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A Review on Nucleopolyhydroviruses (NPV) as Biological Control of Army Worm, *Spodoptera litura*

Zulnorain Sajid[1*], Muhammad Ramzan[2], Muhammad Mamoon Shafiq[3], Muhammad Usman[3], Ghulam Murtaza[3], and Vikas Pareek[4]

[1]Department of Entomology, PMAS Arid Agriculture University, Rawalpindi, Pakistan

[2]State Key Laboratory for Biology of Plant Diseases and Insect Pests, Institute of Plant Protection, Chinese Academy of Agricultural Sciences Beijing, 100193, China

[3]Department of Entomology, University of Agriculture Faisalabad

[4]Head & Assistant Professor, Department of Botany, Gramin Mahila PG College Sikar, Rajasthan, India

*Corresponding Author E-mail: ramzan.mnsua@gmail.com

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ABSTRACT

The current review was written to give the detail information about Nucleopolyhydroviruses (NPV) that consider a major part of integrated pest management. The review of literature was collected from Google Scholar by giving different key words about Nucleopolyhydroviruses (NPV) against Spodoptera litura, previously published data on NPV and from well-known journal sites. Army worm, Spodoptera litura is a destructive pest of many agricultural and horticultural crops all over the world including Pakistan. Different management strategies are adopted to control this destructive pest in Pakistan. Among adopted strategies, insecticides are commonly used against this pest under laboratories as well as fields conditions. The excessive application of insecticides causes insecticides resistance and negative impact on environment resulting health problems (Ramzan et al., 2019c). The application of NPVs against insect pests is an alternative strategy to control pest population, reduce resistance issues, health problems and environmental pollution. The current review of literature shows that Nucleopolyhydroviruses has potential against insect pest population. The biological activity of ucleopolyhydroviruses should be tested against other lepidopteran insect pests that becoming primary pest of agricultural crops or invasive alien species especially Fall armyworm, Spodoptera frugiperda.

Keywords: *Nucleopolyhydroviruses, Biological control, Invasive alien species, Resistance problems, Integrated pest management.*

INTRODUCTION

1.1. Baculoviruses

The crop production decreases worldwide every year due to many factors like weeds, insect pests and pathogens i.e., viruses, bacteria and fungi (Ramzan et al., 2019a). These issues mostly occur in tropical and subtropical where crop grown in monocultures.

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The synthetic chemical pesticides highly used for pest management since few decades. The synthetic insecticides harm the beneficial fauna like entomopathogens. These also affect humans, birds, animals and persist in the environment. Today farmers don't adopt synthetic chemical control due to their non-availability, expensive, persistence in environment and insect resistance. The integrated pest management (IPM) adopted by public which summarize cultural, physiological, chemical and biological control to avoid the insecticides resistance against insects and environmental hazards (Chandler et al., 2011; Rodriguez, Belaich, & Ghiringhelli, 2012).

An alternative control which are ecofriendly and biodegradable must be used. The biological agents like predators, parasitoids and other microbial pesticides such as viruses, fungi, bacteria and nematodes that are ecofriendly and don't harm the beneficial insects have been used since 1940s and 1950s for controlling various insect pests hence not a new technology. The viruses are mixed with other control strategies like integrated pest management (IPM) (Moscardi et al., 2011) for effective pest control. These viruses are infectious for larval and some moth like shrimps, mosquitoes and sawfly (Jeremy et al., 2015). The viruses enter into mammal cell but don't replicate in them so noninfectious for them (Burand & Hunter, 2013; & Theze et al., 2011).

The virions of virus present in two form like occlusion derived viruses (ODV) that present in viral crystalline protein matrix and baculoviruses (BV) which produced from diseased cells. Both virions have cylinder like nucleocapsids which embedded in protein envelope. Due to rod like shape viruses name as baculovirus which called a cane (Rohrmann, 2013[a]). Nucleocapsids varies in size, length about 40 to 60 nanometer (nm) wide and 250 to 300 nm long. The length varies according to size (Ihalainen et al., 2010). It contains protein and viral genome DNA that are embedded in protein capsid. DNA also present in arginine-rich basic protein.

1.2. Classification

Baculoviridae is the main family which composed 66 species, 4 genera (Viral Zone, 2015 & Jump up, 2018) and used as insecticide. The genera named as Alphabaculovirus Betabaculovirus, Gammabaculovirus and last one is Deltabaculovirus. Alphabaculovirus and Betabaculoviruses are specific for lepidopteran while Gamma Baculoviruses and Delta Baculoviruses are used for hymenopteran and dipteran respectively (Rohrmann, 2013; Theze et al., 2011 & Di Lelio, et al., 2014). The word "virus" derived from Latin word poison so-called disease-causing agent and pass through the filter paper. Virus consists of nucleic acid and a protein which are called capsid. The viruses which used as biological agent belong to genus Nucleopolyhedrovirus after which names as nucleopolyhedroviruses (NPV).

There are two subfamilies of the baculoviridae like Eubaculovirinae and Nudibaculovirinae. The main distinguish between these two subfamilies is that eubaculovirinae virions are inclosed in protein matrix while in Nudibaculovirinae no protein matrix surround. Further Eubaculovirinae divided into two genera like nuclear polyhedrosis virus (NPV) and granulosis virus (GV). Both these genera have been used widely for insect pest management. Due to their virulence effect these viruses only cause mortality in class insecta. It has been estimated that 60 per cent viruses of baculoviridae family used against pests that are threat for fiber and food crops.

1.3. Structure of Bucaloviruses

Bucaloviruses are the small viruses that cause many diseases in insects, arthropods and human. These are minute in size, have circular double stranded DNA which codes genes for virus production and transmission. The genetic material exposed through the sunlight, coated by protein known as a polyhedron which protect the material (Rohrmann, 2011, 2013). The toxic particles of the virus enveloped in occlusion bodies (OBs). From crane fly

(Tipula paludosa) belong to order Diptera, the disease which name as polyhedrosis reported as known as nuclear polyhedrosis of spruce sawfly (Gilpinia hercyniae) the order hymenopteran. Another order of bucaloviruses is granulosis which are in the form of granules reported from cabbage butterfly (Pieris brassicae). The baculoviruses, polydnaviruses, ascoviruses and reoviruses are the main pathogens that are used for insect pests' control. Baculoviruses have not been used for insect pests' control but also use in gene therapy and vaccines production (Lapointe et al., 2012; Moscardi et al., 2011; & Pidre et al., 2013).

Several insects eat the material which become fatal for those. The biological control agents are very importance for controlling agriculture and forest insects' pests. These used as biological control agent for Lepidoptera, Hymenoptera, Diptera, Neuroptera, Coleoptera, Trichoptera, Crustacea and mites. The Lepidoptera species mostly affected with entomopathogenic viruses like nucleopolyhedroviruses (NPV). The more than 11 various families that are fatal for the insects among them baculoviridae (nucleopolyhedroviruses and Granulas virus) has been extensively used due to ecofriendly, effectiveness, stability and comprehensive environmental safety. NPV recorded first time in 1913 as filterable virus. These viruses infect the insects and mites with their virion. The virion attached many years with their hosts due to presence of occlusion bodies. The viruses are rod shaped, multiply in hosts and invade inside the hosts and at last insect die. In 1940 these used in crop fields as biopesticides.

NPV recorded from insects like *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* in India. Many authors have been reviewing the development of baculoviruses worldwide (Moscardi et al., 2011; & Rodriguez et al., 2012).

1.4. Characteristics of Nucleopolyhedroviruses

Nucleopolyhydroviruses are extensively used in integrated pest management due to following reasons; species specific, have no negative effect on non-target hosts like mammals, birds, fish etc., safer for beneficial insects like predators and pollinators, ecofriendly, don't harm the plant or human and good natural control (Ahmad et al., 2011). These are compatible with integrated management strategies or tactics. These can be developed at farm and industrial level as reported by (Inceoglu et al., 2006). Some factors like narrow spectrum, low speed to kill the pest and non-availability of sample limit the use of viruses globally. The toxicity of insecticides has been enhanced through Baculoviruses. But still no such type of baculovirus-based insecticide has been improved or accessible (Beas-Catena et al., 2014).

1.5. Host Range

There are several viruses' families but Baculoviruses belong to family, baculoviridae. The order of insects from which more than 700 baculoviruses has been secluded are Lepidoptera, Hymenoptera, and Diptera. Majority of viruses have been isolated from lepidoptera and small quantity from Hymenoptera and Diptera. (Ahmad et al., 2011) reported that more than 50 products of viruses have been used globally for pest management. The various products of viruses have been used against forest, agricultural and vegetables insect pests.

1.6. Transmission

The virus can be transmitted vertically as well as horizontally from infected to healthy hosts. This mode of transmission either vertically or horizontally play key role in the ecology of bacaloviruses. Spreading of virus from infected populations to healthy populations termed as horizontal transmission. In horizontal transmission, infected hosts die and viral particles released from those cadavers into soil and foliage. Huge amount of occlusion bodies (OBs) released from the dead cadavers and fed during the feeding of susceptible hosts. The host plants polluted with the defecation of infected populations and become the major source of further transmission.

1.7. Infectious process (larvae or adult etc.)

There are large number of pathogens such as parasites, predators and parasitoids which change their host behavior during infection (Van Houte et al., 2013). All these pathogens are altering their behavior like toxoplasma-infected rodents losing their innate behavior and loathing the cats, ants climbing into grass leaves, infected with lancet liver fluke. Gordian worm infected crickets and grasshoppers are also change behavior.

Baculoviruses can only infect the larval stages of insects. Infection occurs orally when the insect host feeds on OB contaminated plant parts or from soil. There are two virion such as occlusion derived virus (ODVs) and budded virions (BVs), which cause infection in the insect host tissues. The horizontal transmission between insect hosts spread by ODVs while BVs are responsible for systemic spread of the infection within a host. The OBs and food particles travel through the foregut and enter the midgut of the larvae, where they initiate infection. After post entry, ODVs are spread throughout the midgut within about 10 minutes. After spreading into midgut, ODVs can breach the peritrophic membrane, enter the midgut epithelial cells and subsequently establish efficient systemic infections (Passarelli, 2011). Lepidopteran larvae have alkaline (pH10-11) midgut juices and the baculoviruses have evolved to exploit this alkaline microenvironment. The alkalinity of the midgut enables dissolution of OBs, releasing the embedded occlusion derived virions (ODVs) in the midgut lumen. These ODVs are released within about 10 minutes of post entry into the midgut. The liberated ODVs can then breach the peritrophic membrane, enter the midgut epithelial cells and subsequently establish efficient systemic infections (Passarelli, 2011; & Federici, 1997).

1.8. Symptoms

After transmission in their hosts, viruses show some behavioral changes. First larvae movement stops and feeding also reduce. The infected larvae stuck and hang to vegetables. The body becomes swollen and glossy. The difference between NPV and GVs is that NPV have angular polyhedral and GV look like ellipsoidal.

1.9. Life cycle

Autographa californica multicapsid nucleopolyhedrovirus (AcMNPV) is the mostly studied virus. AcMNPV belong to family Baculoviridae. This virus isolated from alfalfa looper which belong to order lepidoptera. The larvae eat viral occlusion bodies, occlusion bodies (OBs) release the occlusion derived viruses (ODVs) which attack on the midgut of the larvae recorded by many researchers. These attached on the specific site of midgut like epithelial cells. Then viruses enter into the cytosol, nucleus and show the gene expression. The budded viruses (BV) invade the closely attached tracheal system or hemocytes of the insects. Then the AcMNPV spread throughout the insect body and viruses digest the insect tissues with the help of some activating enzymes, helpful for the multiplication of the viruses in the environment for further attachment.

2. *Spodoptera litura*

Cotton and vegetables are affected by various insect pests but armyworm is the major one that cause severe losses in both (cotton and vegetables) (Zhou et al., 2012; & Ramzan et al., 2019b). It spreads throughout the cotton growing areas of the world like Africa, Asia and Europe (El-Helaly, 2013), especially causes losses in southern and northern areas of Pakistan (Ahmed et al., 2016; Saleem et al., 2016; & Ramzan et al., 2019) and India (Noma et al., 2010; & Naik et al., 2017). The main crops affected by armyworm are cotton, okra, maize, berseem, cauliflowers, alfalfa, potato, cucurbits, sweet potato, legumes, groundnut and many others host in Pakistan (Maqsood et al., 2016; El-bendary & El-Helaly, 2013; & Saleem et al., 2016). It attacks on various crop parts such as stem, leaf, flower as well as seed pods also affected. The economic losses in the form of yield occurred due to severe attack of the pest (Ahmad et al., 2018).

2.1. Biology of *Spodoptera litura*

Eggs are laid in groups underside the host leaves. First instars are gregarious and then solitary in later stages. Larvae are 40 mm in length and pale to dark green in colour without any body hair. Adults are 20 and 30 mm long in length with gray-brown forewings and pale hind wings (Ramaiah & Maheswari, 2018). The time period of life cycle changes throughout the year according to regions. There are 12 generations in a year. A generation is complete in one month (CABI, 2018). There were slight morphological differences between *S. litura* male and female. The wing length of male is smaller than female and orbicular spot also present on each wing (EPPO, 2015). The period of life cycle can varies with respect to host plants as reported by many previous researchers (Ramzan et al., 2019a, b)

3. Integrated pest management options

The various control methods like cultural, mechanical, physical, biological, botanical and chemical had been adopted for the management of various crop pests (Srivastava et al., 2015; & Ramzan et al., 2019a). The resistance of different insecticides classes has been reported in the armyworm (Sayyed et al., 2012). Resistance can cause due to excessive applications of synthetic insecticides for the management of *Spodoptera litura* (Carasi et al., 2014). There are many disadvantages of improper use of insecticides against pest like environmental pollution, reduction of biological fauna, health problem as well as resistance. There is need to control insect pests by using an alternative technique to insecticides that should be ecofriendly. Entomopathogens play key role in insect pest management and NPVs is major part of these.

3.1. Control of *Spodoptera litura via* Nucleopolyhedroviruses (NPVs)

NPVs had considered an important part of integrated pest management against various insect pests (Tang et al., 2011) especially lepidopteran (Ahmad et al., 2018). A study was conducted to check the toxicity of NPV against 2^{nd} and 4^{th} instars larvae of *S. litura* belonging to different areas. Three concentrations of NPV were prepared such as NPV-34 × 10^9, NPV-23 × 10^9 and NPV-12 × 10^9 POB ml^{-1}. The study concluded that maximum (83.28%) larval mortality was recorded at high concentration. It was observed that Multan population was recorded less susceptible followed by Layyah (LY) and Faisalabad (FSD). The dire3ct impact of NPV was recorded on eggs, larvae, pupae and adults. The developmental duration of each stage was reduced by application of NPV (Yasin et al., 2020). Many early researchers (Arti & Yogita, 2014; Shaurub et al. 2014; Ayyub et al. 2019 Nawaz et al., 2019; & Zhang et al., 2015) had reported the similar findings about the mortality of larvae at different concentrations.

Another experiment was performed to evaluate the combine effect of NPV, chlorantraniliprole and *Azadirachta indica* against different instar larvae of *Helicoverpa armigera* and *S. litura*. The study showed that high mortality was recorded in combine applications while low in individual application (Wakil et al., 2012).

Another study was conducted to check the efficacy of V-*Splt*NPV against different larval instars (second, third and fourth) using various doses such as 1×10^4- 1×10^8 OBs/ml. It was observed that NPV showed high (37.65–96.82%) pathogenicity at early stage of larvae. The mortality percentage was increased with increase in time and decreased with increase in larval age (Kumar et al. 2011; & Khattab 2013). The combine application of NPV with insecticides can enhanced the pathogenicity against larvae. 100% mortality was also recorded under greenhouse conditions against second instars larvae (Ayyub et al., 2019). Many studies have been conducted to isolates the NPVs from various lepidoptera pests especially *S. litura* (Alexandre et al., 2010; Laarif et al., 2011; Kumar et al., 2011, 2012; Wu et al., 2012; Khattab, 2013; Pachippan et al., 2012; & Noune & Hauxwell, 2015).

Future recommendations

Conflict of interest

Authors declare no conflict of interest.

Authors contributions

All authors have equal contribution in writing this review.

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