

# Insecticide resistance in field populations of *Leucinodes orbonalis* (Lepidoptera: Crambidae) in India

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**Abstract**—The status of insecticide resistance in field populations of eggplant fruit and shoot borer, *Leucinodes orbonalis* (Guenée) (Lepidoptera: Crambidae) from the major vegetable growing regions of India was determined during the cropping seasons of 2009–2010 and 2010–2011. Six commonly used insecticides: carbaryl, chlorpyrifos, deltamethrin, endosulfan, fenvalerate, and profenofos were tested against *L. orbonalis* larvae. The resistance ratios (RR) at the lethal dosage (LD)<sub>50</sub> levels were estimated as  $RR = LD_{50} \text{ field strain} / LD_{50} \text{ susceptible strain}$ . The *L. orbonalis* populations exhibited widespread resistance to tested insecticides. The highest average RR in the two-year study was observed in the assays of populations with deltamethrin (21.50–82.42-fold) followed by assays conducted with endosulfan (24.47–68.26-fold), chlorpyrifos (22.17–63.14-fold), carbaryl (39.18–49.09-fold), and fenvalerate (14.00–44.66-fold); and the lowest average RRs were observed in the assays with profenofos (16.65–39.43-fold). The high levels of LD<sub>50</sub> values can be attributed to the long-term indiscriminate use of these insecticides in eggplant (*Solanum melongena* Linnaeus; Solanaceae) growing regions.

## Introduction

Eggplant (*Solanum melongena* Linnaeus; Solanaceae) is known as brinjal or “aubergine” in South Asia, Southeast Asia, and South Africa. India is one of the largest producers of eggplant in the world with 71 100 ha plantation and production around 56 300 tonnes (National Horticulture Board 2014). It is an important cash crop for poor farmers, who cultivate two or three crops per year. Farmers start harvesting fruits at about 60 days after planting and continue to harvest until 90–120 days after planting, thereby providing a steady supply of food for the family and stable income for most of the year. The major constraint for eggplant cultivation and production is the brinjal fruit and shoot borer, *Leucinodes orbonalis* (Guenée) (Lepidoptera: Crambidae), which is the most serious and destructive pest causing yield loss of 70–92% of the total production (Roy and Pande 1994; Dhamdhare *et al.* 1995; Rahman 1997; Haseeb *et al.* 2009).

Damage to the eggplant due to *L. orbonalis* starts in the nursery, and the first symptom of infestation is the appearance of wilted and drooping shoot (shoot damage). At the initial stage, when eggplant fruits have not yet developed, larvae bore into the tender shoots, feed inside, and then tunnel downwards, killing growing points in the process. Larvae also feed on flowers, reducing fruit set and yield. The damaged flower buds drop without blossoming and fruits show visible circular exit holes. The infestation continues until the last harvest is carried on in a subsequent season.

The management practices for control of *L. orbonalis* include the host-plant resistance, mechanical control, biological control, spraying sex pheromone, and insecticides. Insecticides such as bio-pesticides, botanicals, and chitin synthesis inhibitors have been evaluated against *L. orbonalis* (Chatterjee and Roy 2004; Sharma *et al.* 2004; Mishra and Dash 2007) and are being used in some regions of India besides the

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conventional insecticides. An average of 4.6 kg of insecticide (active ingredient) per hectare per season is sprayed on eggplant at a cost of US\$179.60/ha; which is the highest quantity applied to any vegetable crop in India (Choudhary and Gaur 2009). Despite the application of insecticides the eggplant fruits sold in the market are still of inferior quality, because of the infestation of *L. orbonalis* larvae. This approach of increased dependence on pesticides and calendar-based sprays lead to higher costs of production but has not resulted in adequate control of the pest.

Although field management issues have been known for a long time, insecticide resistance in *L. orbonalis* has not been studied and reported. Ali (1994) reported resistance to pyrethroid insecticides in *L. orbonalis* in Bangladesh. Resistance to carbamate and pyrethroid insecticides in *L. orbonalis* was reported in two districts of Bangladesh by Rahman and Rahman (2009). Currently, information on insecticide susceptibility/resistance in Indian populations of *L. orbonalis* is scant, though insecticides belonging to different groups are being used on eggplant crop across India. The objective of our study was to study development of resistance in field-collected field populations of *L. orbonalis* to six conventional insecticides belonging to different groups: carbamate (carbaryl), cyclodiene (endosulfan), organophosphorus (chlorpyrifos and profenofos), and synthetic Pyrethroid insecticides (deltamethrin and fenvalerate).

## Materials and methods

### Insect rearing

The study was carried out for two years during 2009–2010 and 2010–2011. The larvae (second and third instar) of *L. orbonalis* were collected from different locations in northern, central, and southern India, where brinjal is grown as a major crop (Fig. 1). The larvae collected from the field were reared on the modified semi-synthetic diet in the laboratory following standardised procedures at  $25 \pm 1$  °C,  $65 \pm 5\%$  relative humidity, and 9:15 (light:dark) photoperiod until pupation (Anand 2003). Healthy pupae were stored in plastic vials (4 cm diameter and 5 cm height) with a filter paper disc at the bottom and a lid with a mesh window. The emerged adults were released in the sex ratio

of 1:1 in the breeding cages (35 cm height and 15 cm diameter) containing purple paper and wire mesh for egg laying and fed with 10% honey solution and vitamins (Asian Vegetable Research and Development Center 1999). Eggs laid on the purple paper and wire mesh were transferred to plastic containers (9 cm height and 8 cm diameter) and were placed in an incubator at  $28 \pm 1$  °C. The neonates that emerged from eggs were allowed to develop until their third instar and used for insecticide assay. The laboratory susceptible colony used in the study was kept under laboratory conditions for 36 generations.

### Insecticides

The following technical grade insecticides were used for bioassays of *L. orbonalis*: carbaryl (90% w/w), chlorpyrifos (90% w/w), deltamethrin (90% w/w), endosulfan (94% w/w), fenvalerate (90% w/w), and profenofos (90% w/w). These insecticides were purchased from Accu-Standard (New Haven, Connecticut, United States of America).

### Assay procedure

Initially, the larvae of *L. orbonalis* were collected from Jalna, India considering its proximity to the laboratory and availability of infestation. This population was tested with different doses of each insecticide to establish the concentration range where mortality was between 10% and 100% and based on the results obtained seven to eight concentrations of each insecticide were fixed for the bioassays. One microlitre of acetone-based insecticide dilution was manually applied on the dorsal mesothorax of individual using a Hamilton syringe (Hamilton, Reno, Nevada, United States of America) starting from a lower to a higher concentration (Armes *et al.* 1992). Third instars were used in the bioassays and the treated larvae were placed on fresh artificial diet in 25-well insect rearing trays at a temperature of  $26 \pm 1$  °C, humidity of  $60 \pm 5\%$  and 10:14 (light:dark) hours. At least three replicates with 10 larvae/concentration of each insecticide were used and a total of 210 insects were used per insecticide. Acetone alone was used in untreated control with three replicates. Larval mortality was assessed after five days of the topical application of the insecticide. Larvae were considered dead if they

**Fig. 1.** *Leucinodes orbonalis* study localities in India. Location names are followed by state name in parentheses or brackets. Northern India: 1 – Jaipur (Rajasthan), 2 – Karnal (Haryana), 3 – Ludhiana (Punjab), 4 – Varanasi (Uttar Pradesh). Central India: 5 – Anand (Gujarat), 6 – Bhubaneswar (Orissa), 7 – Jalna (Maharashtra), 8 – Nasik (Maharashtra), 9 – Raipur (Chhattisgarh), 10 – 24 Parganas (West Bengal). Southern India: 11 – Coimbatore (Tamil Nadu), 12 – Dharwad (Karnataka).



were unable to move in a coordinated manner when prodded with a blunt needle.

### Data analysis

Data from the replicates were pooled and dose-mortality,  $LD_{50}$ , and their fiducial limits were

computed by probit analysis using POLO-PC (Finney 1971). Resistance ratio (RR) was calculated using the following formula:

$$\text{Resistance ratio (RR}_{50}\text{)} = \frac{LD_{50} \text{ of the field strain}}{LD_{50} \text{ of the susceptible strain}}$$

## Results

### Carbaryl

The LD<sub>50</sub> values for the populations of *L. orbonalis* collected from different locations of India were between 1.829–6.480 µg/larva (Table 1). The highest LD<sub>50</sub> value was observed in the population collected from Anand, India, and the lowest LD<sub>50</sub>s were from in the populations collected from Karnal and Jalna, India, during 2009–2010 and 2010–2011. The RR was found to be highest (39.18 and 49.09-fold) in the population collected from Anand and the lowest RR was observed in the population collected from Karnal, during 2009–2010 and 2010–2011 (Table 1).

### Chlorpyrifos

The assays with populations collected from Ludhiana, India exhibited the highest chlorpyrifos LD<sub>50</sub> and the Jalna population exhibited the lowest LD<sub>50</sub> during 2009–2010 and 2010–2011, respectively (Table 2). The populations of *L. orbonalis* showed varying levels of RRs in the chlorpyrifos assays during 2009–2010 and 2010–2011, respectively. The highest RRs (RR<sub>50</sub>) of 58.88 and 67.40-fold were observed in the

population collected from Ludhiana, and the lowest RR was in the population collected from Jalna (Table 2).

### Deltamethrin

The LD<sub>50</sub> values for different populations exposed to deltamethrin were between 0.034–0.135 µg/larva (Table 3). The maximum LD<sub>50</sub> value of was observed in the assays done with the population collected from Dharwad, India. During 2010–2011, the LD<sub>50</sub> values followed similar trends as observed during 2009–2010. During 2009–2010 and 2010–2011, the RR was highest for the population collected from Dharwad followed by populations collected from Raipur, Jaipur, and Varanasi, India (Table 3).

### Endosulfan

During 2009–2010 and 2010–2011, the highest LD<sub>50</sub> value was observed in the population of *L. orbonalis* collected from Bhubaneswar, India, followed by assay results of populations collected from 24 Parganas, Jalna, and Dharwad, India (Table 4). The lowest LD<sub>50</sub> value was observed in the assays with populations collected from Ludhiana, during 2009–2010 and 2010–2011. When the

**Table 1.** Response of *Leucinodes orbonalis* larval populations to carbaryl during 2009–2010 and 2010–2011.

Locality number	Location	State	n	Year 2009–2010		Year 2010–2011	
				LD <sub>50</sub> (95% CI)	RR <sub>50</sub>	LD <sub>50</sub> (95% CI)	RR <sub>50</sub>
Northern India							
1	Jaipur	Rajasthan	240	3.187 (2.149–4.575)	22.76	4.454 (3.522–5.587)	35.74
2	Karnal	Haryana	240	1.829 (1.495–2.207)	13.06	2.291 (1.730–2.940)	17.36
3	Ludhiana	Punjab	240	2.703 (1.918–3.700)	19.31	2.670 (1.855–3.889)	20.23
4	Varanasi	Uttar Pradesh	240	3.063 (2.531–3.695)	21.88	3.630 (2.872–4.532)	27.50
Central India							
5	Anand	Gujarat	240	5.485 (3.536–8.090)	39.18	6.480 (4.208–9.550)	49.09
6	Bhubaneswar	Orissa	240	4.503 (3.201–6.217)	32.16	5.181 (3.533–7.384)	39.25
7	Jalna	Maharashtra	240	1.876 (1.569–2.232)	13.40	1.953 (0.781–3.049)	14.80
8	Nasik	Maharashtra	240	3.746 (2.732–4.840)	26.76	3.761 (2.582–5.049)	28.49
9	Raipur	Chhattisgarh	240	3.739 (2.548–5.331)	26.71	3.910 (2.720–5.485)	29.62
10	24 Parganas	West Bengal	240	2.274 (1.315–3.456)	16.24	2.361 (1.044–4.109)	17.89
Southern India							
11	Coimbatore	Tamil Nadu	240	2.778 (1.668–4.293)	19.84	3.540 (2.173–5.435)	26.82
12	Dharwad	Karnataka	240	5.403 (4.214–6.879)	38.59	5.689 (4.172–7.504)	43.10
	Laboratory		240	0.140 (0.094–0.191)		0.132 (0.081–0.170)	–

$\chi^2$ , chi-square goodness-of-fit as determined using POLO-PC and departures from an expected model based on heterogeneity factor >1.0; CI, confidence intervals at 95% level; RR<sub>50</sub>, resistance ratio = LD<sub>50</sub> value of population/LD<sub>50</sub> value of susceptible (laboratory) population; n, total number of larvae used during the bioassay; LD, lethal dose expresses in µg/larva.



**Table 2.** Response of *Leucinodes orbonalis* larval populations to chlorpyrifos during 2009–2010 and 2010–2011.

Locality number	Location	State	n	Year 2009–2010		Year 2010–2011	
				LD <sub>50</sub> (95% CI)	RR <sub>50</sub>	LD <sub>50</sub> (95% CI)	RR <sub>50</sub>
Northern India							
1	Jaipur	Rajasthan	240	0.800 (0.619–1.014)	50.00	0.831 (0.641–1.059)	55.40
2	Karnal	Haryana	240	0.931 (0.706–1.204)	58.19	0.970 (0.730–1.266)	64.67
3	Ludhiana	Punjab	240	0.942 (0.709–1.218)	58.88	1.011 (0.754–1.317)	67.40
4	Varanasi	Uttar Pradesh	240	0.776 (0.514–1.102)	48.50	0.831 (0.520–1.257)	55.40
Central India							
5	Anand	Gujarat	240	0.603 (0.462–0.762)	37.69	0.626 (0.492–0.784)	41.73
6	Bhubaneswar	Orissa	240	0.493 (0.329–0.687)	30.81	0.511 (0.409–0.631)	34.07
7	Jalna	Maharashtra	240	0.336 (0.169–0.546)	21.00	0.350 (0.252–0.552)	23.33
8	Nasik	Maharashtra	240	0.545 (0.362–0.760)	34.06	0.592 (0.453–0.748)	39.47
9	Raipur	Chhattisgarh	240	0.676 (0.545–0.829)	42.25	0.761 (0.610–0.939)	50.73
10	24 Parganas	West Bengal	240	0.598 (0.404–0.839)	37.38	0.702 (0.485–0.979)	46.80
Southern India							
11	Coimbatore	Tamil Nadu	240	0.751 (0.580–0.952)	46.94	0.780 (0.536–1.106)	52.00
12	Dharwad	Karnataka	240	0.749 (0.490–1.076)	46.81	0.901 (0.513–1.457)	60.07
	Laboratory		240	0.016 (0.011–0.022)		0.015 (0.012–0.027)	–

$\chi^2$ , chi-square goodness-of-fit as determined using POLO-PC and departures from an expected model based on heterogeneity factor >1.0; CI, confidence intervals at 95% level; RR<sub>50</sub>, resistance ratio = LD<sub>50</sub> value of population/LD<sub>50</sub> value of susceptible (laboratory) population; n, total number of larvae used during the bioassay; LD, lethal dose expresses in µg/larva.

**Table 3.** Response of *Leucinodes orbonalis* larval populations to deltamethrin during 2009–2010 and 2010–2011.

Locality number	Location	State	n	Year 2009–2010		Year 2010–2011	
				LD <sub>50</sub> (95% CI)	RR <sub>50</sub>	LD <sub>50</sub> (95% CI)	RR <sub>50</sub>
Northern India							
1	Jaipur	Rajasthan	240	0.091 (0.063–0.129)	45.50	0.117 (0.072–0.170)	78.00
2	Karnal	Haryana	240	0.061 (0.033–0.109)	30.50	0.082 (0.043–0.158)	54.67
3	Ludhiana	Punjab	240	0.070 (0.048–0.102)	35.00	0.074 (0.039–0.129)	49.33
4	Varanasi	Uttar Pradesh	240	0.075 (0.052–0.109)	37.50	0.096 (0.059–0.140)	64.00
Central India							
5	Anand	Gujarat	240	0.056 (0.040–0.077)	28.00	0.062 (0.043–0.102)	41.33
6	Bhubaneswar	Orissa	240	0.034 (0.024–0.047)	17.01	0.039 (0.026–0.054)	26.00
7	Jalna	Maharashtra	240	0.046 (0.031–0.066)	23.00	0.048 (0.031–0.070)	32.00
8	Nasik	Maharashtra	240	0.056 (0.033–0.094)	28.00	0.062 (0.031–0.105)	41.33
9	Raipur	Chhattisgarh	240	0.113 (0.073–0.168)	56.50	0.139 (0.089–0.202)	92.67
10	24 Parganas	West Bengal	240	0.043 (0.028–0.064)	21.50	0.048 (0.031–0.071)	32.00
Southern India							
11	Coimbatore	Tamil Nadu	240	0.048 (0.030–0.075)	24.00	0.055 (0.029–0.085)	36.67
12	Dharwad	Karnataka	240	0.135 (0.091–0.198)	67.50	0.142 (0.077–0.249)	97.33
	Laboratory		240	0.002 (0.001–0.002)	–	0.0015 (0.0011–0.0021)	–

$\chi^2$ , chi-square goodness-of-fit as determined using POLO-PC and departures from an expected model based on heterogeneity factor >1.0; CI, confidence intervals at 95% level; RR<sub>50</sub>, resistance ratio = LD<sub>50</sub> value of population/LD<sub>50</sub> value of susceptible (laboratory) population; n, total number of larvae used during the bioassay; LD, lethal dose expresses in µg/larva.

LD<sub>50</sub> values were compared with the LD<sub>50</sub> value of susceptible laboratory colony, the RR was found to be highest for the population collected from

Bhubaneswar, India (69.64 and 74.01-fold) followed by 24 Parganas, Jalna, and Dharwad, during 2009–2010 and 2010–2011, respectively (Table 4).

**Table 4.** Response of *Leucinodes orbonalis* larval populations to endosulfan during 2009–2010 and 2010–2011.

Locality number	Location	State	<i>n</i>	Year 2009–2010		Year 2010–2011	
				LD <sub>50</sub> (95% CI)	RR <sub>50</sub>	LD <sub>50</sub> (95% CI)	RR <sub>50</sub>
Northern India							
1	Jaipur	Rajasthan	240	3.870 (2.767–5.071)	30.23	3.690 (2.158–5.730)	35.36
2	Karnal	Haryana	240	3.943 (3.253–4.735)	30.80	4.498 (3.193–6.113)	40.89
3	Ludhiana	Punjab	240	2.821 (0.974–5.016)	22.04	2.960 (1.221–4.891)	26.91
4	Varanasi	Uttar Pradesh	240	3.630 (2.667–4.669)	28.36	3.756 (2.580–5.042)	34.15
Central India							
5	Anand	Gujarat	240	3.771 (1.821–6.066)	29.46	4.337 (2.584–6.387)	39.43
6	Bhubaneswar	Orissa	240	8.414 (2.640–14.443)	69.64	8.170 (5.768–11.417)	74.01
7	Jalna	Maharashtra	240	5.044 (2.424–8.416)	39.41	5.411 (2.772–8.814)	49.19
8	Nasik	Maharashtra	240	4.275 (3.118–5.559)	33.40	4.312 (3.045–7.787)	39.20
9	Raipur	Chhattisgarh	240	4.001 (1.834–6.702)	31.26	4.111 (3.084–5.235)	37.37
10	24 Parganas	West Bengal	240	6.004 (4.196–8.092)	46.91	7.049 (4.588–10.163)	64.08
Southern India							
11	Coimbatore	Tamil Nadu	240	3.387 (2.166–4.711)	26.46	3.581 (2.249–5.064)	32.55
12	Dharwad	Karnataka	240	4.592 (2.547–7.076)	35.88	4.612 (1.916–8.173)	41.93
	Laboratory		240	0.128 (0.096–0.165)		0.110 (0.100–0.181)	–

$\chi^2$ , chi-square goodness-of-fit as determined using POLO-PC and departures from an expected model based on heterogeneity factor >1.0; CI, confidence intervals at 95% level; RR<sub>50</sub>, resistance ratio = LD<sub>50</sub> value of population/LD<sub>50</sub> value of susceptible (laboratory) population; *n*, total number of larvae used during the bioassay; LD, Lethal dose expresses in µg/larva.

**Table 5.** Response of *Leucinodes orbonalis* larval populations to fenvalerate during 2009–2010 and 2010–2011.

Locality number	Location	State	<i>n</i>	Year 2009–2010		Year 2010–2011	
				LD <sub>50</sub> (95% CI)	RR <sub>50</sub>	LD <sub>50</sub> (95% CI)	RR <sub>50</sub>
Northern India							
1	Jaipur	Rajasthan	240	2.355 (1.804–1.006)	42.05	2.143 (1.628–2.805)	41.21
2	Karnal	Haryana	240	2.366 (1.886–2.978)	42.25	2.448 (1.685–3.034)	47.08
3	Ludhiana	Punjab	240	2.102 (1.679–2.633)	37.54	2.446 (1.910–3.260)	47.04
4	Varanasi	Uttar Pradesh	240	0.962 (0.677–1.336)	17.18	0.971 (0.664–1.325)	18.67
Central India							
5	Anand	Gujarat	240	1.521 (1.085–2.017)	27.16	1.550 (1.064–2.140)	29.81
6	Bhubaneswar	Orissa	240	0.942 (0.620–1.383)	16.82	0.970 (0.495–1.722)	18.65
7	Jalna	Maharashtra	240	0.923 (0.641–1.310)	16.48	0.908 (0.700–1.160)	17.46
8	Nasik	Maharashtra	240	1.860 (1.377–2.499)	33.21	1.906 (0.870–2.940)	36.65
9	Raipur	Chhattisgarh	240	2.066 (1.623–2.614)	36.89	2.090 (1.411–3.081)	40.19
10	24 Parganas	West Bengal	240	1.994 (1.270–3.086)	35.61	2.036 (1.318–2.861)	39.15
Southern India							
11	Coimbatore	Tamil Nadu	240	0.769 (0.630–0.932)	13.73	0.763 (0.608–0.928)	14.67
12	Dharwad	Karnataka	240	1.345 (0.916–1.782)	24.02	1.050 (0.716–1.508)	20.19
	Laboratory		240	0.056 (0.043–0.071)	–	0.052 (0.038–0.068)	–

$\chi^2$ , chi-square goodness-of-fit as determined using POLO-PC and departures from an expected model based on heterogeneity factor >1.0; CI, confidence intervals at 95% level; RR<sub>50</sub>, resistance ratio = LD<sub>50</sub> value of population/LD<sub>50</sub> value of susceptible (laboratory) population; *n*, total number of larvae used during the bioassay; LD, lethal dose expresses in µg/larva.

### Fenvalerate

The bioassays with fenvalerate indicated that the RRs were in the range of 13.73–42.25 and 14.67–47.08-fold when the LD<sub>50</sub>s of field-collected

populations were compared with LD<sub>50</sub> of the laboratory population, during 2009–2010 and 2010–2011 (Table 5). During 2009–2010, the population from Karnal had the highest LD<sub>50</sub> and

**Table 6.** Response of *Leucinodes orbonalis* larval populations to profenofos during 2009–2010 and 2010–2011

Locality number	Location	State	<i>n</i>	Year 2009–2010		Year 2010–2011	
				LD <sub>50</sub> (95% CI)	RR <sub>50</sub>	LD <sub>50</sub> (95% CI)	RR <sub>50</sub>
Northern India							
1	Jaipur	Rajasthan	240	0.656 (0.424–0.940)	24.30	0.829 (0.51–1.27)	33.16
2	Karnal	Haryana	240	0.581 (0.404–0.778)	21.52	0.613 (0.45–0.78)	24.52
3	Ludhiana	Punjab	240	0.503 (0.128–1.015)	18.63	0.592 (0.28–0.98)	23.68
4	Varanasi	Uttar Pradesh	240	0.446 (0.296–0.608)	16.52	0.575 (0.41–0.75)	23.00
Central India							
5	Anand	Gujarat	240	0.734 (0.508–1.014)	27.19	0.859 (0.60–1.17)	34.36
6	Bhubaneshwar	Orissa	240	0.591 (0.212–1.101)	21.89	0.828 (0.43–1.38)	33.12
7	Jalna	Maharashtra	240	0.432 (0.306–0.570)	16.00	0.557 (0.22–0.99)	22.28
8	Nasik	Maharashtra	240	0.775 (0.495–1.132)	28.70	0.921 (0.63–1.29)	36.84
9	Raipur	Chhattisgarh	240	0.478 (0.340–0.631)	17.70	0.490 (0.25–0.78)	19.60
10	24 Parganas	West Bengal	240	0.909 (0.638–1.229)	33.67	1.130 (0.61–1.85)	45.20
Southern India							
11	Coimbatore	Tamil Nadu	240	0.853 (0.558–1.188)	31.59	0.930 (0.70–1.21)	37.20
12	Dharwad	Karnataka	240	0.576 (0.211–1.065)	21.33	0.579 (0.39–0.80)	23.16
	Laboratory		240	0.027 (0.021–0.034)		0.025 (0.019–0.032)	

$\chi^2$ , chi-square goodness-of-fit as determined using POLO-PC and departures from an expected model based on heterogeneity factor >1.0; CI, confidence intervals at 95% level; RR<sub>50</sub>, resistance ratio = LD<sub>50</sub> value of population/LD<sub>50</sub> value of susceptible (laboratory) population; *n*, total number of larvae used during the bioassay; LD, lethal dose expresses in µg/larva.

the lowest LD<sub>50</sub> value was observed in the population collected from Coimbatore, India followed by Jalna and Bhubaneshwar (Table 5).

### Profenofos

The highest LD<sub>50</sub> value of profenofos was observed in the population collected from 24 Parganas and the lowest observed LD<sub>50</sub> was in Jalna population (Table 6). The RR values were 16.0–33.67 and 22.28–45.20-fold higher when LD<sub>50</sub> values of field-collected populations were compared with the LD<sub>50</sub> of the laboratory colony (Table 6).

### Discussion

Understanding the susceptibility of insect populations to insecticides plays a key role in insecticide resistance management and for developing new strategies for pest control. Evolution of resistance to insecticides can drive the development and application of new chemical control measures in pest management. Although there are several studies that demonstrate field efficacy of against *L. orbonalis*, there are no reports available in literature on insecticide resistance in populations of *L. orbonalis* from India. Most commonly used insecticides in India, such

as carbaryl, chlorpyrifos, endosulfan deltamethrin, fenvalerate, and profenofos were used in the assays of this study. The topical assays were conducted with populations collected during 2009–2010 and 2010–2011. The assay data of populations when compared with the susceptibility data of laboratory colony demonstrated RRs of 13–97-fold across insecticides. The RRs were higher than 40-fold in fenvalerate, deltamethrin, and chlorpyrifos assays done with populations from northern India. The higher levels of resistance in populations from northern India may be due to high use of pesticides in the northern states of Punjab, Haryana, Uttar Pradesh, and Rajasthan. The data on state-wise insecticide use in on eggplant are not available, but these four northern states accounted for more than 50% of national pesticide consumption, every year from 2005 to 2010 (<http://ppqs.gov.in/PMD.htm>). In a survey conducted in three intensively eggplant growing villages of Uttar Pradesh, it was observed that quinalphos, cypermethrin, and endosulfan are the most preferred insecticides by eggplant growers (Shivalingaswamy *et al.* 2003). This survey is indicative of the fact that these groups of insecticides are most preferred among eggplant growers across northern India.

When the RRs were compared between the years 2009–2010 and 2010–2011, no shifts were observed in the assays conducted with carbaryl, chlorpyrifos, endosulfan fenvalerate, and profenofos. However, there was an increase in RRs of few populations in assays conducted with deltamethrin. This could be due to the fact that these populations may not have been collected from same physical location or the use of deltamethrin may have increased in these specific locations during past two to three seasons.

The assays clearly demonstrated that the *L. orbonalis* populations have decreased susceptibility to commonly used insecticides. Our study reported here is the first report from India that provides a comprehensive analysis of the susceptibility of *L. orbonalis* populations to commonly used insecticides. Ali (1994) reported resistance to pyrethroid insecticides in *Helicoverpa armigera* (Hübner) (Lepidoptera: Noctuidae) and *L. orbonalis* populations from Bangladesh and concluded that adequate control of *L. orbonalis* was not observed with pyrethroids due to their continuous use. Though no other information is available on insecticide resistance among populations of *L. orbonalis* in India, there are several published reports on insecticide resistance in polyphagous pests such as *Helicoverpa armigera* and *Spodoptera litura* (Fabricius) (Lepidoptera: Noctuidae). Resistance to endosulfan in populations of *H. armigera* was reported by Kapoor *et al.* (2002), Ramasubramanian and Regupathy (2004), and Bhosale *et al.* (2008). In our study the assays with chlorpyrifos and profenofos demonstrated RRs of up to 63- and 40-fold, respectively, during 2009–2010 and 2010–2011. Similar observations were reported in by Chaturvedi (2004) in *H. armigera* populations. Field-collected populations of *H. armigera* also exhibited resistance to deltamethrin (Dhingra *et al.* 1988; Kranthi *et al.* 2004; Ishtiaq *et al.* 2012) and fenvalerate (Venkataiah *et al.* 1990; Lal 1998; Borad *et al.* 2001). Resistance to insecticides such as was also reported in populations of *S. litura* by Rao and Dhingra (1996), Armes *et al.* (1997), Shafiq Ansari *et al.* (2002), Sahoo *et al.* (2007), and Venkateswarlu *et al.* (2005). These studies indicate that widespread resistance to these insecticides is prevalent in the insect populations.

In our study, a high level of resistance was observed to synthetic pyrethroids followed by

Organochlorine. This may be due to proportionately heavy use of synthetic pyrethroids in eggplant. The results obtained from this study provide baseline information on susceptibility to some commonly used insecticides in eggplant. The information can be used for developing or modifying existing pest management modules in eggplant for effective management of fruit and shoot borer, which can be used with cultural practices, pheromone traps, and technologies such as Bt eggplant. Most importantly, it strongly suggests us that these insecticides need to be discouraged for pest management in eggplant.

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## References

- Ali, M.I. 1994. Circumstantial evidence supports insect resistance in Bangladesh. *Resistance Pest Management Newsletter*, **61**: 3–4.
- Anand, M. 2003. Studies on mass culturing of the brinjal shoot and fruit borer, on semi-synthetic diet. *In Proceedings of the National Symposium on Frontier Areas of Entomological Research, Edited by B. Subrahmanyam, Indian Agricultural Research Institute, New Delhi, India. Pp. 563–564.*
- Armes, N.J., Jadhav, D.R., and King, A.B.S. 1992. Pyrethroid resistance in the pod borer, *Helicoverpa armigera*, in southern India. *In Proceeding Brighton crop protection conference, pests and diseases. British Crop Protection Council, Farnham, United Kingdom. Pp. 239–244.*
- Armes, N.J., Wightman, J.A., Jadhav, D.R., and Rao., G.V.R. 1997. Status of insecticide resistance in *Spodoptera litura* in Andhra Pradesh, India. *Pesticide Science*, **50**: 240–248.
- Asian Vegetable Research and Development Center. 1999. Rearing of egg plant fruit and shoot borer, *L. orbonalis*. A slide set and illustrated guide. Asian Vegetable Research and Development Center, Shanhua, Taiwan. Pp. 99–486.
- Bhosale, S.V., Suryawanshi, D.S., and Bhede, B.V. 2008. Insecticide resistance in field population of American bollworms, *Helicoverpa armigera* (Hübner). (Lepidoptera: Noctuidae). *Pestology*, **32**: 19–22.
- Borad, P.K., Patel, C.C., Patel, J.R., and Chavda, A.J. 2001. Studies on insecticide resistance in *Helicoverpa armigera* (Hubner) in Gujarat State. *Indian Journal of Plant Protection*, **29**: 97–100.



- Chatterjee, M.L. and Roy, S. 2004. Bio efficacy of some insecticides against brinjal shoot and fruit borer, *Leucinodes orbonalis* (Guen.) and effect of Novaluron on natural enemies of brinjal pests. *Pestology*, **28**: 52–56.
- Chaturvedi, I. 2004. A survey of insecticide resistance in *Helicoverpa armigera* in central and south Indian cotton ecosystems, 1999 to 2003. *Resistance Pest Management Newsletter*, **14**: 36–40.
- Choudhary, B. and Gaur, K. 2009. The development and regulation of Bt brinjal in India (eggplant/aubergine). *International Service for the Acquisition of Agri-Biotech Applications Brief*, **38**: 1–102.
- Dhamdhare, S., Dhamdhare, S.V., and Mathur, R. 1995. Occurrence and succession of pests of brinjal, *Solanum melongena* Linn. at Gwalior (Madhya Pradesh), India. *Journal of Entomological Research*, **19**: 71–77.
- Dhingra, S., Phokela, A., and Mehrotra, K.N. 1988. Cypermethrin resistance in the populations of *Helicoverpa armigera* Hubner. *Proceedings of National Academy of Sciences*, **58**: 123–125.
- Finney, D.J. 1971. *Probit analysis*. Cambridge University Press, Cambridge, United Kingdom.
- Haseeb, M., Sharma, D.K., and Qamar, M. 2009. Estimation of the losses caused by shoot and fruit borer, *Leucinodes orbonalis* Guen. (Lepidoptera: Pyralidae) in brinjal. *Trends in Biosciences*, **2**: 68–69.
- Ishtiaq, M., Mushtaq, A., Saleem, M., and Razaq, M. 2012. Monitoring of resistance in *Spodoptera exigua* (Lepidoptera: Noctuidae) from four districts of the southern Punjab, Pakistan to four conventional and six new chemistry insecticides. *Crop Protection*, **33**: 13–20.
- Kapoor, S.K., Singh, J., Russell, D., Singh, B., and Kalra, R.L. 2002. Susceptibility change of *Helicoverpa armigera* Hubner to different insecticides in Punjab. *Pesticide Research Journal*, **14**: 177–180.
- Kranthi, S.R., Kranthi, S., Behere, G.T., Dhawad, C.S., Wadaskar, R.M., Banerjee, S.K., et al. 2004. Recent advances in insecticide resistance management (IRM) strategies for sustainable cotton pest management in India. *Lead papers of National Symposium on cotton*, Central Institute for Cotton Research, Nagpur, India. Pp. 221–231.
- Lal, R. 1998. Response of *Helicoverpa armigera* (Hübner) to pyrethroids. *Pesticide Research Journal*, **10**: 129–131.
- Mishra, N.C. and Dash, D. 2007. Evaluation of synthetic and neem based pesticide schedules against shoot and fruit borer (*Leucinodes orbonalis* Guen.) on brinjal. *Journal of Plant Protection and Environment*, **4**: 93–96.
- National Horticulture Board. 2014. Indian horticulture database [online]. Ministry of Agriculture, Government of India. Available from [http://nhb.gov.in/area-pro/NHB\\_Database\\_2015.pdf](http://nhb.gov.in/area-pro/NHB_Database_2015.pdf) [accessed 4 February 2016].
- Rahman, A.K.M.Z. 1997. Screening of 28 brinjal line for resistance/tolerance against the brinjal shoot and fruit borer. *Annual Report. Entomology Division, Bangladesh Agricultural Research Institute, Joydebpur, Gazipur, Bangladesh*.
- Rahman, M.M. and Rahman, M.M. 2009. Study on the development of resistance in brinjal shoot and fruit borer against different insecticides. *World Journal of Zoology*, **4**: 137–143.
- Ramasubramanian, T. and Regupathy, A. 2004. Magnitude and mechanism of insecticide resistance in *Helicoverpa armigera* Hub. population of Tamil Nadu, India. *Asian Journal of Plant Sciences*, **3**: 94–100.
- Rao, G.R. and Dhingra, S. 1996. Relative susceptibility of different larval instar *Spodoptera litura* (Fabricius) to some synthetic pyrethroids. *Journal of Entomological Research*, **20**: 103–108.
- Roy, D.C. and Pande, Y.D. 1994. Damage to brinjal by Lepi. Pyraustidae and economics of its insecticidal control. *Indian Journal of Agricultural Research*, **28**: 110–120.
- Sahoo, S.K., Kapoor, S.K., and Singh, B. 2007. Insecticide resistance management in *Spodoptera litura* (Fabricius) (Lepidoptera: Noctuidae) in Punjab, India. *Pesticide Research Journal*, **19**: 226–230.
- Shafiq Ansari, M., Asif, M., and Azizur Rahaman, M.D. 2002. Evaluation of insecticides on tobacco caterpillar *Spodoptera litura* (Fab.). *Pestology*, **25**: 31–33.
- Sharma, J.D., Nagreta, D.S., and Nath, A. 2004. Management of brinjal shoot and fruit borer, *Leucinodes orbonalis* Guen. and persistence of betacyfluthrin in brinjal fruits. *Pesticide Research Journal*, **16**: 139–143.
- Shivalingaswamy, T.M., Rai, S., Wahundeniya, I., Cork, A., Ammaranan, C., and Talekar, N.S. 2003. Development of an integrated pest management strategy for eggplant fruit and shoot borer in South Asia. *Technical Bulletin TB 28. Asian Vegetable Research and Development Center, Shanhua, Taiwan*.
- Venkataiah, M., Subbrathnam, G.V., and Rosaiah, B. 1990. Development of resistance by gram pod borer to fenvalerate. *Indian Journal of Plant Protection*, **18**: 285–286.
- Venkateswarlu, U.U., Madhumathi, I., and Rao, P.A. 2005. Relative toxicity of novel insecticides against insecticide resistant Guntur, strain of *Spodoptera litura* (Fab.) on cotton in Andhra Pradesh. *Pesticide Research Journal*, **17**: 33–35.