



## Evaluation and validation of HearSNPV for fruit borer, *Helicoverpa armigera* (Noctuidae: Lepidoptera) management on tomato

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### ABSTRACT

Larvae of the tomato fruit borer, *Helicoverpa armigera* (Hübner) are susceptible to nucleopolyhedrovirus (NPV). Based on mortality-time, HearSNPV of 3<sup>rd</sup> instar *H. armigera* larvae at  $5 \times 10^4$  OBs/ml was found optimum for larval kill. This is when lab data were subjected to probit analysis and Kaplan-Meier survival estimates with Cox's regression analysis. Based on number of days for 50% larval mortality, volume of HearSNPV required and virulence, 0.0097 OBs/mm<sup>2</sup> is recommended against 3<sup>rd</sup> instar larvae. Early (1<sup>st</sup>, 2<sup>nd</sup> and 3<sup>rd</sup>) larval instars were relatively more susceptible than late (4<sup>th</sup>, 5<sup>th</sup> and 6<sup>th</sup>). The HearSNPV dose when validated in large tomato fields (2 ha). Based on number of surviving *H. armigera* larvae and tomato fruit infestation, 2–3 applications at  $2.47 \times 10^{11}$  OBs/ha (500 litres of water/ha) at 15-d intervals from blooming stage (45-d) were effective. The HearSNPV applications proved as effective as recommended insecticides, indoxacarb, 14.5 SC (Avant ®) at 1 ml/litre and endosulfan 35 EC (Endocil®) at 2 ml/litre of water. Hear SNPV also proved economical and compatible with bioagents namely entomopathogenic fungus *Metarhizium anisopliae* (Metchnikoff), parasitoid, *Campoletus chloridae* (Uchida) and trap cropping with marigold, (*Tagetes patula* L.) when tested at 3 locations in 1 ha fields. The benefit: cost (B: C) ratio (2.58: 1) was better in HNP compared to FPP (2.28:1). These results led to recommending HearSNPV against the fruit borer for sustainable tomato yields.

**Key words:** HearSNPV, *Helicoverpa armigera*, IPM, Tomato, Validation

The fruit borer, *Helicoverpa armigera* (Hübner) (Lepidoptera: Noctuidae) is a polyphagous pest inflicting annual crop damages in India worth US \$ 1 billion. The problem of this pest is due to direct attack on fruiting structures, voracious feeding habit, high mobility and fecundity. In addition, the pest undergoes facultative diapause, has nocturnal behavior, long distance migration, host selection is by learning and exhibits genetic variability on different hosts (Cunningham *et al.* 1999, Subramanian and Mohankumar 2006). *H. armigera* has developed resistance to varying folds to almost all insecticides (Arms *et al.* 1996, Yaqoob *et al.* 2006).

In this context, the insect viruses are of immense utility (Ignoffo and Couch 1981). Owing to their host specificity and high virulence to susceptible insect hosts, baculoviruses

are one of the most efficient biocontrol agents for insects. Nucleopolyhedroviruses (NPVs) are the most suitable and effective pest control agents. This is due to infectivity, lethality, storage stability and environmental safety (Cunningham *et al.* 1999). They are particularly attractive as bio-insecticides for two reasons: safety for vertebrates and other non-target fauna and high pathogenicity.

Though the effectiveness of *H. armigera* single embedded NPV (HearSNPV) has been established, it is sparingly used in Asia. One of the main reasons for under-utilization of HearSNPV is confusion over correct treatment concentration as commercial companies recommend  $2.47 \times 10^{11}$  OBs/ha while Research Institutes,  $14.82 \times 10^{11}$  OBs/ha (Moorthy *et al.* 2003). Farmers are uncertain about the effective dose. Also, baculoviruses typically involve slow speed of kill, inconsistent results and generally affected larvae are difficult to easily trace in the field. NPV spray applications are not systematically applied targeting early instar larvae during early morning or evening. Often with NPV suspension, jaggery (sugar cake made from sugarcane) and boric acid powder or teepol (liquid detergent soap) although recommended are not added. Jaggery acts as a phagostimulant and enhances palatability of NPV to larvae,

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and boric powder serves as an antioxidant, protecting NPV from UV light. Generally jaggery constitutes 1% and boric acid powder 1.5% by volume in the HearSNPV. The NPV recommended doses for application under greenhouse or field conditions vary. Information on compatibility of HearSNPV with recommended insecticides - for example, on tomato - is lacking (Kambrakar *et al.* 2007). The combined effect of HearSNPV with other Integrated Pest Management (IPM) tools remains virtually unstudied. So the goals of this study were to determine effective dose in laboratory and validate it under field conditions.

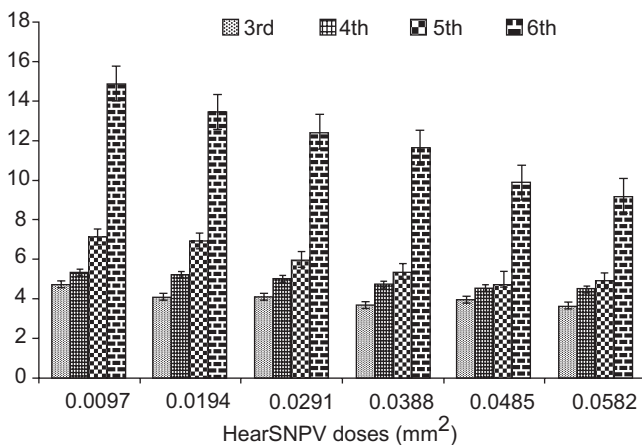
## MATERIALS AND METHODS

### Virus and insects

Larvae of *H. armigera* collected from malvaceous vegetable okra, *Abelmoschus esculentus* (L) were mass reared in the laboratory (at 25±1°C, 65% RH and 12:12 h L: D) on chickpea (*Cicer arietinum* L.) based artificial diet (Gupta *et al.* 2004). A stock culture of NPV was maintained in the laboratory on fifth instar *H. armigera* larvae (Gopali and Lingappa 2001). A *H. armigera* NPV (HearSNPV) isolated, characterized and identified by Bio-Control Research Laboratory (BCRL), Bangalore as HaSSR-2 strain was utilized for the study. This strain of HearSNPV is of the single embedded nucleocapsid type (Analytical Report 2000).

### Laboratory evaluation

Freshly prepared HearSNPV formulation from BCRL was used to determine LT<sub>50</sub> in the laboratory at six concentrations (Fig.1) on 5<sup>th</sup>, 7<sup>th</sup> and 10<sup>th</sup> d after treatment, with a control (Table 1). Virus suspension was distributed uniformly over the entire diet surface by a polished blunt end of a glass rod (6 mm). Before fixing the doses, bracketing



Histograms indicate 50% larval mortality on different days. Vertical bars at tip of each histogram are Mean± standard errors.

Fig 1 Probit analysis of time-mortality response of HearSNPV from BCRL to different instars of *H. armigera* larvae

Table 1 Efficacy of BCRL HearSNPV on *H. armigera* larvae in laboratory on 5<sup>th</sup> day

Treatment (OBS/mm <sup>2</sup> )	% larval mortality of instars			
	3 <sup>rd</sup>	4 <sup>th</sup>	5 <sup>th</sup>	6 <sup>th</sup>
0.0097	56.66 (48.83) <sup>d</sup>	46.11 (33.18) <sup>c</sup>	30.00 (42.76) <sup>d</sup>	10.00 (18.37) <sup>d</sup>
0.0194	70.66 (57.20) <sup>c</sup>	63.33 (52.73) <sup>cd</sup>	30.00 (33.20) <sup>c</sup>	13.33 (21.40) <sup>c</sup>
0.0291	80.00 (63.47) <sup>bc</sup>	70.00 (56.85) <sup>bc</sup>	32.33 (34.97) <sup>c</sup>	20.00 (26.55) <sup>b</sup>
0.0388	83.33 (66.05) <sup>b</sup>	76.66 (61.12) <sup>bc</sup>	50.00 (44.99) <sup>b</sup>	26.66 (31.06) <sup>a</sup>
0.0485	89.99 (71.59) <sup>a</sup>	83.33 (66.05) <sup>b</sup>	60.00 (50.77) <sup>a</sup>	31.33 (34.03) <sup>a</sup>
0.0582	89.99 (71.59) <sup>a</sup>	89.99 (71.59) <sup>a</sup>	56.66 (48.82) <sup>a</sup>	33.33 (35.27) <sup>a</sup>
Control	0.0 (0.062) <sup>e</sup>	0.0 (0.62) <sup>e</sup>	0.0 (0.062) <sup>d</sup>	0.0 (0.62) <sup>e</sup>
SEm	0.93	0.66	0.50	0.44
CD@1%	6.04	4.31	3.25	2.84

Mean of 10 larvae/treatment/replication, Figures in the parentheses are Angular transformed values, Means followed by same letters in each column are not significantly different by LSD at 1%

was done to ensure that the doses selected followed the time-mortality response curve. Bracketing is a preliminary study to find out the dosage range of an insecticide giving different levels of mortality (5-95%) before conducting the bioassay (Finney 1952). Third, fourth, fifth and sixth instar larvae of uniform age and size were released on to the diet 20 min after surface treatment. Each concentration had 30 larvae (10 larvae × 3 replicates). After inoculation, larvae were incubated at 25±1°C in a laboratory incubator. Observations on larval settlements on the diet were checked from 1-d and larval mortality was recorded from 3-d through 10-d at 24 hr intervals. For determining the efficacy of the virus, time-mortality relationships were considered.

### Field efficacy

Field experiments on HearSNPV were conducted in the organic farming villages at the outskirts of Bangalore North (12° 58' N, 77° 38' E 980 AMSL), Karnataka, South India. The vegetable growers are recognized and are adopted by the Association for Promotion of Organic Farming (APOP) and the same villages have also been adopted by the BCRL, Sriramanahalli. There is a memorandum of understanding among vegetable growers, APOP and BCRL to cultivate vegetables following organic farming practices. To validate the effective dose of HearSNPV in large scale and test combination of IPM tools with HearSNPV, field trials were also conducted in neighboring group of villages where farmers do not adopt organic farming practices. Laboratory studies were conducted at Department of Agricultural Entomology,

University of Agricultural Sciences, GKVK and BCRL, Bangalore. All studies were conducted from Jan 2007 to Jun 2009.

The efficacy of the formulated HearSNPV was tested in a farmer's tomato field (SG 687-Namdhari hybrid) in Rajankunte, at the outskirts of Bangalore during Oct-Dec 2008. The small plot trial was laid out in a randomized block design with four replications. The plot size was 4 × 5 m and a gangway of 1 m was allowed all around the plots. Spacing of the plants on a row was 50 cm, whereas the gap between the rows was 1 m. Recommended agronomic practices were followed (Package of Practices 2007). Three rounds of sprays were given at 15-d intervals during evening hours, commencing the first 45-d after sowing when the plants were at flowering stage and the incidence of early instar larvae of the pest was observed. The treatments studied in these experiments were 2.47 × 10<sup>11</sup> occlusion bodies (OBs)/ha + jaggery (1%) + teepol (1.5%) at 1 ml HearSNPV, 4.94 × 10<sup>11</sup> OBs/ha + jaggery + teepol at 2 ml HearSNPV, 7.41 × 10<sup>11</sup> OBs/ha + jaggery + teepol at 3 ml HearSNPV. Insecticides, viz. endosulfan, 35% EC (Endocil®) at 2 ml/liter of water, indoxacarb 14.5 SC (Avant®) at 1 ml/liter of water and Control / untreated. In the spray solution, jaggery constituted 1% and teepol 1.5% by volume. Pre-treatment count was observed at day zero of each spraying on the number of larvae /10 randomly-selected plants and percent incidence of damaged fruits from ten plants/treatment. Post-treatment observations of the same variables were made at 5, 7 and 10-d after each application with same sample size and replication.

#### Validation of HearSNPV dose

The effective concentration of HearSNPV from the small-plot field trial was validated in a large 2-ha farmer's tomato (SG687-Namdhari hybrid) field in Rajanukunte during Jan to Mar 2009, with a 0.25-ha control plot- 0.8 km away where no pesticides or HearSNPV were applied. The HearSNPV from BCRL was applied to the tomato crop by knapsack sprayer thrice at 15-d intervals commencing from flowering (45-d-old) at 2.47 × 10<sup>11</sup> OBs/ha. The larval numbers and infested fruits were counted before and also 5 and 7-d after application of viral suspensions (Zahid *et al.* 2008).

#### Comparison between HearSNPV plot and farmer's practices plot

To compare farmer's practices with HearSNPV, tomato fields (SG 687-Namdhari hybrid) in Rajankunte, Devanahalli (13° 13' 57" N and 77° 41' 57" E) and Doddaballapura (N 12° 58' 35" and E 77° 36' 0") villages at the outskirts (40 km) of Bangalore were selected and observations recorded during Apr-Jun 2009. The experiment was conducted in 2-ha plots at each of the three aforementioned locations. Each treatment was applied to 1-ha. In HearSNPV sprayed plot (HNP) three rounds of HearSNPV at 2.47 × 10<sup>11</sup> OBs/ha with jaggery and teepol were applied at 15-d intervals during evening when a

threshold level of 1 larva/plant was observed.

In farmer's practices plot (FPP) indoxacarb 14.5 SC (Avant®) at 1ml/lit of water, endosulfan (Endocil®) at 2 ml/lit of water and Astra at 2 ml/lit of water were applied sequentially. At the later stage very negligible numbers of leaf miner (*Liriomyza trifolii* Burgess) were found in HNP and FPP plots. The crop in both the plots was also infected by late blight caused by *Phytophthora infestans* which was effectively controlled by spraying Curzite at 3 g/l of water. In both the plots first application commenced when the crop was in flowering stage, i.e. 45-d-old plants.

Pre-treatment counts observed at zero d of each spraying were on the number of larvae/10 randomly-selected plants and % incidence of damaged fruits on 10 plants. Post-treatment counts were observed at 5, 7 and 10-d after each application in each quadrat on number of infested fruits and *H. armigera* larvae on 10 plants/5 m<sup>2</sup> × quadrat 4.

The agronomic practices for both HearSNPV sprayed and farmer's practices plots were recorded and yield at harvest (tonnes) from each plot (only marketable fruits are considered as yield). The benefit: cost (B: C) ratio was also determined.

To determine if HearSNPV and farmer's practices are compatible with IPM tools, observations on the numbers and activities of fruit borer natural enemies were recorded in both the plots. Two rows of marigold (*Tagetes patula* L.) (two whorls of orange petals) as border rows were maintained along the borders in both the plots. The plants bloomed 12-d before tomato crop flowered (Srinivasan *et al.* 1994).

#### Statistical analysis

Analyses of variance (ANOVA) were carried out using Statistical Package for Social Sciences (Spss, Inc., 1999 version 10.00) and means were separated by Least Significance Difference (LSD). The Kaplan-Meier survival function was determined and survival curves compared using Cox's proportional Hazards Model (Hosmer and Lemeshow 1999). All percentage data were transformed to angular values (Angular transformation was effected to linearize sigmoid distributions and equalize variances of proportions) before analysis. The larval counts were also transformed to  $\sqrt{x+0.5}$  values. The Probit analyses (Finney 1952) to determine LT<sub>50</sub> values were also carried out in a SPSS. Chi-square was performed as goodness of fit test and computed means from HearSNPV sprayed and farmer's practices fields were subjected to the paired t-test to determine whether the number of *H. armigera* larvae and percent fruit infestation differed statistically.

## RESULTS AND DISCUSSION

#### Laboratory evaluation

The 50% mortality of 3<sup>rd</sup> instar *H. armigera* larvae was obtained on 4.75-d at 0.0097 OBs / mm<sup>2</sup>, for 4<sup>th</sup> instar, it was obtained on 5.35-d. However, for 5<sup>th</sup> and 6<sup>th</sup> instars, 7.16 and

14.9-d, respectively were required. At the highest concentration of 0.0582 OBs / mm<sup>2</sup>, 4.93 and 9.22-d were required to get 50% larval mortality (Fig 1). As the age of the *H.armigera* larvae increased, the days required for 50% kill also increased. Between the lowest and the highest concentration of HearSNPV tested, the time required for 50% kill of 3<sup>rd</sup> instar was within 5-d.

Kaplan Meier estimates of the survival function of 3<sup>rd</sup> instar *H. armigera* larvae showed less than five days, i e least survival with HearSNPV from the BCRL at 5×10<sup>4</sup>OBs/ml. Paired comparison between different concentrations of BCRL source using Cox's Regression Model showed that there were significant differences between concentrations of HearSNPV. These data analyses (not included here) revealed that HearSNPV at 5 × 10<sup>4</sup> OBs/ml proved the best against 3<sup>rd</sup> instar *H. armigera* larvae (Doddabasappa 2009).

Though data on larval mortality was recorded 5, 7 and 10-d after treatment, data pertaining to 5-d alone is presented in Table 1. At the same concentration, for the 4<sup>th</sup>, 5<sup>th</sup> and 6<sup>th</sup> instars, <50% larval mortality was obtained. At higher concentration of 0.0194 OBs /mm<sup>2</sup>, >50% larval kill for 3<sup>rd</sup> (70.66%) and 4<sup>th</sup> (63.33%) instars were obtained. The >50% larval kill of 5<sup>th</sup> instars was obtained at higher concentrations of 0.0388 OBs/mm<sup>2</sup>. As the age of *H. armigera* larvae increased, the concentration to get >50% larval kill within 5-d also increased. At 0.0582 OBs/mm<sup>2</sup> concentration maximum mortality (90%) of 3<sup>rd</sup> and 4<sup>th</sup> instars and 56.66 and 33.33%

of 5<sup>th</sup> and 6<sup>th</sup> instar larvae, respectively were obtained. Based on 50% larval kill HearSNPV at 0.0097 OBs/mm<sup>2</sup> proved the best against 3<sup>rd</sup> instar larvae only from an economic stand point. From the stand point of performance 0.0582 OBs/mm<sup>2</sup> proved ideal.

A comparison of LT<sub>50</sub> values at six different doses revealed that HearSNPV at 0.0097 OBs/ mm<sup>2</sup> was effective to get 50% kill of 3<sup>rd</sup> instar larvae within 5-d, although LT<sub>50</sub> at 3.65-d was obtained with a concentration of HearSNPV at 0.0582 OBs/ mm<sup>2</sup>, i e with a six-folds increase in the viral spray suspension. Therefore it is advisable to recommend HearSNPV at 0.0097 OBs/ mm<sup>2</sup> against 3<sup>rd</sup> instar *H. armigera* larvae. The larval kill is achieved at the lowest dose of HearSNPV.

#### Field efficacy

##### Number of fruit borer larvae

Data indicated that HearSNPV at the three concentrations tested required two applications to maintain <1 larva /plant (<EIL). It was observed that one application of indoxacarb 14.5 SC (Avant®) or endosulfan (Endocil®) was adequate in protecting the tomato crop (Table 2). Indoxacarb proved superior over other chemicals as there were statistically significant (P<0.05 by LSD) differences in the number of surviving *H. armigera* larvae 5-d after application (data of 5-d and 7-d not given in Table 2). However, after two weeks, a fresh infestation of *H.armigera* was noticed. So, second

Table 2 Field efficacy of HearSNPV against fruit borer on tomato in small plots

Treatment (OBs/ha)	Mean borer larvae						% infested fruits					
	1 <sup>st</sup> spray		2 <sup>nd</sup> spray		3 <sup>rd</sup> spray		1 <sup>st</sup> spray		2 <sup>nd</sup> spray		3 <sup>rd</sup> spray	
	Pretreat- ment	10 <sup>th</sup> day	Pretreat- ment	10 <sup>th</sup> day	Pretreat- ment	10 <sup>th</sup> day	Pretreat- ment	10 <sup>th</sup> day	Pretreat- ment	10 <sup>th</sup> day	Pretreat- ment	10 <sup>th</sup> day
2.47 × 10 <sup>11</sup>	1.57 (1.42)	0.9 (1.17) <sup>ab</sup>	0.84 (1.15)	0.47 (0.98) <sup>b</sup>	0.42 (0.95) <sup>b</sup>	0.3 (0.88) <sup>b</sup>	20.92 (26.60)	13.27 (21.3) <sup>b</sup>	11.3 (19.3) <sup>a</sup>	8.00 (16.4) <sup>b</sup>	7.1 (15.32) <sup>b</sup>	1.44 (6.66) <sup>a</sup>
4.94 × 10 <sup>11</sup>	1.35 (1.34)	1.02 (1.22) <sup>ab</sup>	0.69 (1.08) <sup>bc</sup>	0.45 (0.96) <sup>b</sup>	0.32 (0.9) <sup>ab</sup>	0.2 (0.82) <sup>ab</sup>	15.34 (22.90)	11.26 (19.4) <sup>ab</sup>	10.36 (18.75) <sup>a</sup>	7.01 (15.1) <sup>ab</sup>	6.00 (14.0) <sup>ab</sup>	0.86 (5.2) <sup>a</sup>
7.41 × 10 <sup>11</sup>	1.47 (1.4)	1.0 (1.21) <sup>ab</sup>	0.42 (0.94) <sup>ab</sup>	0.15 (0.80) <sup>a</sup>	0.12 (0.78) <sup>ab</sup>	0 (0.7) <sup>a</sup>	19.35 (26.08)	10.15 (18.3) <sup>ab</sup>	9.87 (18.27) <sup>a</sup>	6.61 (14.8) <sup>ab</sup>	4.58 (11.9) <sup>ab</sup>	0.52 (3.65) <sup>a</sup>
Endosulfan (Endocil®) 35 EC at 2ml/liter	1.42 (1.37)	0.67 (1.08) <sup>a</sup>	0.37 (0.93) <sup>a</sup>	0.22 (0.84) <sup>a</sup>	0.1 (0.76) <sup>ab</sup>	0 (0.7) <sup>a</sup>	23.88 (28.74)	9.72 (17.9) <sup>ab</sup>	8.79 (17.15) <sup>a</sup>	5.24 (12.46) <sup>a</sup>	3.33 (10.19) <sup>a</sup>	0.32 (2.56) <sup>a</sup>
Indoxacarb, (Avant) 14.5 SC at 1ml/liter	1.62 (1.45)	0.72 (1.10) <sup>a</sup>	0.35 (0.91) <sup>a</sup>	0.05 (0.73) <sup>a</sup>	0 (0.7) <sup>a</sup>	0 (0.7) <sup>a</sup>	19.47 (26.16)	8.54 (16.9) <sup>a</sup>	6.82 (15.13) <sup>a</sup>	4.92 (12.75) <sup>a</sup>	4.15 (11.53) <sup>a</sup>	0.13 (1.01) <sup>a</sup>
Control	1.45 (1.39)	1.57 (1.42) <sup>c</sup>	1.40 (1.37) <sup>d</sup>	1.72 (1.48) <sup>c</sup>	1.87 (1.53) <sup>c</sup>	2.50 (1.72) <sup>c</sup>	22.97 (28.36)	16.82 (24.2) <sup>c</sup>	17.95 (24.83) <sup>b</sup>	18.35 (25.17) <sup>c</sup>	18.74 (25.63) <sup>c</sup>	21.25 (27.35) <sup>b</sup>
SEm ±		0.08	0.04	0.03	0.04	0.05		1.07	1.39	0.68	1.16	0.73
CD @5%	NS	0.24	0.14	0.10	0.13	0.15	NS	3.24	4.21	2.05	3.50	2.21

Mean of 10 plants/treatment/replication, Figure in the parentheses are Angular transformed values, Means followed by same letters in each column are not significantly different by LSD at 5%

application of insecticides was required to maintain to <1 larva /plant (<EIL).

#### Fruit infestation

Based on fruits infestation, indoxacarb 14.5 SC (Avant ®) at 1 ml/lit of water realized the best results with 12.87 % fruit infestation 5-d after first application (data of 5 and 7-d not given). The trend in the efficacy of chemicals during second spray, however, was different. The efficacy of HearSNPV at  $2.47 \times 10^{11}$  OBs/ha was not significantly ( $P < 0.01$  by LSD) different from indoxacarb (Avant ®) 5-d after second application.

As found in the present study Rabindra and Subramanian (1974) also reported differential instar susceptibility of *H. armigera* to HearSNPV. Several workers have evaluated under field conditions insecticides and HearSNPV on different crops or on different varieties of the same crop, with varying results (Singh *et al.* 2002, Rabindra and Jayaraj 1995).

Gupta *et al.* (2007) assessed effectiveness of the most virulent isolate (Samba) on tomato and chickpea. Application of HearSNPV alone at  $3.0 \times 10^{12}$  OBs/ha in tomato and  $1.5 \times 10^{12}$  OBs/ha in chickpea resulted in significant suppression of the pest. HearSNPV was found compatible with the recommended insecticide endosulfan, *Bacillus thuringiensis* and egg parasitoid, *Trichogramma pretiosum* (Riley) in combined as well as sequential applications.

#### Validation of HearSNPV

HearSNPV gave 96% suppression of *H. armigera* population, recording 0.1% fruit damage after 3 applications. In control plot, 46% fruit damage with a mean of 2.50 larvae/plant (n=150 plants) was recorded. In HearSNPV plot, *H. armigera* larvae were not noticeable.

HearSNPV at  $2.47 \times 10^{11}$  OBs/ha after two applications at 15-d intervals effectively suppressed *H. armigera* population. The larval numbers/plant at the end reached less than 1 larva/ plant, i.e. below EIL. There were statistical differences ( $t=1.62$ ,  $P > 0.05$  by t test) between two observations. In the control plot on an average 2.8 larvae/plant and 26% fruit infestation (mean of 10 plants/5 m<sup>2</sup> quadrat  $\times$  4) was recorded.

In the present study, repeated HearSNPV applications proved cheaper than insecticidal applications. Mandal *et al.* (2003) reported the highest B: C ratio in monocrotophos application against *H. armigera* on chickpea and pigeonpea (Rao and Reddy 2003). Moorthy *et al.* (2003) tested IPM module using marigold as trap crop and three sprays of HearSNPV at 250 LE/ha given at weekly intervals (starting from the flowering stage) in 17 tomato fields around Bangalore, Karnataka, during 2001-02. IPM significantly reduced the insecticide sprays. In the test plots the larval density on an average varied from 0-0.3 larvae /10 plants. Thus, the large field test validated the small plot field trial. The data supported that HearSNPV be directed at early instar

*H. armigera* (2<sup>nd</sup> or 3<sup>rd</sup>) larvae at  $2.47 \times 10^{11}$  OBs/ha. The HearSNPV applications are to be repeated at 10-15 d intervals in fruiting season.

Results suggested that two or more applications of either HearSNPV or in combination with insecticides like endosulfan (Endocil®) or indoxacarb 14.5 SC (Avant ®) are required to bring down the *H. armigera* larval numbers below EIL (i.e. 1 larva/plant). Field trials also indicated that 2-3 applications of HearSNPV at  $2.47 \times 10^{11}$  OBs/ha proved effective both based on number of surviving larvae and fruit infestation.

#### Comparison between insecticides and IPM tools

The mean numbers of surviving larvae were less in FPP (1.63 larvae/10 plants) after three insecticide applications compared to that in HNP plots (2.25 larvae/10 plants) ( $t=0.41$ ;  $df=9$ ;  $P > 0.05$ ) (Table 3). The mean percent fruit infestation was less (7.02 /10 plants) in FPP compared to 8.60/10 plants in HNP ( $t=1.13$ ;  $df=9$ ;  $P > 0.05$ ). The natural enemies, viz. *C. chloridae* and *M. anisopliae* caused, on an average, 9% larval mortality in HNP. In FPP, where insecticides were applied, the natural enemies of *H. armigera* larvae were not found. In HNP throughout the blooming period of tomato *H. armigera* eggs were found on marigold. This suggested that application of HearSNPV did not deter moths to lay eggs on marigold. However, insecticides deterred *H. armigera* moths to lay eggs on marigold in FPP.

The costs incurred for plant protection in HNP amounted to ₹ 15 937 (1£ = ₹ 80 and 1US\$ = ₹ 47) and in the FPP ₹ 71 250 (22.36% higher). The total numbers of sprays were more in FPP compared to HNP. Based on economics the FPP plot was on par with HNP (Table 3). When percent fruit infestation, mean borer larvae/10 plants and yields were compared with costs, there were statistically significant differences in the crop yields ( $t=7.40$ ;  $df=12$ ;  $P < 0.05$ ) in HNP and FPP. However, the benefit cost ratio worked out to be 2.58 in the HearSNPV sprayed plot compared to the 2.28 in farmer practice plot (Table 3).

Application of HearSNPV is recommended because in HearSNPV treated plot larvae observed were infected with either green muscardine fungus, *M. anisopliae* (2-11%, average =6%) and in addition 1-4% (average=3%) showed parasitization mainly by *Campoplex chloridae* (Uchida). More importantly as the trials were conducted in organic farming villages HearSNPV is compatible with organic farming practices. Around Bangalore, usually farmers apply insecticides 9-12 times /tomato season (= 4 months), incurring the cost of insecticides and labor that worked out to be much higher than 3 HearSNPV applications. Whereas, in HearSNPV sprayed plot, with the other components of pest suppression like natural enemies, less cost was incurred. The economics of treatments were also calculated by earlier workers with respect to HearSNPV (Moorthy *et al.* 2003). The most favourable B: C ratio was obtained from plots treated with

Table 3 Field efficacy of HearSNPV in comparison with Farmer's practices and IPM tools (Mean of plots from three locations)

Days after spraying		Mean borer larvae/10 plants	
		HearSNP sprayed plot (HNP) <sup>f</sup>	Farmer's practice plot (FPP) <sup>g</sup>
1 <sup>st</sup> spray	Pretreatment	4	3.8
	7th day	1	1
2 <sup>nd</sup> spray	Pretreatment	3.5	2
	7th day	0.5	0.5
3 <sup>rd</sup> spray	Pretreatment	3.5	2.5
	7th day	1	0
	Mean	2.25	1.63
		Mean infested fruits (%) 10 plants	
1 <sup>st</sup> spray	Pretreatment	20.05	19.47
	7th day	11.3	8.54
2 <sup>nd</sup> spray	Pretreatment	8.91	6.82
	7th day	7.1	4.92
3 <sup>rd</sup> spray	Pretreatment	2.76	2
	7th day	1.44	0.37
	Mean	8.60	7.02
		Plant protection cost (Rs)	
Number of sprays (Insecticides and Fungicides)		6	11
Insecticides used (lit/ha)		1.80	13
Insecticides cost (₹/ha)		4 687	46 875
Fungicides used (kg/ha)		11	15.5
Fungicides cost (₹/ha)		11250	24375
% fruit infestation		8.30	7.11
Mean borer larvae		2.25	1.60
Total cost for plant protection (₹/ha)		15937	71250
Cost of cultivation (₹/ha) (agronomic + Plant protection)		261 160	335 222
<sup>h</sup> yield (tonnes/ha)		84.5	95.75*
Gross returns (₹/ha)		676,000	766 000
Net returns (₹/ha)		414 840	430 777
B:C ratio		2.58	2.28

Each mean = 10 plants/Quadrat; <sup>a</sup>mean borer larvae in both plots, cal. t-value = 0.41 table t = 1.74 (P > 0.05); <sup>b</sup>mean infested fruits in both plots, cal. t-value = 1.13, table t = 1.74 (P > 0.05); <sup>c</sup>spray repeated after 15-d; <sup>d</sup>natural enemies recorded; <sup>e</sup> no natural enemies recorded; <sup>f</sup>*H. armigera* eggs not found on marigold after 1<sup>st</sup> application; <sup>g</sup>*H. armigera* eggs found on marigold throughout; <sup>h</sup>yield cal. t-value = 7.4 (P > 0.05), table t = 2.13; <sup>i</sup>1₹ = ₹ 80 and 1US\$ = ₹ 47; <sup>j</sup>based on cost of HearSNPV in the market

endosulfan (1:6.06) followed by Neem Seed Kernel Extract (NSKE) (1:5), HearSNPV at 450 LE/ha (1:3.6) than HearSNPV at 350 LE/ha (1:3.02).

The IPM module using HearSNPV sprays and marigold as a trap crop was successfully validated. Insecticides like indoxacarb 14.5 SC (Avant®) or endosulfan (Endocil®) have quick knockdown effect on *H. armigera* larvae compared

to slow kill of larvae in HNP. So sustenance of natural enemies occurred in HNP, but not in FPP. HearSNPV was compatible with *M. anisopliae* fungus and *C. chloridae*.

The vegetable growers of Rajanakunte, Dibbur, Devanahalli and Doddaballapur at the outskirts of Bangalore were convinced that the application of HearSNPV can be harmoniously blended with organic manures, bio-pesticides and other non-chemical tools. This opinion was expressed by farmers when they were interacted with.

Increased public awareness of pesticide hazards and legislative restrictions against introduction of new pesticides would eventually limit the use of chemical pesticides especially in tropical countries like India. To suppress serious pests like *H. armigera*, HearSNPV has been proven by this investigation to be promising, non-polluting and compatible with IPM tools. HearSNPV at  $2.47 \times 10^{11}$  OBs/ha proved effective against 3<sup>rd</sup> instar *H. armigera* larvae. *H. armigera* larval infestation was driven below the 1 larva/plant threshold after second application of HearSNPV at  $2.47 \times 10^{11}$  OBs/ha and with indoxacarb (Avant®) and endosulfan (Endocil®). Application of HearSNPV at  $2.47 \times 10^{11}$  OBs/ha conferred multiple advantages as *H. armigera* larvae were found infected with *M. anisopliae* and parasitoid *C. chloridae*. HearSNPV worked out much cheaper than insecticidal applications. The cost of applying for one spray incurred ₹ 600 compared to indoxacarb (₹ 3 500) and endosulfan (₹ 1 400). This study shows that in these test plots, HearSNPV is competitive with chemical pesticides despite a significant loss in yield. HearSNPV self perpetuates in the field is compatible with select natural enemies and these attributes confers long term benefits to the growers.

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