

ENVIRONMENTAL FACTORS AFFECTING ENTOMOPATHOGENIC FUNGI IN THE SOIL

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ABSTRACT - In the last decade, chemical pesticide substitutes for the organochlorine compounds have been less persistent and effective as soil insect controls resulting in more soil pests with greater importance in agriculture. Concurrently, environmental quality has become a major issue in the use of certain chemical pesticides in the soil resulting in more cancellations and restrictions for use. Because of environmental concerns and the astronomical increase in pesticide developmental costs, agrochemical companies worldwide are, for the first time, making a serious commitment to the development of biopesticides for soil insect control. Fungi within the Zygomycotina, the Ascomycotina and the Deuteromycotina occur in many soil habitats where many insects spend their lives permanently or, most frequently, temporarily in the soil. From this association, 11 fungal genera have evolved as pathogens of soil-inhabiting insects. The genera *Beauveria*, *Metarhizium* and *Paecilomyces*, commonly infect all stages of many insect species and have been scrutinized as potential biopesticides. However entomopathogenic fungi of the soil, just as subterranean insects, are affected by abiotic factors, mainly temperature, relative humidity or soil water content, agrochemicals and soil composition. Biotic factors such as other soil microorganisms, soil arthropods and even plants influence their survival. The interactions between the microflora and the fauna of the soil are numerous and are greatly influenced by physiochemical factors of the habitat.

Index terms: biopesticides, abiotic and biotic factors, soil pests.

FATORES AMBIENTAIS QUE AFETAM FUNGOS ENTOMOPATOGÊNICOS NO SOLO

RESUMO - Na última década, pesticidas químicos utilizados como substitutos dos compostos organoclorados, provaram ser menos persistentes e eficientes no controle dos insetos-pragas. Como resultado, houve incremento dessas pragas, com maior prejuízo para a agricultura. Ao mesmo tempo, a qualidade ambiental tornou-se uma questão relevante no uso de certos pesticidas químicos, ocorrendo a diminuição e limitação do seu uso. Devido à preocupação com o meio ambiente e o inflacionamento dos custos envolvidos com o desenvolvimento de pesticidas, as empresas agroquímicas estão se comprometendo seriamente com o desenvolvimento de biopesticidas para controlar esses insetos-pragas. Fungos de *Zygomycotina*, *Ascomycotina* e *Deuteromycotina* ocorrem em diversos ambientes onde muitos insetos-pragas passam as suas vidas permanentemente, ou, o que é mais comum, temporariamente no solo. Desta associação, onze gêneros de fungos evoluíram como patógenos dos insetos-pragas. Os gêneros *Beauveria*, *Metarhizium* e *Paecilomyces* infetam comumente todos os estágios de muitas espécies de insetos e foram detectados potencialmente como biopesticidas. Entretanto, fungos entomopatogênicos, como os insetos subterrâneos, são afetados por fatores abióticos, principalmente temperatura, umidade relativa do ar, ou conteúdo de água no solo, agroquímicos e composição do solo. Fatores bióticos como outros microorganismos, artrópodes, e mesmo as plantas, influenciam a sua sobrevivência. As interações entre microflora e fauna são numerosas e influenciadas consideravelmente por fatores fisiológicos do ambiente.

Termos para indexação: biopesticidas, fatores bióticos e abióticos, insetos-pragas no solo.

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INTRODUCTION

Entopathogenic fungi occur throughout the fungal kingdom on all developmental stages of many insect species. In the soil, the close contact of the insect, particularly the larval stage, with mycopathogens frequently results in wide spread disease of the host population by one or more fungi. Eleven fungal genera contain species that are considered true pathogens, of which, a few are being scrutinized as microbial control agents (McCoy et al. 1988, Keller & Zimmermann 1989, McCoy 1990). The most common fungi being considered as microbial control agents for soil insects are species in the genera *Beauveria*, *Metarhizium* and *Paecilomyces*.

The interactions between the physical and biological factors of the soil are complex and greatly influence the soil habitat. These soil characteristics, affect the survivorship of entomopathogenic fungi as well as their ability to infect a given host. Realistically, performance of fungi as biological or microbial control agents of soil arthropods will change from one location to another according to soil characteristics. This paper presents a brief review of the abiotic and biotic factors that influence the survivorship of entomopathogenic fungi in the soil. For a more detailed understanding of this phenomena, readers should refer to Lai (1984), Keller & Zimmermann (1989) and Duniway & McCoy (1990).

INFLUENCE OF ABIOTIC FACTORS

Of the many abiotic (physical) factors that affect the function of entomopathogenic fungi in the soil, temperature, soil moisture, soil type, and agrochemicals appear most critical.

Temperature is a key factor in the growth and development of all organisms. For entomopathogenic fungi, the mean optimum temperature for growth and conidial germination normally is between 20 and 25° C, the maximum is about 35° C and the

minimum 5° C. (Roberts & Campbell 1977). However, the optimal temperature for function in the soil can differ depending on the fungal strain. Strains of *Metarhizium* with lower temperature thresholds have been selected and isolated from insects found in soils from temperate regions with prolonged low temperatures (Rath & Yip 1989) and strains of *Beauveria bassiana* have been identified from insects collected in tropical soils that have greater conidial survivorship at high temperatures (McCoy & Boucias 1989). Obviously, the optimal temperature for a given fungal strain is crucial to where they will be used as a microbial control agent.

Soil moisture is of important in maintaining fungal growth and conidial viability in soil. In addition, water is a factor in the migration or movement of the fungal pathogen and its host. Soil surface moisture compared to moisture at different depths will vary greatly in both space and time. Fungal conidia tend to survive well in conditions of high temperature and high humidity or low temperature and low humidity, but as higher temperatures and low humidity survival drops dramatically (Duniway & McCoy 1990). Generally speaking, conidial germination in soil will occur at virtually all moisture levels but a given fungal strain will have an optimum level. Water saturated clay loam soils (water holding capacity >35%) appear to inhibit host infection by *B. bassiana* while no inhibition occurred in sandy soil (Duniway & McCoy 1990).

The first mycelial granular commercial formulation of *Metarhizium anisopliae* (BIO 1020) was developed recently by Bayer AG for the control of soil insects (Andersch et al. 1990). The granule requires rehydration by soil moisture in order to sporulate, therefore, abiotic factors will affect conidiation kinetics of the mycelial granules in different soils. Without conidiation, there can be no host infection and no pest control. Studies by Storey et al. (1990) showed that temperature and soil moisture are key factors to conidiation in the soil. In addition, inoculum

density per given volume of soil was affected significantly by the number of granules applied to the soil.

Water in the form of rain also has a influence on the vertical movement of entomopathogenic fungi in different soils. Naturally, the distribution of the fungal conidia within the soil will impact on the effectiveness of the mycopathogen as a biological control. For example, maintenance of a high conidial density at or near the soil surface is imperative for infecting neonatal larvae entering the soil from a plant host.

Research suggests that the movement of conidia differs according to the composition of the soil type (Keller & Zimmermann 1989). Studies by Storey & Gardner (1987) with *B. bassiana* conidia formulated as a wettable powder showed that the percentage of fungal propagules recovered from the upper 5 cm of soil columns was positively correlated with sand composition and negatively with clay or silt. In tests with the above formulation of *B. bassiana*, McCoy (1989) applied the fungus to Candler fine sandy soil (no organic matter) with daily sprinkler irrigation (1.3 cm, daily). Results showed a gradual decrease in fungal propagule density in time in the upper 5 cm of soil suggesting a leaching effect. Unformulated conidia of *Nomuraea rileyi* were retained by a silt-loam soil probably because they adhered to the clay or organic particles (Ignoffo et al. 1977).

Since dry conidia are hydrophobic in nature, they are difficult to suspend in water. When applied to the soil the conidia are often formulated in a water suspension using a detergent such as Tween. It is possible to alter the hydrophobic properties of the conidia by adding a detergent, which may change their behavior in the soil and thus affect the way they attach to soil particles and the insect host (Duniway & McCoy 1990). Also, this may affect how they obtain the chemical cues necessary for conidial germination.

Pesticides applied on or in the soil for control of plant pathogens (fungicides),

insects (insecticides) or weeds (herbicides), as well as, fertilizers can directly or indirectly effect the survivorship of entomopathogenic fungi in the soil (McCoy et al. 1988, Keller & Zimmermann 1989). Although fungicides appear most disruptive, the degree to which pesticides affect fungal survival differs greatly. In pest management, the side-effect of pesticides should always be considered.

INFLUENCE OF BIOTIC FACTORS

The interaction between entomopathogenic fungi and their insect host can be influenced by biotic factors such as other soil microorganisms, soil arthropods, and even plant exudates.

Many soil-inhabiting insects have evolved a microflora on the outside of the cuticle and this microflora has conferred protection against other fungi that would normally be pathogenic. In experiments with *Metarhizium anisopliae* and its curculionid host, Schabel (1976) detected complete suppression of conidial germination on the non-sterile adult cuticle suggesting strong antibiosis. Surface-sterilized insects resulted in high conidial germination.

Fungistasis is a major factor in survival of conidia in nonsterile soil. Numerous investigations have shown that conidial germination and pathogenicity of many entomopathogenic fungi are suppressed in non-sterile soil, while germination and mycelial growth occurred in sterile soil (Clerk 1969, Lingg & Donaldson 1981, Keller & Zimmermann 1989, Duniway & McCoy 1990). Fungistatic effect has been attributed to actinomycetes (Huber 1958), soil bacteria and protozoa (Fargues et al. 1983) and various soil fungi (Walstad et al. 1970). Blastospores, conidia, germ tubes of germinating conidia, and mycelia appear susceptible to microbial attack while non-germinated resting spores of the entomophthorales appear most resistant. The addition of a carbon and/or nitrogen source to nonsterile soil dramatically reduced

conidial of *B. bassiana* by enhancing growth of antibiotic producing microbes (Lingg & Donaldson 1981).

Small soil arthropods, especially mites and Collembola, have been reported as being important in the dispersal of entomopathogenic fungi in soil (Keller & Zimmermann 1989). Fungistatic factors associated with certain arthropods can also influence the survival of entomopathogenic fungi. Storey & McCoy (1990), found that the venom alkaloids of the fire ant, *Solenopsis invicta*, can delay conidial germination and induce the hyphal body phase in *B. bassiana*, *in vitro*. In addition, fungistatic components, independent of venom alkaloids, were detected in non-sterile compared to sterile ant mound soil.

Little is known about the effect of plant roots and root exudates on the activity of entomopathogenic fungi. However, Zimmermann (1984) working on the control of black vine weevil with *M. anisopliae* found that the pathogen was less effective in cyclamen culture than in that of other plants.

In conclusion, it would appear that successful use of entomopathogenic fungi as classical biological control agents or biopesticides for soil-inhabiting insects depends on careful experimentation focused on a particular agricultural practice and on the chemical, nutritional and moisture inputs of substrates into soil systems.

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