

Relative toxicity of different novel insecticides against *Spodoptera litura* (Fab.) (Lepidoptera: Noctuidae).

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Abstract: A laboratory experiment was conducted to find out the relative toxicity of different novel insecticides against *Spodoptera litura*. The insecticides tested were found to be more effective by leaf dip method than topical application method. Of all the insecticides tested, emamectin benzoate was found superior with relative toxicity of 85 and 51 times at LC_{50} and LC_{90} respectively, by leaf dip method and it was 40 and 35 times at LC_{50} and LC_{90} values respectively by topical application method compared to cypermethrin. The order of toxicity was emamectin benzoate followed by indoxacarb, chlorantraniliprole, flubendiamide, spinosad and novaluron.

Key words: *Spodoptera litura*, insecticides, LC_{50} , LC_{90} and relative toxicity

INTRODUCTION

Tobacco caterpillar, *S. litura* is a polyphagous noctuid pest which was reported on about 112 cultivated plants (Seth and Sharma, 2001) and is capable of causing 25.8 to 100 per cent losses in different crops based on crop stage and its infestation level in the field (Dhir *et al.*, 1992). A number of insecticides have been proved to be effective against this pest, but their extensive use has resulted in development of resistance, that has led to sporadic outbreaks of the pest and thus leading to the failure of crops (Ahmad *et al.* 2007). Keeping this in view, the present study was planned to determine toxicity of new synthetic molecules with diversified mode of action against this pest in laboratory to provide the organised guidance for the selection of pesticides for the management the pest.

MATERIALS AND METHODS

Insect Culture: The egg masses of *S. litura* collected from the castor plants were maintained on castor leaves in laboratory at $25 \pm 5^{\circ}\text{C}$ and $75 \pm 5\%$ relative humidity by placing them in an incubator. Every day, the hatched larvae were changed to new rearing jars containing fresh leaves. Leaves and the

containers were cleaned and sterilized regularly using one per cent formalin solution before using them for rearing of test insect. In case of the grown up larvae, the number of larvae were limited to 20-25 to give sufficient space. When the larvae reached pre-pupal stage, they were transferred to jars containing a layer (15-20 cm) of fine sterilized moist sand. After three to four days of pupation they were taken out and kept in a jar lined inside with butter paper. The emerged adults were fed with honey solution through a cotton swab which was replaced daily. The egg batches laid on butter paper were collected day wise and maintained on the castor leaves and thus overlapping generations of the test insect was maintained. Thus obtained F_1 generation were utilized for bioassay studies.

Preparation of Insecticidal Solutions:

Insecticides; emamectin benzoate 5 SG (Proclaim, Syngenta Crop Protection limited), indoxacarb 14.5 EC (EI Dupont India Pvt. Ltd), spinosad 45 EC (De-Nocil Crop Protection Ltd), novaluron 10 EC (Indofil Chemicals Company), flubendiamide 39.35 SC (Bayer Crop Science India Ltd), chlorantraniliprole 18.5 SC (El. Dupont India Ltd) and cypermethrin 25 EC (Gujarath Pesticides Pvt. Ltd) were obtained from the respective manufacturers. The proprietary

products were used to prepare one per cent stock solution in distilled water from which further concentrations were prepared subsequently by serial dilution technique.

Bioassay methods adopted: The toxicity of the new insecticides was tested by conducting two methods of bioassay *viz.*, topical bioassay and leaf dip method of bioassay in order to know the contact and oral toxicity, respectively. One microlitre of the respective insecticidal solution was applied on the dorsum of second thoracic segment by micro droplet applicator (Hamilton micro droplet syringe). For the leaf dip method, leaf discs were cut from castor leaves and were dipped in the test concentration and shade dried. The treated leaf discs were fed to the third instar larvae. Each concentration was replicated thrice and ten larvae were released per replication. A control treatment was maintained by feeding the larvae with untreated leaf discs.

Data Collection and Analysis: Mortality of the larvae was recorded at 24, 48, 72, 96, 120, 144 hours after treatment respectively, for all insecticides. The experiments were conducted with a wide range of concentrations initially followed by a narrow range so as to get mortality in the range of 10 to 90 per cent and the data was subjected to probit analysis using EPA1.5 software and the respective LC_{50} , LC_{90} and other parameters were calculated.

Assessment of Relative Toxicity of New Insecticides against *S. litura*: The relative toxicity of the seven insecticides was calculated by comparing with cypermethrin at LC_{50} and LC_{90} level to which the pest has developed resistance. The relative toxicity of new insecticides was assessed by dividing LC_{50} and LC_{90} values of cypermethrin with the corresponding values of the new insecticides at 144 HAT.

RESULTS AND DISCUSSION

Emamectin Benzoate

The LC_{50} values of emamectin benzoate were 0.001 and 0.0004 with relative toxicity values of 40 and 85 by topical and leaf dip methods respectively. The LC_{90} values were 0.02 and 0.007 with relative toxicity values of 35 and 51.4 by topical and leaf dip methods

respectively (Table 1 and 2). The LC_{90} value of emamectin benzoate was 0.02 and 0.007 which was lower than the recommended concentration 0.04 (CIB & RC) for the control this pest. This states that the pest has not developed resistance to insecticide. Emamectin benzoate stood first in LC_{50} and LC_{90} values both by the topical and leaf dip methods.

Emamectin benzoate belongs to the avermectins group and act as chloride channel activators (Teran-Vargas *et al.*, 1997). The insecticide is photostable and has translaminar efficacy with lack of cross resistance with many commercial insecticides (Mrozik, 1994 and White *et al.*, 1997). This could be one of the reasons for its high toxicity against the targeted pests. Its foliar application as insecticide was observed many folds toxic as compared to diet incorporation against important lepidopteran pests (Argentine *et al.*, 2002). Gupta *et al.* (2004); Prasad *et al.* (2007); Dhawan *et al.* (2009); Birah *et al.* (2008) and Sharma and Pathania (2014) has also reported that emamectin benzoate was more toxic to *S. litura* larvae by leaf dip method than by topical application method. This derive support from Decher *et al.* (1990) and Jansson and Dybas (1998) who reported that polar nature of emamectin benzoate is a limiting factor its penetration through the insect cuticle by topical application method *i.e.*, lipophilic. Hydrophilic nature of the toxicant results in reaching the target site faster due to its better solubility in digestive enzymes and haemolymph.

Indoxacarb

The LC_{50} values of indoxacarb were 0.003 and 0.001 with relative toxicity values of 13.3 and 34 by topical and leaf dip methods respectively. The LC_{90} values were 0.03 and 0.02 with relative toxicity values of 23.3 and 18 by topical and leaf dip methods respectively (Table 1 and 2). The LC_{90} value of indoxacarb was 0.03 and 0.02 by topical and leaf dip methods, respectively which was three and two folds by topical and leaf dip methods higher than recommended concentration 0.01 per cent (CIB & RC) for the control this pest.

Indoxacarb was reported to be more effective following ingestion than after topical treatment (Song *et al.*, 2011) this was correlated with its action as a sodium channel blocker insecticide. Gunning

and Devonshire (2002) reported that indoxacarb was found to be more toxic to pyrethroid resistant population of *H. armigera* due to enhanced level of carboxyl esterase which would activate indoxacarb in to more potent metabolite decarbo methoxylate JW-64 which could also be applicable to *S. litura* that has received multiple sprays of pyrethroids and developed resistance before the introduction of novel molecules. From present findings it was also evident that indoxacarb is more toxic through leaf dip method than the topical application method which were in conformity with Gupta *et al.* (2004); Prasad *et al.* (2007); Dhawan *et al.* (2009) and Sharma and Pathania (2014).

Flubendiamide

The LC₅₀ values of flubendiamide were 0.005 and 0.002 with relative toxicity values of 8 and 17 by topical and leaf dip methods respectively. The LC₉₀ values were 0.09 and 0.02 with relative toxicity values of 7.8 and 18 by topical and leaf dip methods respectively (Table 1 and 2). The LC₉₀ value of flubendiamide was 0.09 by topical method, which was 3.2 folds higher than recommended concentration 0.025 per cent (CIB & RC) for the control this pest. But the LC₉₀ value by leaf dip method was lower than that of the recommended concentration for the control of this pest. This may be due to the effectiveness of flubendiamide by oral ingestion method.

The superiority of flubendiamide may be due to the absence of the cross resistance to other chemical classes of insecticides. Nauen *et al.* (2007) has reported flubendiamide as a new option for the control of multi-resistant noctuid pests and an excellent choice in resistant management strategies for lepidopteran pests in general. From present findings it was also evident that flubendiamide was more toxic through leaf dip method than the topical application method which were in conformity with Gupta *et al.* (2004); Prasad *et al.* (2007); Dhawan *et al.* (2009) and Sharma and Pathania (2014).

Chlorantraniliprole

The LC₅₀ values of chlorantraniliprole were 0.005 and 0.001 with relative toxicity values of 8 and 34 by topical and leaf dip methods respectively. The

LC₉₀ values were 0.08 and 0.014 with relative toxicity values of 8.75 and 25.7 by topical and leaf dip methods respectively (Table 1 and 2). The LC₉₀ value of chlorantraniliprole was 0.08 by topical method which was 2.7 folds higher than recommended concentration 0.03 per cent (CIB & RC) for the control this pest. The LC₉₀ value of leaf dip method is lower than that of the recommended concentration for the control of this pest. This may be due to the effectiveness of the insecticide through orally than the contact method. From present findings it was also evident that chlorantraniliprole was more toxic through leaf dip method than the topical application method which were in conformity with Dhawan *et al.* (2009); Karuppaiah and Srivatsava (2013) and Kumar *et al.* (2014).

Spinosad

The LC₅₀ values of spinosad were 0.01 and 0.005 with relative toxicity values of 4 and 6.8 by topical and leaf dip methods respectively. The LC₉₀ values were 0.24 and 0.11 with relative toxicity values of 2.9 and 3.3 by topical and leaf dip methods respectively (Table 1 and 2). The LC₉₀ value of spinosad was 0.24 and 0.11 by topical and leaf dip method respectively which was 6 and 2.75 fold higher than recommended concentration 0.04 per cent (CIB & RC) for the control this pest.

From present findings it was evident that spinosad was more toxic through leaf dip method than the topical application. Rana *et al.* (2002) reported that among the three methods of bio assay techniques *viz.*, residual film method, direct spray on filter paper and larval dip followed by feeding larvae with treated food, third method was found to be more sensitive than other two methods. The present LC₅₀ and LC₉₀ values by topical application method were on a par with the findings of Gupta *et al.* (2004) and the leaf dip method values were on a par with the findings of Karuppaiah and Srivatsava (2013) at LC₅₀ but higher at LC₉₀.

Novaluron

The LC₅₀ values of novaluron were 0.02 and 0.002 with relative toxicity values of 2.0 and 17 by topical and leaf dip methods respectively. The LC₉₀ values were 0.19 and 0.07 with relative toxicity values of

3.7 and 5.1 by topical and leaf dip methods respectively (Table 1 and 2). The LC_{90} value by topical and leaf dip method was lower than that of the recommended concentration. Though the

insecticide has recorded low toxicity than other insecticides tested, the absence of resistance development is one of the major reasons for its wide use against the pest.

Table 1
Toxicity of different novel insecticides against *S. litura* by topical application method

| Chemical | LC_{50} | Relative Toxicity | Ficidual limits | LC_{90} | Relative Toxicity | Ficidual limits | χ^2 | b (Slope \pm SE) |
|---------------------|-----------|-------------------|-----------------|-----------|-------------------|-----------------|----------|-------------------------|
| Emmamectin Benzoate | 0.001 | 40 | 0.0006-0.0015 | 0.02 | 35 | 0.008-0.07 | 5.40 | 1.19 \pm 0.16 |
| Indoxacarb | 0.003 | 13.3 | 0.0019-0.0036 | 0.03 | 23.3 | 0.02-0.12 | 2.19 | 1.04 \pm 0.09 |
| Chlorantraniliprole | 0.005 | 8 | 0.003-0.006 | 0.08 | 8.75 | 0.03-0.07 | 3.06 | 0.94 \pm 0.16 |
| Flubendiamide | 0.005 | 8 | 0.004-0.008 | 0.09 | 7.8 | 0.04-0.18 | 2.20 | 1.04 \pm 0.15 |
| Spinosad | 0.01 | 4 | 0.006-0.02 | 0.24 | 2.9 | 0.08-0.73 | 5.40 | 1.19 \pm 0.16 |
| Novaluron | 0.02 | 2 | 0.014-0.025 | 0.19 | 3.7 | 0.1-0.4 | 3.62 | 1.29 \pm 0.14 |
| Cypermethrin | 0.04 | 1.0 | 0.03-0.06 | 0.7 | 1.0 | 0.31-1.59 | 1.35 | 1.02 \pm 0.18 |

Table 2
Toxicity of different novel insecticides against *S. litura* by leaf dip method

| Chemical | LC_{50} | Relative Toxicity | Ficidual limits | LC_{90} | Relative Toxicity | Ficidual limits | χ^2 | b (Slope \pm SE) |
|---------------------|-----------|-------------------|-----------------|-----------|-------------------|-----------------|----------|-------------------------|
| Emmamectin Benzoate | 0.0004 | 85 | 0.0002-0.0005 | 0.007 | 51.4 | 0.003-0.02 | 1.98 | 0.95 \pm 0.15 |
| Indoxacarb | 0.001 | 34 | 0.0007-0.002 | 0.02 | 18 | 0.007-0.04 | 2.45 | 1.00 \pm 0.16 |
| Chlorantraniliprole | 0.001 | 34 | 0.0006-0.0014 | 0.014 | 25.7 | 0.006-0.03 | 5.49 | 1.20 \pm 0.17 |
| Flubendiamide | 0.002 | 17 | 0.001-0.002 | 0.02 | 18 | 0.008-0.03 | 1.12 | 1.27 \pm 0.18 |
| Novaluron | 0.002 | 17 | 0.0016-0.0038 | 0.07 | 5.1 | 0.03-0.16 | 3.00 | 1.01 \pm 0.12 |
| Spinosad | 0.005 | 6.8 | 0.003-0.007 | 0.11 | 3.3 | 0.05-0.26 | 3.49 | 1.15 \pm 0.14 |
| Cypermethrin | 0.034 | 1.0 | 0.025-0.046 | 0.36 | 1.0 | 0.2-0.7 | 0.86 | 1.18 \pm 0.20 |

Novaluron belongs to the class of chitin synthesis inhibitors sub structural type benzophenyl ureas are compounds with selective properties, affecting the larval stage. They act mainly by ingestion but in some species they suppress fecundity. From present findings it is evident that novaluron is more toxic through leaf dip method than the topical application method which were in conformity with Sharma and Pathania (2014). The present LC_{50} and LC_{90} values were higher than that were reported by Dhawan *et al.* (2007); Dhawan *et al.* (2009) and Kumar *et al.* (2014). The relative toxicity values were lower than the values reported by Prasada Rao (2008) compared it to cypermethrin and Dhawan *et al.* (2007) and Kumar *et al.* (2014) who reported in comparison with chlorpyrifos. This might be due to the continuous use of novaluron

against *S. litura* that has increased the selection pressure which resulted in the development of the resistance.

Cypermethrin

The LC_{50} values of cypermethrin were 0.04 and 0.034 and the LC_{90} values were 0.7 and 0.36 by topical and leaf dip methods respectively. The LC_{90} value of cypermethrin was 0.7 and 0.36 by topical and leaf dip method of application which is 6.4 and 3.3 folds respectively higher than recommended concentration 0.11 per cent (CIB & RC) for the control this pest.

Increase in the levels of resistance to cypermethrin by *S. litura* on cotton from the earlier report (Mayuravalli *et al.*, 1987) to date. The present

findings are in proximity with Kranthi *et al.* (2001) reported high levels of resistance against cypermethrin by *S. litura* strains from both North and South India. In the strains from major districts of Andhra Pradesh it ranged between 45-148 folds. Of all the central Indian strains, those from Mahabubnagar had exhibited highest resistance against cypermethrin. But from the present findings it was evident that there was decrease in the levels of resistance against cypermethrin in *S. litura* on cotton compared to reports of Kranthi *et al.*, 2002 and Prasada Rao (2008) which may be probably due to decreased selection pressure with significant decrease in the use of cypermethrin as result of wide spreading *Bt* cotton.

CONCLUSIONS

The novel insecticides have different target sites and are superior in safety for beneficial insects, humans and animals compared over conventional insecticides. These findings may not reflect field efficacy since these are based on a linear response to a variety of dosages under laboratory conditions, where as field efficacy was influenced by several other factors including coverage and environmental conditions. Coupled with their efficacy against targeted pests and selectivity towards natural enemies, these new molecules can greatly reduce the number of sprays applied per season. Since these new chemicals are mostly contact and stomach poisons, they are reported to be highly efficient in the field. Since, growers have a wide range of alternatives in the form of old and new chemicals, the best strategy would be to use effective compounds as one of the components of pest management strategy.

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