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Utilizing Nuclear Polyhedrosis Virus for the Biological Management of Chickpea Pod Borer (*Helicoverpa armigera*) in Organic Chickpea Production



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ABSTRACT

Chickpea (Cicer arietinum L.) is one of the most important organically cultivated pulse crops of our country. The pod borer, Helicoverpa armigera Hubner (Lepidoptera: Noctuidae) is the strategic pest that determines its production quantum to a great extent. Since last few years the organic agriculture has been getting attention of consumers as well as growers because of safety concerns. The pod borer causes huge losses in chickpea crop. It can be effectively managed through synthetic insecticides; however, they are prohibited under organic crop production system. Under such situation, nuclear polyhedrosis virus (NPV) is one of the promising crop protection practices for managing this pest. The Chickpea crop was monitored continuously for two successive cropping seasons (2019-20 and 2020-21) to get the native nuclear polyhedrosis virus. Due to its non-availability, the viral formulation was procured from the market and evaluated against pod borer under field conditions. The survey showed that there was no natural infection of the nuclear polyhedrosis virus in pod borer larvae at the research area of an organic farm. Bioefficacy field trials showed that NPV formulation was effective in controlling this pest during the 2021-22 and 2022-23 crop seasons. The findings indicated that NPV at a concentration of 1.5 ml per liter of water reduced 59.4% of the pest population after 7 days of spray, which reached up to 64.0% after 10 days. A 2.0 ml per liter concentration caused a higher reduction after 7 and 10 days. There was no positive correlation between male adult catch and ETL with weather factors. The formulation did not cause any type of toxicity on the crop and it was found safer for natural enemies viz., honeybees, yellow wasps, and Chrysoperla sp. It is concluded that HaNPV application can contribute in effective management of chickpea pod borer. In addition, being natural this virus safe for ecosystem and hence it can be an ideal approach under an organic crop production system to provide a protective umbrella against this pest without any hazardous effects on crops as well as beneficial insects.

Keywords: Chickpea, organic, pod borer, HaNPV, economic threshold level, yellow wasps, Chrysoperla sp., phytotoxicity, weather conditions

Introduction

Organic agriculture has been gaining the attention of the farming community, consumers, government as well as stakeholders in our country since the nineties. Out of several reasons for its adoption, human health problems are of prime concern. During the seventies, to increase crop production for the food security of the population of our country the usage of chemical pesticides, herbicides, etc. had been excessive, which in succeeding years had created health problems. Haryana as well as Punjab states contributed to a great extent in the central pool of food grains. Malwa region of Punjab is well known as a cancer belt.

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Likewise, agrochemical consumption in Haryana has increased many folds which is responsible for the deterioration of soil, environment, and human health. To overcome such problems and ensure the availability of chemical-free healthy agricultural commodities, conventional agriculture has now been shifting towards organic agriculture. The total area under organic agriculture is about 4.4 million hectares with a production of 3.6 million tonnes (https://apeda.gov.in/apedawebsite/organic/Organic_Products.htm).

The chickpea has also been growing as one of the organic crops. It covers about 98.86 lakh hectares with production of 107.37 lakh tonnes. In Haryana, 0.36 lakh hectares are under this crop [1]. It has also been cultivated as an organic crop due to its nutritional richness. It contains a good amount of carbohydrates, protein, and fat [2].

Organic cultivation of this crop encounters infestation of several insect pests of which *Helicoverpa armigera* (Hubner) (Lepidoptera: Noctuidae) is a key pest [3] responsible for great crop damage in terms of quantity as well quality [4,5,6,7].

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This pest may infest the crop at any growth stage, however, being more severe when attacks during flowering and pod formation stages [8]. For this pest, different economic threshold levels have been determined to adopt management strategies [7,9,10]. Under the organic crop production system, its management through chemical insecticides is prohibited. Collection and destruction of the infected pods as well as grownup larvae, intercropping of coriander, monitoring *Helicoverpa* by pheromone traps, installation of bird perches, sprinkling or application of turmeric powder added puffed rice or cooked rice in the morning or evening hours for attraction of birds and use HaNPV are suitable operations for management of pod borer under organic cropping system. HaNPV being one of the suitable management approaches for controlling the pod borer, efforts have been made in this study to isolate the native nuclear polyhedrosis virus from naturally infected *H. armigera* larvae of field population and its bioefficacy evaluation under field conditions for consecutive two seasons.

Materials and Methods

Survey and collection of *H. armigera* nuclear polyhedrosis virus-infected larvae

During 2019-20 and 2020-21, the chickpea (var. HC-5) crop was cultivated at a research farm, Deendayal Upadhyay Centre of Excellence for organic Farming, Chaudhary Charan Singh Haryana Agricultural University, Hisar (Latitude 29°08'11.3"N and Longitude 75°42'15.6"E). The crop was kept unsprayed during the complete crop season. It was surveyed at regular intervals from February to April month. Four pheromone traps per acre were installed in the cropped area to detect the presence of gram pod borer, *Helicoverpa armigera* in the field. Male moths trapped in the traps were assessed visually. The natural infection of gram pod borer larvae with nuclear polyhedrosis virus was assessed in cropped areas by selecting ten spots per acre.

Field trials of *H. armigera* nuclear polyhedrosis virus formulation

During the succeeding two seasons i.e., 2021-22 and 2022-23, HaNPV formulation (trade name - SunBio HaNPV of M/s Sonkul Agro Industries Private Limited, Nashik) containing 1x10° polyhedral occlusion bodies/ml) was procured and tested for the management of pod borer using its different concentrations. There were five treatments in four replications including untreated control. Each plot was of 5 m x 5 m sizes and a randomized block design was adopted to execute field trials. The insect population was assessed on randomly selected 10 plants to judge the economic threshold level. At the economic threshold level (average one 2nd instar larva per plant) the crop was sprayed with viral formulation during evening hours and the spray was repeated after ten days. Insect pest population was recorded before spray and then after 3, 7, and 10 days of spray. Percent pest population reduction was worked out over control (pest population in control plot - pest population in treated plot/pest population in control plot × 100). Data were statistically analyzed using the online statistical package OPSTAT of the university.

Effect of *H. armigera* nuclear polyhedrosis virus formulation on beneficial insect and crop

To determine the effect of formulation on beneficial insects, the population of honeybees, yellow wasps, and *Chrysoperla* sp. in HaNPV-applied plots was recorded before and after 7 as well as 10 days of spray.

To find out the toxic effect of the formulation on the sprayed crop, after $10 \text{ days of } 2^{\text{nd}} \text{spray randomly chosen five plants were assessed. Some of the common symptoms such as plant yellowing, undergrowth, burning, etc. were considered for this purpose.$

Results

Survey and collection of *H. armigera* nuclear polyhedrosis virus-infected larvae

The chickpea pod borer (*H. armigera*) infestation on the crop was started in the first week of March 2020 which was continued till the third week of April 2020. During 2nd year, field was regularly surveyed from February to March 2021 which indicated that the *H. armigera* field population was not infected with NPV and hence there was no larval mortality due to viral infection in both years. Field population reared in the laboratory to detect NPV latent infection revealed no infection of this virus.

Field trials of *H. armigera* nuclear polyhedrosis virus formulation

The presence of *H. armigera* in the crop was detected using pheromone traps. The trapped adult moth population ranged from 7.7 to 25.75 and 5.75 to 25.0 during 2022 and 2023, respectively (Fig. 1). The infestation of the pest reached ETL (1.3 and 1.0 larva per plant) during the first fortnight in both years (Figure 1) and spray was initiated at this time. The findings of 2021-22 and 2022-23 divulged that the tested formulation was effective in reducing the population of chickpea pod borer. 1.5 ml per liter concentration of viral formulation showed a 64.0 percent population reduction; however, the highest pest population reduction (70.3%) was achieved when 2.0 ml per liter concentration was applied (Table 1).

The relationship between pest incidence and prevailing weather conditions during the crop season 2022 and 2023 was correlated. In the correlation study, temperature (maximum and minimum), relative humidity (morning and evening), wind speed, sunshine hours, evaporation, and rainfall did not show a positive correlation with adult catch and economic threshold level of the pest in both years as indicated in table 2. During the trial period 2022, there was lesser rainfall as shown in Figure 2.

Effect of *H. armigera* nuclear polyhedrosis virus formulation on beneficial insect and crop

In HaNPV sprayed plots there was no adverse effect on the population of honeybees, yellow wasps, and *Chrysoperla* sp. (Table 3).

Discussion

During two years' survey, it was noted that there was no natural infection of entomopathogenic virus in the field population of chickpea pod borer. Similarly, monitoring of chickpea pod borer, *H. armigera* at the Organic research farm, Institute of Agricultural Sciences, Bundelkhand University, Jhansi in Uttar Pradesh was done with the help of pheromone trap during the *rabi* season of 2021-2022. It was noted that there were adult male moth activities from mid-February to 3rd week of March. In the pheromone traps the moth trapping was higher from 05.00 to 07.00 hours in morning [11].

In the present study, spray of nuclear polyhedrosis virus resulted in to reduction of the larval population of *H. armigera* in chickpea crops. The research findings of earlier researchers are in line with our results where a greater reduction in the population of chickpea pod borer and an increment in crop yield was noted [12,13,14,15].

Bioefficacy of the nuclear polyhedrosis virus indicated higher larval mortality of *H. armigera* in all chickpea varieties which reflected in lesser pod damage [16,17,18]. Application of nuclear polyhedrosis virus at 300 larval equivalents per hectare reduced pod borer population by 78.7 percent and enhanced production [19].

Field evaluation of HaNPV at different larval equivalent concentrations reduced population of $\it H. armigera$ (Hubner) in chickpea crop up to 60.5 per cent during $\it rabi$ seasons. The crop yield was $1610 \, kg$ in treated plot as compared to $1480 \, kg$ / ha in control [20, 21].

The larval numbers per plant were 2.52 and 2.56 when two and three sprays of 250 larval equivalent per hectare were performed as compared to 11.23 and 12.0, respectively in control [11].

Spray of a few isolates of HaNPV (1.5×10^{12} POB/ha) reduced chickpea pod borer larval population (31.14 - 61.88%) as well as pod damage (15.86 - 54.52%) over the control. The chickpea yield was 718.3 to 983.6 kg per hectare when compared with the control ($693.3 \, \text{kg/ha}$) as reported by [22].

In a field experiment the spray of *Helicoverpa* nuclear polyhedrosis virus at a concentration of 0.1 g per liter of water for integrated pest management practice against pod borer in chickpea could minimize 68.2% pod damage. Further, there was 1832.2 kg grain yield as compared to 1116.6 kg per hectare in control and the benefit-cost ratio was 2.11 [23].

Spraying 500 ml concentrations of HaNPV formulation (Helicop 2% AS) in the chickpea field had lowered down 77.78 and 82.71% percent pod borer populations after the first and second spray, respectively. The formulation increased yield (18.53 q/ha) in comparison to the control (13.90 q/ha) as reported by [24]. HaNPV spray at 1.5 ml per liter concentration resulted in higher chickpea yield as compared to unsprayed control plots [25]. A bioefficacy study conducted in Bangladesh during the *rabi* cropping season of 2004-05 divulged that this viral biopesticide reduced pod borer damage, enhanced yield, and improved benefit-cost ratio [26].

It is reported that 350 LE per hectare of *H. armigera* nuclear polyhedrosis virus was highly effective in reducing the chickpea pod borer larval population and enhancing yield [27].

Earlier researchers evaluated various HaNPV isolates against *H. armigera* in the chickpea field for two years at Dharwad, Karnataka, and noted that the Gulbarga and Coimbatore isolates outperformed the others. These isolates demonstrated superior efficacy against mortality of *H. armigera* larval resulted in higher chickpea yield. The incremental benefit-cost ratio was also highest for the Gulbarga and Coimbatore isolates, followed by the Dharwad isolate [28]. Our study indicated that tested viral formulation was safer for crop as well as beneficial insects. This study is useful for organic growers where they can apply such viral formulation for the management of chickpea pod borer without any side effect on ecosystem.

Conflict of Interest: The authors declare that they have no conflict of interest.

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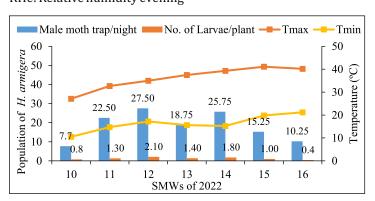
Abbreviation

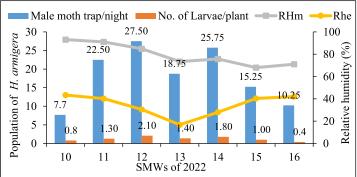
CCS HAU: Chaudhary Charan Singh Haryana Agricultural University

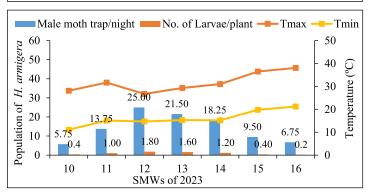
HaNPV: Helicoverpa armigera nuclear polyhedrosis virus

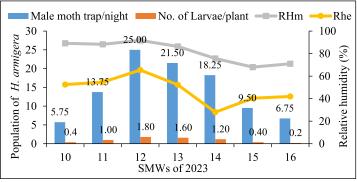
LE: Larval equivalent POB: Poly occlusion body

SMW: Standard Meteorological Week RHm: Relative humidity morning RHe: Relative humidity evening









 $\textit{Fig. 1: Male moth trappped and economic threshold level of H. armiger a in chick peasure of the property o$



Fig. 2: Rainfall during both years

Table 1: In vivo bioefficacy of nuclear polyhedrosis virus formulation against H. armigera of chickpea (Average of two years)

HaNPV dose (mL per litre water)		Larv	ae / 10 plants	Per cent pest population reduction over control		
	Pre spray	3 Days after spray	7 Days after spray	10 Days after spray	7 Days after spray	10 Days after spray
0.5	7.4	8.9	9.2	9.1	41.5	49.0
1.0	9.8	11.3	8.4	8.2	46.5	53.8
1.5	7.8	10.0	6.5	6.4	59.4	64.0
2.0	8.5	11.6	5.7	5.3	64.8	70.3
Control	10.3	13.9	15.8	17.5	0	0
CD at 5%	-	-	-	-	8.2	5.7

Table 2: Correlation between H. armigera adult moth catch and economic threshold level

Treatment		Weather parameter									
		Temp. Max.	Temp. min.	RH morning	RH evening	Wind Speed	Sunshine hour	Evaporation	Rainfall	Accumulative rain	
Adult moth catch	Corr.	-0.01	-0.09	0.21	-0.2	0.03	-0.09	-0.26	0.12	-0.2	
	Abs Corr.	0.01	0.09	0.21	0.2	0.03	0.09	0.26	0.12	0.2	
	Sig. level	NS	NS	NS	NS	NS	NS	NS	NS	NS	
Economic threshold level	Corr.	-0.16	-0.28	0.35	-0.17	-0.06	-0.2	-0.42	0.12	-0.28	
	Abs Corr.	0.16	0.28	0.35	0.17	0.06	0.2	0.42	0.12	0.28	
	Sig. level	NS	NS	NS	NS	NS	NS	NS	NS	NS	

n-2=12, 5% Sig. level

 ${\it Table\,3:} \textit{Effect of HaNPV on population of beneficial insects}$

	Population of beneficial insects							
HaNPV dose (ml per litre water)	Honeybee		yellow wasp		Chrysoperla sp.			
	Pre spray	10 DAS	Pre spray	10 DAS	Pre spray	10 DAS		
0.5	1.00	0.75	0.50	0.75	0.75	0.50		
1.0	0.75	0.50	0.75	0.75	0.50	0.50		
1.5	0.50	0.75	0.75	1.00	0.25	0.50		
2.0	0.50	0.50	0.75	0.75	0.75	0.75		
Control	0.75	0.75	0.75	1.00	0.50	0.75		
C.D. at 5%	NS	NS	NS	NS	NS	NS		

 $Average\,of\,4\,replications$

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