*Research Journal of Agriculture and Forestry Sciences* _____ ISSN 2320-6063
Vol. **1(8),** 30-33, September **(2013)** *Res. J. Agriculture and Forestry Sci.*

***Review Paper***

# Nuclear Polyhedrosis Virus (NPV), A Potential Biopesticide: A Review

**Erayya[1], Jaba Jagdish[2], Sajeesh P.K[1] and Vinod Upadhyay[1]**
[1]Department of Plant Pathology, GBPUAT, Pantnagar,-263145, Uttarakhand, INDIA
[2]Department of Entomology, GBPUAT, Pantnagar-263145, Uttarakhand, INDIA

**Available online at: www.isca.in**
Received 19th August 2013, revised 29th August 2013, accepted 2nd September 2013

## Abstract

*The indiscriminate use of pesticides for the last 40 years has almost eliminated natural enemies from many crop eco-systems. This scenario has led many countries to consider the potential of biological control as a component of pest management. At present, the world market for microbial pesticides is in excess of US $ 125 million per annum which is still less than 1 percent of the total global market for agrochemical crop protection of $ 20-25 billion. The viruses belonging to 11 families which are pathogenic to insects. Baculoviruses are associated with the orders of Lepidoptera Hymenoptera, Diptera, Neuroptera, Coleoptera, Trichoptera, Crustacea and mites (The virions are rod-shaped, 40-70 nm X 250-400 nm, comprising a lipoprotein envelope around a protein capsid containing DNA-protein core. Baculoviruses are one of the best alternative to chemical pesticides asthey are environment friendly. NPV is known for high epizootic levels and is naturally occurring, self-perpetuating, safe to natural enemies due to host specificity and environmental friendly, because of its obligate nature and host specificity. Nuclear polyhedrosis viruses recorded in India includes Helicoverpa armigera, S. litura, S. exigua, Amsacta moorei, Agrotis ipsilon, A. segetum, Anadividia peponis, Trichoplusia ni, Thysanoplusia orichalcea, Adisura atkinsoni, Plutella xylostella, Corcyra cephalonica, Mythimna separata and Phthorimaea operculella.*

**Keywords:** Nuclear polyhedrosis virus, bio-pesticide, baculovirus, characterization.

## Introduction

The development of biological control has followed no master plan but has surged on, based on insights, luck, personal endeavour and institutional momentum. The indiscriminate use of pesticides for the last 40 years has almost eliminated natural enemies from many crop eco-systems. It is estimated that only a small portion of pesticides applied to the crop reaches the target while the major portion reaches the non-target. This scenario has led many countries to consider biological control as a vital component of integrated pest management.

Biopesticides mean the use of fungi, bacteria, viruses, protozoa and nematodes through inundative or inoculative release for the biological control of insect pest, diseases, weeds and nematodes in agriculture, medicinal, veterinary, forestry and horticultural eco-systems. At present, the world market for microbial pesticides is in excess of US $ 125 million per annum which is still less than 1 percent of the total global market for agrochemical crop protection of $ 20-25 billion. The market is dominated by *Bacillus thuringiensis* (80%) followed by nematodes (13.3%) and others (6.67%). During last decade government of India spent nearly Rs.14, 926 million for bio-control programme in different crops.

In India, Madhya Pradesh, Maharasthra, Uttar Pradesh, Andhra Pradesh and Karnataka are the major states adapting IPM strategy. It is, therefore, clear that biological suppression of crop pests needs proper encouragement. Insect Viruses are submicroscopic, obligate, intracellular, pathogenic entities, The viruses belonging to 11 families which are pathogenic to insects. Baculoviruses are associated with the orders of Lepidoptera Hymenoptera, Diptera, Neuroptera, Coleoptera, Trichoptera, Crustacea and mites (The virions are rod-shaped, 40-70 nm X 250-400 nm, comprising a lipoprotein envelope around a protein capsid containing DNA-protein core. The capsid and core is known as the nucleo-capsid[1]. These viruses are specific and often highly virulent to their hosts. They are restricted in their pathogenecity to the class insecta; they are often genus or species specific. Approximately 60 per cent of the 1200 known insect viruses belong to the family Baculoviridae and it is estimated that such viruses could be used against nearly 30 per cent of all the major pests of food and fibre crops. HaNPV is an important biopesticide, but, it is having certain constraints like longer duration of target killing. Presuming that a refined multiplication method of HaNPV is available, commercial utilization of HaNPV has several limitations including its bio-efficacy under field conditions. The germicidal action of UV radiation of sunlight and substrate environment has greater influence in reducing the potency of viruses to kill the pest.

NPV is known for high epizootic levels and is naturally occurring, self-perpetuating, safe to natural enemies due to host specificity and environmental friendly. Since, NPV is an obligate parasite, it multiply only in living larvae. So, mass production of NPV is tedious joband require skilled labours[2].

Nuclear polyhedrosis viruses recorded in India includes *Helicoverpa armigera, S. litura, S. exigua, Amsacta moorei, Agrotis ipsilon, A. segetum, Anadividia peponis, Trichoplusia ni, Thysanoplusia orichalcea, Adisura atkinsoni, Plutella xylostella, Corcyra cephalonica, Mythimna separata* and *Phthorimaea operculella.*

## Baculoviruses (Fam: Baculoviridae)

The virions are rod-shaped, 40-70 nm X 250-400 nm, comprising a lipoprotein envelope around a protein capsid containing DNA-protein core. Baculoviruses are large, circular and double-stranded DNA genomes ranging from 81.7 to 178.7 kb [3]. They are pathogenic to Lepidoptera, Hymenoptera, and Diptera[4].

The capsid and core is known as the nucleo-capsid. The baculoviridae family having two sub-families, one is *Eubaculovirinae* in which the virions are embedded in a crystalline matrix of protein and another one is *Nudibaculovirinae* not embedded in a crystalline matrix. These viruses are specific and often highly virulent to their hosts. They are restricted in their pathogenicity to the class insecta; they are often genus or species specific. Approximately 60 per cent of the 1200 known insect viruses belong to the family Baculoviridae and it is estimated that such viruses could be used against nearly 30 per cent of all the major pests of food and fibre crops.

Baculoviruses viruses are highly host specific and non-pathogenic to beneficial insects as well as other non-target organisms[5]. Low toxic residual effect and low pest resistance development is the advantage over conventional pesticides[5]. *Eubaculovirinae* includes two genera, nuclear polyhedrosis virus (NPV) and granulosis virus (GV). For the expression of recombinant proteins and biological control of insect pests they are extensively studied[6]. Baculovirus preparations registered for pest control in various parts of the world are mentioned in table 1 and records of formulations of NPV in India are given in table 2.

## Pathological Changes

In NPV behavioral changes may occur first as the larvae hang upside down. The integument often changes in colour and the insects become flaccid and fragile. At death rupture of the dody wall releasing the IBS is a common feature. In case of *Oryctes* the body becomes translucent and hindgut may completely evaginate. NPV and GVs can be distinguished by their size difference. The GV capsules are ellipsoidal in outline while the NPV polyhedra are angular or spherical.

## Characterization of NPV isolates

HaNPV isolates collected from different locations of Karnataka, Tamil Nadu and Andhra Pradesh and characterised them using molecular tools. DNA profiling revealed significant difference between the isolates[7]. Based on similarity matrix, co-efficient and dendrogram analysis they devided the isolates into two major clusters. Similarly, variation among the different isolates of NPV isolated from Egyptian cotton worm, *Spodoptera littoralis* was reported by Cherry C.L. and Summers M.D.[8].

Minor genotypic differences of eight geographically different isolates of potato tuber moth granulosis virus (PTMGV) was analysed by restriction endonuclease studies which indicated the existence of three distinct but closely related genotypes[9].

All the HaNPV isolates appeared as clear, irregular six sided objects with rounded edges, phase-bright under phase contrast[10].

HaNPV isolated from different areas of Tamil Nadu and compared with the isolates of Gujarat and Rajsthan[11]. The diameter of the POB ranged between 1.734 to 2.006 µm. The least being recorded in Ooty isolate while, the other extreme was observed in Rajasthan isolate. However, there was no significant difference among the isolates with respect to the POB size. Further, the time mortality response studies proved that the Ooty isolate recorded shorter LT50 values as compared to other isolates at all tested concentrations made an effort to characterize the NPV of *H. armigera, Spodoptera litura* (F.)*, Amsacta albistriga* (W,) and *Mythimna separata* (W.)[12]. The samples were collected from the same ecosystem. The DNA fragment profiles of Pst1 and Hind-III showed that the genome size ranged between 69.30 to 125.30 kbp. The size and number of fragments in each isolate varied drastically. Each isolate showed their unique fragments of varied size and the variation in the genome size was mainly due to the genetically controlled factors as well as the climatic factors prevailing in that region.

**Approaches in Biotechnology in for increasing the effectiveness of micro-organisms:** Widespread acceptance and use of baculoviruses has never been achieved due to the very slow kill, which is characteristic of wild type viruses. Hence, a favourable option in microbial control with baculoviruses would be to use recombinant baculoviruses which can skill insects more efficiently in shorter time. The baculovirus genome is amenable for genetic engineering and enabled cloning of several proteins of insecticidal value, *viz.,* toxins, juvenile hormone esterase, PTTH, mellitin, trehalase, fungal insecticidal protease, scorpion and mite toxins in to NPV (table 3).

## NPV Quality Control

Various problems are encountered in the production of entomopathogenic cultures with respect to quality. Firstly, the quality of water used in preparing the formulations including presence of chlorine. So, for large-scale production distilled water should be used. It would be useful to try out the possibility of using de-mineralized water for formulations.

A staining method for counting the polyhedra in NPV can be used a quality control test for NPV. In the case of fungal pathogens, if the virulence has to be retained, it is necessary to

periodically pass through the host culture. For quality control of Bt formulations, diet-incorporation method was generally used, which was made simpler. Regulatory oversight of microbial biopesticides still in its infancy. Although further advanced than that of microbial Biocontrol agents, the majority of countries worldwide have no specific registration procedure for microbial pest control agents. Because of their association with conventional chemical pesticides, many countries have adopted the basic chemical model for biopesticides registration. This requires the generation of date that are inappropriate for biological organisms on one hand and yet neglect some important biological features of these agents on the other. Probably the most advanced regulatory procedure for microbial pesticides is that of the US Environmental Protection Agency (EPA). Standardized guideline of quality control procedures for biocontrol agents not yet available. So, there is need of manufactures to develop their own quality control procedures. There are reports that many fungal, bacterial and viral products produced in developing countries are failing to fulfil required standards.

**Table-1**
**Baculovirus preparations registered for pest control in various parts of the world**

| Insect species | Trade name of the product | Country | Year of registration |
|---|---|---|---|
| **Granulosis viruses** | | | |
| *Adoxophyes orana* | Capex | Czechoslovakia | 1989 |
| *Agotis segetum* | Agrovir Germany 1990 | Germany | 1990 |
| *Cydia pomonella* | Madex | Czechoslovakia | 1987 |
| **Nuclear polyhedrosis viruses** | | | |
| *Heliothis* Spp. | Biotrol VHZ and Virion H | USA | 1973 |
| *Heliothis* spp. | Elcar | USA | 1975 |
| *Lymantria dispar* | Gypcheck | USA | 1978 |
| *Mamestra brassicae* | Mamestrin | Finland | 1988 |
| *Neodiprion lecontei* | Lecont-virus | USA/Canada | 1982/1983 |
| *Orgyia seudotsugata* | Biocontrol-1 and Virtuss | USA | 1976 |

**Table-2**
**Nuclear polyhedrosis viruses recorded in India**

| Pest | Crop |
|---|---|
| *H. armigera* | Chickpea and others |
| *S. litura* | Tobacco and others |
| *S. exigua* | Tom ato and others |
| *Amsacta moorei* | Pulses |
| *Agrotis ipsilon, A. segetum* | Potato and others |
| *Anadividia peponis* | Gourds |
| *Trichoplusia ni* | Potato and others |

**Table-3**
**Development of genetically engineered baculoviruses**

| Virus | Foreign gene | Host insect |
|---|---|---|
| BmNPV | Toxin from *Andractomus australis* | *Bombyx mori* |
| zHPV | Toxin 34 from *Pyemotes tritici* | *Heliothis zea* |
| AcMNPV | Toxin 21 A from *Pyemotes tritici* | *Trichoplusia ni* |
| AcMNPV and HvJHE | Neurotoxin from Spider | *S.frugiperda* |

## Conclusion

NPV is being one of the important biopesticide, as it is eco-friendly, having less residual toxicity, compatible with many chemical pesticides, self-perpetuating nature. Hence, NPV can be implemented as one of major component in IPM programme. But, there is a scope to develop quality control guidelines and methodologies, systematic registration policies, to identify effective stains and to develop UV (Ultraviolet) resistant strains. In addition, the guidelines and training for implementation of biocontrol agents should be made available.

## References

**1.** Steinhaus E.A., Principals of Insect Pathology, McGraw Hill, New York, 757 (**1949**)

**2.** McIntosh A.H., Rice W.C. and Ignoffo C.M., Genotypic variants in wild type populations of Baculovirus. In:maramorsoch K. (ed), *Biotechnology in Inverterbrate pathology & cell culture,* Academic Press, New York, 305-325 (**1987**)

**3.** Zhang C.X., Ma X.C. and Guo Z.J., Comparison of the complete genome sequence between C1 and G4 isolates of the *Helicoverpa armigera* single-nucleocapsid nucleopolyhedrovirus, *Virol.,* **333**, 190–199 **(2005)**

**4.** Adams J.R., and McClintock J.T., Nuclear polyhedrosis viruses of insects, In J. R. Adams and J. R. Bonami (Eds.), Atlas of invertebrate viruses, Boca Raton, FL, USA: CRC Press, 87–204 **(1991)**

**5.** Barreto M.R., Guimaraes C.T., Teixeira F.F., Paiva E. and Valicente F.H., Effect of Baculovirus spodoptera isolates in *Spodoptera frugiperda* (J.E. Smith) (Lepidoptera: Noctuidae) larvae and their characterization by RAPD, *Neotropical Entomology*, **34(1)**, 67–75 **(2005)**

**6.** Ma X., Xu H., Tang M., Xiao Q., Hong J. and Zhang C., Morphological, phylogenetic and biological characteristics of Ectropis obliqua single-nucleocapsid nucleopolyhedrovirus, *J. Microbiol.*, **44**, 77–82 (**2006**)

**7.** Kambrekar D.N. and Kulkarni K.A., Field performance of *Helicoverpa armigera* nuclear polyhedrosis virus of the fruit borer, *H.* Armigera (Hubner) on chickpea, *Pest Magt. Econ. Zool.,* **13(2),** 289- 296 **(2005)**

**8.** Cherry C.L. and Summers M.D., Genotypic variation among wild isolates of two nuclear polyhedrosis viruses isolated from *Spodoptera littoralis*, *J. Invert. Pathol*., **46**, 289-295 **(1985)**

**9.** Vickers J.M., Cory J.S. and Entswistle P.F., DNA characterization of eight geographic isolates of granulosis virus forn the potato tuber moth (*Phtorimae operculella*) (Lepidoptera, Gelechiidae), *J. Invert. Pathol*., **57**, 334-342 **(1991)**

**10.** Charmi Shailesh Patel, Janardan J. Jani, Vipulkumar B. Parekh, Vijay B. Darji and Piyush R. Vaishnav, Genetic diversity and differentiation of *Helicoverpa armigera* nuclear polyhedrosis virus isolates from India, *Phytoparasitica,* **37**, 407–413 **(2009)**

**11.** Somasekar, S., Jayapragasam, M. and Rabindra, R. J., Characterization of five Indian isolates of the nuclear polyhedrosis virus of *Helicoverpa armigera*, *Phytoparasitica*, **21(4)**, 333-337 **(1993)**

**12.** Rabindra R.J. and Rajasekharan B., Insect cell cultures in Biotechnology and Pest control, In Biotechnological Perspectives in Chemical Ecology of Insects, (T. N. Anantakrishnan ed.) Oxford and IBH Co. Pvt. Ltd. New Delhi, 223-239 **(1996)**