Microbial Management of Crop - Pest

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ABSTRACT

In India most of the farmers depends upon synthetic pesticides for protecting their crops form pest attack. These pesticides not only caused environmental pollution, but also causing health hazardous to human being and domestic animals. This could be prevented by using bio-intensive integrated pest management (BIPM) were microbial insecticides place an important role. Here I have discussed about the important bacterial, fungal, viral, protozoan and nematode - based insecticides.

INTRODUCTION

With the globalization and liberalization of agriculture the microbial management of the crop pest has attained immense importance. All the microbial insecticides can be intensively used without any possibility of development of resistance. In view of their specificity and safety, there will be least environmental, ecological and health hazards. The use of microbes is highly sustainable and can contribute to increase the productivity and profitability in agriculture. Providing safe, economical and reliable control of crop pests is not only a contribution to sustainable agriculture but also a question of mere survival of a huge number of poor farmers in India. Microbial pesticide would offer a new method to control insecticide resistant pest population of *Helicoverpa armigera*, could prevent the rise of secondary pest problems, extend the life span of chemical insecticides, while protected the efforts and investment of the farmers and help in organic – production of vegetables, fruits and pulses etc. for domestic consumption and export (Pawar and Borikar, 2005). India is endowed with a rich bio-diversity of several viral, bacterial, fungal, protozoan and entomopathogenic nematodes of crop pests, which offer great scope in the microbial control of crop pests. Microbial pesticides are safe to mankind and animals, do not pollute the environment, do not kill beneficial parasites and predators and generally pests do not develop resistance to these microbes. In view of these , several organisms are currently being developed in India as eco-friendly biopesticides. In microbial management pathogens are utilized which may be virus, bacteria, fungi, protozoans and nematodes. Use of these pathogens may vary considerably between crops and locations depending upon climate, symptomatology and economic threshold of crop damage. In general the pathogens function naturally in the environment as population suppressors. Microbial management of H. armigera in different crops has been very extensively reviewed by Pawar and Borikar (2005).

RESULTS AND DISCUSSION

Insect viruses

Work on insect viruses in India was initiated as early as 1968 with the report of nuclear polyhedrosis virus (NPV) from *Helicoverpa armigera* (Patel *et al.,* 1968), a pest of national importance and *Spodoptera litura* (Dhandapani *et al.,* 1992), a polyphagous pest attacking several crops. Since then studies on insect viruses have progressed rapidly and several viruses were reported to occur in insect pests, most of them from the order Lepidoptera. These comprises of Nuclear Polyhedrosis Virus (NPV), granulosis virus (GV) and cytoplasmic polyhedrosis virus (CPV).

Development of NPV

Of the different types of viruses, the NPV has the greatest potential since they are more virulent, killing the insects much faster than the GV and CPV. The NPV of *Helicoverpa armigera* (HaNPV) has been studied very extensively to evaluate its efficacy as a viral pesticide. HaNPV, a single embedded virion type has a high virulence against *Helicoverpa armigera* and the techniques of mass production, formulation (Chandel *et. al.*, 2001, Grzywacz *et. al.*, 2005, Rabindra *et. al.,* 2005 and Saxena and Ahmad, 2003, 2005) and field use have been developed. The virus at a dose of 1.5-3 x 10^{12} polyhedral occlusion bodies (POB)/ha effectively controlled *Helicoverpa armigera* on crops like chickpea (Pal *et. al.,* 1998), pigeonpea (Ahmad and Saxena, 2001), groundnut (Muthuswami *et al.,* 1993), sunflower (Ahmad and Saxena, 2001), sorghum (Dhandapani *et al.,* 1993), cotton (Dhandapani *et al.,* 1987) and tomato (Mistry *et al.,*1984). Rabindra and Jayaraj (1988) used several adjuvants to increase the efficacy of the virus. Aqueous leaf extract of *Vitex negundo* increase the efficacy of the virus in the field (Rabindra *et al.,* 1991). Several new strains of NPV of *Helicoverpa armigera* (Ahmad *et. al.,* 2001), *S. litura* and *Amsacta albistriga* with increased virulence have been isolated.

S. litura a polyphagous defoliator pest has been controlled with NPV on cotton , tobacco, banana, black gram and cauliflower. A wettable powder formulation of

Table 1. Transgenic plants with pest resistance

Crop	Gene	Pest
Cotton	*B.t.* toxin Cry IA(b)	Boll worm
Maize	Syn Cry 1A (b)	*Ostrinia nubilalis*
Tomato	Cry 1 A (c)	Fruit borer
Rice (Japonica)	Modified Cry 1A (b)	Striped stem borer + *C. medinalis*
Tobacco	Cry 1 A (a)	—
Cauliflower	Cry 1 A (a)	—
Rice (Indica)	Syn Cry 1 A (b)	*Scirpophaga incertulas, Cnaphalocrosis medinalis Chilo suppressalis, Marasmia patnalis*

the virus was found effective in reducing the damage to groundnut plants (Rabindra *et. al., 2001*). The red hairy caterpillar *A. albistriga*, a gregarious seasonal pest causing extensive damage to groundnut can be controlled with NPV could induce and epizootic of the viral disease in field populations of the pest resulting in long term control of the pest (Rabindra *et. al*., 2001). The safety of NPV to several beneficial organisms like the silk worm *Bombyx mori*, honey bees *Apis cerana indica* and parasitoids and predators have been established.

Development of granulosis virus

A granulosis virus was found to control *Chilo infuscatellus* effectively (Easwaramoorthy and Santhalaxmi, 1988). The virus was found to be safe to the egg parasitoids *Trichogramma chilonis a*nd *Trichogramma japonicum*. New strains of granulosis virus was also reported from *C. infuscatellus* (Easwaramoorthy and Cory, 1990). A granulosis virus was reported from the rice leaf folder *Cnaphalocrocis medinalis* from Kerala. Recently a GV from the diamondback moth *Plutella xylostella* was reported.

Baculovirus of *Oryctes rhinoceros* L.

Attempts were made to utilize the baculovirus for the suppression of the coconut rhinoceros beetle *O. rhinoceros* and the results are encouraging (Mohan *et al*., 1993). Release of infected beetles spread the disease to subsequent generation of adults and larvae in breeding sites.

Bacillus thuringiensis

The spore forming, crystalliferous and ubiquitous bacterium *B. thuringiensis* (B.t.) is one of the earliest microbial insecticides to be commercially produced world wide. In India, this bacterial insecticide has been used in the management of several insect pests notably the diamond back moth *P. xylostella* larvae on cruciferous vegetable (Ashokan *et al*., 1996). Other insect pest controlled by *Bt* are *H. armigera* on pigeonpea and chickpea (Saxena *et. al.,* 1992 and 1995), *Autographa nigrisigna* in chickpea (Saxena and Ahmad, 1997), *Diacrisia obliqua* (Saxena *et. al*., 1992), *S. litura* on cabbage (Dutta and Sharma, 1997)and fruit borers of okra (Sathpathy and Panda, 1997). There are several reports however indicating the ineffectiveness of *Bt* preparations particularly against the noctuids. Delfin was ineffective in controlling *H. armigera* on chickpea (Saxena *et. al*., 1995). Therefore, it is that new strains of *Bt* with increased host spectrum and efficacy are developed. A few indigenously prepared *Bt* formulations are not as effective as the imported preparations. There is an urgent need to isolate indigenous *Bt* strains with high pesticidal activity, develop techniques of mass production and stable formulations and patent them. Since, insects can develop resistance to the *Bt* toxin, this bacterial insecticide should be used judiciously. Commercial formulations of *Bacillus thuringiensis* (Bt.) such as thuricide, dipel, delfin etc., have provided high mortality of *H. armigera* in laboratory as well as in field conditions (Saxena and Ahmad, 1998).

FUNGAL PATHOGENS

Fungal pathogen particularly *Beauveria bassiana, Metarhizium anisopliae, Verticillium lecanii* and *Nomuraea rileyi* have been found to be promising in the control of several agricultural pests (Lingappa *et. al*., 2005). *B. bassiana* and *M. anisopliae* were found effective against *H. armigera* and other pest of pulses (Saxena, *et. al.,* 1989 and Saxena and Ahmad, 2002) and *S. litura* (Gopalakrishnan and Narayanan, 1989), the sweet potato weevil *Cylas formicarius* , the termite *Odontotermis brunneus* and *O. obesus* (Khader Khan *et. al.,* 1993). *M. anisopliae* when applied to manure pit suppressed the population of *O. rhinoceros* larvae. The white halo fungus *V. lecanii* effectively controlled the coffee green bug *Coccus viridis* in coffee plantations (Easwaramoorthy and Jayaraj, 1978) and addition of glycerol as a humectant increased the efficacy of the fungus.

N. rileyi a fungal pathogen active against several lepidopteran insects was found to occur in epizootic form on *H. armigera* (Gopalakrishnan and Narayanan, 1989), *S. litura* and *Spodoptera exigua* (Phadke *et al.,* 1978) suppressing the pest under field conditions. While *N. rileyi* killed *Achaea janata* larvae, it was not pathogenic to *Telenomus proditor* an egg parasitoid of *A. janata* (Phadke and Rao, 1978). Saxena and Ahmad (1997a) reported *Beauveria bassiana* is effective against *H. armigera* infesting chickpea. Saxena and Chaudhary (2002) recorded two new entomogenous fungi infesting larvae of *H. armigera* in fields. They are identified as *Aspergillus flavus* and *Aspergillus niger*. *A. flavus* was found 80% pathogenic to 3rd and 4th instar larvae of *H. armigera* in laboratory (Saxena, *et al*., 2005 and Pandey *et. al*., 2007). *Nomuraea rileyi* has been established as a viable pathogen of *H. armigera* which is suitable for commercialization, as it is readily amenable to the mass production (Vimala devi, 2001). These reports clearly indicate the scope of entomopathogenic fungus for pest control in India.

ENTOMOPATHOGENIC NEMATODES (EPN)

Entomopathogenic nematodes popularly called as EPNs offer great scope for the management of many lepidopterans insect pests (Hussaini, 2001). In India , the work on EPN started in 1960 with the use of DD-136 for the control of pests of rice, sugarcane and apple. In 1970 other workers studied in the laboratory and field, the life cycle and compatibility of DD-136 with insecticides and fertilizers. Our eco-system is rich in diversity of EPN. A field survey in several locations in Gujarat during 1997 reveled 4 species of *Heterorhabditids* and 11 of *Steinernematids*. A survey in vertisols revealed abundance of *Steinernema feltiae*. Recently two new species of entomopathogenic nematodes (EPN) *i.e., Steinernema masoodi* and *S. seemae* have been reported from the larvae of *H. armigera* at Kanpur (U.P.), which is causing 67% mortality of *H. armigera* larvae in the laboratory.

PROTOZOA .

Two protozoans *Nosema* sp. and *Vairiomorpha* sp. are reported on *H. armigera*.

BIOTECHNOLOGY AND BIOPESTICIDES

There are tremendous opportunities for genetic improvement of microbial pathogens for virulence, persistence, host range, speed of kill and stability in storage. The nuclear polyhedrosis viruses have been prime targets for genetic improvement of microbial pathogens for virulence, persistence, host range, speed of kill and stability in storage. Recently, a strain of *S. litura* NPV has been isolated with a characteristic restriction profile which was more virulent than the standard Coimbatore strain by over 100 times. Development of *in vitro* systems for replication of baculoviruses in insect cells has opened new avenues for genetic improvement as well as genetic engineering of NPV with foreign genes to enhance its virulence.

***Bt* in Transgenics**

The advantages of insect resistant transgenic plants are well known. The first results of insect control by plants engineered with the crystal protein gene were demons trated in tomato, cotton and tobacco. Subsequently, several other *Bt* crops have been constructed (Table 1). 1997, over 70 transgenic crops have been approved for commercialization in nine countries plus the E.U. Many are approved for growing and human consumption particularly in the U.S.A. and Canada, whereas some are for import and human consumption of the product and over 10 crops were pending approval. Of the 80 crops or so approved or pending approval in eight countries, 21 are *B.t.* transgenic crops dominated by corn and followed by potato and cotton. The developing countries that commercialized transgenic crops are Mexico and South Africa.

Bt. expressing transgenic corn, cotton and potatoes were grown in over 2 million acres in the U.S. during 1996 with excellent results. It is estimated that by 1997 the area would have increased to 2.4 million acres. However, there are two major concerns: a. Outbreak of less susceptible secon dary pests and b. Possibility of development of resistance.

Genetic Improvement of Entomopathogenic Fungi

Culture conditions can influence the characteristics of fungul spores and can be manipulated to increase the efficiency. Blastospores of *B. bassiana* from nitrogen limited culture had higher concentration of carbohydrate and lipid and were significantly more virulent (lower LT_{50}) towards the rice green leaf hopper than blastospores from carbon limited culture (Lane *et. al.,*1991). Growth of *B. bassiana, M. anisopliae* and *Paeciomyces farinosus* on agar-based media with low water activity or with high concentration of glycerol encouraged accumulation of polyols in conidia that are more pathogenic at lower RH than produced on control media. Chemical mutagenesis, parasexual cycle, protoplast fusion and direct genetic manipulation could be used. Mutants of *M. anisopliae* and *P. farinosus* have been generated which are significantly more virulent (reduced LT_{50}) at low RH than parentals.

FUTURE OF MICROBIAL PESTICIDES IN INDIA

The baculoviruses particularly NPVs have tremendous scope for development as microbial pesticides for the management of *H. armigera* (Saxena and Ahmad, 2005) and *S. litura* on crops like cotton, gram and groundnut. The GV of *P. xylostella* is another promising candidate. However, commercial availability of quality formultions is the immediate need. Multinational companies who have the production capabilities are not interested in view of the limited market potential. A few indigenous small producers who ventured into commercial production of NPV of *H. armigera* and *S. litura* did not succeed in producing quality virus. Most of the samples tested had extremely low virus content if not no virus at all. Many had unacceptable levels of spores of microsporidians. NPV production is done *in vivo* in respective host larvae and hence producers should employ properly trained manpower for production of host insects as well as the virus.

Most of the *B.t.* products now being sold in India are very expensive since they are imported. One or two products developed indigenously in India are not as effective as the imported ones. A few institutions in India including the TNAU and the BARC have isolated indigenous *B.t.* strains which are as potent as the standard HD 1. There is ample scope for development of techniques of indigenous production and formulation of native *Bts*. Species of entomopathogenic fungi like *Metarhizium, Beauveria, Verticillium* and *Nomuraea* isolated from different agroecosystems in India have shown promise in pest control. We should genetically improve these strains and develop techniques of mass production and stable formulations. India with the rich biodiversity of organisms, has tremendous potential for development of native strains of microbials well adapted to the Indian subcontinent.

There has been some success in the use of pathogens such as *Bacillus thuringiensis, N. rileyi* and *Helicoverpa armigera* nuclear polyhedrosis virus (*Ha*NPV). However, the relatively high cost and rapid inactivation by ultraviolet light often lead to poor performance under field conditions. In the case of *Ha*NPV, the difficulty in obtaining consistently high levels of purity and virulence necessary to achieve satisfactory control have limited its usefulness. The increasing prevalence of resistance to insecticides and awareness of environmental concerns has given a new impetus to the development of suitable microbial insecticides for use in IPM of *H. armigera*.

Educational programmes in institutions should give special emphasis on development of biorational pest control technologies like microbial control. There is a need to develop inter-institutional collaborations in order to pool all the resources available in different institutions and develop microbial pest control technologies which would ultimately reduce the pressure on chemical insecticide use.

In view of the new patent laws, it is very important that we take special care to preserve our entomopathogenic microbial wealth. We should characterise all indigenous microbial species with pest control potential and develop a suitable mechanism to patent them in due course of time. Microbial management is a very broad concept. A number of strategies and techniques are involved. Many pathogens operate unsuspected and it is amazing how complex bio-ecological interaction is going on. For the promotion of microbial pesticides, their intensive commercial production and formulation with strict enforcement of quality control measures are necessary.

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