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Short Communication

Bioassay of *Bacillus thuringiensis* var. *kurstaki* against okra shoot and fruit borer *Earias vittella* Fabricius

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Okra *(bhendi), Abelmoschus esculentus* (L.) Moench is one of the most important vegetable crop grown extensively in India along with Gujarat state throughout the year. Different insect species are known to attack okra of which shoot and fruit borer *Earias vittella* Fabricius act as major constraint in achieving potential yield (Rai *et al*., 2014; Halder *et al*., 2015). The microbial insecticide, Halt 5 % W.P. (55000 S.U./mg) is the product of bacterium, *Bacillus thuringiensis* var. *kurstaki* (serotype H 3a, 3b, 3c) can be eco-friendly alternative to eco-destabilizing chemical insecticides, against *Earias vittella* Fab. a noxious lepidopteran pest infesting okra all over India.

The bioassay of the *Bacillus thuringiensis* var.*kurstaki* (*B.t.k.)* against five day old larvae of okra shoot and fruit borer (*E. vittella* Fab.) was carried out in the laboratory at Department of Entomology, College of Agriculture, JAU, Junagadh (India). The larvae of the insect was mass reared using the technique developed by Patel and Patel (1986). The insect larvae were reared on artificial diet developed by Thamankar *et al.,* (1992). Five day old larvae from second generation used for the bioassay. In this bioassay the poisoned food technique developed by Navon *et al.,* (1990) was applied. The commercial formulation trade name Halt 5 % WP (*B.t.k*. 55000 S.U./mg) was used. A series of seven concentrations of the product viz., 25, 50, 100, 200, 400, 800 and 1600 µg/ml of distilled water were prepared as the stock solution. Bioassay trays of size (13cm L X 8.5 cm B X 2.3 cm H) having 24 cavity was used. Each cavity of 1.7 cm diameter and 1.5 cm depth were filled with the artificial diet and 1 ml suspension from the series of concentrations was poured in the artificial diet and dried excess of liquid at room temperature. The uniform aged larvae of five day old previously starved for 12 hr were released individually and under each replication 10 larvae were tested for all the test dose in three repetitions. The bioassay tray were covered with plastic transparent stickers having micro pores to prevent the escape of larvae from the wells. Trays were then kept at 28 $\pm$ 2 °C for 7 days in BOD incubator. After consumption of treated food, larvae were provided with fresh pods of okra during subsequent rearing. Observations on larval mortality (dead and moridund) were recorded at 24 hrs interval up to 7 days.

The potency of *B.t.k.* (serotype H 3a, 3b, 3c) traded as Halt 5 % WP having $5X10^7$ spore/mg was measured by means of mortality which it produced in 5 day old (uniform aged) larvae of *E. vittella* using the artificial food contamination bioassay technique.

The data given in Table: 1 revealed that the mortality of the insect larvae was not induced by the different concentrations of *B.t.k.* used within 24 hr (1day) after ingestion of the contaminated food. The larval mortality was initiated on 2nd day and it was significantly highest (63.95%) in highest concentration (1600 µg/ml). While minimum mortality (9.54%) was recorded in the lower concentration (25 µg/ml). The mortality of the insect larvae was increased very fast on 3rd day and reached at maximum on 4th day and there after it was not increased. The highest 99.73% mortality was caused by concentration 1600 µg/ml. Further it was found that there was no significant difference in the mortality induced by concentration 1600 and 800 µg/ml. It means the potency of *B.t.k.* was maximum at concentration of 800 µg/ml. The bioassay conducted by Navon *et al.,*

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Table 1. Bioassay of *Bacillus thuringiensis* var. *kurstaki* against five day old larvae of *E. vittella*

Sr no	Dose (µg/ml)	Larval mortality (%) day after treatment				
		1st day (24hr)	2nd day (48hr)	3rd day (72hr)	4th day (96hr)	5th day (120hr)
1	25	0.85*	18.00	25.00	38.15	38.15
		(0.02)	(9.54)	(17.86)	(38.16)	(38.16)
2	50	0.85	19.31 (10.93)	32.05	44.30	44.30
		(0.02)		(28.16)	(48.78)	(48.78)
3	100	0.85	20.20	38.20	57.00	57.00
		(0.02)	(11.92)	(38.24)	(70.34)	(70.34)
4	200	0.85	32.20	57.15	62.25	62.25
		(0.02)	(28.55)	(70.58)	(78.52)	(78.52)
5	400	0.85	35.40	57.15	70.13	70.13
		(0.02)	(33.56)	(70.58)	(88.45)	(88.45)
6	800	0.85	45.0	65.81	83.40	83.40
		(0.02)	(50.0)	(83.21)	(98.68)	(98.68)
7	1600	0.85	52.10	75.95	87.00	87.00
		(0.02)	(63.95)	(94.11)	(99.73)	(99.73)
8	Control	0.85	0.85	0.85	0.85	0.85
		(0.02)	(0.02)	(0.02)	(0.02)	(0.02)
	SEM ±	0.001	1.17	2.0	1.70	1.70
	CD at 5 %	0.02	3.36	5.76	4.89	4.89
	CV %	0.01	8.50	9.25	6.20	6.20

*Are sin transformation

Figure in parenthesis are the retransformed value

(1990) with the strain of *B.t.k.* HD-263 (3a,3b) against neonate larvae of *E. insulana* on artificial diet gave 100 % mortality with LC_{50} 0.18 µg/ml of diet. The larval mortality of 88.45 %, 78.52 % and 70.34 % induced due to concentration of 400, 200 and 100 µg/ml respectively. The larval mortality decreased with decreased concentration of the *B.t.k.* and it was increased with increased time. The Probit analysis (Table 2) indicated that the LC_{50} was 46.67 µg/ml at about 96 hr after ingestion of the diet. Shalaby *et al.,* (1986), conducted bioassay with five concentrations of *B.t.k.* viz., 80, 160, 240, 320 and 400 IU/ml incorporated in the artificial diet of 1st instar larvae of pink boll worm resulted in 76.80% mortality on 48 hr of treatment with LD_{50} value 11.29 IU/ml larvae.

Table 2. Probit analysis of *Bacillus thuringiensis* var. *kurstaki* against five day old larvae *Earias vittella*

Name	Chi^2	Slope	LC_{50} (µg/ml)	Fiducial limit (95%)		Regression equation
				lower	upper	
Halt 5 % WP (*B.t.k.* 55000 s.u./mg)	2.45	1.40	46.671	31.691	65.871	Y=2.66+1.40x

* Table value of Chi^2 (P (0.05)=11.1)

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