Assessment of insecticides and Cry2AB toxin resistance development in Karnataka population of Plutella xylostella (Linn.)

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Abstract: Insecticidal resistance studies against third instar larvae of DBM (Plutella xylostella L.) were carried out to know the rate of development of resistance from F₁ to F₃ generations in Karnataka population. The third instar larvae obtained from field were subjected to bioassay studies with tested against acephate, cypermethrin, spinosad, cartap hydrochloride and Cry2AB toxin using leaf dip method to calculate LC₅₀ values. The LC₅₀ values of the insecticides were further used to quantify the resistance in P. xylostella of parental generation (F₀) from Karnataka field population. The survivals from F₂ generation were reared to next generation (F₁). Resistance development studies was assessed from F₁ to F₃ generation with third instar larvae in every generation with a concentration that caused 80.00 % mortality for all the test insecticides and Cry2Ab toxin. Results revealed that ((0.17 folds) no resistance was developed against acephate in F₂ generation. In case of cypermethrin 2.33 folds. Resistance studies further revealed that 1.50 folds resistance was developed against spinosad, 2.28 folds against cartap hydrochloride and Cry2Ab toxin 1.49 folds resistance was recorded in F₂ generation. The rate of development of resistance from F₁ to F₂ generations increased in all the test insecticides and Cry toxin, except against acephate in Karnataka population. This data will be useful in the development of insecticide resistance management approach for DBM.

Keywords: DBM, Insecticide Resistance, Karnataka, Toxin

INTRODUCTION

India is the world's largest cauliflower (Brassica oleracea var. botrytis L) grower and second largest cabbage (B. oleracea var. capitata L) grower next to China occupying an area of 2,32,800 and 2,78,800 hectares, respectively (Uthamasamy et al., 2011). Diamondback moth (DBM), Plutella xylostella (L) (Lepidoptera: Yponomeutidae), is important pest of cruciferous crops and ubiquitous in nature (CIE, 1967). In India, DBM was reported in 1914 on cruciferous vegetables and is now the most devastating pest of cole crops in the states of Punjab, Haryana, Himachal Pradesh, Delhi, Uttar Pradesh, Bihar, Andhra Pradesh, Tamil Nadu, Maharashtra and Karnataka (Uthamasamy et al., 2011). The infestation of the pest increases gradually from first fortnight of August and leads to total loss of the crop (Dhaliwala et al., 2010). In India it causes significant economic losses up to 50% with an estimate of US$ 168 million per year. Absence of effective natural enemies and rapid development of insecticide resistance to many classes of insecticides, which account for 30-50% of the total cost of production are considered to be the major causes of increasing pest status of DBM in most parts of the country. DBM occupies second position in being resistant to 82 compounds of insecticides (APRD, 2012) and to be the first species to develop field resistance to Bacillus thuringiensis (Bt) Cry toxins, and is one amongst three insect species to have developed field resistance to Bt based spray products (Talekar and Shelton, 1993). It is documented that resistance is inevitable within a span of two to three years for following the introduction of a new insecticide. Recent examples of field resistance developed to relatively selective new compounds, include indoxacarb, avermectins, spinosad, Bt-based products, benzyl ureas and chlorantraniliprole (Furlong et al. 2013). Hence, the present study was undertaken for quantifying the resistance levels in DBM from Karnataka against four commonly used insecticide groups with diverse modes of action and one entomocidal toxin.

MATERIALS AND METHODS

Laboratory investigations were carried out during 2011-2012 in the Bt Lab, Department of Entomology, Col-
lege of Agriculture, Rajendranagar, Hyderabad.

**LC₅₀ Calculation**

**Test Insect Population:** Cabbage cultivar “Charmant” nursery was raised in the greenhouse and one month old seedlings were transplanted in the main field and raised without any insecticide application. DBM, larvae were collected from farmers’ cabbage fields in and around Hyderabad, to establish culture. Leaves were harvested daily washed with tap water and provided as feed to the larvae. Larvae were allowed to pupate in the jars and the pupae were kept in petri plates and placed in a cage for moth emergence. The emerged adult moths were allowed to lay eggs on mustard seeds. Adults were provided with 10% honey solution fortified with multivitamins and protein on a cotton swab for better egg laying. Mustard seedlings with eggs of DBM were collected from the cage and kept in glass jars for hatching. The neonates were reared on insecticide free cabbage leaves. At every successive instar, the larvae were shifted to clean jars and fresh cabbage leaves were provided. Larvae in the third instar stage were used in bioassay studies.

**Test insecticides and Cry toxin:** To determine the LC₅₀ values of insecticides and Cry toxin against DBM larvae, four insecticides viz., acephate (Organophosphate), cypermethrin (Synthetic pyrethroid), spinosad (Spinosyn), cartap hydrochloride (Neriestoxin) and Cry2Ab toxin were used. 100 ml of one per cent stock solution of all the above test insecticides were used for the preparation of serial dilutions. Initially broad range concentrations were tested for each test insecticide and toxin depending on the 20 to 80% mortality observed, narrow range concentrations were tested. A control was also maintained at each time of experimentation and the mortality data was corrected by using modified Abbott’s formula (Flemings and Ratnakaran, 1985). Bioassay was repeated for treatments wherein control mortality exceeded 20%.

**Stock solution preparation for Cry toxin:** The technical formulation of Cry2Ab (3.93 mg/g) was supplied by Central Institute of Cotton Research, Nagpur. 100 mg of the toxin was dissolved in 5 ml distilled water to obtain a stock solution of 60 µg/ml concentration. The stock solution was subjected to serial dilutions to obtain different concentrations and a drop of Tween-80 was added. Similarly a drop of Tween-80 was added to control also.

**Bioassay:** Bioassays were conducted with third instar larvae of *P. xylostella* by using a standard leaf dip method (Sayyed et al., 2000). A bioassay was conducted to deduce the LC₅₀ of all the four test insecticides and Cry2Ab toxin, this LC₅₀ concentration was used in assessment of resistance in Karnataka populations of DBM from parental generation F₀ to F₇ generation.

The leaf discs (5 cm) were used for bioassay studies. The leaf discs were dipped in 10ml of aqueous solution of various concentrations of test insecticides and Cry toxin, whereas, control leaf discs were immersed in distilled water having a drop of Tween-80 for about fifteen seconds and shade dried before transferring onto a moistened filter paper in a petri plate. Ten third instar larvae were released on each treated leaf disc in each concentration. Each treatment was replicated thrice. Larval mortality was recorded at 24, 48, 72 hours after treatment (HAT) by counting the larvae as dead or moribund when they did not resume activity after repeated proddings. The mortality at 72 HAT was considered as end point for the assessment of toxicity of test insecticides and Cry toxin (Fisk and Wright, 1992). LC₅₀ values of all test insecticides and Cry2Ab toxin were determined by probit analysis (Finney, 1971). The calculated LC₅₀ was used in quantifying the resistance in different populations by inducing selection pressure.

**Quantification of insecticidal resistance in Karnataka population:** To assess the resistant levels in Karnataka DBM population, larvae were collected from Kolar District of Karnataka and reared on insecticide free cabbage leaves in the laboratory. Larvae in the third instar were used for bioassay studies.

**Bioassay and lab selection:** The larvae obtained from the field collected population were designated as F₀ population and the subsequent generations (obtained from previous generations) were designated as F₁ (First generation), F₂ (Second generation), F₃ (Third generation). The process of selection pressure for insecticides and Cry toxin was initiated in the parental generation (F₀) and continued up to F₅ generation. The calculated LC₅₀ values of each insecticide and Cry2Ab toxin was subjected to preliminary bioassay. Individual DBM population was subjected to five concentrations (LC₅₀, two concentrations higher than LC₅₀ and two concentrations lower the value of LC₅₀) of each individual insecticide and Cry2Ab toxin and a control with ten third instar larvae per treatment and replicated thrice. Larval mortality was recorded at 24, 48 and 72HAT. The concentration (LC₅₀) that gave 80% mortality was selected from the preliminary bioassay and the survivals at other concentrations were rejected. Using this LC₅₀ concentration of all the test insecticides and Cry2Ab subsequent bioassays were conducted with Karnataka DBM populations using 100 third instar larvae per treatment (individual insecticide and Cry2Ab) and replicating the same thrice for inducing selection pressure from the parental generation (F₀) onwards along with a control. The survivals in the bioassay were raised to first generation (F₁), again during third instar F₁ larvae were subjected to bioassay in the above mentioned manner till F₅ generation. The concentrations were adjusted in subsequent generations depending on the per cent larval survivals obtained in the previous generation.

**Assessment of insecticidal resistance in *P. xylostella*:** The degree of development of resistance through different generations was determined by working out LC₅₀ values in each generation and thus comput-
ing the resistance ratio (RR) by dividing the LC50 value for F0 generation with LC50 value of the F1 generation (Arora, 2003).

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\text{Resistance ratio (RR)} = \frac{\text{LC50 value of F2 generation}}{\text{LC50 value of F1 generation}}
\]

RESULTS AND DISCUSSION

The initial LC50 calculated for the DBM population collected around Hyderabad cabbage agro-ecosystem for all the four test insecticides and Cry2Ab was acephate (0.1%), cypermethrin (0.008%), spinosad (0.003%), cartap hydrochloride (0.01%) and Cry2Ab (0.3µg/ml), these LC50 values were used for assessing the resistance development for Karnataka DBM population.

Resistance development in DBM against acephate:
The concentrations of acephate which was used in bioassay varied from 0.01% to 0.20% in F1, F2 and F3 generations. The LC50 which was used for inducing selection pressure varied in all three F1, F2 and F3 generations. The documented LC50 against third instar larvae of DBM collected from Karnataka was 0.20%, 0.15%, 0.10%.

The calculated LC50 values obtained in F1, F2 and F3 generations were 0.027%, 0.022% and 0.015%.

Resistance ratios obtained in F2 and F3 generations in comparison to F1 generation were 0.81 and 0.55 folds.

The results obtained in the present study indicated there is no development of resistance to acephate because the resistance ratios being less than one (Table 1). The LC50 values reduced from F1 to F3 generation whereas lowest LC50 was recorded in F1 generation (0.015%).

The results obtained in the present study indicate that the resistance ratios of F2 and F3 generations were less than one which indicated there is no development of resistance to acephate in Karnataka population. In the present study DBM did not develop resistance against acephate. DBM population sampled from Karnataka if got exposed to more selection pressures with acephate for several generations, than resistance might have been developed similar to that of Sannaveerapanavar (1995) who documented 100 fold resistance to acephate in DBM population collected from Bangalore. Calibration of resistance ratios would have been precise with that of laboratory strain maintained for several generations without insecticidal exposure as recorded elsewhere.

Resistance development in DBM against cypermethrin:

LC50 concentration of 0.008% was obtained in bioassay using DBM population sampled from cabbage agro-ecosystem in and around Hyderabad the same was used to obtain the survivals of DBM in Karnataka population with cypermethrin. Concentrations in the range of 0.002% to 0.064% were used bioassays in all three different generations (F1, F2 and F3) to get LC30 and LC50 against DBM third instar larvae of Karnataka population. The calculated LC50 for applying selection pressure were 0.016%, 0.064%, 0.032% in F1, F2 and F3 generations, respectively. The LC50 recorded from the bioassays were 0.003 %, 0.011 % and 0.007% in F1, F2 and F3 generations respectively. Resistance ratios in F2 and F3 generations over the F1 generation were 3.66 and 2.33 (Table 1). The highest median lethal concentration for cypermethrin was documented in Karnataka population LC50-0.011% in F2 generation. In general the median lethal concentration following increasing trend up to F2 generation for Karnataka population and then decreased in F3 generation. Enzymatic role of mixed function oxidases coupled with target site nerve insensitivity (kdr) (Holden, 1979; Gammon, 1980) are regarded as the most common mechanisms of resistance by DBM to synthetic pyrethroids, reduced insecticide penetration through the cuticle is also cited to occur. The low magnitude of resistance development in the present study might be due to the less usage of synthetic pyrethroids (cypermethrin) in cabbage agro-ecosystem.

Resistance folds developed by DBM against cypermethrin are in accordance with other reports -144 fold against cypermethrin in DBM at Panipat (Haryana) and -115 fold resistance to pyrethroids at Delhi and Karnataka (Saxena et al., 1989), 25 fold resistance against pyrethroids (Raju and Singh, 1995), 2650 fold resistance against cypermethrin by DBM population sampled from Bangalore (Sannaveerapanavar, 1995). In the present study there was a decrease in susceptibility pattern upto F2 generation for Karnataka populations followed by increase in the susceptibility in the F3 generation (as depicted by increase in LC50 values) showing a moderate level of resistance development. Further studies are required with regards to calibration of variation in enzyme titer viz., mixed function oxidases, glutathione S transferases using specific substrates that play vital role in resistance development against synthetic pyrethroids as reported in other insects as such. The present results are in corroborations with Liu et al. (1982) who reported resistance ratio of 21 fold for Peng Hu strain that is very similar to many populations of three ecological zones in Pakistan which fell in moderate resistance and 899 fold for Ban-Chu strain in China that is very high resistance zone. Khaliq et al. (2007) reported the LC50 (mg ml−1at 48 h) values of cypermethrin for 18 field populations of P. xylostella from three different zones in Pakistan ranged from 0.19-1.88. The reason for low toxicity of pyrethroids was due to the evolution of multiple resistance mechanisms. Resistance to cypermethrin was also documented in all populations of third instar by Liu et al. (1981) in Taiwan , Chawla and Joia (1991) in Punjab, and Yu and Nguyen (1992) in USA and Perez et al. (2000) in Nicaragua.

Resistance development in DBM against spinosad (Spinosyns):
The median lethal concentration of 0.003% for spinosad was recorded in the bioassay con-
ducted with DBM larval population from Hyderabad. Using this LC50 bioassays were conducted with third instar larvae of DBM collected from Karnataka population to get 80% larval mortality and for inducing selection pressure for resistance development. Concentrations ranging from 0.0015% to 0.004% were used in bioassays studied in F1, F2, F3 generations, respectively. LC50 obtained was 0.0035% in F1, F2 generations and 0.004% in F3 generation of Karnataka population. The LC50 values calculated were 0.002%, 0.002% and 0.003% in F1, F2 and F3 generations (Table 1).

Resistance ratios in DBM population of F2 and F3 generations in relation to F1 generation were 1.00 and 1.50 fold. Studies indicates that resistance was developed against spinosad in Karnataka population.

Resistance development studies for the Karnataka population of DBM against spinosad showed neutral results. Susceptibility pattern for Karnataka population showed miscellaneous results (as depicted by LC50 values) ultimately showing negligible resistance development. The reason for the P. xylostella populations either developing moderate resistance or no resistance may be due to the fact that spinosad being a novel insecticide and the usage pattern and selection pressure by this insecticide is relatively new in cabbage agro ecosystem, alternatively the pest was never predisposed to spinosad sprays in this areas as such. The findings of the present studies are in corroborarion with the findings of Peter et al. (2000), Dey and Som-Choudhary (2001) and Vadodaria et al. (2000), who earlier reported the higher field efficacy of spinosad against P. xylostella.

The present data confirms the findings of Zhao et al. (2002) who determined the toxicity ratio of 1.3 to 1.2 from seven zones and 0.8 to 316 from six zones in Geneva. Kao and Cheng (2001) reported LC50 of 24.06 ppm and 26.77 ppm from Lu Chu and His-hu strain, respectively, in China during 2001. Shelton et al. (1996) showed tolerance ratio of more than 100 to spinosad in DBM population of California (USA), which indicated high levels of resistance than the present study. Walker et al. (2002) showed no significant resistance in field population of diomandback moth (New Zealand) which is in conformity with the findings of the present study.

The present study indicated that population of P. xylostella in Karnataka remain susceptible to spinosad. However, resistance to spinosad occurred in Hawaii (2000), Georgia (2001) and California (2002) as a consequence of multiple years of extensive application. A major reason for the rapid resistance development to spinosad in Hawaii was the lack of suitable alternatitives and the unsynchronized use of insecticide classes that led to continuous population exposure to spinosad as it happened in South East Asia (Sayyed et al., 2003, 2004) and North America (Zhao et al., 2002).

The propensity for the selection of spinosad resistance may have arisen from pre - existence of resistance al-

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### Table 1: Toxicity of insecticide and toxin to third instar larvae of *P. xylostella* in different generations of Karnataka population.

<table>
<thead>
<tr>
<th>Generation</th>
<th>Heterogeneity (χ²)</th>
<th>Regression equation</th>
<th>LC50 (%)</th>
<th>Fiducial limits</th>
<th>Resistance ratio</th>
<th>Slope ± S.E</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acephate</td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>F1</td>
<td>0.904</td>
<td>Y=6.4154+0.9019x</td>
<td>0.027</td>
<td>0.007 - 0.50</td>
<td>1.00</td>
<td>0.9019 ± 0.2429</td>
</tr>
<tr>
<td>F2</td>
<td>6.058</td>
<td>Y=6.6575+0.9977x</td>
<td>0.022</td>
<td>0.006 - 0.39</td>
<td>0.81</td>
<td>0.9976 ± 0.2460</td>
</tr>
<tr>
<td>F3</td>
<td>4.940</td>
<td>Y=7.2742+1.2379x</td>
<td>0.015</td>
<td>0.015 - 0.26</td>
<td>0.55</td>
<td>1.2379 ± 0.2597</td>
</tr>
<tr>
<td>Cypermethrin</td>
<td></td>
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<td></td>
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<tr>
<td>F1</td>
<td>0.706</td>
<td>Y=8.3308+1.3049x</td>
<td>0.003</td>
<td>0.001 - 0.04</td>
<td>1.00</td>
<td>1.3049 ± 0.2933</td>
</tr>
<tr>
<td>F2</td>
<td>0.633</td>
<td>Y=7.5252+1.2999x</td>
<td>0.011</td>
<td>0.006-0.017</td>
<td>3.66</td>
<td>1.2999 ± 0.2797</td>
</tr>
<tr>
<td>F3</td>
<td>1.380</td>
<td>Y=8.2334+1.4813x</td>
<td>0.007</td>
<td>0.003-0.010</td>
<td>2.33</td>
<td>1.4813 ± 0.3017</td>
</tr>
<tr>
<td>Spinosad</td>
<td></td>
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<tr>
<td>F1</td>
<td>1.839</td>
<td>Y=21.5207+6.2115x</td>
<td>0.002</td>
<td>0.001-0.002</td>
<td>1.00</td>
<td>6.2115 ± 1.2354</td>
</tr>
<tr>
<td>F2</td>
<td>0.375</td>
<td>Y=15.4870+3.9491x</td>
<td>0.002</td>
<td>0.002-0.003</td>
<td>1.00</td>
<td>3.9491 ± 1.0790</td>
</tr>
<tr>
<td>F3</td>
<td>0.517</td>
<td>Y=14.9325+3.8552x</td>
<td>0.003</td>
<td>0.002-0.003</td>
<td>1.50</td>
<td>3.8552 ± 1.0609</td>
</tr>
<tr>
<td>Cartap Hydrochloride</td>
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<td></td>
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<tr>
<td>F1</td>
<td>1.307</td>
<td>Y=7.9558+1.3867x</td>
<td>0.007</td>
<td>0.002-0.011</td>
<td>1.00</td>
<td>1.3867 ± 0.4478</td>
</tr>
<tr>
<td>F2</td>
<td>0.966</td>
<td>Y=9.8129+2.6246x</td>
<td>0.015</td>
<td>0.010-0.018</td>
<td>2.14</td>
<td>2.6246 ± 0.6762</td>
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<tr>
<td>F3</td>
<td>2.249</td>
<td>Y=10.2740+2.9525x</td>
<td>0.016</td>
<td>0.009-0.020</td>
<td>2.28</td>
<td>2.9525 ± 0.8741</td>
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<tr>
<td>Cry2Ab</td>
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<tr>
<td>F1</td>
<td>6.597</td>
<td>Y=7.3013+3.3705x</td>
<td>0.208</td>
<td>0.160-0.249</td>
<td>1.00</td>
<td>3.3705 ± 0.5906</td>
</tr>
<tr>
<td>F2</td>
<td>1.758</td>
<td>Y=6.6862+3.2750x</td>
<td>0.306</td>
<td>0.237-0.361</td>
<td>1.47</td>
<td>3.2750 ± 0.7060</td>
</tr>
<tr>
<td>F3</td>
<td>0.563</td>
<td>Y=6.2574+2.4814x</td>
<td>0.311</td>
<td>0.217-0.385</td>
<td>1.49</td>
<td>2.4814 ± 0.6713</td>
</tr>
</tbody>
</table>
levels from the past use of organochlorine insecticides as the mode of action of organochlorine and spinosad as GABA/nicotinic acetyl choline receptor as a target (Ortells and Lunt 1995, Massol et al., 2000). However, the possibility of P. xylostella carrying spinosad resistance allele may have been introduced from other areas via transportation of cabbage also cannot be ruled out (Khalig et al. 2007). Our results indicated that P. xylostella from Karnataka were susceptible to spinosad. From this it can be construed that spinosad can be used commercially as an alternative to particularly those insecticides against which P. xylostella has developed resistance.

Globally, results pertaining to development of resistance manifested by DBM against spinosad showed an 22.4- and 60.7-folds increase in resistance ratios over the parental DBM strain and laboratory susceptible strain in conjunction to the results the genetics of resistance in this strain revealed to be autosomal, incomplete recessive and probably polygenic in association with fitness costs, showed no cross resistance towards abamectin, fipronil, chlorfenpyr and indoxacarb and target site sensitivity was attributed for spinosad resistance (Xiang and Dong, 2006).

Sayyed et al. (2008) also reported field collected DBM population from Pakistan showed a rise in resistance ratio to the tune of 283-, 13- and 67-folds against spinosad, indoxacarb and aceatomprid over the parental population, moreover genetics of resistance to spinosad in this population was inherited as autosomal, incompletely recessive trait coupled with fitness cost attributes. Log dose regression analysis of F1 reciprocal crosses between laboratory susceptible strain and field population of DBM collected from Cameron Highlands revealed that spinosad resistance was inherited as codominant. At the highest dose of spinosad tested, resistance was verge of being completely recessive and at the lowest dose it was incompletely dominant. (Sayyed et al., 2004)

**Resistance development in DBM against cartap hydrochloride (Nereistoxin)**: The median lethal concentration of 0.01% for cartap was recorded in the bioassay conducted with DBM larval population from Hyderabad. Using this LC50 bioassays were conducted with third instar larvae of DBM collected from Karnataka populations to get 80% larval mortality and for inducing selection pressure for resistance development. Concentrations ranging from 0.005% to 0.045% were used in bioassays studied in F1, F2, F3 generations, respectively. LC50 obtained were 0.025%, 0.030% and 0.03%, in F1, F2 and F3 generations, respectively. The LC50 calculated for Karnataka population were 0.007 %, 0.015 %, 0.016 % in F1, F2 and F3 generations (Table 1).

Resistance ratios in DBM population of F3 and F4 generations in relation to F1 generation were 2.14 to 2.28 fold for Karnataka population. The results clearly showed that the resistance ratios were more than one in F1 generation, which indicates that resistance developed against cartap hydrochloride in Karnataka population.

Studies pertaining to resistance development for the P. xylostella populations from Karnataka showed considerable decrease in susceptibility pattern over the three generations and developed moderate resistance over a magnitude of 2 folds.

Branco and Gatehouse (2001) calculated the resistance ratio for a cartap hydrochloride resistant field strain of DBM that ranged from 2.8 to 7.1. On contrary, Vastrad et al. (2004) documented low to moderate level of resistance to cartap hydrochloride against all the DBM populations sampled from three locations that were monitored continuously and for DBM populations sampled from 12 locations that were monitored randomly in Karnataka.

The present study is in accordance with that of Chandrasekharan and Reghupathy (1996) who found resistance levels (expressed as % survival) varied from 17.9 to 52.4 against cartap hydrochloride and Vastrad et al. (2002) who recorded the moderate survival percentage (1.11) of DBM treated with cartap hydrochloride. Sannaveerappanavar and Virakatham (1997) reported the development of resistance to cartap hydrochloride at recommended field concentrations.

The results of the present studies, by and large, fall in line with those obtained by Joia and Udeaan (1997) who obtained LC50 values 0.015% to 0.020% with cartap against a multi-resistant P. xylostella population from Punjab. In the present study in LC50 of cartap in comparison with doses of other test insecticides and Cry2Ab was in the range of 0.007-0.029 which proves the effectiveness of cartap in the management of DBM.

In another study conducted elsewhere using a strain of DBM showing 14.2- resistance folds to cartap and no cross resistance to abamectin implicating the rotation of both these insecticides cartap and abamectin as a successful pest management strategy (Ninsin, 2015).

**Resistance development in DBM against Cry2Ab:** The median lethal concentration of 0.3 μg/ml for Cry2Ab was recorded in the bioassay conducted with DBM larval population from Hyderabad. Using this LC50 bioassays were conducted with third instar larvae of DBM collected from Karnataka population to get 80% larval mortality and for inducing selection pressure for resistance development. Concentrations ranging from 0.1 μg/ml to 1.0 μg/ml were used in bioassays studied in F1, F2, F3 generations, respectively. LC50 obtained was 0.4 μg/ml, 0.5 μg/ml, 0.4 μg/ml in F1, F2 and F3 generations for Karnataka population. The LC50 calculated for Karnataka population were 0.208 μg/ml, 0.306 μg/ml and 0.311 μg/ml in F1, F2 and F3 generations (Table 1).

Resistance ratios in DBM population of F2 and F3 generations in relation to F1 generation were 1.47, 1.49 fold. The results clearly showed that the resistance ratios were more than one in F1 generation, which indicates that resistance developed against Cry2Ab in Karnataka population. The resistance development in P.
**References**


Furlong, Michael J, Wright, Denis J, Dosdall, Lloyd M..2013. *Diamondback moth Ecology and Management:...*


