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Resistance ratio geographical variation in the susceptibility of *P. xylostella* to different biorationals

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Abstract
Five different bioretionals, *viz.*, entomopathogenic fungi such as *Beauveria bassiana* (1.15% WP) and *Metarhizium anisopliae* (1.15% WP), *Bacillus thuringiensis* (0.5% WP), Azadirachtin (300 ppm) and Spinosad (45 SC) were evaluated against DBM larvae. The data on resistance profile of *P. xylostella* population collected from different geographical locations *viz.* NBII, Bangalore, Akola, Sindkhed Raja, Karanja, Aurangabad and Coimbatore at ambient temperature on the basis of larval mortality revealed Karanja with higher resistance to all biorationals. The NBAII, Bangalore populations was used as the laboratory susceptible culture for comparison with populations from Akola, Sindkhed Raja, Karanja, Aurangabad and Coimbatore locations. Population from Coimbatore registered lowest LC_{50} value for Spinosad and *Metarhizium anisopliae*, whereas, Sindkhed Raja registered lowest LC_{50} value for *Bacillus thuringiensis* and Azadirachtin and Aurangabad population revealed lowest LC_{50} value for *Bacillus thuringiensis* and *Beauvera bassiana*.

Keywords: Biorationals, diamondback moth, persistence and toxicity

1. Introduction

Cauliflower is one of the most popular vegetable crops in India. Among the various vegetable crops, Cauliflower is much preferred by the growers because of its assured yield and transportable capacity. The Cauliflower thus becomes a commercial crop and occupies a major area under vegetable cultivation. The importance of cauliflower is for its vitamins A and C, minerals (like iodine, iron, copper, potassium, sulpher etc) used in salad, vegetables, cooked curries, pickles and dehydrated forms and for its medical properties.

The diamond back moth, *Plutella xylostella* (L.) (Lepidoptera: Yponomeutidae) is considered as the most damaging insect pest of cruciferous crops worldwide. *P. xylostella,* is commonly known as diamondback moth because the adult male back forms three yellow diamonds at rest when the wings are folded. DBM larvae are voracious defoliators with a potential to destroy the entire crop if left uncontrolled [1] and can reduce the produce quality and marketability [2].

Biorational would be a key for designing management strategy in a better way for sustained and economically feasible production and would translate into attainment of higher production and productivity in cabbage and cauliflower in a cost effective way. Thus, present investigation was conducted to evaluate five different biorational viz., entomopathogenic fungi such as *Beauveria bassiana* (1.15% WP) and *Metarhizium anisopliae* (1.15% WP), *Bacillus thuringiensis* (0.5% WP), Azadirachtin (300 ppm) and Spinosad (45 SC) against *P. xylostella* larvae

2. Material and Methods

2.1 Insects

P. xylostella population collected from Akola district in Maharashtra was used for the bioassay during 2015-16. At least 10-12 second instar (3 day old) larvae of *P. xylostella* from the F1 population were used for the bioassay.

2.2 Biorationals

Beauveria bassiana 1.15% WP @ 4 gL-1, *Metarhizium anisopliae* 1.15% WP @ 4 gL-1, *Bacillus thuringiensis* 0.5% P @ 2 gL-1, Azadirachtin 300 ppm @ 5 mlL-1 and Spinosad 45

45 SC @ 0.32 mlL-1 were evaluated for their persistence and toxicity at the recommended doses.

2.3 Bioassays
Second instar larvae of *P. xylostella* were exposed to these treated and untreated leaves and allowed to feed. The set of experiment was replicated twice. The observations on the mortality of the larvae were recorded regularly at an interval of 24 hrs up to 5 days for data on larval mortality whereas, up to 15 days for assessment of toxicity up to adult emergence. Moribund larvae and deformed pupa or adult at any stage of the observation were treated as dead.

2.4 Data Analysis
The mortalities recorded in each treatment were converted to corrected per cent mortalities using Abbott's formula [3] and used for further computation of persistent toxicity (PT) values. PT values refer to the product of average percentage residual toxicity (t) and the period (p) for which toxicity is observed [4]. The average residual toxicity was calculated by adding the values of corrected percentage mortalities at 1, 7, 10 and 14 days of observations and then dividing the total by number of observations [4] delineated as under.

3. Result
In case of entomopathogenic fungi, *M. anisopliae* and *B. bassiana* against *Plutella xylostella* collected from different locations, order of efficacy (based on LC_{50}) was Karanja ($1.2x10^5$ cfu/g &$1.6x10^4$ cfu/g) followed by Sindkhed Raja ($5.7x10^4$ cfu/g &$8.1x10^3$cfu/g) after that Akola ($5.3X10^4$ cfu/g & $8.0x10^3$ cfu/g) then Aurangabad ($5.3x10^4$ cfu/g &$6.9x10^3$ cfu/g) then Coimbatore ($2.8x10^4$ cfu/g &$1.2x10^4$ cfu/g) whereas, Lab populations of NBAII, Bangalore ($2.6x10^4$ cfu/g &$6.3x10^3$ cfu/g, respectively) was most susceptible.
In case of *B. t* the order of efficacy was Karanja ($2.5X10^5$ cfu/g) followed by Coimbatore ($1.4X10^5$ cfu/g) then Akola ($1.1X10^5$ cfu/g) next Sindkhed Raja ($1.0X10^5$cfu/g) after that Aurangabad ($1.0X10^5$ cfu/g) and last NBAII, Bangalore ($4.7X10^4$ cfu/g). In case botanicals, Azadirachtin the order of efficacy was Karanja (0.70 ppm) followed by Aurangabad (0.40 ppm) then Akola (0.27 ppm) then Coimbatore (0.17 ppm) followed by Sindkhed Raja (0.16 ppm) and last NBAII, Bangalore (0.083 ppm). Efficacy trend of Spinosad in descending order was NBAII, Bangalore (0.0082 ppm) then Coimbatore (0.031 ppm) after that Aurangabad (0.034 ppm) then Sindkhed Raja (0.039 ppm) followed by Akola (0.047 ppm) and last Karanja (0.12 ppm).

3.1 Geographical variation in the susceptibility of *P. xylostella* to different biorationals.

Table1: Resistance profile of *P. xylostella* population collected from different geographical locations.

Biorationals	***Metarhizium anisopliae* (1.15%WP)**	***Beauveria bassiana* (1.15%WP)**	***Bacillus thuringiensis* (0.5%WP)**	**Azadirachtin (300ppm)**	**Spinosd (45SC)**
Akola	$5.3X10^4$ cfu/g	$8.0x10^3$ cfu/g	$1.1X10^5$ cfu/g	0.27 ppm	0.047 ppm
Sindkhed Raja	$5.7x10^4$ cfu/g	$8.1x10^3$cfu/g	$1.0X10^5$ cfu/g	0.16 ppm	0.039 ppm
Karanja	$1.2x10^5$ cfu/g	$1.6x10^4$ cfu/g	$2.5X10^5$ cfu/g	0.70 ppm	0.12 ppm
Aurangabad	$5.3x10^4$ cfu/g	$6.9x10^3$ cfu/g	$1.0X10^5$ cfu/g	0.40 ppm	0.034 ppm
Coimbatore	$2.8x10^4$ cfu/g	$1.2x10^4$ cfu/g	$1.4X10^5$ cfu/g	0.17 ppm	0.031 ppm
NBAII, Bangalore	$2.6x10^4$ cfu/g	$6.3x10^3$ cfu/g	$4.7X10^4$ cfu/g	0.083 ppm	0.0082 ppm

3.2 Discussion
The data on resistance profile of *P. xylostella* population collected from different geographical locations at ambient temperature on the basis of larval mortality. Population from Coimbatore registered lowest LC_{50} value for Spinosad and *M. anisopliae*, *whereas*, Sindkhed Raja registered lowest LC_{50} value for *B.t* and Azadirachtin and Aurangabad population revealed lowest LC_{50} value for *B.t* and *B. bassiana.*
According to, [5] reported LC_{50} of spinosad was 24.06 and 26.77 ppm from Lu-Chu strain and His-hu strain of DBM from China, respectively.
According to, [6] observed development of a higher resistance levels to synthetic pyrethroids (2314 to 4613 fold), organophosphates (5 to 2726 fold) and carbamates (36 to 108 fold) as compared to endosulfan (16-fold) and *B. thuringiensis* (2 to 15-fold) in *P. xylostella*.
According to, [7] in the pathogenisity studies reported effectiveness of *M. anisopliae* @ $1X10^8$ spore/ml which recorded the larval mortality of 100, 90, 56.7 and 46.7 per cent against 1st 2nd 3rd and 4th instar larvae of *Helicoverpa armigera,* respectively.
According to, [8] and [9] reported variable toxicity ratios in the range of 1.0 to 12.0fold, during survey for spinosad resistance in *P. xylostella* populations from Pakistan.
According to, [10] recorded a resistant development in a field population of *P. xylostella* from farmers' fields in the Cameron Highlands, Malaysia, which indicated resistance ratios of 813, 79, 171, 498 and 1285-fold for indoxacarb, fipronil, spinosad, deltamethrin and *Bt* toxin Cry1Ac, respectively.
According to [11], screened eight isolates of the entomopathogenic fungi *B. bassiana* and *M. anisopliae* indigenous to Benin for virulence against larvae. The *B. bassiana* isolates tested were Bba14, Bba5644, Bba5645, Bba5653, Bba5654, and Bba5655, and *M. anisopliae* isolates were Ma178 and Ma182. The isolate Bba5653 caused 94% mortality of DBM larvae, and the mortality was significantly higher than that caused by any of the other isolates.
According to, [12] studied the acute toxicity of emamectin and spinosad on *P. xylostella* collected from three different locations in Tamil Nadu. The LC_{50} values were found to be 0.066 and 2.12 ppm for emamectin and spinosad, respectively.
According to, [13] reported that the selection of the field population of *H. armigera* from Pakistan with Cry1Ac from G1 to G5 increased the resistance ratio (RR) to 160- fold in comparison with the unselected field population. Interestingly, selection with Cry1Ac also increased RR for deltamethrin (125-fold), chlorpyrifos (650-fold), profenofos (2840-fold), cypermethrin (9830-fold), spinosad (370-fold), indoxacarb (3090-fold), and abamectin (1330-fold).
According to, [14] (2013) reported that the isolate ESALQ-447 of *B. bassiana* (at a concentration of 5 x 106 conidia/ml) and the mixture with castor bean oil (at a concentration of 2% and

at 70 days in storage) reduced larval viability to 40% and pupal viability to 60.8 and 67.6%, respectively of the diamondback moth under greenhouse condition

According to, [15] reported toxic effect of methanol and hexane extracts of neem leaf against diamondback moth (DBM), *Plutella xylostella*. The toxic effect of the two extracts was evaluated at six different concentrations, viz. 0.5%, 1%, 1.5%, 2%, 2.5% and 3%. Mortality of larvae was significantly higher at even the lowest concentration of 0.5% (61.67%) as compared to control (13.33%). Complete larval mortality with neem methanol extract (NME) was recorded at 3% concentration. Mortality during early larval stages was significantly higher as compared to control at all the NME concentrations, and more than 50% larvae died during first two larval stages at 3%, 2.5% and 2% NME concentrations.

4. Summary

In experiment, studied on resistance profile of *P. xylostella* population collected from different geographical locations on ambient temperature only for larval mortality. The NBAII, Bangalore collected populations used as a laboratory susceptible culture and compare with Akola, Sindkhed Raja, Karanja, Aurangabad and Coimbatore locations. In above the Karanja culture shown higher resistance of all biorationals because the larvae collected from those area/field where continuous use of higher molecules.

In case of entomopathogenic fungi, *M. anisopliae* and *B. bassiana* against *P. xylostella* collected from different locations, order of efficacy (based on LC_{50}) was Karanja ($1.2x10^5$ cfu/g & $1.6x10^4$ cfu/g) followed by Sindkhed Raja ($5.7x10^4$ cfu/g & $8.1x10^3$cfu/g) after that Akola ($5.3X10^4$ cfu/g & $8.0x10^3$ cfu/g) then Aurangabad ($5.3x10^4$ cfu/g & $6.9x10^3$ cfu/g) then Coimbatore ($2.8x10^4$ cfu/g & $1.2x10^4$ cfu/g) whereas, Lab populations of NBAII, Bangalore ($2.6x10^4$ cfu/g & $6.3x10^3$ cfu/g, respectively) was most susceptible. In case of *B. thuringiensis* the order of efficacy was Karanja ($2.5X10^5$ cfu/g) followed by Coimbatore ($1.4X10^5$ cfu/g) then Akola ($1.1X10^5$ cfu/g) next Sindkhed Raja ($1.0X10^5$cfu/g) after that Aurangabad ($1.0X10^5$ cfu/g) and last NBAII, Bangalore ($4.7X10^4$ cfu/g). In case botanicals, Azadirachtin the order of efficacy was Karanja (0.70 ppm) followed by Aurangabad (0.40 ppm) then Akola (0.27 ppm) then Coimbatore (0.17 ppm) followed by Sindkhed Raja (0.16 ppm) and last NBAII, Bangalore (0.083 ppm). Efficacy trend of Spinosad in descending order was NBAII, Bangalore (0.0082 ppm) then Coimbatore (0.031 ppm) after that Aurangabad (0.034 ppm) then Sindkhed Raja (0.039 ppm) followed by Akola (0.047 ppm) and last Karanja (0.12 ppm).

5. Conclusions

- In case of entomopathogenic fungi, M. anisopliae (1.15% WP) and *B. bassiana* (1.15% WP) against *P. xylostella* collected from different locations, ascending order of efficacy (based on LC_{50}) was Karanja> Sindkhed Raja > Akola > Aurangabad > Coimbatore whereas, Lab populations of NBAII, Bangalore was most susceptible.
- In case of *B. thuringiensis* (0.5% WP) the increasing trend of toxicity for *P. xylostella* popuations was found in Karanja> Coimbatore> Akola> Sindkhed Raja> Aurangabad > NBAII, Bangalore.
- In case Azadirachtin (300 ppm) the increasing trend of toxicity for *P. xylostella* popuations was found in Karanja> Aurangabad> Akola> Coimbatore> Sindkhed Raja> NBAII, Bangalore
- Efficacy trend of Spinosad (45 SC) in descending order was NBAII, Bangalore > Coimbatore > Aurangabad > Sindkhed Raja> Akola > Karanja.

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