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Advances in Insect Biocontrol

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Introduction:

Biological control can be defined as the use of natural enemies to reduce the damage caused by an insect pest population. It is a tactic or approach that fits into an overall insect pest management system (IPMS) and represents a potential alternative strategy to combat with the insect pest damages to the agriculture & forestry agriculture. Manipulation of natural enemies under BC approach for insect pest control can be achieved by Introducing, Augmenting or conserving naturally available bio agents. BC Approaches draw theory and practice together, indicating where advancement in understanding may contribute for improvement in management. There is an ample scope for further improvement through advanced research in bio control field like

- Chitinase-producing bacteria
- Entomopathogenic nematodes as biocontrol agents in orchards
- Advances in formulation and application of entomopathogenic fungi (EPF) in Storage
- Double strand RNA delivery system for plant sap-feeding insects.

Plant protection is based on a good deal of knowledge of agro-ecosystem as well as information about the identification of target insect pests, assessment of damage caused, preventive measures, interaction of plant-environment-pest and use of effective bio control agent. To reduce the crop losses we mostly dependent on the use of chemical pesticides, but the major problem of using these chemicals are that target organisms often develop resistance to them, impact of pesticides on non-target organisms, as well as on human health, potential harm that these chemicals cause to the environment and to animal health, leaching into ground water damages both aquatic environments and drinking water resources.

Recent advances in biocontrol:

1. **Chitinase-Producing Bacteria and Their Role in Biocontrol:**

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Chitin is an important component of the exteriors of insects and fungi. Upon degradation of chitin by a number of organisms, severe damage and even death may occur in pests whose external surfaces contain this polymer. Chitin synthesis is limited to insects and fungi, many of which are cause damage to plant so this molecule is a logical target for pest control. Chitinolytic microorganisms are likely to play an important role as bio-control agents which may become a potential alternative to chemical fungicides and insecticides. Chitinolytic enzymes will become a more obvious and important solution towards overcoming the environmental and human hazards that result from the application of synthetic pesticides.

Mode of Action:

Microbial Chitinases weaken and degrade the cell walls of many pests and pathogens, thereby exhibiting anti-fungal, insecticidal or nematicidal activity. Degradation of chitin is catalyzed by chitinases, which are found in the organisms containing chitin, mainly insects, crustaceans, and fungi. These enzymes are detected in viruses, bacteria, protista, higher plants, and animals. Chitinolytic bacteria decompose chitin in both aerobic and anaerobic conditions and are found in a wide range of habitats. In the soil and rhizosphere, bacteria use chitin from insects and fungi as a carbon and nitrogen source. Chitinases have also been demonstrated to affect insect growth, both feeding rate and body weight of larvae decrease if they are in contact with chitinases which ultimately leads to death. *Orgyia pseudotsugata* peritrophic membrane was degraded by chitinases and later this effect was also observed in vivo with *Spodoptera littoralis* with chitinase from *Serratia marescens*. These symptoms are attributed to the weakening of the peritrophic membrane that lines the gut epithelium of the larvae, the main component of which is chitin.

2. **EPN as Biocontrol Agents of Insect Pests in Orchard:**

EPNs are increasingly being used as biological control of insect pests due to their successful biocontrol activity against various economically important insect pests. These EPNs kill insects with the aid of a mutualistic symbiosis with a bacterium *Xenorhabdus spp.* and *Photorhabdus spp.* for EPN genera *Steinernema* and *Heterorhabditis*. Discovery of many efficacious isolates/strains and significant advances in mass production and formulation technology during last decade make it popular BC agent. They can easily be produced in liquid culture. The outcome of an EPN application is influenced to a great extent by environmental parameters such as temperature, moisture, soil type, salinity and organic content of soil, exposure to ultraviolet light, means of application, agrochemicals etc. Despite the growing success of EPNs as biological control agents of an increasing number of target insect pests there are some limitations.

Limitation

Limiting factors for using them in orchards IPM program are low-temperature & desiccation reduced control activity & persistence, when applied against aboveground pest stages. Higher costs and inferior performance of EPNs compared with chemical insecticides in many situations. There is inconsistent performance of EPNs due to their large dependence on a variety of biotic and abiotic parameters (Tim Belien, 2018).

3. **Recent Advances in Formulation and Application of EPF for Biocontrol of Stored-Grain Insects:**

Several liquid and dry formulations of EPF strains were developed and used against storage insects but commercially registered products are missing. There is increasing evidence that EPF have great potential for control of insects in the storage environment but isolating new and effective strains, with the improved forms of natural diatomaceous earth (DE) needed. The oil-based liquid formulations have no environmental side-effects or harmful toxic effects on treated plants or plant products since they are mainly composed of natural substances. Globally, oil-based liquid formulations such as invert emulsions (water-in-oil type) are the most promising and most appropriate for formulation of effective strains of EPF. Invert emulsions emulsion contain the necessary water for conidial germination during application under hot dry storage conditions. Dried conidia of EPF were thoroughly mixed with other natural dusts then applied either by mixing thoroughly with the grains these combinations enhanced the efficacy compared to the use of conidia alone. The EPF has synergistic effect with DE which attributed to the abrasion effect of DE particles on insect cuticle, so penetration of applied fungal hyphae increases and thus infection increases (Batta, 2016).

Therefore, in order to achieve an effective control of these insects, the following potential areas of future research are i) Screenings for new effective strains of EPF are needed in addition to new effective formulations ii) Applying the most effective strains and formulations selected in the previous step under storage conditions iii) We have to optimize the proportions of ingredients in the selected formulations iv) Integrating the products of the most effective formulations, after registration and commercialization, in the IPM programs of stored-grain insects.

4. **Double strand RNA Delivery System for Plant Sap-Feeding Insects**

This dsRNA-mediated gene silencing is also known as RNA interference (RNAi) represents a potential tool for the management of sucking insect pests. Introduction of exogenously synthesized dsRNA into an organism are triggering highly efficient gene silencing through the degradation of endogenous RNA homologous. Deliveries of dsRNA to piercing-sucking insects are major challenge. Effective and practical use of RNAi as molecular biopesticides for biocontrol of insects in the environment requires that dsRNA to be delivered *in vivo* through ingestion. Therefore, the key challenge for molecular biologists in developing insect-specific molecular biopesticides is to find effective and reliable methods for practical delivery of stable dsRNAs such as through oral ingestion.

Oral delivery of dsRNA into Insects:

RNAi is a well described gene regulatory mechanism wherein exogenous dsRNA is introduced into the cells which cause targets degradation of host cell mRNAs (complementary to the dsRNA). Delivery of dsRNAs by injection is tedious and impracticable for developing a successful biopesticide. One of the first bioassays inducing RNAi through oral ingestion of dsRNA through artificial diet in a chewing insect was demonstrated in the Western corn rootworm resulting in larval stunting as well as mortality. Other method of oral delivery are by droplet feeding in diamond back moth *larvae*, blood meal in tsetse fly, dsRNA-soaked paper disks in termite and through dsRNA sprayed leaves in weevils etc.

Sap feeding Hemipteran delivery of dsRNA must be into vascular tissues which is very difficult task. Transgenic plants expressing species-specific dsRNA were used to silence genes in the cotton bollworm indicating a steady progress towards RNAi technology.

Vegetable Mediated Delivery of dsRNA

Delivery of exogenously synthesized dsRNA to the BMSB (*Halyomorpha halys)* by using a vegetable delivery system in a natural diet and significant induction of the RNAi mechanism was first time successful demonstrated. The use of highly attractive vegetables, fruits which are not used for human consumption, may be used for delivery of dsRNA, for development of biopesticide. For biocontrol of BMSB, *Phaseolus vulgaris* were selected as the medium for delivery of dsRNA. A solution of green food coloring was mixed at 1:10 ratio with water to imitate dsRNA. Slender green beans were trimmed from the calyx, inverted and immersed into food coloring solution. Successful feeding on the bean with green food coloring was evidenced by green excreta, which indicated the material had been delivered orally and had passed through the gut before excretion. This demonstrated that BMSB-specific dsRNA was successfully transported through the bean vascular tissues to the style. RNAi has emerged as a potential tool in functional gene regulation that may be of great importance in future years for the control of insect pests. RNAi biopesticide development is dependent upon the ability to deliver the specific dsRNA to the insect. This study successfully demonstrated the use of a vegetable, green bean, to deliver dsRNA designed to specifically impact and reduce an insect pest of global importance, BMSB by reducing JHAMT and Vg gene expression

Slow progress in RNAi biopesticide development**:**

- Lack of efficient delivery methods
- The *in vivo* testing through methods of ingestion,
- Efficiency of gene regulation by the RNAi machinery
- Durability of resistance
- Feasibility for application under field conditions (Ghosh *et al* 2017).

Conclusion:

Novel biocontrol agents can serve as alternatives to pesticides and there is less possibility of resistance development against this approach. Integration of novel biocontrol agent in plant protection makes our production sustainable and ecologically friendly. They provide a significant advancement towards safer ecological pest management. These products have started to appear in the market and should be a priority goal for the future. We need more research in this area to make our dream practically possible and socially acceptable among farmers and consumers.

Reference

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