

RESISTANCE MONITORING OF HELICOVERPA ARMIGERA TO INSECTICIDES ACROSS LOCATIONS OF KARNATAKA

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ABSTRACT

This study evaluates the toxicity of eight insecticides against *Helicoverpa armigera* (Hubn.). These include conventional and newer molecules which are being used on a large scale in six districts (Raichur, Kalaburagi, Bidar, Dharwad, Ballari, Bengaluru and Gangavathi) of north eastern Karnataka. The results revealed that the least LC_{50} value was observed in chlorantraniliprole 18.5%SC (0.17- 0.39 ppm-2014-15; 0.19-0.43 ppm- 2015-16; and 0.70-0.94 ppm- 2016-17. Maximum LC_{50} value was observed with chlorpyriphos 20%EC (35.16-41.08; 37.35-43.27; and 36.02- 41.94 ppm). The order of toxicity was chlorantraniliprole > emamectin benzoate > flubendiamide > spinosad > thiodicarb > methomyl > profenophos > chlorpyriphos. These results reveal that rotation of conventional insecticides along with the new insecticides might be more effective.

Key words: Helicoverpa armigera, insecticide toxicity, N-E Karnataka, topical bioassay LC₅₀ values, chlorantraniliprole, chlorpyriphos, resistance management

The pest *Helicoverpa armigera* (Hubn.), also known as the cotton bollworm is classified as one of the top 100 world invasive species (Kontsedalov et al., 2012). This is a cosmopolitan insect and has gained importance as a major destructive pest (Dinsdale et al., 2010), and its control up to desired level has become difficult (McCaffery et al., 1998). Crops such as cotton, chickpea, tomato, sunflower, okra, pea, tobacco, potato, egg plant are particularly affected by H. armigera. Due to its tremendous damage to crops, the use of insecticides constitutes the main control strategy. However, the indiscriminate use of insecticides has resulted in the development of resistance (Ferre and Vann, 2012). Resistance to a wide range of insecticides in H. armigera had been reported (McCaffery et al., 1998). Moderate to high level of resistance to conventional insecticides (chlorinated hydrocarbons, organophosphates, carbamates and pyrethroids) as well as to neonicotinoids and insect growth regulator (IGR) had been reported in field populations (Nauen and Bretschneider, 2002). Indiscriminate use of broad spectrum insecticides has resulted in secondary pest outbreaks and development of resistance (Kranthi et al., 2002; Ahmad et al., 2007). Hence, the insecticide resistance must be continuously monitored and must form an integral part of chemical control. The use of the new chemistry insecticides has increased now. These

were found highly effective in controlling *H. armigera* as compared to conventional ones (Razaq et al., 2005), but a low level of resistance to these in *H. armigera* is known (Ahmad et al., 2007). In the present study, the degree of resistance in *H. armigera* against both conventional and new chemistry insecticides has been evaluated using topical bioassay.

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MATERIALS AND METHODS

The 5th and 6th instar larvae of H. armigera were collected from fields of seven districts (Raichur, Kalaburagi, Bidar, Dharwad, Bellary, Bangalore and Gangavathi) during 2014-15, 2015-16 and 2016-17. The distance between locations are approximately 200-250 kms. From Bangalore, the larva was collected during 2014-15 and 2015-16. About 400-500 larvae were collected by walking through a plot randomly of selected host crop from each location and larvae were reared in the insecticide resistance laboratory at the UAS, Raichur during cropping season from 2014 to 2017. Rearing was done on semisynthetic wheat germ based diet $(25\pm 2^{\circ}\text{C}, 65\pm 5\%\text{RH}, 14:10 \text{ hrs light:})$ dark photoperiod). Diet was replaced after 24 hr, and pupae were collected on sequential days. The adults that emerged from larvae were kept in perspex oviposition cages (45x 25x 30 cm) with two sides covered with muslin cloth to maintain ventilation. These were fed on a solution containing sucrose (10%), vitamin solution (20 ml) and methyl 4- hydroxybenzoate in soaked cotton wool hanging in the oviposition cages. Commercial formulations of profenophos 50%EC, emamectin benzoate 5%SG, spinosad 45%SC, methomyl 40%SP, chlorpyriphos 20%EC, thiodicarb 70%SP, flubendiamide 37.9%SC and chlorantraniliprole 18.5%SC were used. Newly moulted third instar larvae (30-40 mg) from F₁ laboratory cultures were exposed to these insecticides using topical bioassay method (IRAC; http://www.irac-online.org/ resources/methods. asp). Serial dilutions as ppm the active ingredient of these insecticides were prepared in distilled water. Number of larvae used for each location varied from 75-120, larvae after the treatments were reared in the semisynthetic diet and observation on the mortality vs. dose response was observed after 48hr exposure. Larvae were regarded as dead when they were not able to move when probed with a blunt probe or brush. Mortality data were corrected by Abbott's formula where necessary and analyzed by probit analysis. Estimation of LC₅₀ values and their 95% fiducial limits (FL) was done by probit analysis using the SPSS.

RESULTS AND DISCUSSION

The data on the LC_{50} values of profenophos to the populations of H. armigera given in Table 1 reveal that the values varied from 26.41 to 33.28 ppm; least LC₅₀ value was observed in Dharwad population (26.41 ppm) and maximum with that of Bangalore (33.28 ppm) (2014-15); in 2015-16, these values varied from 28.20 to

Bellary

Bangalore

Gangavathi

34.7 ppm; least being with Dharwad population (28.20 ppm) and maximum with Raichur one (34.77 ppm). Similarly, the least value was observed in Dharwad population (26.71 ppm) and maximum with Raichur one (33.28 ppm) in 2016-17. As regards chlorpyriphos, the values ranged from 37.28 to 41.08 ppm; least with Gangavathi (35.16 ppm-2014-15); and in 2015-16, with the larvae from Dharwad (37.35 ppm); LC₅₀ did not vary between Dharwad and Ballari populations during 2016-17. Maximum LC₅₀ values and slopes was obtained with profenophos followed by chlorpyriphos and these were least effective. For emamectin benzoate, LC50 values across locations did not vary much, with overlapping fiducial limits (0.28 to 0.39 ppm), least value being with Bellary population (0.28 ppm) followed by Dharwad (0.29 ppm), and Bangalore (0.39 ppm) during 2014-15; in 2015-16, it varied from 0.26 to 0.47 ppm, and the least LC₅₀ value was observed in Gangavathi population (0.26 ppm). More or less similar results were obtained in 2016-17. Brevault et al. (2009) observed maximum mortality with emamectin-benzoate (33.33 mg a.i L-1) of a high level (99.3 \pm 0.8%) in 2nd instar. Hirooka et al. (2007) obtained a much lower LC₅₀ value (0.049 mg a.i. L-1) for emamectin in a laboratory reared susceptible strain. Gupta et al. (2005) concluded that emamectin benzoate was more toxic than indoxacarb and spinosad.

As regards spinosad, in 2014-15 LC₅₀ values varied from 0.41 to 0.58 ppm; with the least being observed in Kalaburagi population (0.41 ppm) and maximum with that of Bangalore (0.58 ppm). In 2015-16, these varied from 0.38 to 0.53 ppm, least with Bidar population

33.43

34.79

38.56

35.68

39.72

41.52

38.22

41.99

39.11

17.10

20.23

22.32

19.55

26.34

20.29

20.76

22.85

20.08

 1.03 ± 0.13

 1.86 ± 0.22

 2.05 ± 0.20

 1.85 ± 0.25

 1.73 ± 0.56

 2.15 ± 0.29

 2.04 ± 0.29

 2.49 ± 0.15

 2.29 ± 0.10

Insecticides	T4:	Year	n	LC ₅₀	95% (FL)		Clarat C.F.		D
	Location			(ppm)	LL	UL	Slope± S.E	χ2	1
		2014-15	90	29.25	18.2	37.75	1.72 ± 0.38	1.63	0.86
	Raichur	2015-16	120	34.77	28.43	43.49	2.25 ± 0.74	2.05	1.00
		2016-17	90	33.28	25.66	40.61	1.75 ± 0.54	1.15	0.96
		2014-15	105	31.42	20.41	40.82	1.56 ± 0.43	2.14	0.93
	Kalaburagi	2015-16	120	32.25	22.5	44.59	1.85 ± 0.31	1.80	0.85
		2016-17	75	30.76	19.73	41.71	2.05 ± 0.25	1.93	1.00
		2014-15	75	30.28	19.86	42.25	1.29 ± 0.79	1.85	0.56
	Bidar	2015-16	75	32.07	21.95	46.02	1.55 ± 0.50	2.13	0.93
		2016-17	75	30.58	19.18	43.14	1.55 ± 0.40	2.05	1.00
		2014-15	75	26.41	17.78	32.54	2.04 ± 0.22	1.76	0.88
Profenophos 50%EC	Dharwad	2015-16	90	28.20	19.87	36.31	1.93 ± 0.40	2.05	1.05

90

75

90

105

90

120

90

75

2016-17

2014-15

2015-16

2016-17

2014-15

2015-16

2016-17

2014-15

2015-16

2016-17

26.71

27.60

29.39

27.90

33.28

31.04

27.39

28.45

26.96

Table 1. Toxicity to insecticides in field collected populations of H. armigera

1.09 1.00 (contd.)

0.95

0.55

1.10

1.05

1.02

1.01

1.04

2.41

1.52

1.79

1.25

1 95

1.55

1.98

2.00

								(Table	1 contd.)
	Raichur	2014-15	75	0.35	0.16	1.26	1.09± 0.85	3.74	0.54
		2015-16	105	0.44	0.21	1.37	1.44 ± 1.05	2.15	0.65
	** 1.1	2016-17	75	0.39	0.16	1.30	1.50 ± 0.50	1.95	0.75
	Kalaburagi	2014-15	90	0.38	0.18	1.59	1.18 ± 0.46	2.96	0.77
		2015-16	105	0.47	0.23	1.55	1.35 ± 0.58	2.74	0.49
Emamectin benzoate	Bidar	2016-17 2014-15	75 75	0.43 0.32	0.18 0.15	1.48 0.93	1.26 ± 0.28 1.66 ± 0.21	2.05 1.78	1.00 0.91
	Diuai	2014-13	90	0.32	0.13	1.01	1.08 ± 0.21 1.08 ± 0.76	1.78	0.57
		2015-10	90	0.33	0.17	0.94	1.00 ± 0.70 1.00 ± 0.34	2.14	1.05
	Dharwad	2014-15	90	0.29	0.13	3.04	1.55 ± 0.31	2.04	0.59
		2015-16	75	0.34	0.16	0.88	2.05 ± 0.20	2.05	0.74
5%SG		2016-17	90	0.30	0.11	0.81	1.92 ± 0.11	1.96	0.78
	Bellary	2014-15	75	0.28	0.21	1.02	1.19 ± 0.52	2.55	0.72
		2015-16	90	0.31	0.15	0.97	2.11 ± 0.44	1.75	1.05
		2016-17	75	0.28	0.10	0.91	2.01 ± 0.25	1.55	1.00
	Bangalore	2014-15	105	0.39	0.24	1.22	2.04 ± 0.73	2.08	0.56
		2015-16	105	0.35	0.14	1.08	1.89 ± 0.51	2.41	0.83
	Composyathi	2016-17	 75	0.20	0.10	0.92	1 25 1 02	2.16	1.00
	Gangavathi	2014-15 2015-16	75 75	0.38 0.26	0.19 0.18	0.83	1.35 ± 1.02	2.16 1.59	1.00 1.10
		2015-10	75 75	0.20	0.18	1.05 1.00	1.68 ± 0.73 1.36 ± 0.16	1.39	1.10
	Raichur	2014-15	90	0.22	0.13	0.71	1.96 ± 0.10 1.96 ± 0.11	2.18	0.76
	Raichai	2015-16	90	0.52	0.39	0.79	2.05 ± 0.08	2.24	0.76
		2016-17	75	0.48	0.34	0.72	2.05 ± 0.08	1.75	0.75
	Kalaburagi	2014-15	75	0.41	0.33	0.65	1.81 ± 0.09	1.95	0.54
		2015-16	75	0.44	0.35	0.62	1.93 ± 0.15	2.00	0.93
		2016-17	75	0.40	0.30	0.55	1.93 ± 0.15	2.25	0.56
	Bidar	2014-15	75	0.46	0.29	0.81	1.59 ± 0.47	1.88	0.48
		2015-16	102	0.38	0.19	0.91	1.75 ± 0.30	1.75	0.81
		2016-17	90	0.33	0.15	0.82	1.75 ± 0.30	1.54	1.03
	Dharwad	2014-15	75	0.49	0.38	0.56	2.17 ± 0.17	2.41	0.91
Spinosad 45%SC		2015-16	90	0.43	0.21	0.55	2.05 ± 0.15	2.09	1.02
		2016-17	90	0.39	0.16	0.46	2.05 ± 0.15	1.76	0.82
	Bellary	2014-15	75 75	0.58	0.43	1.39	1.55 ± 0.15	1.95	0.85
		2015-16	75 75	0.51	0.38	1.12	2.15 ± 0.22	1.93	0.79
	Dangalara	2016-17 2014-15	75 105	0.47 0.51	0.33 0.38	1.03 0.70	2.15 ± 0.22 1.66 ± 0.12	2.58 2.13	1.16 0.73
	Bangalore	2014-13	75	0.51	0.38	0.70	1.00 ± 0.12 1.93 ± 0.10	1.82	0.73
		2016-17					1.75± 0.10		
	Gangavathi	2014-15	75	0.45	0.31	1.02	1.56 ± 0.18	2.03	0.71
	S 8	2015-16	105	0.47	0.30	1.05	1.95 ± 0.05	2.05	1.10
		2016-17	90	0.43	0.25	0.96	1.95 ± 0.05	3.05	0.68
	Raichur	2014-15	90	14.13	10.17	15.88	2.55 ± 0.15	2.09	0.93
		2015-16	105	15.62	11.14	18.79	1.85 ± 0.10	1.98	1.05
		2016-17	75	18.34	13.96	21.15	1.85 ± 0.10	1.11	0.93
Methomyl 40%SP	Kalaburagi	2014-15	90	15.27	12.09	18.26	1.75 ± 0.23	2.26	0.79
		2015-16	105	16.76	13.06	21.17	2.45 ± 0.31	2.05	1.00
	D: 1	2016-17	75	17.05	13.72	22.04	2.45 ± 0.31	2.05	0.59
	Bidar	2014-15	90	16.29	11.86	20.43	1.89 ± 0.28	1.77	1.01
		2015-16	90	17.78	12.77	22.45	2.04 ± 0.10	2.14	0.95
	Dharwad	2016-17 2014-15	45 75	18.07 14.65	13.43 11.96	23.32 20.42	2.04 ± 0.10 1.58 ± 0.34	2.61	0.69
	Duaiwau	2014-13	75 75	16.14	12.59	20.42	1.38 ± 0.34 2.15 ± 0.20	1.89 2.52	0.84 1.14
		2015-10	60	16.14	13.25	23.00	2.13 ± 0.20 2.15 ± 0.20	1.85	0.77
	Bellary	2010-17	90	16.56	12.33	17.37	1.73 ± 0.28	3.05	0.70
	Donary	2015-16	75	18.05	13.3	20.28	1.88 ± 0.37	2.79	0.70
		2016-17	90	15.91	11.80	19.66	1.88 ± 0.37	1.77	1.15
	Bangalore	2014-15	90	17.10	13.56	19.08	2.11 ± 0.18	1.79	0.58
	5	2015-16	90	18.59	14.41	20.99	1.95 ± 0.25	2.00	1.09
		2016-17							
	Gangavathi	2014-15	75	13.10	11.86	17.30	1.66 ± 0.35	1.78	0.66
		2015-16	105	14.59	12.23	19.21	2.05 ± 0.22	1.93	0.78
		2016-17	75	14.88	12.89	20.08	2.05 ± 0.22	2.06	1.04

(contd.)

0.62

(contd.)

								(Table	$1\ contd.)$
	Raichur	2014-15	90	0.18	0.11	0.38	2.73 ± 0.24	3.52	0.87
Chlorantriniliprole 18.5%SC		2015-16	105	0.23	0.12	0.41	2.34 ± 0.43	2.59	0.59
		2016-17	75	0.70	0.59	1.17	1.76 ± 0.25	1.74	0.95
	Kalaburagi	2014-15	90	0.25	0.17	0.45	1.86 ± 0.12	2.61	0.76
		2015-16	105	0.43	0.23	0.63	2.19 ± 0.10	3.04	0.90
		2016-17	75	0.94	0.71	1.52	2.25 ± 0.15	2.95	0.88
	Bidar	2014-15	75	0.19	0.13	0.43	1.54 ± 0.59	3.16	1.00
		2015-16	75	0.20	0.13	0.45	1.86 ± 0.21	2.76	1.05
		2016-17	75	0.76	0.56	1.28	3.05 ± 0.20	1.86	1.00
	Dharwad	2014-15	75	0.17	0.10	0.29	2.19 ± 0.26	1.72	0.68
		2015-16	105	0.24	0.10	0.51	2.00 ± 0.10	1.95	1.11
		2016-17	90	0.71	0.61	1.34	2.10 ± 0.36	3.90	0.92
	Bellary	2014-15	75	0.22	0.09	0.36	1.85 ± 0.32	2.58	1.05
		2015-16	90	0.25	0.08	0.39	1.58 ± 0.26	2.44	0.74
		2016-17	90	0.75	0.58	1.40	2.11 ± 0.22	3.20	1.05
	Bangalore	2014-15	90	0.39	0.22	0.55	2.05 ± 0.25	1.86	0.58
		2015-16	90	0.27	0.17	0.49	2.22 ± 0.13	3.15	0.64
		2016-17							
	Gangavathi	2014-15	75	0.20	0.11	0.64	2.13 ± 0.51	2.29	1.06
		2015-16	75	0.19	0.11	0.28	2.09 ± 0.38	2.83	0.85
		2016-17	75	0.74	0.60	1.30	2.00 ± 0.31	1.99	1.13

(0.38 ppm) and maximum with that of Bengaluru. The observations from 2015-16, revealed the least value in Dharwad population (0.33 ppm) and maximum with Raichur (0.48 ppm). Ahmad et al. (2005) found spinosad (1 ppm) toxic to 2nd instar larvae. Kranthi et al. (2000) observed that the toxicity of spinosad was relatively less variable falling within LD₅₀ range of 0.023 to 0.24 μg /larvae and LD₉₀ of 0.27 to 4.33 μg / larvae. With methomyl LC₅₀ varied from 13.10 to 17.10 ppm, least observed in Gangavathi population (13.10 ppm) and maximum in Bangalore (17.10 ppm) during 2014-15. In 2015-16, similar results were obtained. Ahmed et al. (1990) reported that the egg mortalities were more with methomyl @ 1%. LC₅₀ values of thiodicarb varied from 10.73 to 15.15 ppm (2014-15) with least values being in Gangavathi population (10.73 ppm); in 2015-16 these varied from 11.61 to 16.02 ppm, with least being again with Gangavathi population (11.61 ppm). Lowest LC₅₀ value was observed in Dharwad population (12.44 ppm) in 2016-17 (Table 1). Prasad Rao and Grace (2008) reported that LC₅₀ value of thiodicarb was 1.86 µg/ larvae, much higher than that of spinosad, emamectin benzoate and methomyl. The lower level of toxicity and higher level of resistance was also observed earlier by Gunning et al. (1996). The effect of thiodicarb on the larval population of H. armigera was found to be moderate (Ramasubramanian and Regupathy, 2003).

 LC_{50} values of flubendiamide ranged from 0.18 to 0.29 ppm, with the least value being with Gangavathi population (0.18 ppm) and maximum with that of Bellary (0.29 ppm) in 2014-15; least value during 2015-16 was observed in Dharwad population (0.41 ppm); while in 2016-17, it was the least in Dharwad population (0.99 ppm). Naresh Kanwar et al. (2012)

found in their studies, flubendiamide 480 SC was relatively more toxic (relative toxicity was calculated over novaluron); and flubendiamide was 6.41x and lufenuron was 2.73x more toxic (Nikam et al., 2015). LC₅₀ values of chlorantraniliprole in 2014-15 varied from 0.17 to 0.38 ppm and with the least value being with Dharwad population (0.17 ppm) and maximum with that of Bangalore (0.39 ppm). The least value was observed with Gangavathi population (0.19 ppm) and maximum with Kalaburagi population (0.43 ppm) during 2015-16; while the least value was observed in Dharwad population (0.71 ppm), and maximum with Kalaburagi population (0.94 ppm) (2016-17) (Table 1). In laboratory studies, LC_{50} for rynaxypyr (0.1 ppm) were significantly lower compared to indoxacarb and cypermethrin in tobacco budworm (Anonymous, 2007); in third instar larvae of *H. armigera* in okra, susceptibility increased after five generations. Joshua et al. (2008) in bioassay against bollworm obtained LC₅₀ values ranging from 0.038 to 0.089 µg/ml of diet. Thus, bioassay results showed varying degrees of toxicity to insecticides in the populations of H. armigera and the order of toxicity of insecticides chlorantraniliprole > emamectin benzoate > flubendiamide > spinosad > thiodicarb > methomyl > profenophos > chlorpyriphos. If used in rotation with the new insecticides, insecticide resistance management can be better.

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