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Base line toxicity of combination insecticides against okra shoot and fruit borer, *Earias vittella* (Fabricius) (Lepidoptera: Noctuidae)

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Abstract
The present laboratory studies were conducted to generate information on relative toxicity of different readymix insecticides Novaluron 5.25% SC + Indoxacarb 4.5% SC, Profenophos 40% EC + Cypermethrin 4% EC, Indoxacarb 14.5% SC + Acetamiprid 7.7% SC, Pyriproxifen 5% EC + Fenpropathrin 15% EC against okra fruit borer *Earias vittella* (Fab.) using larval dip and fruit dip method in 2016-17. From present investigations Indoxacarb 14.5% SC + Acetamiprid 7.7% SC readymix insecticide formulation emerged as the most toxic insecticide formulation for management of *Earias vittella*.

Keywords: *Earias vittella,* readymix insecticides, relative toxicity

Introduction
In a recent scenario readymix formulations of two or more insecticides are commercially marketed in India and many of these products have been tested for their efficacy against different pests of crops [1-4]. Work on efficacy of readymix formulations of insecticides against okra shoot and fruit borer [5-7] revealed that due to their unique mode of actions these insecticides are highly effective. Currently, few readymix insecticide formulations like Novaluron 5.25% SC + Indoxacarb 4.5% SC, Profenophos 40% EC + Cypermethrin 4% EC, Indoxacarb 14.5% SC + Acetamiprid 7.7% SC and Pyriproxifen 5% EC + Fenpropathrin 15% EC are readily available in the market. At present there is very little information available on bioefficacy and persistence toxicity of these readymix insecticide products against okra fruit borer *Earias vittella.* Taking into account an experiment was framed to know the bioefficacy of four readymix insecticide formulations against shoot and fruit borer *E. vittella* on okra.

2. Material and methods
2.1 Insecticides
The present investigation on "toxicity of some readymix products against *Earias vittella* (Fab.)" were conducted in the Department of Entomology, Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Akola (M.S.) during the year 2016-17.

Table 1: Details of insecticides used in experiment

Sr. No.	Readymix insecticide formulation	Trade name	Manufacturer	Field Dose (ml / L)
1	Novaluron 5.25% SC + Indoxacarb 4.5% SC	Plethora	Adama, Pvt. Ltd.	1.75
2	Profenophos 40% EC + Cypermethrin 4% EC	Profex super	Nagarjuna Agrichemicals	2.00
3	Indoxacarb 14.5% SC + Acetamiprid 7.7%SC	Kite	Gharda Chemicals	1.00
4	Pyriproxifen 5% EC + Fenpropathrin15% EC	Sumiprempt	Sumitomo India Pvt. Ltd.	1.00

2.2 Rearing of insect for bioassay studies
For establishing the culture of *E. vittella* in laboratory, rearing methodology with some modifications in rearing material and their dimensions were followed [8].

Initially the infested okra fruits due to *E. vittella* along with larvae were collected from unsprayed okra fields. They were supplied with fresh okra pieces till they undergo pupation in the rearing container. The pupae were separated and kept in for adult emergence**.** The male and female moths were transfered in beljar provided with 10% honey-water solution. The oviposition chambers were covered with black cloth in order to ensure sufficient darkness hours for mating and oviposition of the females. A rolled piece of non- absorbent cotton was kept in the oviposition chamber as ovipositional substratum. The eggs laid by the females were collected daily in incubation jars having size of 15 cm diameter and 18 cm height, kept in BOD incubator at 28±2 °C temperature and 70±5 per cent relative humidity. The newly hatched larvae were reared in small plastic containers (25 cm diameter and 10 cm height) on fresh cut pieces of okra fruits. Fresh food was supplied daily to the larvae and proper hygienic conditions were maintained throughout the rearing.

2.3 Bioassay methods

Larval dip[9] and fruit dip [10] methods of bioassays were followed in order to determine the lethal concentrations and relative Toxicity (RT) values of different readymix formulations against *Earias vittella.*

2.4 Bioassay with larval dip method

Starved third instar larvae were gently dipped in the insecticidal solution of known concentrations for 5 seconds and placed on double layered tissue paper in a petri dish to soak excessive wetness in an aerated condition. They were transferred after 5 min. in plastic boxes (3.5cm height X 6.5cm width X 8cm length) and fed with cut fresh okra pieces. The diet was changed every day in morning. The mortality on each concentration was recorded at a regular interval up to 72 hours.

2.5 Bioassay with fruit dip method

In this method, tender fresh okra fruits were dipped in stock solutions of different insecticide concentrations for 5 seconds using sterilized forceps and placed in petri dishes having double layered tissue paper to soak excessive wetness on the fruits for 5 minutes under aerated conditions. Ten third instar larvae of *Earias vittella* were starved and gently transferred using camel hair brush on 4 to 5 treated okra fruits in plastic boxes (10cm height X 16cm width X 25cm length). The peduncles of the fruits were wrapped with wet cloth to prevent fruit from drying during feeding of the larvae. The mortality on each concentration was recorded at a regular interval up to 72 hours.

2.6 Data analysis

2.6.1 Relative toxicity (RT)

The results were expressed as percentage mortality on each concentration. The mortality data so obtained were converted into corrected per cent mortality [11]. The corrected per cent mortality data, thus, obtained were subjected to the Probit analysis [12] verified using a software (EPA Probit) to compute LC_{50} value for each insecticide from which relative toxicities of different insecticides were determined as below.

$$\text{Relative Toxicity (RT)} = \frac{LC_{50} \text{ of less toxic compound}}{LC_{50} \text{ of more toxic compound}}$$

3. Results

3.1 Relative Toxicity (RT) Studies

The results from the two bioassay studies in larval dip method and fruit dip method on relative toxicities of the four readymix insecticide products against *Earias vittella* are given below.

3.1.1 Relative toxicity (RT) by larval dip method

The results of bioassay studies through larval dip method (table 2) revealed that the LC_{50} of Indoxacarb 14.5% SC + Acetamiprid 7.7% SC was lowest (0.036%) than the rest of the other readymix formulations tested indicating its high toxicity to the third instar larvae of *E. vittella*. The slope of probit test for all the four readymix products was in the range of 1.014 to 2.427 indicated homogeneity in the larval population used for bioassay studies.

It was observed that Novaluron 5.25% SC + Indoxacarb 4.5% SC, Pyriproxifen 5% EC + Fenpropathrin 15% EC and Profenophos 40% EC + Cypermethrin 4% EC were found 1.91, 1.83 and 1.11 fold less toxic than Indoxacarb 14.5% SC + Acetamiprid 7.7% SC readymix insecticide product. From these results the four insecticide readymix products studied for their relative toxicities can be arranged in their decreasing order of toxicities as [Indoxacarb 14.5% SC + Acetamiprid 7.7% SC] > [Profenophos 40% EC + Cypermethrin 4% EC] > [Pyriproxifen 5% EC + Fenpropathrin 15% EC] > [Novaluron 5.25% SC + Indoxacarb 4.5% SC].

3.1.2 Relative toxicity (RT) by fruit dip method

The LC_{50} of Indoxacarb 14.5% SC + Acetamiprid 7.7% SC was lowest (0.038%) than the rest of the other readymix formulations tested indicating its high toxicity to the third instar larvae of *E. vittella* (table 3). The slope of probit test for all the four readymix products was in the range of 1.26 to 2.466 indicated homogeneity in the larval population used for bioassay studies.

The relative toxicities were determined by comparing LC_{50} values with most toxic insecticide readymix product i.e. Indoxacarb 14.5% SC + Acetamiprid 7.7% SC. It was observed that Novaluron 5.25% SC + Indoxacarb 4.5% SC, Pyriproxifen 5% EC + Fenpropathrin 15% EC with and Profenophos 40% EC + Cypermethrin 4% EC were found 2.02, 1.63 and 1.13 fold less toxic than Indoxacarb 14.5% SC + Acetamiprid 7.7% SC readymix insecticide product. From these results the four insecticide readymix products studied for their relative toxicities can be arranged in their decreasing order of toxicities as [Indoxacarb 14.5% SC + Acetamiprid 7.7% SC] > [Profenophos 40% EC + Cypermethrin 4% EC] > [Pyriproxifen 5% EC + Fenpropathrin 15% EC] > [Novaluron 5.25% SC + Indoxacarb 4.5% SC].

Table 2: Relative Toxicity (RT) of selected insecticides to *Earias vittella* by larval dip method

Sr. No.	Insecticides	LC_{50} (%)	Fiducial limits (Lower-Upper)	LC_{90} (%)	Fiducial limits (Lower-Upper)	Heterogeneity (x^2)	Slope ± SE	Relative Toxicity (RT)
1	Novaluron 5.25% SC + Indoxacarb 4.5% SC	0.069	0.055-0.089	0.232	0.175-0.361	3.259	2.427	1.91
2	Profenophos 40% EC + Cypermethrin 4% EC	0.040	0.030-0.051	0.184	0.134-0.292	7.299	1.928	1.11
3	Indoxacarb 14.5% SC + Acetamiprid 7.7% SC	0.036	0.028-0.045	0.162	0.115-0.271	4.612	1.944	1
4	Pyriproxifen 5% EC + Fenpropathrin15% EC	0.066	0.039-0.113	1.204	0.569-3.666	4.042	1.014	1.83

Table 3: Relative Toxicity (RT) of selected insecticides to *Earias vittella* by fruit dip method

Sr. No.	Insecticides	LC_{50} (%)	Fiducial limits (Lower-Upper)	LC_{90} (%)	Fiducial limits (Lower-Upper)	Heterogeneity (x^2)	Slope ± SE	Relative Toxicity (RT)
1	Novaluron 5.25% SC + Indoxacarb 4.5% SC	0.077	0.062-0.095	0.256	0.192-0.399	3.88	2.466	2.02
2	Profenophos 40% EC + Cypermethrin 4% EC	0.043	0.033-0.055	0.184	0.136-0.284	6.952	2.036	1.13
3	Indoxacarb 14.5% SC + Acetamiprid 7.7% SC	0.038	0.029-0.048	0.183	0.127-0.320	6.26	1.865	1
4	Pyriproxifen 5% EC + Fenpropathrin 15% EC	0.062	0.039-0.099	0.634	0.342-1.567	4.816	1.26	1.63

4. Discussion

The relative toxicity studies revealed that there was no considerable difference recorded in the LC_{50} values of a same readymix insecticide product in two different bioassay methods (larval dip and fruit dip) against the third instar larvae of *E. vittella*. A very little difference in the LC_{50} values of insecticides tested by larval dip and surface diet bioassay methods against *Helicoverpa armigera* (Hub.) [13]. The closer LC_{50} values in leaf dip, larval dip and vial assay methods of bioassay against *Plutella xylostella* (L.) support the findings of the present investigations [9]. The identical slopes produced in both the bioassay methods (larval and fruit dip) under investigations suggests that any of these two bioassay methods can be employed for testing the efficacy of the four readymix insecticides against the third instar larvae of *E. vittella*. The leaf dip and topical method of bioassay against *P. xylostella* larvae also arrived at the same conclusion [14]. The larval immersion (dip) method was quicker and more practical for use by extension workers [15]. Since this method did not allowed batch treatment of larvae and required immense care at handling of individual larvae, it seemed more time consuming and laborious than fruit dip method. The fruit dip method of bioassay employed in the present study was found practically less laborious and less time consuming. Moreover, it resembles field application and hence can be preferred for bioassay studies against *E. vittella* larvae.

As far as the toxicities of the selected readymix insecticide formulations are concerned, Indoxacarb 14.5% SC + Acetamiprid 7.7% SC recorded the lowest LC_{50} value with highest relative toxicity values in both larval dip and fruit dip methods of bioassay. It is suggested that the 3rd instar larvae of *E. vittella* were equally susceptible to this readymix insecticide even though the method of bioassay was changed. Similar results were also evidenced in rest of the other three readymix insecticides studied in the present investigation. Indoxacarb is considered as highly toxic to the lepidopteran larvae on crops in field. One of the bioassay studies in laboratory reported that Indoxacarb14.5 SC was relatively more toxic to the 3 to 4 days old larvae of *H. armigera* than Betacyfluthrin 2.5 SC, Lamdacyhalothrin 5 SC and Endosulphan 35 EC [16]. In a readymix combination with Acetamiprid (having different mode of action) this insecticide performed even better resulting into highest relative toxicity value of the ready mix product as observed in the present studies.

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