ABioefficacy of *Bacillus thuringiensis* var kurstaki against eggplant borer, *Leucinodes orbonalis* (Pyraustidae: Lepidoptera)

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Abstract
Median lethal concentration (LC_{50}) value of Dipel was lesser with 83.93, 190.0 and 310.42 ppm against II, III and IV instar larvae than Delfin which had 245.24, 372.32 and 536.34 ppm against the II, III and IV instar *Leucinodes orbonalis* larvae, respectively. Testing the efficacy of formulated commercial *B. t* products revealed that, Delfin with a maximum concentration of 0.25 per cent persisted for longer duration of 9 and 6 days on shoots and fruits respectively while, Dipel persisted for shorter period of 7 and 5 days respectively.

Keywords: Effectiveness, persistence, commercial *B. t* products, *Leucinodes orbonalis*.

INTRODUCTION

Eggplant is a hardy plant compared to other vegetables. Because of the hardiness it can be successfully grown in very dry areas under rainfed conditions or with minimum irrigation facilities. Eggplant can be kept more than one year in productive stage by pruning at the end of the season. Brinjal, a versatile vegetable is one of the most popular and economically important vegetables among small-scale farmers and low-income consumers of the entire universe. Area and production of brinjal crop in the world are 5,56,000 hectares and 89,79,000 tonnes respectively. Out of the total area, 90 per cent is in Asia and the production is 77,91,000 million tones (FAO, 2000). In India, brinjal is cultivated in 0.50 million hectares and the production is 7.85 million tonnes, while in Tamil Nadu the area is 7,189 ha and the productivity 13.08 tonnes (Anon., 2000).

Such an economically important commercial crop is reported variedly to be infested by 142 species of insects, 4 species of mites and 3 species of nematodes (Sohi, 1966), 26 pests (Vevai, 1970), 23 species of insects and 19 diseases (Gowda and Veeresh, 1984), 50 insect pests (Nair, 1967), more than 36 pests (Regupathy et al., 1997) from the time of its planting to harvest. Rizvi (1996) highlighted nine major pests, mites and nematodes causing economic losses in brinjal. Among them the Internationally known eggplant fruit and shoot borer (EFSB) *Leucinodes orbonalis* Guenee (Pyraustidae: Lepidoptera) is considered to be the most serious pest of brinjal in all parts of India (Isahaque and Chaudhuri, 1984).

MATERIALS AND METHODS

The commercial *Bacillus thuringiensis* var kurstaki (B.t.k.) formulations viz., Delfin 25 WG and Dipel 8L were mostly preferred by the farmers in managing many of the lepidopteran pests on various crops. Further B.t.k was reported to be effective against *L. orbonalis* under field condition (Abhilash, 2000). Hence, these products were tested against the second, third and fourth instar larvae of *L. orbonalis* under laboratory condition. The details of the products tested are given below:
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<table>
<thead>
<tr>
<th>B.t.k. products</th>
<th>Doses tested against</th>
<th>Company</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>II instar</td>
<td>III instar</td>
</tr>
<tr>
<td>Delfin 25 WG</td>
<td>50 - 300 ug/l</td>
<td>2 - 2 ug/l</td>
</tr>
<tr>
<td>Dipel 8L</td>
<td>0.1 - 1.0 ml/l</td>
<td>0.5 - 2.5 ml/l</td>
</tr>
</tbody>
</table>

Table 1: Dosage mortality response of different instar of *L. orbonalis*

<table>
<thead>
<tr>
<th>S No.</th>
<th>Instar</th>
<th>No. of larvae</th>
<th>$\chi^2$ (n-2)</th>
<th>B±SE</th>
<th>LC$_{50}$ (ppm)</th>
<th>LC$_{90}$ (ppm)</th>
<th>Fiducial limit (ppm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>a. Delfin</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1. II</td>
<td>90</td>
<td>6.07</td>
<td>2.07</td>
<td>+0.1857</td>
<td>245.24</td>
<td>211.97</td>
<td>283.82</td>
</tr>
<tr>
<td>2. III</td>
<td>90</td>
<td>5.82</td>
<td>2.49</td>
<td>+0.2176</td>
<td>372.32</td>
<td>327.53</td>
<td>419.93</td>
</tr>
<tr>
<td>3. IV</td>
<td>90</td>
<td>3.41</td>
<td>1.69</td>
<td>+0.2199</td>
<td>536.34</td>
<td>454.05</td>
<td>645.59</td>
</tr>
<tr>
<td>b. Dipel</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1. II</td>
<td>90</td>
<td>8.35</td>
<td>2.34</td>
<td>+0.2119</td>
<td>83.93</td>
<td>73.76</td>
<td>94.20</td>
</tr>
<tr>
<td>2. III</td>
<td>90</td>
<td>2.04</td>
<td>2.51</td>
<td>+0.256</td>
<td>190.01</td>
<td>165.78</td>
<td>213.79</td>
</tr>
<tr>
<td>3. IV</td>
<td>90</td>
<td>3.10</td>
<td>2.8</td>
<td>+0.2748</td>
<td>310.00</td>
<td>279.20</td>
<td>344.95</td>
</tr>
</tbody>
</table>

All the lines are significantly a good fit P (<0.05)

Bioassay with dipping method

Brinjal nodal region of the size (3 x 0.7 cm) was cut using a sterile blade, and dipped for 30 seconds with various concentrations viz., 150, 160, 170, 180, 190, 200 and 210 ppm of *B.t.k* formulations containing 0.01 per cent of *Tween 20* as sticking agent. Then treated nodes were shade dried and used for bioassay. Nodal regions treated with distilled water and sticker served as control. The treated and control nodes were dried in the shade and used for bioassays. Larval mortality was measured after 6, 2, 18, 24, 30, 36, 42, and 48 h post treatment. LC$_{50}$ and LC$_{90}$ were calculated using probit analysis (Finney, 1971).

Persistence of *Bacillus thuringiensis* on brinjal plants

Persistence of the B.t.k. formulations was tested by applying the products on plants in simulated field tests. The brinjal plants (60 DAT) were sprayed with commercial *B.t.k* formulations viz., Delfin 25 WG at 0.5, 0.1, 0.15, 0.2 and 0.25 concentrations along with 0.01 per cent *Tween 20* using a handsprayer. Utmost care was taken to ensure uniform deposition of spray fluid. The nodal region and fruits were sampled immediately after spraying and at 24 h interval for ten days. Twenty nodal regions and ten fruits were maintained per replication. The nodal region and fruits was brought to the laboratory and placed in disposable cups at one per cup.

$$\%OAR = \frac{\% \text{ Larval mortality by feeding treated surface after respective days}}{\% \text{ Larval mortality by feeding treated surface immediately}} \times 100$$

The neonate larvae were released on the nodal region while second instar larvae of *L. orbonalis* of uniform size on the fruits kept inside disposable cups covered with muslin cloth, secured tightly with a rubber band. After 24 h of feeding, the larvae were transferred individually to surface sterilized (carbendazim 250 mg in 250 ml of sterile distilled water) potato disc (1 cm dia). A set of larvae fed similarly on untreated nodal region and fruit served as control. The observation on the mortality of larvae was recorded at 24 h interval for ten days. The percent original activity remaining (OAR) was determined as followed by (Praveen, 2000).

RESULTS

Effectiveness of *B.thuringiensis* products on *L. orbonalis*
Bioefficacy of *Bacillus thuringiensis* var kurstaki

**Figure 1:** Effectiveness of B.t.k.(Delfin) against second instar of *Leucinodes orbonalis* on eggplant shoot.

**Figure 2:** Effectiveness of B.t.k.(Delfin) against second instar of *Leucinodes orbonalis* on eggplant fruit.

**Figure 3:** Effectiveness of B.t.k(Dipel) against second instar of *Leucinodes orbonalis* on eggplant fruits.

**Figure 4:** Effectiveness of B.t.k (Dipel) against second instar of *Leucinodes orbonalis* on eggplant shoot.

**Figure 5:** Per cent of original activity of Delfin remaining on eggplant shoot.

**Figure 6:** Per cent of original activity of Delfin remaining on eggplant fruit.

**Figure 7:** Per cent of original activity of Dipel remaining on eggplant shoot

**Figure 8:** Per cent of original activity of Dipel remaining on eggplant fruit
Bioassay were carried out to find LC$_{50}$ and LC$_{90}$ of two commercial B.t. products Delfin and Dipel 8L against II, III and IV instars of *L. orbonalis*. Bioassays with Dipel on egg plant nodes resulted in LC$_{50}$ values of 84, 190 and 310 ppm or th three larval stages, respectively compared to LC$_{50}$ values of Delfin of 245, 372 and 536 ppm. The LC$_{50}$ values of Dipel were significantly lower than those of Delfin against all larval stages (Table 1).

**Persistence of B.thuringiensis products**

Persistence of Delfin and Dipel was found by measuring over 10 days the daily the mortality of second instar larvae feeding on fruits and of first instar larvae feeding on shoots of potted plants sprayed with different concentrations of the two Bt products. For Delfin, the highest mortality (92%) of first instars was observed on shots treated with the highest concentration (0,25 %) and on the day of treatment (Fig. 1). For the lowest concentration (0,05%), start mortality was 79 %. Effect declined during the following days with a decline of around 9.81% pr day for all concentrations. All dosages gave more than 50 % control of larvae within the first 3 days of application. Effects on second instar larvae on fruits were lower (Fig. 2). For the Dipel 8L product, the highest mortality (97%) was also found at the highest concentration on the day of treatment and declined during the following days as it did for all other concentrations (Fig. 3 & 4). Calculating the decline of activity per day, the decline was around 17.05 % pr day. Decline of effect was measured as the slope of mortality as function of day after application. These slopes were not significantly different for the two products at different dosages, but significantly different between the two products at any dosage.

As a result, the effect of Dipel sprayed at the lowest dosages were close to zero after 6 days for I instar on shoot and after 5 day for II instar on fruits. For the highest concentration, effect was close to zero after 8 and 6 days, respectively. (Fig. 7 & 8) The effect of Delfin sprayed at the lowest dosages were close to zero after 6 days for I instar on shoot and after 5 day for II instar on fruits. (Fig. 2) For the highest concentration, effect was close to zero after 7 and 6 days, respectively. (Fig. 5, Fig. 6).

**DISCUSSION**

**Efficacy of B. thuringiensis commercial products to *L. orbonalis***

Among the *B. thuringiensis* commercial products tested, Dipel had minimum LC$_{50}$ value of 83.93, 190.01 and 310.00 ppm than Delfin with 245.24, 372.32 and 536.34 ppm when bioassayed against II, III and IV instars of *L. orbonalis* respectively. Though Yin (1993) and Abilash (2000) observed that Delfin 50 WG at 10 per cent concentration was most effective against *L. orbonalis* in reducing fruit damage in brinjal however, on comparing with Dipel, the response was significantly less.

Efficacy of *B. t.* products against similar fruit borer, *Earias vitella* Fabricius was also reported. Kharbade et al. (1998) observed 100 per cent mortality with certain commercial *B. t.* products and LT$_{50}$ was shorter (29.07 h) for first instar larvae of *E. vitella* with 2 g/lit of Delfin than other *B. t.* products formulations. Li et al. (1996) reported that increased concentrations of *B. t.* products formulations resulted in higher mortality against *H. armigera*. However, Thomas and Poinar (1973) observed no significant difference between Dipel 0.1 per cent alone and Dipel + econeem 0.2 per cent combination against *E. vitella* which might be due to antifeedant principle of neem and hence decreased the effect of *B. t.* being a stomach poison. Spictrurin @ 2 ml took six days to cause 48.8$\%$ while Delfin 1.0 per cent 81.11 and Biobit at 1.0 per cent 88.89 per cent mortality in five days after spraying on *E. vitella*.

**Persistency of B. t. products**

The persistency of *B. t.* products on shoot and fruit was determined as per cent original activity remaining (% OAR). Results of the experiments conducted under pot culture condition indicated that persistence of Delfin 50 WG and Dipel 8L varied significantly which may be due to physical characteristics of the formulations (Pinnock et al. 1971). Delfin at higher test concentrations persisted longer on shoots than fruits, since the OAR on shoots was extended up to nine days. Findings of Li et al. (1996) stating that a *B. t.* product with increased application rate prolonging the persistency was in support of the present investigation since 0.25 per cent effected highest mortality and persistency.
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Lesser persistency on the fruits may be due to growth dilution factor as the fruits grow faster than shoots. However, considerable mortality of *L. orbonalis* larvae could be achieved within 78 and 48 hours exposure after application of Delfin and Dipel respectively before significant loss in OAR. Persistency of Bactospeine, Dipel, experimental strain Hill B.t. 9 and strains HD-1, HD-229 and HD 241, upto 12 days, especially bactospeine maintaining its superiority over other treatments was reported earlier (Mohan, 1988). The persistence of *B.t* on plants and in soil was limited to few weeks as the ultraviolet in sunlight inactivated (Mallapadidam, 1991).

References


