

## Role of Entomopathogenic Fungi in Insect Pest Management

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### SUMMARY

Entomopathogenic fungi are considered to play an important role as biological control agent of insect populations. Entomopathogenic fungi were among the first organisms to be used for the biological control of pests. Entomopathogenic fungi (EPF) are widely distributed with both restricted and wide host ranges which have different biocontrol potentials against arthropods insects and plant pathogenic fungi. A very diverse array of fungal species is found from different classes that infect insects. These insect pathogenic species are found in a wide range of adaptations and infecting capacities including obligate and facultative pathogens. Spreading of fungal diseases is common in many insect species while some species may not be affected. In 1980s, the first insect pathogenic studies were carried out and their focus was to find the methods of disease management of the silkworm. Bassi in 1835, first time formulated the germ theory by the use of white muscardine fungus on the silkworm that was then named in his honor as *Beauveria bassiana*. This silkworm disease gave the idea of using insect infecting fungi for the control of insect pest management. Many commercial products are available globally that are formulated by utilization of less than ten species of fungi

### INTRODUCTION

A group of fungi that kill an insect by attacking and infecting its insect host is called entomopathogenic fungi. The main route of entrance of the entomopathogen is through integument and it may also infect the insect by ingestion method or through the wounds or trachea. The divisions of fungi are Ascomycota, Zygomycota and Deuteromycota, and the divisions Oomycota and Chytridiomycota were also included in the previous classification of fungi. More than 700 species of fungi from around 90 genera are pathogenic to insects. EPF are a major component of IPM techniques as biological control agents against insect pests and other arthropods and are an integral part of mycoinsecticides in horticulture, forestry and agriculture. The mass production of Hyphomycete fungi is cheap. Its storage is very easy and it is disciplined on a wide range of humidity and temperature. Insect pathogenic fungi are being developed and produced in mass production globally to control a wide range of harmful insects of crops. More than 300 insect infecting species are present in Cordyceps that is best known ascomycete. Chalkbrood disease in bees occurs by the genus *Ascosphaera*, which has dimorphism in sexuality. The spores of *Ascosphaera*, in an unusual manner of infecting species are ingested by the bee larvae that germinate in gut causing infection. The most important insect infecting species occur in *Aspergillus*, *Beauveria*, *Metarhizium*, *Hirsutella*, *Aschersonia*, *Culicinomyces*, *Lecanicillium*, *Paecilomyces*, *Tolypocladium* and *Sorosporella*. Mostly, these genera have a linkage with one or many genera that can be verified with biological studies or by the molecular studies presenting the genetic relationship between telemorphs and anamorphs.

### Biology and Pathogenesis

The entomopathogenic fungi have life cycles that synchronize with insect host stages and environmental conditions. During life cycle insect pathogenic fungi, an infective spore stage are required generally that germinates on the host cuticle. It forms a germ tube penetrating the host cuticle the infecting fungal spores then increase in number, causing toxin production that in the end kills the insect.

### Classification of Entomopathogenic Fungi:

Division	Class	Order	Family	Genus
<i>Zygomycota</i>	Zygomycetes	Entomophthorales	Entomophthoraceae	<i>Entomophaga</i>
				<i>Entomophthora</i>
				<i>Erynia</i>
				<i>Eryniopsis</i>
				<i>Zoophthora</i>

				<i>Massospora</i>
				<i>Strongwellsea</i>
				<i>Furia</i>
				<i>Tarichium</i>
				<i>Pandora</i>
			Neozygitaceae	<i>Neozygites</i>
<i>Ascomycota</i>	Sordariomycetes	Hypocreales	Clavicipitaceae	<i>Beauveria</i>
				<i>Metarhizium</i>
				<i>Nomuraea</i>
				<i>Lecanicillium</i>
				<i>Cordyceps</i>
				<i>Cordycepioideus</i>

Host plays major two types of roles for fungus that is either active (under favorable situations, fungi need to contact with host) or passive (as it is the main source of nutrition for fungus). It is difficult to describe the role of environmental factors for land-living fungi. In most cases, for all developmental stages of fungi, high humidity is required while for infection, temperature is limiting within certain ranges. The optimum temperature for Hyphomycetes is between 20-30°C while for Entomophthorales, it is considered as 15-25°C. The optimum temperature for all stages of fungus development is different for individuals of same species. Some species require high humidity than others such as *Lecanicillium lecanii* requires about 16 h of 100% RH to kill whiteflies at leaf level. While some species require low relative humidity, such as *Entomophthora muscae*, that causes infection in Diptera. It is considered difficult to have an estimate of the actual humidity due to variability in microclimate. Soil moisture, organic matter in soil and pH are important for infection.

### Specificity and Host Range

In the population of a particular host, individuals are not equally susceptible to infection. Different species of hosts are also not equally susceptible to infection from a particular species or strain of the pathogen. On the one hand, different species or even strains of the fungus may display different levels of virulence and parasitic specialization against a specific host. Many other hosts have been reported in the recent years including species of Lepidoptera, Coleoptera, Diptera, Hymenoptera and Homoptera. The prevalent hosts of *M. anisopliae* are mostly coleopteran species that include more than 70 scarab species. The insect pathogenic fungus, *Zoophthora radicans* has been reported from over 80 species of insects of Coleoptera, Lepidoptera, Diptera and Homoptera. Generally, fungi produce toxin and have the capacity to infect vertebrates so it called allergens. Thus, fungi are an important source of allergic reactions in humans. However, the fungi that cause infections in insects are not allergic to human beings. Over all, vertebrates are not susceptible to the insect infecting fungi. In exception cases *Aspergillus flavus* is famous pathogen of vertebrates. This is why; they are not used for commercial production.

### Mode of Action and Host Reactions

The mode of action of Entomopathogenic fungi varies and kills the insect by different ways such as causing starvation to toxin production. These Entomopathogenic fungi produce many toxins and extracellular enzymes such as proteases and chitinases. Entomopathogenic fungi need to penetrate through the cuticle into the insect body to obtain nutrients for their growth and reproduction. Entry into the host involves both enzymic degradation and mechanical pressure as evidenced by the physical separation of lamellae by penetrated hyphae. The process of infection starts by contact of the spores to the host cuticle. Sometimes, conidium attaches to the cuticle or secrete mucus for adhesion during its germination and swelling. Some infective sessile spores (Capilliconidia) have an adhesion drop at spore end to aid attachment for insect penetration. Some structures and general processes are involved in the penetration of host cuticle and the mechanisms of each fungus may also differ. Development of certain structures from conidia to aid attachment and germ tube penetration is occurring by *M. anisopliae*. The infection pegs (Appressoria), aid the germ tube penetration. After the penetration of germ tube through the cuticle and insect epidermis, the fungus multiplication is occur into the body cavity of insect. In some Entomophthorales,

this multiplication may be by the protoplasts while in some hyphomycete (e.g., *M. anisopliae*), blastospores are involved in the initial proliferation. With the presence of cuticle, some other humoral and cellular defense methods are used by the insect against invasion of fungus. Toxins are produced by some insect pathogenic fungi and more of them aid to increase pathogenesis and play an insecticidal role. While some other such fungi produce antimicrobial metabolites. Destruxins of *Metarhizium* species are among such fungal metabolites that increase the pathogenicity of fungus. The toxins Beauvericin, Bassianolide, Isarolides, and Beauverolides have been isolated from *B. bassiana* infected hosts. Toxins Destruxins (DTXs) and Cytochalasins have been isolated from *M. anisopliae* infected hosts. The toxins have shown to have diverse effects on various insect tissues. DTX depolarizes the lepidopteran muscle membrane by activating calcium channels. In addition, function of insect hemocytes can be inhibited by DTX. In the initial stages of infection, no considerable behavioral symptoms are observed. But, some days before death, symptoms start to appear such as reduced co-ordination, feeding (e.g., grasshopper and locust infected with *M. anisopliae*) and activity. Some other behavioral responses include behavioral fever, increased feeding (e.g., Colorado potato beetles infected with *B. bassiana*), positive or negative photo- or geotropism and altered mating. “Behavioral fever”, is another response of fungal infection in which the insect changes its body temperature by basking in the sun or using warm surfaces for their positioning e.g., response of locusts and grasshoppers in the infection of *M. anisopliae* and *B. bassiana*.

### **Transmission and Dispersal**

Different strategies are used by the fungi to enhance the chances of encountering new host. For the groups producing abundant spores such as Hyphomycetes, rain, wind and invertebrates help in transmission of spores. Major source of spore distribution is Wind. Growing hyphae out of insect cadaver is also a major source of conidia dispersion. For the transmission of terrestrial fungi, High humidity and moisture are required for germination and sporulation. Fungus ability to infect multiple stages of life of an insect is helpful in disease spreading. For example, winged insects can spread spores and ultimately the disease in insect populations.

### **Interactions between Pathogens and Other Natural Enemies**

Many insects have numerous species of natural enemies such as parasitoids, predators and other pathogens. They have a competition among them for same food resource. There may be chances that a single insect may be infected by one or more natural enemies or pathogens. An interaction is found among them such as natural enemies can feed upon host that is already infected with pathogen. While on the other hand, Entomopathogenic fungus may infect natural enemies or other non-target insects. Another aspect is that these natural enemies and non-target insects become a source of fungal spore's dispersion.

### **Epizootiology and Its Role in Suppressing Pest Populations**

Insect outbreaks are quickly reduced by the use of entomopathogenic fungi through epizootics. A vital factor in controlling insect pest population is natural epizootics. Epizootiology occurs in different environments.

### **Epigeal Environment**

Epigeal environment is suitable for epizootics. Many sources of infection are in this environment such as from infected insects to non-infected insects, from the conidia on leaves to those insects feeding on leaves and through dispersion of spores in the air surrounding. Major occurrence is mostly found in aphid species.

### **Soil Environment**

Natural epizootics mostly prevail due to the hidden nature of soil living insects. The soil may act as a storage place for spores. It is seen that soil is major inoculum source of many Hyphomycetes. Most of the epizootics have been noted in the stages of insect that live in soil. Many researchers have reported the spores present in the soils are of the genera *Metarhizium*, *Beauveria*, *Tolypocladium* and *Isaria*. *Cordyceps* spp. (Ascomycetes) has been found on soil surface from the insect cadavers. During the spring, for epizootics initiation, presence of spores in the upper layers of soil surface is important in the epigeal environment.

### Aquatic Environment

This environment is quite from soil environment. The Entomopathogenic species that infect aquatic insects have motile spores, zoospores. Disease can be affected by a wide range of biotic and abiotic factors such as pH, salinity, organic and inorganic pollutants and water movement. Another major influencing source is temperature affecting disease and epizootiology.

### Development as Microbial Control Agents

When the living organisms are used for control pests, this is called as Inundative biological control. Some characteristics of Entomophthoralean fungi suggest that these can be used for Inundative biological control. For this purpose these are produced in masses and commercial production is also done. These are mostly produced due to wide host range, ease of production, shelf life, application and persistence. *B. bassiana* is broadly used commercially to use against a broad range of insect pests such as banana weevils, pine caterpillars, European corn borer and greenhouse aphids etc. These formulations are in the form of wettable powders. Commercial products of *M. anisopliae* are also available against a wide range of pests such as locusts and grasshopper. Formulations of *I. fumosoroseus* are also available against thrips, whiteflies, aphids and spider mites. Several other products are also available in many nations.

### CONCLUSION

Since centuries ago, fungi have always been used for medicinal and other beneficial proposes and they are just as important nowadays. In this review, we summarized the advantages and applications of fungi as biopesticides, attempted and collected the know-ledge about the entomopathogenic fungi as biocontrol agents. Though, a number of studies have been done for the improvements in production, pesticide formulation and practical application, even then many improvements are required to search, study and implementation. Improvement in strains by the use of guides and selections will be a best strategy in the future. The use of microbial insecticides should be a contribution towards all fields of agriculture, sustainable agriculture, forestry and horticulture. It should be cared that the Entomopathogenic fungus should not destroy beneficial natural fauna in the environment. Genetic and proteomic studies are expected to be the main tools for the future development of the entomopathogenic fungi as biocontrol agents; however, in the near future, there will be a wider array of techniques become available to biologists which will enable us to take full advantage of entomopathogenic fungi.

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