The Andhra Agric. J 55(4):481-483, 2008

Effect of Certain Plant Oils on the Viability and persistent Toxicity of *Bacillus thuringiensis* var, *kurstaki Kurstak* against Spodoptera litura (Fab.)

G Rajesh Babu, P V Krishnayya, P Arjuna Rao and V Srinivasa Rao
Department of Entomology, Agricultural College, Bapatla - 522 101, Andhra Pradesh.

ABSTRACT

The field persistent toxicity of *Bacillus thuringiensis var*, *kurstaki Kurstak* (B t K; Delfin WG, Serotype-3a, 3b, SA 11) in combination with plant oils such as cottonseed oil (*Gossypium hirsutum L.*) neem oil (*Azadiracgata india A. Juss.*), sesamum oil (*Sesamum indicum L.*), citronella oil (*Cymbopogan winterianus Stapf.*) and karanj oil (*Pongamia glabra Vent.)* against third instar larvae of *Spodoptera litura* (Fab.) revealed at one, three and five days after treatment that B.t.k 0.2% + neem oil 5% recorded significantly the highest larval mortality (65.0 to 77.5%), whereas at seven and nine days after treatment, significantly the highest larval mortality was observed in B.t.k 0.2% + cottonseed oil 5% (40.0 and 13.3%, respectively). The combinations of B.t.k with plant oils tested for viability revealed that B.t.k. 0.2% + cottonseed oil 5% recorded the highest number of viable spores at three, five, seven and nine days after application that ranged from 16.3 x 10^4 to 35.3 x 10^4, when compared to B.t.k. combinations with neem oil 5%, citronella oil 5%, karanj oil 5% and sesamum oil 5% (7.3 to 30.0 x 10^4).

Key words : *Bacillus thuringiensis*, Persistent Toxicity, Plant Oils, Viability.

Bacillus thuringiensis Ber.(B.t), a gram-positive bacterium containing insecticidal -endotoxin, is one of the successfully exploited bioagent constitutes over 90 per cent of total biopesticides market (Chopra, 2001). B.t.is highly effective against lepidopteran pests, but with limitations like shorter field efficacy due to U V inactivation in the sunlight (Morris, 1977).

Similary, plant oil *viz.,* neem oil (*Azadirachta indica* A. Juss.), karanj oil (*Pongamia glabra* Vent.), conttonseed oil (*Gossypium hirsutum* L.), citronella oil (*Cymbopogan winterianus* Stapf.) and sesamum oil (*Sesamum indicum* L.) though proved for their insecticidal efficacy have shorter field persistence.

Preparation of insecticide formulations using certain oils as adjuvants or synergists either to improve the insecticidal efficacy, persistence or penetration is in vogue. Hence, the present laboratory study was conducted to evaluate certain plant oils on the viability and persistence toxicity of B.t. against *Spodoptera litura* (Fab.)

MATERIAL AND METHODS

The field persistent pathogenicity due to viable B.t. var. *kurstaki Kurstak* (B.t.k) spores as spray in combination with different plant oils was evaluated against third instar larvae of *S.litura* on cauliflower in the laboratory (27+ 4°C).

B.t.k. in the form of commercial formulation, Delfin WG, (Serotype-3a, 3b, SA 11) and plant oils such as cottonseed oil (*G.hirsutum*), neem oil (*A.indica*), sesamum oil (*S.indicum*), citronella oil (*C.winterianus*) and karanj oil (*P.glabra*), obtained from the local market were used.

Cauliflower leaves were brought from the treated tagged plants in field at different intervals i.e. one, three, five, seven and nine days after treatment and were fed to laboratory reared *S.litura* larvae for 24 hours and from the next day onwards untreated leaves were given as food.

The persistent pathogenicity in terms of corrected per cent larval mortality using Abbott's formula was recorded and subjected to ANOVA in completely randomized design with three replications using 20 larvae per replication. The persistent toxicity values were calculated by multiplying the average persistent pathogenicity with the period for which mortality was observed.

The field viability of B.t.k. spores was also evaluated by viable spore count method (Justin *et al.,* 1999) at one, three, five, seven and nine days after treatment in the field.

To assess the field viability of B.t.k. spores in combination with plant oils, the leaf discs (1cm diameter) were cut from the treated tagged plants with the help of sterile cork borer and the leaf discs were asecptically transferred to sterile conical flasks containing 100 ml distilled water. Theflasks were vigorously shaken for 15 minutes in the shakers and serial dilutions were made till 1x10^{-3} and pasteurized

Table 1. Field persistent pathogenicity of *Bacillus thuringiensis* var. *kurstaki* (B.t.k.) in combination with plant oils against third instar larve of *S.litura*.

Trt. No.	Treatments	Per cent larval mortality after different days after application					Persistence toxicity
		1	3	5	7	9	
T_1	B.t.k 0.2%	48.2 (44.0)^{d}	35.1 (36.3)^{d}	31.7 (34.3)^{d}	15.0 (22.6)^{c}	1.7 (4.3)^{b}	56.4
T_2	B.t.k. 0.2% + Cotton seed oil 5%	53.4 (47.0)^{c}	47.3 (43.4)^{c}	45.0 (42.1)^{c}	40.0 (39.2)^{a}	13.3 (21.3)^{a}	76.0
T_3	B.t.k 0.2% + Neem oil 5%	77.5 (61.8)^{a}	68.4 (55.8)^{a}	65.0 (53.8)^{a}	30.0 (33.3)^{b}	3.3 (8.6)^{b}	102.2
T_4	B.t.k 0.2% + Sesamum oil 5%	56.6 (48.8)^{c}	43.9 (41.5)^{c}	40.0 (37.1)^{c}	20.0 (26.5)^{c}	1.7 (4.3)^{b}	68.9
T_5	B.t.k. 0.2% + Citronella oil 5%	67.2 (55.1)^{b}	61.2 (51.4)^{b}	60.0 (50.8)^{b}	30.0 (33.3)^{b}	3.3 (8.6)^{b}	91.5
T_6	B.t.k 0.2% + Karanj oil 5%	56.8 (48.9)^{c}	45.6 (42.5)^{c}	45.0 (42.1)^{c}	20.0 (26.6)^{c}	1.7 (4.3)^{b}	71.4
T_7	Untreated check	0.0	0.0	0.0	0.0	0.0	
	F test	Sig.	Sig.	Sig.	Sig.	Sig.	
	SED	1.3	1.0	1.4	1.9	5.2	
	CD (P = 0.05)	2.7	2.1	3.0	4.1	11.2	

Sig. : Significant;
Note : Values in parentheses are angular trans formed values.
In each column values with similar alphabets do not vary significantly.

at 65^{0}C for 10 minutes.Then the spores obtained were inoculated on nutrient agar medium (Gainey *et al.*, 1956) under laminar flow chamber.

The number of viable spores were determined by counting the B.t.k. colonies developed from them on nutrient agar medium (Aneja 1993) using the formula,

No.of cells mL^{-1} = No.of colonies / (Amount plated X Dilution).

RESULTS AND DISCUSSION

The data revealed that there was decrease in mortality of S.*litura* from one day to nine days after application of B.t.k. and its combinations with plant oils (Table 1). The leaves brought from the field at one, three and five days after spraying of different treatments when fed to third instar larvae of S.*litura*, indicated that B.t.k. 0.2% + neem oil 5% was the most efficacious in resulting significantly the highest mortality of S.*litura* larvae (77.5 to 65.0%) followed by B.t.k. 0.2% + citronella oil 5% (67.2 to 60.0%).The combinations of B.t.k. 0.2% (48.2 to 31.7%). On seventh and ninth day after spraying, B.t.k. in combination with cottonseed oil recorded significantly the highest larval mortality (40.0%) were on par with each other, while the lowest mortality was recorded in B.t.k. 0.2% (48.2 to 31.7%). On seventh and ninth day after spraying, B.t.k. in combination with cottonseed oil recorded significantly the highest larval mortality (40.0 and 13.3% respectively) compared to other treatments (30.0 to 3.3%). This may be due to the better U.V. protectant activity of cottonseed oil compared to other plant oils.

The data also revealed that there was a decrease in number of viable spores of B.t.k. from one day to nine days after spraying (Table 2). The number of colonies developed on agar medium by the viable spores from the field collected leaf discs indicated that B.t.k. 0.2% + cottonseed oil 5% resutled in significantly highest number of B.t.k. viable spores 35.3 x 10^{4} to 16.3 x 10^{4} at three to nine days after application. In other treatments at seven and nine days after spraying the number of viable spores of B.t.k. were very less. This was evident from higher larval mortality in B.t.k. 0.2% + cottonseed oil 5% compared to other treatments.

Table 2.Effect of certain plant oils on the viability of *Bacillus thuringiensis* var. *kurstaki* (B.t.k.) on cauliflower.

Trt. No.	Treatments	Per cent larval mortality after different days after application				
		1	3	5	7	9
T_1	B.t.k 0.2%	40.0	26.7^{c}	18.7^{c}	13.3^{c}	6.7^{b}
T_2	B.t.k. 0.2% + Cotton seed oil 5%	40.7	35.3^{a}	27.7^{a}	22.7^{a}	16.3^{a}
T_3	B.t.k 0.2% + Neem oil 5%	41.3	30.0^{b}	22.7^{b}	17.7^{b}	7.3^{b}
T_4	B.t.k 0.2% + Sesamum oil 5%	39.7	28.3b^{c}	20.7^{b}	15.3b^{c}	7.7^{b}
T_5	B.t.k. 0.2% + Citronella oil 5%	40.7	29.3b^{c}	22.0^{b}	17.3^{b}	7.7^{b}
T_6	B.t.k 0.2% + Karanj oil 5%	39.7	27.7b^{c}	19.7^{b}	15.3^{bc}	7.3^{b}
	F test	NS	Sig.	Sig.	Sig.	Sig.
	SED	1.7	1.3	1.4	1.5	1.2
	CD (P = 0.05)	3.7	2.9	3.1	3.2	2.5

Sig. : Significant; NS : Non significant
Note : Values in parentheses are angular trans formed values.
In each column values with similar alphabets do not vary significantly.

With reference to spore viability, combination of B.t.k. with plant oils viz., neem oil, citronella oil, karanj oil, sesamum oil are on par with b.t.k. alone. That indicates the absence of antagonistic activity between B.t.k. and the plant oils, which were mixed just before their application. With special reference to neem, it was earlier reported (Sailaja and Krishnayya, 2003; Devaki and Krishnayya, 2004) that neem was antagonistic to the growth of B.t., when incubated together in the laboratory for different periods of time. However, in the present study it is proved safe for the B.t.k. spore viability and also for the independent action of B.t.k. and the plant oils, when they were mixed just before their spray in the field. But for further understanding the combination effect of plant oils with B.t.k. new explorations have to be attended.

LITERATURE CITED

Aneja K R 1993. Experiments in microbiology, plant pathology and Tissue culture. Wishwa Prakasham, New Delhi, pp: 36-44.

Chopra V L 2001. Biotechnological approaches to biological control of crop pests. Advances in biocontrol, Souvenir released in the symposium on Biocontrol based pest mangement for quality crop protection in the current millennium. July 18-19 2001, PAU, Luchiana. pp: 6-11.

Devaki K and Krishnayya P V 2004. Combination effect of three proprietary formulations of B.t. with neem against Spodoptera litura (Fab.). Indian Journal of Plant Protection 32 (2): 34-36.

Gainey P L, Thomas H L and Miller W A 1956. Basic Bacteriology Laboratory Manual, Burges Publishing Company, Minnesota, USA pp:81.

Justin C G L, Rabindra R J and Swamiappan 1999. Screening of substances as UV protectans for Bacillus thuringiensis Berliner against Plutella xylostella (L.) on cauliflower. Pest Management and Economic Zoology 7(1): 67-72.

Morris O N 1977. Long term study of the effectiveness of Bacillus thuringiensis and acephate combination against the spruce budworm, Choristoneura fumifera (Lepidoptera: Totricidae). Canadian Entomologist 109: 1238 - 1246.

Sailaja K and Krishnayya P V 2003. Efficacy of Bacillus thuringiensis var. kurstaki as influenced by neem against insect pests of cauliflower. Plant Protection Bulletin 55 (1 & 2): 27-29.

(Received on 01.03.2008 and revised on 09.06.2008)