20

Efficacy of Biopesticides and Natural Plant Products for *Heliothis/ Helicoverpa* Control

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Biopesticides based upon entomopathogenic bacteria, fungi and insect viruses such as Nuclear Polyhedrosis Virus (NPV) have the potential to play an important role for the management of cotton bollworm/legume pod borer [*Helicoverpa armigera* (Hübner)]. While there is significant research interest in protozoa and nematodes, these are not as yet sufficiently developed to have a clear role in *H. armigera* control. In addition, this chapter will also review the use of botanical pesticides in the control of *H. armigera*. Here, the definition of biopesticides includes the use of fungi, bacteria, viruses, protozoa and nematodes for the biological control of insect pests (Dent and Jenkins 2000). Natural plant products will cover all crude or refined extracts of any plant or part thereof.

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Why alternatives to existing chemical insecticides are needed?

Viable and sustainable control of *H. armigera* using the conventional approach of relying primarily on chemical insecticides has become increasingly expensive and unreliable over the last two decades. Increasing resistance to chemical insecticides has been the primary cause of control failures. However, the increasing unacceptability of using broad-spectrum insecticides—because of concerns about the environmental impact, safety and residues in food products—has stimulated the search for alternatives to synthetic insecticides.

Biopesticides and plant products offer potentially effective and safe techniques for pest control. The particular advantage of biopesticides derives not only from their capacity to kill the insect pests, in which they act like a conventional chemical pesticide, but also their unique capacity to reproduce and compound the killing action over time (Thomas and Waage 1996). Additionally, many biopesticides show a degree of specificity for controlling *H. armigera* that makes them safer than conventional chemical pesticides, many of which have an adverse impact on non-target fauna and the environment. Biopesticides and natural plant products also have the advantage that they can be produced locally, thus providing a sustainable local resource that can compete cost effectively with increasingly expensive imported chemical pesticides.

Given these apparent advantages of biopesticides and plant products, a question must be asked as to why are these not used by the farmers. Here, we must admit that biopesticides currently have a variety of real limitations related to the speed of kill, cost effectiveness, availability and activity spectrum. However, by examining these constraints in detail, we can seek to identify how research efforts can be focused on overcoming these problems so that the full potential of these valuable resources can be realized in improving *H. armigera* management.

Helicoverpa armigera nucleopolyhedrovirus (HaNPV)

Currently a major focus of interest in biopesticides for the control of *H. armigera* is on the use of nucleopolyhedrovirus (NPV) (Plate 20.1). NPVs are naturally occurring pathogens of *H. armigera*, and have wide distribution in Asia, Africa and Australia. Strains of these viruses have been developed as commercial biopesticides in America, Australia, India, China and Thailand. HaNPV has been shown to be highly effective in controlling *H. armigera* on a range of crops, including legumes (Rabindra et al. 1992; Cherry et al. 2000), oilseeds (Rabindra et al. 1985), cotton (Jones 1994) and vegetables (Ketunuti and Tantichodok 1990; Jones et al. 1998). However, apart from Australia, their use by farmers is still restricted to niche markets and high-value horticultural crops, and they are not yet widely used on major field crops attacked by *H. armigera*. Therefore, there is a need to take a critical look as to why its uptake is so restricted. It may be possible then to identify the technical, commercial,

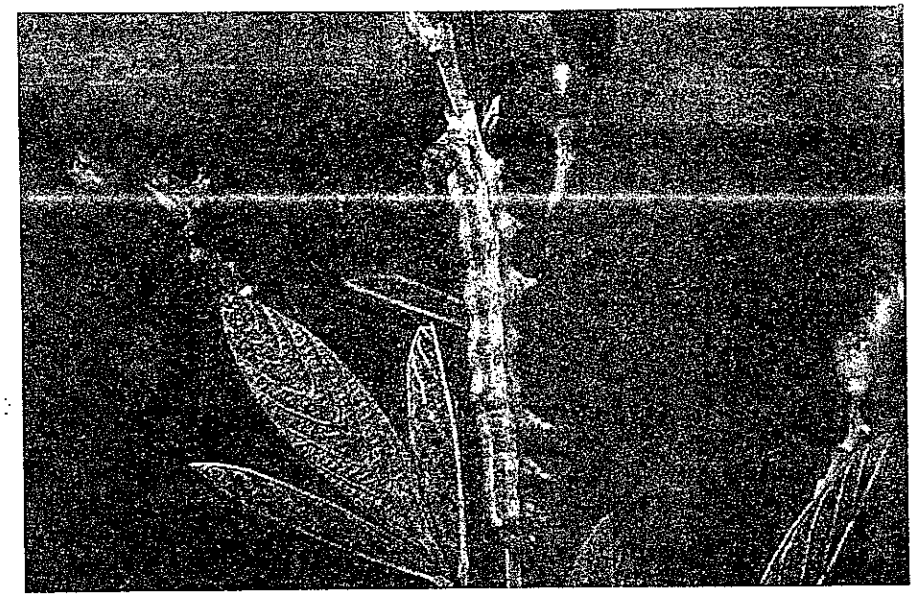

Plate 20.1: *Helicoverpa armigera* larvae infected by nucleopolyhedrosis virus.

perceptual and regulatory constraints that have limited the use of HaNPV. This, in turn, will enable researchers, producers, extension workers and regulators to identify where the future efforts need to be concentrated so as to overcome these constraints. We should be aware that there are still key factors in terms of efficacy, cost and availability that limit the effectiveness of HaNPV. Although we may wish to promote alternatives to existing chemical pesticides, these efforts will be fruitless unless the new solutions we are promoting are acceptable to farmers in terms of efficacy, reliability and cost.

Production of HaNPV

Production is a significant constraint for NPVs, both in volume and cost. The *in vivo* production of NPV in live insects is alien to most agrochemical companies, and many are loath to adopt it. *In vitro* tissue culture is under development (Reid and Weiss 2000). Although significant progress has been reported with HaNPV, a commercially viable system seems 5 to 10 years away at least. However, while mainstream agrochemical companies have avoided NPV (Harris 1997), smaller producers in the USA, Brazil, China and India have shown that *in vivo* mass production in insects is both technically and commercially viable (Entwistle 1998; Moscardi 1999). The cost of HaNPV products, however, still tends to be higher than for synthetic chemicals and reduction in production costs will be crucial to any expansion of the market (Lisansky 1997).

Most commercial mass production of the NPV in India and Asia have been carried out using insects reared in insectaries. This alone, many producers believe, is the way to ensure a quality product with adequate HaNPV titre. However, in Brazil, where two million hectares of soybean is treated annually with *Anticarsia gemmatalis* NPV (AgNPV), most producers use an 'in-field' technique infecting natural host outbreaks in specially raised crops, and this enables them to produce NPV at a cost of US$ 1.26 per ha (Moscardi 1999). This option should also be researched fully in relation to producing HaNPV in countries such as India, Nepal and Africa, where there is a need for low-cost locally produced material.

HaNPV is, in fact, a prime candidate for field-based production. Host larvae typically occur in highly dense populations, often in well-defined seasonal peaks, and are among the most productive in terms of NPV with a late-instar larvae containing $>10^{10}$ progeny virus polyhedra at death. In a series of proof of concept trials in Australia, it has been recently demonstrated that sufficient HaNPV can be generated in one hectare of lucerne to treat 10,000 to 20,000 ha of target crop at a commercial application rate of 5×10^{11} polyhedra ha^{-1}.

Although in-field HaNPV production is an attractive proposition, this virus has one drawback that AgNPV does not have. As is the case for nearly all other NPVs, HaNPV causes rapid breakdown of the host insect cuticle during the final stages of infection, resulting in dispersal of progeny virus particles into the environment. Host cuticle breakdown is probably an adaptation by an NPV to maximize its transmission rate, but it has the unhappy consequence of constraining the economical recovery of virus. In contrast, AgNPV does not disrupt the cuticle of the infected host insect and this allows for the high efficiency recovery of AgNPV in field-based production systems (Moscardi 1997).

But there may be a solution to this problem. In well-studied systems (e.g. *Autographa* and *Spodoptera* NPVs), insect cuticle breakdown has been shown to be due to the expression of enzymes encoded by two viral genes, the chitinase and cathepsin (Hawtin et al. 1997; Thomas et al. 2000). It is likely that in AgNPV, these genes are either absent, non-functional or under-expressed. Since we know that the efficiency of insect cuticle breakdown is highly variable among HaNPVs, it should be possible to identify naturally occurring isolates deficient in chitinase/cathepsin activity for use as virus-seeding material for in-field production systems. The kind of quantitative-diagnostic methods necessary for building large collections of virus isolates for screening purposes have now been developed (Richards and Christian 1999; Christian et al. 2001), and it should be relatively straight forward to develop a program to achieve this objective.

There is also the question as to who should produce the NPV, particularly if it is to get to the poor farmers, whose livelihoods are the most at risk. Currently, international companies in North America and Europe are producing NPV, but it is questionable whether these can ever meet the potentially huge needs in Asia and Africa for low cost products. In both India and Thailand, biopesticide producers are both state funded and commercial companies (Grzywacz and Warburton 1999). These are basically producing for the local market, and are also seeking to meet potentially lucrative export markets. It may be that some form of local or community production may be the only way to meet the needs of the poorest. Here, ICRISAT and its partners have (in IFAD project) pioneered the development of low-cost production systems that can be used by individual farmers or village cooperatives to produce the NPV. However, achieving and maintaining quality control is a serious problem for small producers (Kennedy et al. 1999; Tripp and Arif 2001). Research is needed to determine whether adequate quality control systems can be incorporated in small-scale production systems, and if this approach is to have a significant future.

Efficacy of HaNPV

The problem of slower kill of NPV even now discourages many farmers from adopting NPV. An important question is: can we train farmers to move away from their habit of waiting until the last minute to treat pests? Can we induce them to adopt scouting and appropriate threshold treatment so as to make NPV effective? Much of the IPM training now focuses on getting the farmers to adopt scouting and decision making in order to rationalize the use of agrochemical inputs and reduce the costs. If these efforts succeed, they should induce farmers to successfully utilize biopesticides.

There is also the problem of poor efficacy on some major target crops. It would be valuable to identify the crop factors that favor or impair the functioning of NPV. On some crops, NPV appears to perform very well and low doses can be used, e.g. HaNPV on sorghum (Murray et al. 2000), but on cotton and chickpea, it appears that plant factors impair its efficacy (Forschler et al. 1992; Hoover et al. 1998). Research is needed to identify the precise mechanisms that cause the problem. Only then can we go on to develop appropriate formulations to overcome this problem.

Strain selection also offers considerable potential for improving HaNPV insecticides. Pathogenicity and virulence (taken here to mean lethality and speed of host kill, respectively) are known to vary considerably both between species (Bianchi et al. 2000), and among geographic variants of the same HaNPV (Narang et al. 2001). Presently, we have little idea of the range of genetic variability represented within the HaNPV group, although data are beginning to emerge, which indicates that it is significantly under exploited.

The co-evolution theory predicts that lepidopteran NPVs, which are usually only transmitted following host death, have evolved towards an intermediate level of virulence as there is a trade-off between the kill rate and progeny virus production (May and Anderson 1983). According to this theory,

NPV strains that kill hosts too quickly will be selected because too few progeny viruses are produced to sustain transmission. If this theory holds true, it may be presumed that maladapted 'super virulent' NPV strains will occasionally arise, but these will be quickly out-competed by strains that have optimized virulence and reproduction for maximum transmission. Therefore, there may be scope to screen naturally occurring virus populations for fast-acting HaNPV insecticides. Another key trait for which selection seems possible is the improved stability on the leaf surface and resistance to environmental degradation, particularly to solar inactivation.

In order to identify HaNPV isolates with improved insecticidal characteristics, there is a need to understand more fully the genetic determinants of NPV phenotype. To this end, much is promised by 'functional genomics'. The entire genomic sequence of HaNPV is now known (Chen et al. 2002) and the task of matching gene(s) with complex traits such as virulence probably lies some way off. It may be possible, however, to fast-track this process using genomic markers to facilitate sampling of the range of genetic variation in virus populations, perhaps fast-tracking this process through controlled application of appropriate selection pressure in well-designed experimental systems.

Genetic enhancement

Genetic enhancement provides an alternative means of improving virus activity, and to this end, a variety of recombinant 'rapid action' NPVs have been developed and tested in the laboratory and field. These include NPVs transformed to express insect-selective nerve toxins (Stewart et al. 1991), hormones that disrupt pest development (Bonning and Hammock 1996) and proteases that perforate pest cell membranes, hastening the systemic spread of infection (Harrison and Bonning 2001).

Genetic enhancement has the potential to enhance speed of action of NPV. In a recent study, the median lethal time (LT_{50}) for an *H. zea* NPV strain expressing an insect-selective scorpion toxin was demonstrated at 2.5 days compared to 6 days for the unmodified virus (Treacy et al. 2000). Given that the transgenic era is still young, we may reasonably expect further improvements in NPV performance. One potentially fruitful area of research is the manipulation of the host range, as success here will pave the way for viral insecticides that can be tailored to specific pest complexes.

Nevertheless, it should be borne in mind that serious impediments to the commercial development of recombinant NPV insecticides still remain unsolved. One of these is a cost-effective means of mass production, since feasibility of high volume *in vitro* systems has not yet been established, and *in vivo* production for rapid action NPVs is unlikely to be commercially viable. The other significant barrier is registration. Currently, regulatory oversight of Genetically Modified Organisms (GMOs) is extremely cautious, and this has

undoubtedly extended development time, increased costs, and impacted on commercial confidence of an early return on investment. To date, several prototype products, including a rapid-action *H. zea* NPV insecticide, have been successfully tested in the field, although none has been registered for commercial use. For the present, at least, commercial activity appears to have been entirely stalled.

Formulation

There is a need for improved formulations with long shelf lives at ambient temperatures. Currently, NPV formulations need to be refrigerated or kept in the deep-freeze if they are to maintain long-term viability (Burges and Jones 1998). This is not tenable in an Indian or African context, where many users do not have access to suitable storage. There is a need to develop simple stable formulations that can match the stability of chemical insecticides at more than two years (Couch and Ignoffo 1981; Cherry et al. 1996; Jenkins and Grzywacz 2000). The unpurified aqueous formulations currently produced in India also often smell bad due to bacterially mediated decomposition of insect-derived lipids. Cheap methods are needed for cleaning up or suppressing bacterial action without impairing NPV viability. New formulations are needed to overcome the limited efficacy of NPV on key crops such as cotton and chickpea. The poor UV stability of NPV has, of course, been a significant problem in promoting their use in the tropics where functional persistence times can be as short as a day on unshaded foliage. Here, the improvements in formulation chemistry and the development of cheap UV absorbers compatible with NPV could prove to be a real breakthrough.

Regulation, quality control and safety of HaNPV products

It is important that a regulatory system for biopesticides does not keep out small producers by an excessive registration burden (Jayanth and Manjunath 2000). To date, lack of regulation has encouraged the development of biopesticides in India, but imposition of expensive chemical-type registration systems may halt the progress. However, some registration system is probably essential to prevent the proliferation of substandard products that would destroy the consumer confidence.

It is important with any biopesticide such as NPV to maintain high quality product. To date, in a number of countries, the production of biopesticides has been accompanied by serious failings in quality control. Here, responsible companies are clearly aware that this issue is crucial to long-term growth of these products (Jayanth and Manjunath 2000). But there are reports that many products are clearly failing adequate quality standards and this risks a loss of farmer confidence (Kennedy et al. 1999). To put this in perspective, the same issue has also been identified in respect of chemical pesticides and pheromones,

where adulteration or dilution of active ingredients to save on production costs also has been found to be widespread in countries with inadequate consumer protection apparatus.

In considering the safety of HaNPV, we are on very firm grounds in considering these as among the safest and most environmentally benign of available pest control technologies. There is some fifty years of data on NPVs, both as natural pathogens as augmentative biocontrol agents and as bioinsecticides, and there is no evidence of significant harmful effects on non-target organisms (Black et al. 1997; Cory 2001).

Competing technologies

In examining the future for NPV, we must consider the impact of alternative technologies. The appearance of transgenic plants with insect-resistant traits may be a major tool in overcoming many insect problems, including *H. armigera*. These may be seen as competing technologies to biopesticides such as NPV. However, the need for refugia to slow the development of resistance in pests such as *H. armigera* may itself create new opportunities for biopesticides as key elements in a sustainable transgenic plant strategy.

It has been a constant refrain of the chemical industry that there is no need to look to alternatives, as 'new chemicals' will solve the resistance and environmental problems. It is true that there are some very exciting new chemicals that are much more efficacious and more environmentally acceptable. However, given the propensity of *H. armigera* to develop resistance, it may be considered unlikely that a sustainable chemical-only solution will be developed in the next five to ten years. In fact, the move to look for more selective, environmentally acceptable chemicals may improve the prospects for biocontrol agents, as farmers learn to live without the rapid action broad-spectrum insecticides they have become used to. In addition, new chemicals tend to be much more expensive than the old ones and the increased cost of chemical control may improve the market for biopesticides such as NPV.

Growth sectors for HaNPV uptake

In the insect resistance management program in field crops, an example is from Australia where NPV finds role as part of management program using NPV early in the season to delay the point when contact insecticides need to be used (Murray et al. 2000). Another promising market is in high value horticultural crops. In Thailand, NPV is used to protect crops such as asparagus, okra, and tangerines destined for export (Jones et al. 1998). Here, farmers take advantage of the safety of NPV to the consumers to spray at times close to harvesting, when the use of chemicals would be unacceptable to importing countries. This is especially important in crops destined for countries such as Japan and European Union (EU), where any chemical residues can make the

crop unsaleable. There are also forthcoming EU regulations to make chemical residues an issue in cut flowers as these will aim to protect flower handlers in the supply chain from exposure to high chemical residues. Regulations in the EU and the USA are increasingly banning many existing chemical pesticides for environmental and safety reasons (both for use on crops and as residues in produce). New opportunities may arise for NPV for controlling some highly resistant insect pests, where the existing chemicals have become ineffective and *H. armigera* is likely to be one of the target pests.

Finally, we may suggest that India would be an ideal place to produce many biopesticides such as NPV. These biopesticides are produced using low capital technologies, but with a high labor requirement. In India, lower labor costs and a growing commercial biopesticide sector may make it a major producer of NPVs, both for local use as also for export.

Bacterial and fungal pathogens of *Helicoverpa*

Both bacteria and fungi can be pathogenic and kill *Helicoverpa* (Luthy et al. 1982). Among bacteria, *Bacillus thuringiensis* (*Bt*) is the most researched. It is an aerobic, gram-positive endospore-forming bacterium, and is widespread in natural environments such as compost and soil. It produces large insecticidal protein crystals (ICPs) during sporulation. Its first record goes back to 1901, but its first practical application was made in 1938, which led to the first commercial *Bt* product 'Sporeine' in France (Luthy et al. 1982). A major breakthrough came with the appearance of two commercial *Bt* pesticide products 'Thuricide' and 'Dipel' in 1960s. But the market share of different *Bt* products is quite low, at <1% of the total pesticide market (Navon 2000). *Bt* toxins have been reported to kill insects among the Lepidoptera, Coleoptera, Diptera (Hofte and Whiteley 1989) and nematodes (Feitelson et al. 1992).

The crystalline insecticidal protein (toxins) are referred as *Cry* toxins. These toxins are coded by genes on plasmids or on chromosomes. There are 5 to 6 different toxins of molecular weight 40 to 150 kDa expressed by a given *Bt* strain. Structure of at least three crystal proteins Cry 3A, Cry 1Aa and Cry 2A have been studied by x-ray crystallography (Li et al. 1991, 1996). At sporulation, *Bt* produces an inert polypeptide protoxin, which is often bi-pyramidal. This delta toxin targets the midgut (tubular epithelium) of the insect. Lepidopteran gut has an electrogenic K^+ pump in the apical membrane. The toxin induces pore formation in the membrane, thus causing osmotic imbalance and then vesiculation of the endoplasmic reticulum. This mechanism is specific to target insect species and as mammals lack the receptor sites for *Bt* binding, the Cry toxins are harmless to them.

Much of *Bt* research and product development is focused on the 'toxins' it produces. *Bt* screening programs are based on potency bioassays. In that, the proteins are isolated, purified, and studied for their efficacy to kill insect larvae (Kaur 2000). Formulation additives include wetting agents, stickers,

sunscreens, synergists and phagostimulants, and seem to focus on protecting the toxin (Navon et al. 1997; Navon 2000). Research in this area (generally in private sector) seems to constitute a major cost in *Bt* product development.

Toxin-centered product development and protection of crystal protein from UV inactivation suggests that not only will the products be expensive; they may not be widely effective, particularly in low cost subsistence agriculture in semi-arid tropics. *Bt* products have been indicated to have a narrow host range (Navon 2000). Also, these are most effective on early-instar larvae (Navon et al. 1990), necessitating frequent sprays. Some insects have also been reported to be resistant to *Bt* toxins (Commandeur and Komen 1992). Much of the research on *Bt* has been done by or supported by pesticide industry, for which a stand-alone product on the lines of chemicals is an established model of marketing. Efforts are on to develop strains where toxin is encapsulated or UV protected (Gelernler and Schwab 1993). Use of feeding stimulants to increase ingestion and, in turn, the efficacy of microbes is another researchable area (Gelernler and Schwab 1993). Genetic modification, combining toxin proteins of two or more *Bt* strains in one product, have also been reported (Wu et al. 1994). Much of the work is focused on developing insect-resistant transgenic crops by transferring toxin genes from *Bt* to crop plants, such as *Bt* cotton and *Bt* maize. Looking at the strengths and weaknesses of the *Bt*, it seems likely that we may manage *Helicoverpa* more effectively by using this agent in combination with other management techniques (such as trap crops and natural enemy enhancement) rather than with existing chemical pesticides.

Fungal pathogens

Entomopathogenic fungi have for a long time been recognized as important natural enemies of *H. armigera*. Fifteen fungal species have been reported to be promising myco-insecticides. Species pathogenic to insect pests are *Metarrhizium anisopliae* (Metsch.) (Plate 20.2), *M. flavoviride* (Metsch.), *Nomuraea rileyi* (Farlow) Samson, *Beauveria bassiana* (Balsamo) and *Paecilomyces farinosus*. Rangaswami et al. (1968) discussed the scope of controlling *Helicoverpa* with *M. anisopliae*. *Nomuraea releyi* was isolated as a major pathogen in an outbreak in Mississippi (Smith et al. 1976). Aima (1975) reported 50% *Helicoverpa* mortality due to epizootics of *P. farinosus*. During an *Helicoverpa* outbreak, Abbaiah et al. (1988) isolated *Beauveria* spp. Nurindah and Indrayani (1989) reported *B. bassiana* and *N. releyi* as most important pathogens of *Helicoverpa* in cotton in Indonesia. Both *B. bassiana* and *M. anisopliae* have been reported as major pathogens of *Helicoverpa* in Hunan, China (Jing 1999).

Based on laboratory studies on *M. anisopliae* Gopalakrishnan and Narayanan (1989) reported 80 to 100% mortality of all five instars and pupae of *Helicoverpa* in 2 to 10 days using 1.8×10^9 conidia mL^{-1}. Sixty to 100% mortality of *Helicoverpa* larvae was reported with application of 1.0×10^7 conidia mL^{-1} of *B. bassiana* (Gopalakrishnan and Narayanan (1990). Third- to fifth-instar larvae

Plate 20.2: *Helicoverpa armigera* larva infected by the fungus, *Beauveria bassiana*.

of *Helicoverpa* are more susceptible to infection by *N. releyi* than first- and second-instars (Mohamed et al. 1977).

Mass production of different entomopathogenic fungi may not be difficult. A carrot medium was reported to be suitable for multiplication of *M. anisopliae* and Zapek Dox Broth (containing 2% chitin and 3% molasses) for good growth and sporulation of most entomopathogenic fungi (Srinivasan 1997). For commercial scale production, however, one would require a solid state fermentation system.

Adhesion of fungal spores to host cuticle and their germination is a prerequisite for efficacy of fungal pathogens. It is widely accepted that 90% relative humidity (RH) is required for germination of fungal spores, a big handicap in the widespread use of such biopesticides. However, special formulations of fungi in oil can overcome this problem by creating high RH microclimates around the spores, enabling entomopathogenic fungi to function at low RH environment (Bateman et al. 1993).

On germination, the fungus penetrates the cuticle (setae and intersegmental membrane) of the insect, and grows in hemocoel of the insect and eventually the larvae die. The fungus also grows saprophytically (producing toxins), and in due course, the hyphae re-emerge (cuticle-out) and sporulate. In case of infected larvae, fungal growth is visible, particularly when

humidity is high. Field application of 2.8×10^5 spores mL^{-1} of *B. bassiana* was found to be highly effective in reducing *Helicoverpa* damage in chickpea (Saxena and Ahmed 1997).

Safety of fungi and bacteria

Biopesticides are often developed from species that are ubiquitous in natural environments. Residual effect of the biopesticides is, thus, of less importance. Therefore, the regulatory criteria used for their release as commercial products may not be as stringent as for chemicals. Even so, their effect on beneficial insects, allergenicity and pathogenicity to humans must be evaluated. Several studies involving *Bt* indicate that chemical pesticides are not compatible with some biopesticides. For example, some insecticides have a significant antifeeding effect, while a *Bt* preparation has to be ingested for a desired effect. Benomyl resistant *B. bassiana* strain, without significant loss of pathogenicity to *Helicoverpa*, has been reported by Sandhu et al. (2001). Application of *N. releyi* (10^{12} spores ha^{-1}) along with HaNPV (250 larval equivalents) effectively controlled *H. armigera* in tomato (Srinivasan 1997). In field experiments in India, both NPV and *Bt* in different combinations improved yield from 5 to 73% over control, but application of endosulfan was the most effective (92% more yield over control) (Pawar 1998).

Bacteria and fungi—the journey ahead

Spurious chemicals in the market are a common occurrence, largely due to laxity in the enforcement of regulations. Although, the reason(s) of reported suicides by farmers (particularly in India) are debatable, the significant role of failure of chemicals in protecting crops cannot be ruled out easily. Enhanced awareness about the environmental problems associated with chemical pesticides and their residues in food as health hazard strongly suggest a decline in their use in future. Newer pesticides, being developed to be more selective and safer than the old generation of chemicals, are likely to be much more expensive and beyond the reach of resource-poor farmers. Also, insects may develop resistance to these insecticides. In such a scenario, biopesticides remain quite promising. There is a need to develop strategies on using the biopesticides either alone or in combination with synthetics. Screening *Bt* strains on the basis of their ability to kill larvae instead of rating these on the basis of potency of the toxin they produce is suggested. This may lead to identification of new strains that are effective in managing *H. armigera*.

Another important question is the use of formulations essential in the case of bacteria and fungi. This possibility needs to be examined. It has been argued that formulation is essential. It often plays a role in making a product easier to handle and improving its reliability (Jones et al. 1997). If we can avoid the use of expensive formulations, it should be possible to deliver *Bt* strains to farmers at prices close to those of rhizobial inoculants (<2 US$ per ha). It may then be affordable to spray them frequently, which seems necessary for effective control of *Helicoverpa*. If formulations are not essential, multiplication and distribution of bacteria and fungi can be developed as a small scale enterprise as has been demonstrated in India for rhizobial inoculants. Also, there is a need to shift the focus of research from the 'industry' to 'farmers'. For example, a well-fertilized crop attracts more insect pests than otherwise (Phelan et al. 1995). Can we think of change(s) in fertilizer (particularly N) application schedule in such a way that we achieve desired plant growth/yield, but attract fewer pests? Raising crops for harvesting high yield requires knowledge and timely action, particularly for protecting them from insect pests. Setting up information kiosks at a village level in developing countries can be important in effectively managing *Helicoverpa*.

Natural plant products

Increasing environmental pollution and health hazards associated with the use of synthetic insecticides has resulted in renewed interest in the development and use of botanical products for insect pest management. A large number of plant products, viz. extract of seeds of custard apple, karanj oil, neem oil, neem leaf extract, neem seed kernel extract, tobacco decoction, nicotine sulfate, etc., have been tested against *H. armigera*. So far, neem has dominated the international literature on botanicals. Also, neem has generally been stated as the most effective botanical. Studies on extracts of four botanicals (neem 1%, *Annona* 1%, Jatraofa 1% and Mahua 1%) (Vyas et al. 1999) suggested that neem and *Annona* killed all *Helicoverpa* larvae, while 30 to 70% pupation was noted with Jatraofa and Mahua compared to 83% pupation in untreated control. Field efficacy and subsequent commercial development of botanical insecticides derived from the neem tree have changed our basic assumptions about how a natural product can be useful for plant protection on a commercial scale (Shankar and Parmar 1999; Sharma et al. 1999). A large number of commercial formulations of neem, e.g. Neemguard, Neemax, Achook and Repelin are available in the market (Shankar and Parmar 1999; Vyas et al. 1999).

Neem (*Azadirachta indica* A. Juss) is indigenous to India. It is widespread in many Asian and African countries. Neem derivatives have traditionally been used by farmers against household, agricultural and pests of medical importance. Neem derivatives comprise a complex array of novel compounds with profound behavioral and physiological effects such as repellence, phagodeterrance, growth disruption, oviposition inhibition, etc. Some of these effects are attributed to bitter principles such as azadirachtin, salannin and meliantriol that occur abundantly in the seed kernel (Shankar and Parmar 1999; Sharma et al. 1999). Recently, four new tricyclic diterpenoids have been isolated from the neem bark. These diterpenes possess a wide range of

biological activities including hypocholesterolemic, antitumor, antileukemic, antibiotic, plant cell expansion, cellular division inhibition and insecticidal properties. The complexity of the chemical structures of these precludes their synthesis on a practical scale. Therefore, the use of neem leaf and seed kernel extract and neem oil have been recommended for pest management. While neem is active against a wide range of insect pests, it is known to have little or no effect against beneficial spiders, ladybird beetles, parasitic wasps and predatory mites (Walter 1999).

If we take the international neem conference of February 1996 in Australia (Singh and Saxena 1999) as an indicator, much of the applied research on neem has been reported from India (12 out of 29 papers). Papers on formulations, mechanisms of the action and chemistry of active ingredients were generally from the developed world. Only two of the 29 papers reported some work with *Helicoverpa*. In India, neem has been evaluated against 100 species of insects, 11 nematodes and at least 4 fungi (Singh, 1990). Butani and Mittal (1990) reported that neem seed kernel suspension (NSKE) 3% was as good as malathion 0.05% and DDT (0.2%) in reducing the larval population of *H. armigera*. Sachan and Lal (1990) reported that neem leaf extract, neem seed kernel extract (NSKE), neem oil and karanj oil (*Pongamia glabra*) were field-evaluated against *H. armigera* on chickpea and pigeonpea. Results showed that the use of 5% NSKE was highly effective and almost at par with the recommended insecticide endosulfan (0.07%) for controlling *H. armigera*.

Several experiments conducted with neem leaf and neem seed kernel extract as part of the All India Coordinated Pulses Improvement Project (AICPIP), reported increases in yield (upto 24%) with the use of neem products compared to that of control. Some scientists reported on the positive effect of commercial preparations also (Saxena, H., unpublished).

The way forward for biopesticides and botanical pesticides

Evidence shows that biopesticides and botanical pesticides have considerable potential in integrated management of *H. armigera*. They can provide a safe and effective tool for farmers to use, and can be produced locally in countries of South Asia and Africa, where this pest is prevalent. There are researchable issues that need to be addressed in order to improve the efficacy, availability and cost effectiveness so that they can compete successfully with chemical pesticides.

In developing practical cost-effective biopesticides and botanical pesticides, there is a need to move research from the traditional single discipline approach that has characterized much of public sector biopesticides research into a multidisciplinary approach that characterizes successful industrial R & D. Many biopesticide researchers have adequate skills in the initial stages of pathogen isolation, strain selection, characterization and evaluation, but lack the full range of skills needed to develop promising pathogens into successful

commercial products (Harris and Dent 2000). This downstream work involves a different mix of disciplines, many of them including process engineering, economics, formulation chemistry, packaging and marketing. It is only by including these elements in the development process that cost-effective biopesticides can be developed. As many of these skills reside primarily in the private sector, it is probably only through effective public–private sector research partnerships that real progress will probably be made.

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Heliothis/Helicoverpa Management

Emerging Trends and Strategies for Future Research

Editor

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