during 2008 to 2017 in cotton seasons in Egypt.

Insecticides	season	Bini-swef	fayoum	Sharkia	Dakahlia	Menofia	Gharbia	Kafr-elshakh	Behera
Dipel Df 5.4 % WG <i>Bacillus thuringiensis</i>	2008	1.36	1.00	1.24	1.21	1.01	1.02	1.33	1.16
	2009	1.85	1.99	1.26	1.00	1.73	1.58	1.55	2.52
	2010	0.61	1.18	0.72	0.72	0.93	1.06	1.36	0.84
	2011	1.10	1.56	1.90	2.00	1.60	1.70	1.40	1.50
	2012	2.30	13.80	1.50	2.20	3.50	3.70	2.20	2.50
	2013	12.55	6.35	2.98	4.78	6.77	11.35	16.96	11.38
	2014	5.39	7.06	6.90	9.18	3.58	5.27	3.65	3.39
	2015	4.11	0.64	1.67	1.00	0.98	0.60	4.89	4.30
	2016	2.93	3.99	9.4	2.16	1.89	0.54	0.90	4.54
	2017	0.98	3.19	2.45	2.12	1.04	1.83	1.35	1.52
Spintor 24 % Sc spinosad	2008	0.09	0.91	1.21	2.43	1.92	1.95	2.10	2.47
	2009	0.51	1.08	1.56	1.65	2.41	2.46	2.69	3.15
	2010	0.86	1.34	2.16	3.58	3.15	4.63	4.72	3.63
	2011	1.30	1.45	2.50	1.70	2.20	1.90	1.20	1.60
	2012	0.50	0.40	0.60	0.40	0.60	0.80	1.20	0.80
	2013	1.73	2.42	2.89	1.96	3.04	1.68	1.25	1.56
	2014	1.00	1.49	1.44	1.46	1.42	0.97	1.48	1.48
	2015	1.019	1.64	2.35	1.46	1.30	1.04	1.31	1.45
	2016	0.22	0.03	0.1	0.25	0.13	0.08	0.12	0.66
	2017	0.06	6.12	1.62	0.12	0.23	1.03	0.22	0.11
Radecarectical 0.5 % Ebamectinbenzo at	2008	1.65	1.12	2.56	1.41	1.74	1.00	2.62	2.96
	2009	1.44	1.00	2.28	1.00	1.46	2.11	1.82	2.15
	2010	1.27	1.77	1.00	1.60	1.04	2.98	2.11	1.49
	2011	0.10	0.14	0.16	0.17	0.15	0.20	0.19	0.40
	2012	0.60	13.20	0.70	0.50	0.50	1.00	0.70	1.40
	2013	3.70	3.80	0.51	4.87	5.17	1.16	0.79	1.07
	2014	0.28	0.32	0.09	0.12	0.10	0.19	0.12	0.42
	2015	0.265	0.75	0.07	0.66	0.07	0.212	0.123	0.610
	2016	0.09	0.02	0.16	0.005	0.06	0.007	0.007	0.001
	2017	1.73	0.0069	0.0071	0.23	0.088	1.2	0.22	0.11

During 2006-2008 Cotton Seasons four field strains of the cotton leaf worm, *Spodoptera littoralis* (Boisd.) were collected from different Egyptian Governorate during 2006-2008 cotton seasons for monitoring their resistance to four pyrethroids and six different biocides. In case of pyrethroids, resistance ratios of Karate were very high followed by Sumicidin and Sumi-alpha, while Meothrin showed lower levels of resistance when compared with the other pyrethroids tested during 2006-2008 cotton season. In case of biocides, resistance levels were insignificant during the cotton seasons 2006-2007 and 2008. This, indicate the possibility of using these materials as a replacement to conventional insecticides or in alternative with them in IPM programs Megahed *et.al.* (2013) Biochemical Effects of Certain Bioinsecticides on Cotton Leaf Worm, *Spodoptera littoralis* (Boisd.) (Lepidoptera: Noctuidae). The insecticidal activities of three bioinsecticides (i.e. emamectin benzoate "Proclaim", abamectin "Romacten" and spinosad "Tracer") were evaluated on the 4th larval instar of the cotton leaf worm, *Spodoptera littoralis* by leaf dipping technique as well as determining the biochemical changes in treated insects. These bioinsecticides showed immediate effects with 24hrs-LC50 values of 0.17, 0.23 and 38ppm for emamectin benzoate (Proclaim), abamectin (Romacten) and spinosad (Tracer), respectively. Omayma *et. al.* (2014) Field strains of the cotton leafworm *Spodoptera littoralis* (Boisd.) were collected from three Governorates (Gharbya, Kafr El-Sheikh and Behera) during 2012, 2013 and 2014 cotton seasons for monitoring of its resistance to tested biocides. The results indicate that resistance ratios RR and RC fluctuated from one year to another and from Governorate to another. During three cotton seasons the tested *Bt* (Dipel 2x, Dipel DF, Agerine and Protecto) showed high levels of resistance in all Governorates during 2014 cotton season which (RC) reached (13.93, 9.23 and 15.04), (9.24, 10.55 and 17.93), (6.01, 8.12 and 14.48) and (6.95, 7.08 and 8.34) in Behera, Gharbya and Kafr El-Sheikh respectively. Also, RC fo Radiant compound reached to (14.07, 11.58 and 22.05) in the same Governorates. While Radical recorded medium levels of resistance (2.16, 2.29 and 4.07) and Spintor had low levels of resistance (1.21, 1.73 and 1.66) to the same field strains during cotton season 2014. The previous results suggest that Biocides Radical and Spintor may be recommended as an effective component of the future IPM programs against *Spodoptera littoralis* on cotton fields.

ACKNOWLEDGEMENTS

The authors are grateful for Dr. Ibrahim Abdallah, Associate Professor at Cairo University for his help in submitting the manuscript to this journal.

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