

Research Article

Acute, Persistence Toxicity and Field Evaluation of Spinetoram 12 SC against *Earias Vittella* Fabricius on Okra

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Abstract

The okra shoot and fruit borer, *Earias vittella* (Fabricius), is the major pest on okra which cause extensive damage to the extent of 50 – 60 per cent. Laboratory experiments on acute, persistence toxicity and field evaluation of recent green chemistry biological insecticide spinetoram 12 SC w/v (11.7 w/v), a new class insecticide molecule against third instar larvae of okra shoot and fruit borer, *Earias vittella* (Fab.). LC₅₀ values of spinetoram 12 SC against 3rd instar larvae of *E. vittella* on okra were 4.81, 1.88 and 1.30 ppm; however, LC₉₅ values were 17.79, 9.84 and 6.08 ppm against *E. vittella* after 24, 48 and 72 hour after treatment respectively. In okra spinetoram 12 SC 54 and 45 g a.i./ha were persisted up to 14 days after treatment (DAT) and 36 g a.i./ha persisted for 11 DAT. Based on persistent toxicity index (PTI) values of spinetoram 12 SC, order of relative efficacy (ORE) of the insecticides against *E. vittella* on okra was spinetoram 12 SC 54 g a.i./ha > spinetoram 12 SC 45 g a.i./ha > cypermethrin 25 EC > emamectin benzoate 5 SG > spinetoram 12 SC 36 g a.i./ha > quinalphos 25 EC.

Regarding field experiment in okra, during first season, field application of 54 and 45 g a.i./ha, three times at 15-20 days interval based on economic threshold level were significantly superior in minimizing larval population of *E. vittella* (81.8 and 78.2% reduction respectively). Spinetoram 12 SC provided effective control of *Earias vittella* and is compatible with the predatory coccinellids. It is an effective and excellent tool to be incorporated into integrated pest management for okra crop.

Keywords: Acute toxicity, persistence toxicity, spinetoram, , *Saccharopolyspora spinosad*, *Earias vittella*, okra

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Introduction

Among the total cropped area in India, 2.8 % of area under vegetable cultivation. So, India is the second largest producers of vegetable next to china. Among various vegetables, okra, commonly known as Lady's finger is the most universal and widely grown all over the country. One of the major constraints recognized in its production is the ever-increasing incidence of insect pests, diseases and nematodes, sometimes resulting in substantial yield losses. Due to its tender and supple nature and its cultivation under high moisture & input regimes, okra is more prone to pest attack and at a conservative estimate cause about 35-40% losses. Although there are larger areas under cultivation, productivity remains low. There are many factors for the stagnant or low productivity, and insect pests are one of the major direct causes for yield reduction. Considering the potentiality of vegetable pests as a limiting factor on yield, it is imperative to make more and more scientific information on the effect of newer insecticide molecules including green insecticides, which are available time to time. Insecticides still provide dramatic effects initially, however, repetitive and indiscriminate uses result serious consequences on the biosphere. This condition necessitates for looking at newer insecticide molecules of biological origin. One such new insecticide molecule is spinetoram (derived from soil actinomycetes, *Saccharopolyspora spinosad* Mertz and Yao), a multi component tetracyclic macrolide which belongs to the new chemical class of spinosyn compound [1] and having unique mode of action (Group V insecticides) with an ecotoxicological profile similar to spinosad. Spinetoram has shown outstanding efficacy against codling moth (*Cydia pomonella* Linnaeus), oriental fruit moth (*Grapholita molesta* Busck), army worms (*Spodoptera spp.*), cabbage looper (*Trichoplusia ni* Hubner), western flower thrips (*Frankliniella occidentalis* Pergande), onion thrips (*Thrips tabaci* Lindeman), leaf miners (*Liriomyza spp.*), chilli thrips (*Scirtothrips dorsalis* Hood), fruit borer (*H. armigera*) and many other pests [2, 3].

Residual toxicity resulting from foliar spray of insecticides could be of great significance in indicating an effective period over which an insecticide could persist in biologically active stage under field conditions. The persistence of insecticides depends on many factors: type of insecticide, insecticides physiochemical properties, dose used, host plant and environmental factors. So there is a need to investigate the toxicity of spinetoram 12 SC

persistence against *Earias vittella* on okra plants. However, up till now not much research was made to understand the relationship uniqueness between spinetoram 12 SC and okra shoot and fruit borer, *Earias vittella* which is major pest of okra in tropical situations. Therefore, present investigations were aimed to do research in acute, persistence toxicity and field evaluation of spinetoram 12 SC against *Earias vittella* on okra.

Experimental

Origin and rearing of test insect:

Second to third instar larvae of *E. vittella* were collected from farmers' fields and reared on okra fruits at Insectary, Agricultural College and Research Institute, Madurai. The choice of okra as food was based on earlier reports that developmental time of *E. vittella* was shorter on okra fruits [4]. Fresh and tender okra fruits were collected from potted okra plants grown in Insectary under insecticide free condition. The fruits were cut into pieces of 2.5 cm long and placed in 12 well-cavity trays. Using camel hairbrush, the larvae were transferred on fruits. Filter paper of same size as that of tray was placed over the fruits in order to absorb excess moisture thereby preventing the rotting of fruits. The tray was then covered using plastic lid and fastened by a rubber band. Fresh okra fruit pieces were provided on alternate days.

The boat shaped cocoons were collected and placed in adult emergence cage (30 x 30 x 30 cm). Newly emerged adults were provided with 10 per cent sugar solution fortified with ABDEC multivitamin solution in 5 ml glass vial provided with cotton wool wick to prevent moths from drowning. Four to five tender okra fruits were kept in cages as oviposition substrates. These fruits were removed on next day of egg laying and observed for hatching. The newly hatched first instar larvae were provided with cut pieces of okra fruit [5]. This represented first generation. Similarly, five generations were maintained in the laboratory at $26 \pm 1^\circ\text{C}$, RH $75 \pm 5\%$ and photoperiod 16 : 8 h scoto/photo regimes. Second to third instar larvae of sixth generation were used for laboratory experiments.

Acute toxicity experiment

Experiment on acute toxicity of spinetoram 12 SC against okra shoot and fruit borer, *E. vittella* by Fruit dip bioassay technique of [6] was adopted. Okra fruit disc pieces were surface sterilized in sodium hypochlorite (0.5%), rinsed in sterile water, shade dried, and dipped in spinetoram 12 SC serial concentrations (1.2 ppm, 2.4 ppm, 3.6 ppm, 4.8 ppm, 6.0 ppm and 7.2 ppm) for 30 seconds and shade dried. The treated fruits were transferred to clean plastic containers which lined with moistened filter paper at the bottom. Batches of 30 pre-starved laboratory reared third instar larvae of *E. vittella* were released into each treatment, which was replicated thrice. Larvae released on water treated fruit disc pieces served as control. Observations on larval mortality were fixed till 72 hours of exposure as spinetoram 12 SC tested was lepidoptericides characterized by stomach action showing slower mortality. The cumulative mortality data were observed till 72 h at 24 h interval and corrected by Abbott's formula.

Persistence toxicity experiment

Pot culture experiment was conducted in the Insectary to assess the persistent toxicity of spinetoram 12 SC on shoot and fruit borer in okra. Thirty days old potted okra plants were used for the study. Insecticidal solutions were prepared by dissolving spinetoram 12 SC 0.6 ml, 0.75 ml and 0.9 ml, emamectin benzoate 5 SG 0.34 g, quinalphos 25 EC 1.6 ml and cypermethrin 25 EC 0.4 ml in one liter of water which was equivalent to the field doses. Potted okra plants were sprayed with the insecticides at the respective concentrations at 30 days after sowing (DAS) by using a hand operated sprayer to the point of run-off. After application, treated tender okra fruits were collected separately from the plants starting from first day after treatment (DAT) (2h after spray) and continued 3, 5, 7, 9, 11, 14 and till the mortality due to insecticides on *E. vittella* declined to practically negligible level. In each treatment, treated fruit samples were placed in plastic cups and laboratory reared third instar larvae of *E. vittella* of 20 numbers were released on treated fruits.

After infestation, the containers were placed in a climatic chamber (temperature $25 \pm 1^\circ\text{C}$, relative humidity $70 \pm 10\%$). There were three replications for each insect. Larval mortality was assessed 24 hrs after their confinements by cutting open the fruits under a binocular microscope. Moribund larvae were considered as dead. The per cent mortality was calculated and data were corrected by [7] formula. The product (PT) of average residual toxicity (T) and the period (P) for which the toxicity persisted was used as an index of persistent toxicity. The procedure by [8] and elaborated further by [9, 10] was utilized to calculate the persistent toxicity.

Effect of spinetoram 12 SC against *E. vittella* under field condition

Two field experiments were conducted at farmers' field in Madurai district, Tamil Nadu, India, in the plots of size of 5 X 5 m. The experiments were laid out in a randomized block design at Soorakundu, Melur block and Kokkulam, Chekkanoorani block respectively. Standard agronomic practices as per the recommendations of Tamil Nadu Agricultural University (TNAU) were adopted to maintain healthy okra plants (Hybrid Splendor No. 10). Newer green insecticide molecule spinetoram 12 SC was assessed at various doses (36, 45 and 54 g a. i. / ha) and compared with standard checks (emamectin benzoate 5 SG @ 8.5 g a.i/ha, quinalphos 25 EC @ 200 g a.i/ha and cypermethrin 25 EC @ 50 g a.i/ha) against shoot and fruit borer *E. vittella*. There were three applications at 20 days interval based on ETL of target pests (10% shoot and fruit damage due to *E. vittella*). Thorough coverage of plants (to a run off point) with the spray fluid of 500 l/ha was ensured by using high volume knapsack sprayer with hydraulic cone nozzle. Larval population of *E. vittella* per plant, per cent shoot and fruit damage due to *E. vittella* were assessed from 10 randomly selected plants on pre-treatment, 1, 3, 7 and 10 days after 1st, 2nd and 3rd sprays/treatments (DAT). Marketable fruit yield was recorded from eight harvests and the total fruit yield was represented as quintal/ha.

Statistical analysis

The data from various field experiments were scrutinized by RBD analysis of variance (ANOVA) after getting transformed into $\sqrt{x+0.5}$, logarithmic and arcsine percentage values where appropriate [11]. Critical difference values were calculated at five per cent probability level and treatment mean values were compared using Duncan's Multiple Range Test (DMRT) [12]. The corrected per cent reduction over untreated check in field population was calculated by [13] formula,

$$\text{Corrected per cent reduction} = \left\{ 1 - \frac{T_a \times C_b}{T_b \times C_a} \right\} \times 100$$

where,

- T_a - number of insects in the treatment after spraying
- T_b - number of insects in the treatment before spraying
- C_a - number of insects in the untreated check before spraying
- C_b - number of insects in the untreated after spraying

Results and Discussion**Acute toxicity of spinetoram 12 SC to *E. vittella* on okra**

Data in **Table 1** indicates that per cent mortality of *E. vittella* larvae have positive correlations with the spinetoram concentrations. LC_{50} values of spinetoram 12 SC estimated by fruit dip method to the 3rd instar larvae were 4.81, 1.88 and 1.30 ppm and LC_{95} values were 17.79, 9.84 and 6.08 ppm after 24, 48 and 72 hour after treatment respectively. The probit regression line for spinetoram 12 SC were $Y = 3.121 + 2.774x$, $Y = 4.000 + 2.804x$ and $Y = 4.491 + 3.585x$ after 24, 48 and 72 hours after treatment respectively. As for slope values of the regression line, spinetoram 12 SC had the least slope of 2.77 with 24 hours after exposure, while slope values were 2.80 and 3.59 with 48 h and 72 hours after exposure respectively.

These results corroborate with the findings of [14] who reported that spinosad was the most toxic against *E. vittella* when compared to emamectin benzoate abamectin and indoxacarb and the order of toxicity in terms of LD_{50} ($\mu\text{g larva}^{-1}$) to *E. vittella* was spinosad (0.00188) > abamectin (0.00264) > emamectin benzoate (0.00266) > indoxacarb (0.09270), respectively. However [15] reported that LC_{50} of spinosad was 0.0004 per cent for five day old larvae of *E. vittella*. Differences in the observed values may be due to difference in the age of the larvae used and method of application.

Persistence toxicity of spinetoram 12 SC to *E. vittella* on okra

When spinetoram 12 SC was applied at 54 g a.i./ha, cent per cent mortality of 3rd instar of *E. vittella* larvae was observed upto 3 DAT (**Table 2**). Spinetoram 12 SC at 45 g a.i/ha recorded 98.6, 91.4 and 82.3 per cent mortality at 1,

3 and 7 DAT respectively. More than 50 per cent mortality was observed in spinetoram 12 SC at 36, 45 and 54 g a.i./ha upto 7 and 9 DAT respectively. Persistence for spinetoram 12 SC 36 g a.i./ha was upto 11 DAT and 14 DAT for 45 and 54 g a.i./ha. This was however, upto 7 DAT for emamectin benzoate 5 SG at 8.5 g a.i./ha and cypermethrin 25 EC at 50 g a.i./ha, and upto 5 DAT for quinalphos 25 EC 200 g a.i./ha. There was a reduction in the mortality of *E. vittella* larvae as the time increased and there was no mortality at 21 DAT. The order of relative efficacy (ORE) of the insecticides based on the persistent toxicity index (PTI) values was spinetoram 12 SC 54 g a.i./ha > spinetoram 12 SC 45 g a.i./ha > cypermethrin 25 EC 50 g a.i./ha > emamectin benzoate 5 SG 8.5 g a.i./ha > spinetoram 12 SC 36 g a.i./ha > quinalphos 25 EC 200 g a.i./ha.

Table 1 Acute toxicity of spinetoram 12 SC at different concentrations against *Earias vittella* on okra

Dose (ml/l)	Concentration (ppm)	After 24 h of treatment		After 48 h of treatment		After 72 h of treatment	
		Mean dead larvae \pm SE	Mortality %	Mean dead larvae \pm SE	Mortality %	Mean dead larvae \pm SE	Mortality %
0.01	1.2	3.00 \pm 0.58	10.00	9.70 \pm 0.40	32.33	16.30 \pm 0.40	54.33
0.02	2.4	6.70 \pm 0.40	22.33	18.50 \pm 0.58	61.67	19.77 \pm 0.46	65.67
0.03	3.6	10.33 \pm 0.58	34.33	23.10 \pm 0.58	77.00	22.70 \pm 0.12	75.67
0.04	4.8	13.67 \pm 1.15	45.67	27.30 \pm 0.40	91.00	24.00 \pm 0.58	80.00
0.05	6.0	17.31 \pm 0.58	59.00	29.10 \pm 0.52	97.00	28.30 \pm 0.35	94.33
0.06	7.2	24.10 \pm 0.29	80.33	29.70 \pm 0.52	99.00	29.90 \pm 0.52	99.67
Untreated		1.17	3.90	1.00	3.33	1.00	1.25
LC ₅₀ and Fiducial limit		4.81 (4.07 – 5.68)		1.88 (1.53 – 2.32)		1.30 (0.81 – 2.07)	
LC ₉₅ and Fiducial limit		17.79 (10.76 – 29.34)		9.84 (4.96 – 17.50)		6.08 (4.53 – 8.16)	
Slope		2.77		2.80		3.59	
Regression equation		Y = 3.121 + 2.774x		Y = 4.000 + 2.804x		Y = 4.491 + 3.585x	

Table 2 Persistent toxicity of spinetoram 12 SC to *E. vittella* on okra

Treatments and doses (g a.i./ha)	Corrected per cent mortality at different intervals (days)								P	T	PTI	RE	ORE
	1	3	5	7	9	11	14	21					
Spinetoram12SC 36 g a.i./ha	94.2	89.4	70.3	56.4	31.4	22.9	0.0	0.0	11	60.8	668.6	1.27	5
Spinetoram12SC 45 g a.i./ha	98.6	91.4	82.3	62.6	43.5	36.7	21.6	0.0	14	62.4	873.5	1.65	2
Spinetoram12SC 54 g a.i./ha	100	100	96.6	78.5	63.4	47.6	33.7	0.0	14	74.3	1039.6	1.97	1
Emamectin benzoate 5 SG 8.5 g a.i./ha	96.3	90.5	81.4	62.9	48.3	31.3	0.0	0.0	11	68.5	753.0	1.42	4
Quinalphos 25 EC 200 g a.i./ha	88.4	76.6	73.4	36.6	18.5	0.0	0.0	0.0	9	58.7	528.4	1.00	6
Cypermethrin 25 EC 50 g a.i./ha	100	90.1	79.5	61.3	42.6	31.0	19.6	0.0	14	60.6	848.0	1.60	3

P – Period of toxicity persistence (days); T – Mean per cent mortality; PTI – Persistent toxicity index; RE – Relative efficacy; ORE – Order of relative efficacy

Present findings are in conformity with [16] who found on the basis of PT values, spinosad 0.005% (1026.2) was effective against 1st instar larvae of *E. vittella* and the highest PT value was obtained for spinosad 0.005% (764.9) against 3rd instar larvae of *E. vittella* on okra fruits. [17] also reported that spinosad persisted in cabbage and cauliflower up to 7 and 10 days, respectively following spinosad application at lower and higher dosages. However, spinetoram 12 SC has translaminar activity, thereby providing a relatively prolonged residual action [18].

Field evaluation of spinetoram 12 SC against shoot and fruit borer *E. vittella*

Field experiment observations recorded on 1, 3, 7 and 10 days after treatment (DAT) of two seasons were pooled and given in **Table 3**. Number of larvae of *E. vittella* varied from 4.0 to 4.3 per plant before imposing treatments. In first season experiment, mean data revealed that number of larva ranged from 1.0 to 5.5 larvae per plant due to treatments. Spinetoram 12 SC 54 and 45 g a.i./ha were significantly superior and registered the lowest larval population of 1.0

(81.8% reduction over control) and 1.2 (78.2% reduction over control) per plant, respectively. Spinetoram 12 SC 36 g a.i./ha also contributed moderate reduction in the larval population (1.7 larvae/plant with 69.1% reduction over control). Emamectin benzoate 5 SG at 8.5 g a.i./ha and cypermethrin 25 EC at 50 g a.i./ha registered larval population of 1.9 (65.5% reduction) and 2.4 (56.4% reduction) per plant respectively. Quinalphos 25 EC at 200 g a.i./ha however registered higher larval population 2.8 larvae/plant with 49.1 per cent reduction over control.

Table 3 Effect of spinetoram 12 SC against shoot and fruit borer *E. vittella* on okra – Pooled data of two season experiments

Treatments and doses (g a.i./ha)	Number of larvae per plant on days after treatment					Per cent shoot damage on days after treatment			
	Pre count	1 st season		2 nd season		1 st season		2 nd season	
		Pooled mean of 3 sprays	Per cent reduction over control	Pooled mean of 3 sprays	Per cent reduction over control	Pooled mean of 3 sprays	Per cent reduction over control	Pooled mean of 3 sprays	Per cent reduction over control
Spinetoram 12 SC 36 g a.i./ha	4.1	1.7 ^b	69.1	1.5 ^b	68.1	7.7 ^b	70.0	6.3 ^b	71.8
Spinetoram 12 SC 45 g a.i./ha	4.1	1.2 ^a	78.2	0.9 ^a	80.8	5.0 ^a	80.5	4.1 ^{ab}	81.6
Spinetoram 12 SC 54g a.i./ha	4.0	1.0 ^a	81.8	0.8 ^a	83.0	4.7 ^a	81.7	3.8 ^a	83.0
Emamectin benzoate 5SG 8.5 g a.i./ha	4.1	1.9 ^b	65.5	1.6 ^b	65.9	8.0 ^b	68.8	7.6 ^c	66.0
Quinalphos 25EC 200g a.i./ha	4.4	2.8 ^c	49.1	2.3 ^c	51.0	8.9 ^c	65.3	8.5 ^d	61.9
Cypermethrin 25EC 50g a.i./ha	4.3	2.4 ^c	56.4	2.0 ^c	57.4	8.6 ^c	66.5	8.8 ^d	60.6
Untreated check	4.0	5.5 ^d	-	4.7 ^d	-	25.6 ^d	-	22.3 ^c	-
CD (0.05%)	-	0.15	-	0.13	-	0.007	-	0.02	-
SEd	-	0.07	-	0.06	-	0.003	-	0.01	-

Data are mean values of three replications

Figures were transformed by square root transformation and the original values are given

Means within columns lacking common lower case superscript are significantly different (P<0.05)

In second season field experiment, pooled mean data resulted that number of larva ranged from 0.8 to 4.7 larvae per plant due to all treated experiments. Spinetoram 12 SC 54 and 45 g a.i./ha were equally effective and registered the lowest larval population of 0.8 (83.0% reduction over control) and 0.9 (80.8% reduction over control) per plant, respectively. Lower dose of spinetoram 12 SC 36 g a.i./ha also contributed reasonable reduction in the larval population (1.5 larvae/plant with 68.1% reduction over control). Standard check treatments viz., emamectin benzoate 5 SG at 8.5 g a.i./ha and cypermethrin 25 EC at 50 g a.i./ha recorded larval population of 1.6 (65.9% reduction) and 2.0 (57.4% reduction) per plant respectively. Larval population of 2.3 per plant with 51.0 per cent reduction over control was registered in the treatment with quinalphos 25 EC at 200 g a.i./ha.

Spinetoram is nicotinic acetylcholine receptor (nAChR) allosteric activator [19] and the action on the primary site nAChR may be the reason for the quick knock down effect. These findings are in agreement with [20] who reported that spinetoram 12 SC at 45 g a.i./ha was the most effective in reducing *Spodoptera litura* and *Thrips tabaci* on tomato and *Leucinodes orbonalis* in brinjal.

Spinetoram 12 SC effected marked reduction in the larval population and shoot and fruit damage caused by fruit borer complex. Spinetoram 12 SC at 54 g a.i./ha recorded 81.8 and 83.0 per cent reduction of *E. vittella* in the first and second field experiments, respectively. Spinetoram 12 SC 45 g a.i./ha was on par with spinetoram 12 SC 54 g a.i./ha in reducing the larval population of *E. vittella*. Even though *E. vittella* has the habit of concealed nature inside the shoots and fruits, spinetoram 12 SC active ingredient possesses systemic activity which is the ability of the active ingredient to penetrate plants and move to various parts of the plant and thus plant itself will obtain insecticidal activity [21]. These findings are in agreement with [22] who reported that spinosad 45 SC at 0.1% was the most

effective in reducing the shoot and fruit borer in okra. Spinosad @ 0.005%, 750 ml, 30 g a.i/ha, 75 g a.i/ha and 75 g a.i/ha were effective for the control of *E. vittella* infesting okra as reported by [23-25], respectively. Spinosad with a novel mode of action having ovicidal and larvicidal properties was proved excellent against *E. vittella* by [14]. [26] also reported spinosad 45 SC recorded cent per cent larval mortality of okra fruit borer, *E. vittella*.

Effect of spinetoram 12 SC on fruit yield

Data on marketable yield of okra fruits in field experiments ranged from 34.4 to 52.2 q / ha and from 27.8 to 50.5 q / ha respectively in all treatments (**Figure 1**). The maximum yield in both seasons was recorded in spinetoram 12 SC @ 54 g ai/ha (52.2 and 50.5 q/ha) followed by spinetoram 12 SC @ 45 g a.i /ha (50.1 and 48.2 q/ha) and spinetoram 12 SC @ 36 g a.i / ha (46.5 and 43.1 q/ha) respectively. These were followed by emamectin benzoate (45.5 and 40.1 q/ha), cypermethrin (45.0 and 39.2 q/ha) and quinalphos (43.3 and 38.3 q/ha), compared to untreated check which yielded only 34.4 and 27.8 q/ha fruit in first and second seasons respectively. [27, 28] reported that spraying of spinosad resulted in the highest fruit yield in brinjal compared to other insecticides which was similar to the present observations on fruit yield of okra due to application of spinetoram. In conclusion, spinetoram 12 SC was very effective against *E. vittella* with enhanced fruit yield in okra.

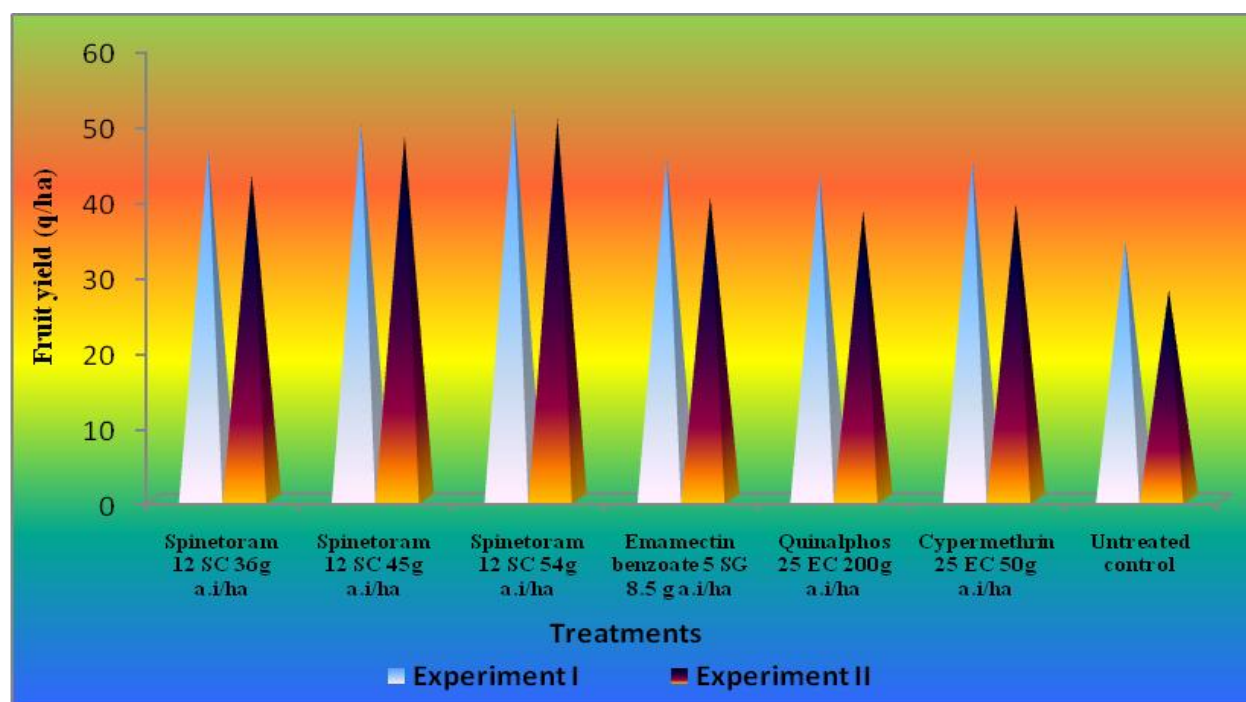


Figure 1 Effect of spinetoram 12 SC on okra fruit yield

Conclusion

Spinetoram 12 SC at 45 and 54 g a.i/ha was significantly superior and very effective against *E. vittella* larval population and shoot and fruit damage, enhanced okra fruit yield and caused no phytotoxic effects on plants. Therefore it is an effective and excellent tool to be incorporated into integrated pest management for okra crop.

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