Interactive Effect of Sublethal Concentrations of Fenvalerate and Various Microbial Insecticides to Larval Instars of *Spilarctia obliqua* (Walker)

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ABSTRACT

The interactive effect of sublethal concentrations of fenvalerate in combination with dipel and biobit applied simultaneously or sequentially to different instars of *Spilarctia obliqua* was evaluated. Mixtures of high concentration of fenvalerate with highest concentration of *Bt* resulted in significantly higher mortality than the sum of the individual components to younger instars. With decrease in the concentration of fenvalerate, there was a reduction in mortality, lower than the sum of the mortalities of individual components. In the sequential application, delay in the application of fenvalerate up to 5 days after treatment with dipel or biobit, resulted in potentiation. However, the degree of synergistic effects differed between instars and concentration of the components. In the sequence of application of fenvalerate followed by *Bt*, potentiation resulted from the combinations at 5-day lag, involving higher concentration of both the components. Between the simultanaous components and sequential combinations, the later application method resulted in a significant increase in mortality, when applied at 5-day lag against all the instars and also at 3-day lag to 4-day old larvae.

Key words : Dipel, Insecticides, *Spilarctia obliqua*

The Bihar hairy caterpillar, *Spilarctia obliqua* (Walker) is a sporadic, yet potentially destructive insect causing appreciable losses in vegetables, pulses and oilseed crops (Gupta, 1983). Wide spectrum of insecticides were evaluated by various workers against this pest (Pradhan *et al.*, 1960; Saini, 1986) and shown a significant change in the susceptibility to different insecticides over the years (Singh *et al.*, 1985; Dhingra, 1995). Such a genetically acquired resistance to commonly used insecticide has given impetus to the search for the alternative method of control and some of the pathogens have shown much promise and offer an excellent alternative to chemical pesticides (Chaudhari *et al.*, 1980). *Bacillus thuringiensis* has shown great potential as insect controlling agent, but due to its inherent limitations, such as slow rate of kill, loss of activity under sunlight, their use has been limited and, used only as supplements or in conjunction with chemical pesticides. When microbial and chemical insecticides are present on a crop, whether applied simultaneously or sequentially, the potential exists for an additive effect if no interaction occurs between the molecules of different modes of activity. However, potentiating effect of such combinations is determined by the nature of the chemical insecticide, concentration levels of both the components, and time duration between their combinations in sequential application (Benz, 1971).

Materials and Methods

Fenvalerate obtained from Searle India Ltd. was formulated from the technical grade. Insecticide emulsions were prepared by using xylene as a solvent and Triton X-100 as emulsifier, being respectively maintained at 5.0 and 0.625% levels in the final concentration. Microbial suspensions were prepared from the commercial formulations of *Bt viz.* dipel and biobit with emulsifier and water.

Field collected larvae were reared in the laboratory on castor leaves (*Ricinus communis* Linn.) in glass jars (20 x 15 cm) at 27°C and 60% RH. Disease free culture was maintained by keeping only 100 larvae in each jar, thus, preventing over crowding and unsanitary conditions. Fresh and clean leaves sterilized with 0.5% formaldehyde were provided everyday after removing the remnant leaves and excreta. The fully grown larvae were allowed to pupate in the leaves itself. The moths emerging after a fortnight were collected and transferred to clean jars containing a suspended cotton swab soaked in honey solution, and pieces of folded papers were provided for oviposition. The egg masses were surface sterilized with 0.1% sodium hypochloride. The freshly hatched larvae of the same batch were removed and kept separately on fresh and tender castor leaves in order to have continuous supply of larvae of various instars.

The concentrations of fenvalerate, dipel and biobit which caused 20, 30 and 40% mortality on all the instars of *S. litura*, when evaluated alone were calculated and used in the formulation of various mixtures. By combining these pre-determined concentrations, the following experiments were conducted.

Simultaneous treatment : The larvae of the test insects were fed mixed suspensions of *Bt* and fenvalerate.

LC_{20} of insecticide mixed with LC_{20}, LC_{30} or LC_{40} of microbial suspension

LC_{30} of insecticide mixed with LC_{20}, LC_{30} or LC_{40} of microbial suspension

LC_{40} of insecticide mixed with LC_{20}, LC_{30} or LC_{40} of microbial suspension

Sequential treatments : The test larvae were first fed with fenvalerate treated leaves followed by microbial suspension after 1, 3 or 5-days.

LC_{20} of insecticide followed by LC_{20}, LC_{30} or LC_{40} of microbial suspension

LC_{30} of insecticide followed by LC_{20}, LC_{30} or LC_{40} of microbial suspension

LC_{40} of insecticide followed by LC_{20}, LC_{30} or LC_{40} of microbial suspension

The test larvae were first fed with microbial suspension treated leaves followed with fenvalerate after 1, 3 or 5 days.

LC_{20} of the microbial suspension followed by LC_{20}, LC_{30} or LC_{40} of insecticide

LC_{30} of the microbial suspension followed by LC_{20}, LC_{30} or LC_{40} of insecticide

LC_{40} of the microbial suspension followed by LC_{20}, LC_{30} or LC_{40} of insecticide

The untreated control and the treated control for both the components treated on the same day were maintained in all experiments. Mortality counts were taken at 24 hrs interval for 2-day exposure period and subsequently mortality data was recorded for 7 days. The mortality against a particular concentration or its combination was corrected by adjusting the test mortality with control mortality using formula Abbott's (1925) formula. Probit analyses was done according to Finney (1971). The synergistic effect was calculated as suggested by Benz (1971) with slight modification in the definition of potentiation to implicate "combined mortality significantly higher than the calculated independent action with zero correlation" [$=M_{m+I} = M_m + M_I (1-M_m/100)$] (Savanurmath & Mathad, 1981).

Mm = Mortality due to microbial insecticide alone

Mi = Mortality due to insecticide alone

M_{m+i} = Mortality due to combination

To find out whether or not the difference between the independent synergism and the combined mortality was significant, Z (Standard Normal variable) - test was employed. The combined per cent mortality lower than the calculated independent action resulted in antagonism; the per cent mortality higher than the independent action, but lower than the sum of the individual effects was characterized as subadditive synergism; while the combined effect of the combination, significantly higher than the calculated

independent action, suggested potentiation, and supplemental effect, if not significant.

Results and Discussion

The data on the interactive effects of simultaneous application of sublethal concentrations of fenvalerate with dipel or biobit against different larval instars of *S. obliqua* is presented in Table 1. Against 6 and 9 days old larvae, only the combinations of LC_{40} of fenvalerate with LC_{30} of dipel gave a combined per cent mortality, significantly higher than the calculated independent

Table 1. Effect of sublethal doses of dipel and biobit with fenvalerate to 6, 9 and 11 days old larvae of *Spilarctia obliqua*

Conc (insecticides)	Cm	Mi	Mm	Mi+m	Action	Mi	Mm	Mm+1	Action
		Dipel				Biobit			
6 days old									
LC20	LC_{20}	30.45	33.33	60.00	Subadditive	30.45	27.93	46.66	Antagonism
	LC_{30}	30.45	33.33	56.66	Subadditive	30.45	31.11	46.66	Antagonism
	LC_{40}	30.45	53.33	73.33	Subadditive	30.45	41.93	53.33	Antagonism
LC30	LC_{20}	31.11	33.33	63.33	Subadditive	31.11	27.93	53.33	Subadditive
	LC_{30}	31.11	33.33	63.33	Subadditive	31.11	31.11	56.66	Subadditive
	LC_{40}	31.11	53.33	76.66	Subadditive	31.11	41.93	66.66	Subadditive
LC40	LC_{20}	43.33	33.33	76.66	Subadditive	43.33	27.93	56.66	Antagonism
	LC_{30}	43.33	33.33	90.32	Potentiation	43.33	31.11	66.66	Subadditive
	LC_{40}	43.33	53.33	100.00	Potentiation	43.33	41.93	63.33	Antagonism
9 days old									
LC20	LC_{20}	26.66	43.33	60.00	Subadditive	26.66	23.33	53.33	Subadditive
	LC_{30}	26.66	40.00	63.33	Subadditive	26.66	27.93	53.33	Subadditive
	LC_{40}	26.66	46.66	74.19	Supplemental	26.66	33.33	63.33	Supplemental
LC30	LC_{20}	33.33	43.33	62.26	Subadditive	33.33	23.33	50.00	Subadditive
	LC_{30}	33.33	40.00	66.66	Subadditive	33.33	27.93	52.91	Subadditive
	LC_{40}	33.33	46.66	76.66	Subadditive	33.33	33.33	66.66	Subadditive
LC40	LC_{20}	40.00	43.33	70.00	Subadditive	40.00	23.33	55.17	Subadditive
	LC_{30}	40.00	40.00	76.66	Subadditive	40.00	27.93	64.37	Subadditive
	LC_{40}	40.00	46.66	90.00	Potentiation	40.00	33.33	73.33	Subadditive
11 days old									
LC20	LC_{20}	33.33	23.33	53.33	Subadditive	33.33	27.93	53.33	Subadditive
	LC_{30}	33.33	36.66	46.66	Antagonism	33.33	33.33	54.37	Antagonism
	LC_{40}	33.33	46.66	66.66	Subadditive	33.33	40.00	66.66	Subadditive
LC30	LC_{20}	36.66	23.33	53.33	Subadditive	36.66	27.93	56.66	Subadditive
	LC_{30}	36.66	36.66	63.33	Subadditive	36.66	33.33	58.74	Subadditive
	LC_{40}	36.66	46.66	76.66	Subadditive	36.66	40.00	76.66	Subadditive
LC40	LC_{20}	36.66	23.33	60.00	Subadditive	36.66	27.93	60.00	Subadditive
	LC_{30}	36.66	36.66	73.33	Subadditive	36.66	33.33	66.66	Subadditive
	LC_{40}	36.66	46.66	83.87	Supplemental	36.66	40.00	80.00	Supplemental

Cm = concentration of microbial insecticide; Mi=Mortality due to insecticide; Mm=Mortality due to microbial insecticide; Mm+1=Mortality due to combination

synergism, expressing potentiation. With decrease in the concentration of fenvalerate, there was a reduction in mortality, lower than the sum of the mortalities of individual components. None of the combinations of fenvalerate and dipel against 11-days old larvae, yielded potentiation. In the simultaneous mixtures involving fenvalerate and biobit, antagonistic effect was observed in all the combinations against 6 days old larvae, with mortality being significantly lower than the calculated independent synergism. Against 9 and 11 days old larvae, none of the combinations gave a combined per cent mortality higher than the sum of the individual mortalities, as synergistic effect ranged from subadditive to antagonism.

Thus, it is clear from the results that simultaneous application of fenvalerate and dipel or biobit; potentiation resulted only from the combinatons of highest concentration of fenvalerate with medium or high concentration of dipel, especially in younger instars. Salama *et al.* (1984) reported that pyrethroids potentiated the activity of *Bt* against *Spodoptera littoralis* and opined that pyrethroids had a mild effect on sporulation process of *Bt* to other insecticides. The hypothesis that application of mixtures of *Bt* and chemical insecticides at reduced concentration would protect the crops as well as either material used alone was attributed to a synergistic effect (Jaques & Laing, 1978).

The results of sequential application of sublethal concentrations of fenvalerate and dipel or biobit at 1,3 or 5 day lag against *S. obliqua* are presented in Table 2 and 3. In the sequential application involving dipel and fenvalerate, both the components synergized each other when applied at 5 day lag, involving higher concentration (LC_{30} or LC_{40}) of one or both the components against all the instars except application of fenvalerate followed by dipel against 11 day old larvae. Sequential application of fenvalerate and biobit in either sequence at 1, 3 or 5 day lag resulted in potentiation, only when the application of second component was made at 5 day lag, especially against 6 days old larvae. However application of biobit at 1 and 3 day lag after treating with fenvalerate, also elicited potentiation. The time taken to 50 % mortality was both concentration and times dependent i.e. higher the concentration of one or both the components, lower the LT_{50}. Longer the time gaps between the application of components, higher the LT_{50}. Among the two sequences, application of dipel followed by fenvalerate at 3 and 5 day lag recorded lower LT_{50} value; while, the reverse sequence recorded lower LT_{50} value at 1 day lag, against all the instars. In contrast, the median lethal time was lower in the sequential application of fenvalerate followed by biobit at all time lags, against all the instars.

Thus, the results clearly indicate that sequential application of dipel or biobit with fenvalerate elicited potentiation, irrespective of the sequence at 5 day lag. Savanurmath and Mathad (1982) reported that post virus infection treatment of endosulfan at 7 day lag, especially in combinations with high concentrationof the virus. Chandrasekhar and Joshi (1984) found that combinations of insecticide and *Bacillus cereus* was more effective when the insecticide was applied 24 hrs before treating with *B. cereus* as compared to those combinations in which treatment of insecticides was made 24 hrs after the application of *B. cereus* against *Trichoplusia ni.* However, the effect is more pronounced in 6 days old larvae, except with dipel; while in 11 days old larvae, the sequence of *Bt* followed by fenvalerate, recorded lower LT_{50}. In the sequential combination of sublethal concentrations of fenitrothion and virus, an increase in fenitrothion concentration decreased median lethal time to larval death (Savanurmath & Mathad, 1981).

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Table 2. Effect of sublethal doses of dipel and fenvalerate in sequential combinations at 1, 3 and 5 days old larvae of *Spilarctia obliqua*

			6 days old				9 days old				11 days old			
			Dipel-Fen		Fen-Dipel		Dipel-Fen		Fen-Dipel		Dipel-Fen		Fen-Dipel	
Cm	Ci	DAT	Action	LT_{50}	Action	LT_{50}	Action	LT_{50}	Action	LT_{50}	Action	LT_{50}	Action	LT_{50}
LC20	LC_{20}	1	+	84.65	++	117.7	++	82.8	++	85.9	++	144.4	++	128.8
	LC_{20}	3	+	104.6	++	138.7	+	103.8	++	125.5	++	157.1	++	179.5
	LC_{20}	5	++	145.6	+	204.6	+++	154.8	+++	168.9	+++	181.2	++	251.7
LC20	LC_{30}	1	+	86.2	+	97.5	+	84.8	++	76.4	+	124.5	++	113.3
	LC_{30}	3	+	94.4	++	124.5	++	101.8	++	101.2	++	144.4	++	150.4
	LC_{30}	5	++	131.3	+++	151.4	+++	136.3	+++	143.3	++	178.6	+	201.0
LC20	LC_{40}	1	+	78.6	-	111.5	+	67.4	++	73.3	+	101.9	++	93.4
	LC_{40}	3	++	112.5	+++	132.7	++	87.0	+++	101.4	++	124.3	++	119.5
	LC_{40}	5	+++	119.5	+++	148.7	+++	102.2	+++	125.0	+++	159.8	+	168.4
LC30	LC_{20}	1	+	87.5	++	110.4	+	83.4	++	83.5	+	120.5	++	115.6
	LC_{20}	3	+	99.9	+++	132.2	++	102.3	++	109.4	++	154.4	++	173.8
	LC_{20}	5	++	1409.0	++	202.4	++	142.7	++	162.6	+++	178.4	+	212.6
LC30	LC_{30}	1	++	71.1	+	83.4	+	78.5	+	73.4	+	112.2	+	104.8
	LC_{30}	3	+	89.9	++	101.1	++	97.6	+	91.5	++	132.3	++	126.5
	LC_{30}	5	+++	110.5	++	143.4	+++	115.8	+++	135.4	+++	156.6	+	161.3
LC30	LC_{40}	1	+	56.3	+	53.5	+	71.1	++	55.7	+	93.3	-	91.4
	LC_{40}	3	++	89.7	+++	79.9	++	83.4	+++	87.4	++	99.6	+	121.1
	LC_{40}	5	+++	96.5	+++	114.4	+++	89.5	++	118.5	+++	138.4	++	144.7
LC40	LC_{20}	1	+	76.5	++	70.1	+	71.1	++	69.8	++	121.5	++	118.4
	LC_{20}	3	+	97.9	+++	101.4	+	112.8	++	95.8	++	111.4	++	131.1
	LC_{20}	5	+++	134.6	+	150.4	++	134.6	++	147.6	+++	174.3	+	169.7
LC40	LC_{30}	1	+	66.5	+	65.0	++	62.2	+	53.7	++	101.4	+	94.5
	LC_{30}	3	++	82.6	++	73.2	++	79.7	++	82.3	++	129.5	++	132.6
	LC_{30}	5	+++	97.4	++	119.	+++	101.5	++	107.1	++	148.3	+	164.1
LC40	LC_{40}	1	+	53.3	+++	58.8	+	59.8	+	48.0	+	83.2	+	81.8
	LC_{40}	3	+++	77.7	++	81.5	++	73.6	+++	77.6	++	95.6	+	112.6
	LC_{40}	5	+++	88.8	++	101.8	+++	93.3	++	93.8	+++	114.6	+	134.7

Cm- Concentration of the microbial insecticide; Ci- Concentration of the insecticide Mm+i-Mortality due to combination; DAT-Days after treatment; - = Antagonism; + = Subadditive; ++ = Supplemental; +++ = Potentiation

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Table 3. Effect of sublethal doses of biobit and fenvalerate in sequential application at 1, 3 and 5 days to 6, 9 and 11 days old larvae of *Spilarctia obliqua*

			6 days old				9 days old				11 days old			
			Dipel-Fen		Fen-Dipel		Dipel-Fen		Fen-Dipel		Dipel-Fen		Fen-Dipel	
Cm	Ci	DAT	Action	LT_{50}	Action	LT_{50}	Action	LT_{50}	Action	LT_{50}	Action	LT_{50}	Action	LT_{50}
LC20	LC_{20}	1	++	109.5	+++	69.9	+	123.6	++	100.2	++	143.2	+	298.8
	LC_{20}	3	++	91.0	+++	73.7	+	118.9	+	91.2	+++	128.8	++	122.9
	LC_{20}	5	+++	125.2	+++	134.9	+	135.3	+	156.9	++	201.5	+	297.1
LC20	LC_{30}	1	++	107.7	+++	72.6	++	+++	+	87.4	+	143.2	++	180.0
	LC_{30}	3	+	97.5	++	90.4	+	124.1	++	100.0	++	143.7	++	132.7
	LC_{30}	5	+++	125.1	++	139.1	++	120.3	++	109.7	++	186.,7	++	176.4
LC20	LC_{40}	1	++	106.6	+++	61.7	++	96.6	++	71.2	++	109.4	+	185.5
	LC_{40}	3	++	93.4	+++	74.9	++	126.3	++	72.6	++	129.5	++	105.5
	LC_{40}	5	+++	104.8	+++	131.6	+++	110.6	+++	122.0	+++	144.6	+++	134.9
LC30	LC_{20}	1	++	95.3	+++	70.2	+	101.9	+	79.4	++	111.7	++	161.1
	LC_{20}	3	++	86.3	+++	82.2	+	120.6	++	68.8	+++	130.8	++	114.3
	LC_{20}	5	+++	124.6	+++	118.7	++	120.3	++	146.9	++	180.4	++	188.1
LC30	LC_{30}	1	+	81.2	+++	60.3	+	89.7	++	73.0	++	106.7	++	113.7
	LC_{30}	3	++	92.6	++	69.9	+	128.7	++	90.3	+++	101.2	++	111.9
	LC_{30}	5	+++	113.4	+++	133.4	++	103.0	+	106.6	++	170.6	++	144.5
LC30	LC_{40}	1	++	84.6	+++	66.0	++	87.2	-	58.5	++	106.5	++	89.6
	LC_{40}	3	++	91.2	+++	41.8	+++	106.1	+	80.8	++	124.1	++	112.3
	LC_{40}	5	+++	105.2	+++	99.5	+++	99.9	++	97.6	+++	144.3	++	134.9
LC40	LC_{20}	1	+	70.3	+++	41.5	+	93.4	+	39.3	++	108.1	++	55.7
	LC_{20}	3	+++	83.2	+++	65.0	+	105.6	-	77.6	+++	120.5	++	75.8
	LC_{20}	5	+++	107.0	+++	70.5	+++	81.6	++	61.8	+++	118.3	++	111.5
LC40	LC_{30}	1	+	78.2	++	52.9	++	91.2	+	64.0	++	88.2	++	79.3
	LC_{30}	3	+++	82.3	++	54.0	+++	97.8	++	54.3	+++	114.2	++	66.6
	LC_{30}	5	+++	116.4	+	88.2	+++	76.3	+++	109.6	+++	132.0	+	156.1
LC40	LC_{40}	1	+	72.9	+++	54.5	+	95.1	+	41.4	+	93.0	+++	34.6
	LC_{40}	3	+++	75.0	++	27.3	+++	91.7	+	67.1	++	101.0	++	48.4
	LC_{40}	5	+++	109.5	++	111.3	+++	99.4	+++	81.4	++	149.2	+++	81.8

Cm- Concentration of the microbial insecticide; Ci- concentration of the insecticide Mm+i-Mortality due to combination; DAT-Days after treatment; - = Antagonism; + = Subadditive; ++ = Supplemental; +++ = Potentiation

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