Chapter 11
Entomopathogenic Fungi

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11.1 Introduction

Biological control, defined as the reduction of pest populations by natural enemies, is an alternative to synthetic chemical pesticides and usually involves human intervention (Hoffmann and Frodsham 1993). Biological control agents are classified as predators, parasitoids, and pathogens (the latter are produced by microorganisms and cause diseases). Many microorganisms, such as biopesticides, have been used because they offer additional benefits beyond their objective function (Glare et al. 2012). Furthermore, they are easy to handle, multiply, and formulate, exhibiting high pest control effectiveness levels. Entomopathogenic fungi (EPFs) are highlighted within this group and have been widely studied.

More than 1000 species included in approximately 100 genera are currently known as EPFs; they affect insects of different orders and their use as biopesticides has increased during the last decades (Shah et al. 2009; Vega et al. 2012). Most EPF species are found in the Hypocreales (Ascomycota) group, which have a wide range of hosts, and Entomophthoromycota (Zygomycota), which are more specific. Although different in some aspects, both groups produce conidia or other asexual spores that are infective units (Furlong and Pell 2005; Roy et al. 2006), and they constitute a fundamental element when developing biocontrollers. An important aspect to consider in the biology of these fungi is that they can act as obligate or facultative arthropod pathogens (Goettel et al. 2005). The latter condition allows them to live as saprophytes, in which they are able to survive at the expense of

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organic matter from the soil or other substrate and as endophytes in plants (Vega et al. 2008).

One of the most relevant EPF groups for practical pest control purposes consists of the *Metarhizium* and *Beauveria* genera. They have been most studied worldwide because they are frequently found in nature and have a large number of hosts, their control is very effective, and they are simple to use in the development of commercial products. They are characterized for producing free conidia from reproductive structures (conidiophores) that are easily transportable by the wind. EPFs are widely distributed in different ecosystems and can be found in different geographic and climatic locations in both cultivated and natural soils (Vega et al. 2008). They can infect at different stages of insect development, such as eggs, larvae, pupae, and adults. Although these fungi are known as pest control agents, they have also demonstrated their ability to protect their host from diseases and limit damage caused by pathogenic microorganisms (Arnold et al. 2003; Ownley et al. 2010).

The aim of this chapter is to review the importance of entomopathogenic fungi in the context of sustainable agriculture and crop production for biological control of pests.

11.2 How Do Entomopathogenic Fungi (EPFs) Behave?

EPFs are related to insects because they use chitin as a source of carbon, which is the main component of their exoskeleton; their conidia adhere to the bodies of the insects and the cycle begins (Fig. 11.1). The life cycle is divided into a parasitic phase, which starts with infection and lasts until the host dies, and a saprophytic phase, which takes place after the death of the insect. In the first phase, conidia make contact with the insect and the germination tube is formed; the appressorium or haustorium, through mechanical (hyphal pressure) and enzymatic (lipases, proteases, and chitinases) action, penetrates the cuticle and a micropore is produced by which it advances toward the interior of the insect and reaches the hemocele (Vega et al. 2012). When it reaches the hemocele, a transition from germinative growth to vegetative growth occurs, in which metabolic changes allow the fungus to utilize the nutrients for growth and reproduction. After producing substantial biomass, the fungus generates toxins and degrading enzymes that saturate the immune system of the insect and alter its metabolism by primarily affecting its nervous system, Malpighian tubules (excretory system), and other organs because it is unable to curtail fungal development (Roberts and Humber 1981). The secondary metabolites produced by EPF have antifungal and antibacterial properties (Wagner and Lewis 2000; Parine et al. 2010). Hundreds of small molecules with insecticidal activity have been identified from EPF, such as destruxins, oosporein, beauvericin, bassianolide, bassianin, beauveriolides, bassiacridin, cordycepin, and ciclosporin among others (Hamil et al. 1969; Susuki et al. 1977; Quesada-Moraga et al. 2006; Wang and Wang 2017). The action of these metabolites induces to the host-mediated behavioral changes of the insect, such as behavioral fever, elevation seeking, reduced or increased activity,

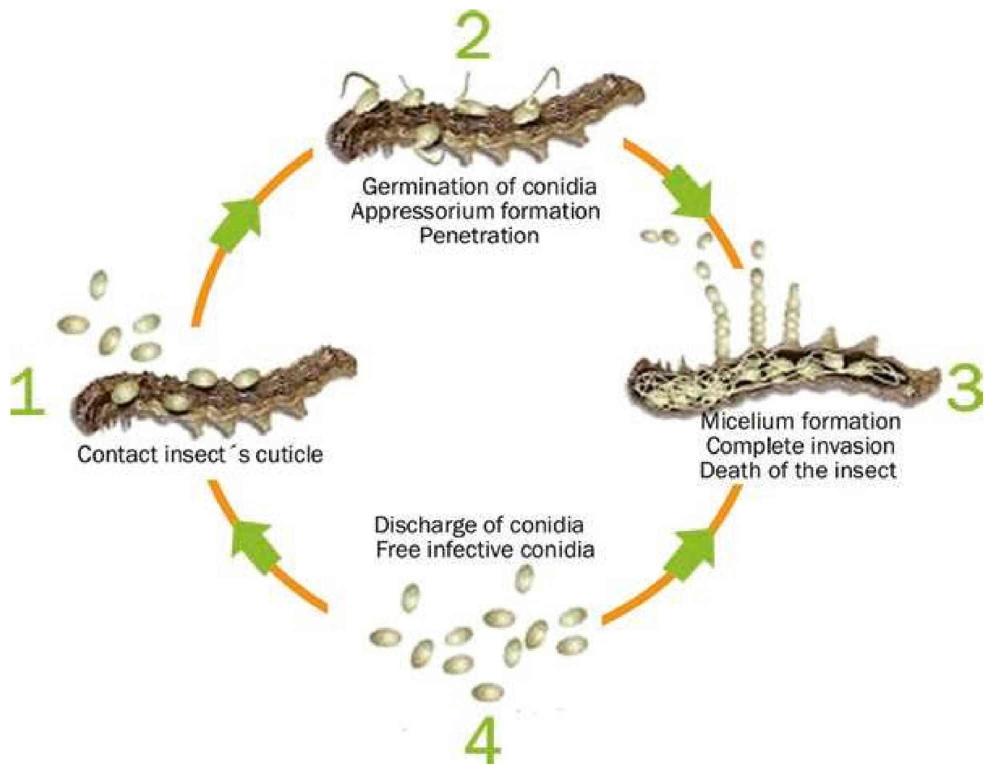

Fig. 11.1 Life cycle of entomopathogenic fungi. (Figure from INIA Quilamapu)

reduced response to semiochemicals, changes in reproductive behavior, and death (Roy et al. 2006).

The death of the insect can occur from the second day after infection, thus initiating the second phase of the cycle. The fungus has the ability to go through the cuticle of the insect and go outside where it continues its saprophytic development on the cadavers, which rapidly produces conidia or other asexual propagules that are converted into new foci of fungal dissemination (Meyling and Eilenberg 2007). EPFs have a life cycle that is synchronized with the insect stages and environmental conditions (Shah and Pell 2003).

11.3 Characteristics of Entomopathogenic Fungi (EPFs) for Biological Pest Control

During the last decades, attention has been focused on EPF development, mainly *Beauveria bassiana* (Balsamo-Crivelli) Vuillemin and *Metarhizium anisopliae* (Metchnikoff) Sorokin as inundative biopesticides (Faria and Wraight 2007). Licensed commercial products exist that have formulations containing conidia and/or mycelium for field application (Faria and Wraight 2007; Lacey et al. 2015). They are very important for the development of sustainable agriculture, significantly contribute to integrated pest management, and respond to an increasing world demand for ecologically compatible and environmentally friendly products. Some characteristics of this ecological implication include:

- Safety: EPFs are usually safe for humans and it is still unknown if they have a negative impact on the environment (Vega et al. 2008). However, there are some reports of problems caused by *Metarhizium* spp. in immunodepressed individuals (Nourrisson et al. 2017).
- Resistance: No reports have yet been found of insects that have developed resistance to EPF. However, there are studies that demonstrate insect response to infection as a result of their ability to produce detoxification enzymes, antibiotic secretions, and immune responses (Serebrov et al. 2006; Vega et al. 2012).
- Selectivity: This point is debatable because strains have been detected that are pathogens of one species; however, with the commercial development of EPF strains in the last few years, some have been found that can be parasites for an important group of insects.
- Persistence: Given the condition of facultative parasite organisms, the possibility of these fungi to persist in the soil as saprophytes awaiting new insects to appear is quite probable.
- Residues: They do not produce residues that can contaminate the environment.
- Mass production: They can be produced in large volumes and easily applied with machinery or irrigation water. These microorganisms can be produced and formulated as mycoinsecticides to be used as bait or in liquid and solid applications. They can be cultured in the laboratory and taken to the field in different substrates and conditions to cause damage to pest insects (Jaronski and Mascarin 2017).
- Handling: Handling products developed based on EPF must be similar to the handling of chemical products; however, few reports exist of human pathologies associated with EPF, which would allow reducing protection requirements during applications.
- Compatibility: They can be applied with chemical insecticides, herbicides, and some fungicides (Yáñez and France 2010; Schumacher and Poehling 2012; Machado 2016). However, it is important to consider that some products, primarily fungicides, could cause problems in its effectiveness. It is therefore necessary to know the chemical groups with which they are compatible.

11.4 Recommendations for Use of Entomopathogenic Fungi (EPFs)

Similarly to chemical products, biological products also require certain use recommendations in order to obtain expected control results. One important recommendation is to try using products developed with native or local strains that are adapted to the conditions where they will be used; this is considered as an essential factor for the success of this type of control under field conditions (Gutiérrez and Maldonado 2010).

EPFs are live organisms and are therefore susceptible to a set of factors that can affect their effectiveness, such as:

Temperature Fungi have a temperature range for optimal growth; most EPFs can germinate and develop adequately between 22 and 28 °C. Outside this range, development problems can occur, which would not allow them to reach the expected results.

Humidity One of the most important conditions for conidial germination is humidity. The majority of EPF species are hydrophilic; they reach high mortality levels in insects when humidity is high. However, some species can also achieve low mycosis levels. This is very relevant when selecting the strains to be used. Humidity on the surface of the integument of the insect, in the foliage, or on the soil can limit germination and penetration of the fungus in the host. Water combined with other environmental factors may or may not limit the persistence and effectiveness of EPF at field level.

Ultraviolet Radiation Solar radiation has wavelengths, such as UVA and UVB, which affect EPF effectiveness. Short wavelengths are able to delay or, in some extreme cases, suppress conidial germination mainly because of the damage that these wavelengths cause in fungal DNA. There is direct damage associated with the formation of photoproducts, such as pyrimidine dimmers, pyrimidine hydrates, and crosslinks between DNA and proteins. There is also indirect damage produced by the appearance of reactive oxygen molecules (e.g., hydrogen peroxide and free radicals), which damage DNA (Diffey 1991; Cerdá-Olmedo et al. 1996). The geographic zone where strains are obtained therefore becomes relevant. Studies conducted with several *Beauveria* sp. isolates showed high variability in the resistance to UVB rays among isolates obtained at different latitudes. Resistance can vary from 0% to 80% and is higher in isolates obtained at lower latitudes (Fernandes et al. 2007).

Synthetic Chemical Pesticides The use of some pesticides can affect EPF effectiveness. Studies conducted by Machado (2016) demonstrated that the tebuconazole molecule inhibits sporulation and germination of *B. bassiana*. Other studies probed that azoxystrobin, benomyl, and chlorothalonil are incompatible with different strains of *Metarhizium anisopliae* (Yáñez and France 2010). Mancozeb applications can also reduce sporulation of this fungus as much as 75%. This same study revealed that some herbicides such as glufosinate ammonium could affect EPF spore production and their performance as biocontrollers. On the other hand, studies that tested the effect of chemical pesticides (fipronil, permethrin, imidacloprid, and amitraz) on EPF concluded that they have no negative effects if applied at the recommended doses (Schumacher and Poehling 2012).

In order to improve control effectiveness using EPF, it is recommended to consider the following aspects:

1. Identify the target pest: It is necessary to have knowledge about the pest to be controlled because many EPFs are specific. Once the pest is identified, the most susceptible stage of the life cycle of the insect must be identified for control.
2. Monitoring: Before and after applying EPF, monitoring must be done with the aim of knowing pest behavior; this will help to determine the best time to make applications and increase the probabilities of success in pest control.
3. Time of application: As previously mentioned, different conditions exist in the medium that affect EPF effectiveness. For this reason, it is recommended to make applications of many EPF-based products first thing in the morning or before sunset. Most fungi require high humidity to be able to infect the insect (80% to 100%), so that natural epizootics are more common during high humidity conditions.
4. How to apply: There are different ways to apply EPF-based products, and the most effective one will depend on the nature of the pest to be controlled, the crop, and the environmental conditions.
5. Application equipment: It is recommended that the equipment be specially designed for the use of EPF because there is the risk of affecting fungal effectiveness when using equipment containing chemical pesticide residues, especially if fungicides have been used.
6. Phytosanitary management of nearby fields: It is also recommended to have knowledge about the phytosanitary management of fields bordering those managed by EPF. Fungicide applications in neighboring fields under wind conditions toward the field threatened the EPF and could affect its effectiveness.
7. Storing the product: Just as for all pesticides, EPF-based products have an expiry date; it is recommended to strictly follow manufacturer conditions because many of these products rapidly lose their viability. On the other hand, storage conditions affect their effectiveness and it is usually recommended to store them in cool, dry, and dark places.
8. Transportation and handling: It is necessary to always take into account that fungi are live organisms affected by the environmental conditions surrounding them. It is therefore recommended to transport and handle these EPF-based products under conditions that allow maintaining their effectiveness, preventing exposure to solar radiation and high temperatures.

11.5 Successful Experiences in the Development and Use of Entomopathogenic Fungi (EPFs)

11.5.1 Experience in Argentina

One of the most important cotton pests of the Americas is the cotton boll weevil (*Anthonomus grandis* Boheman) (Curculionidae), which is found in Argentina since 1993. The National Institute of Agricultural Technology (INTA) has performed evaluations to control it; the objective was to select native isolates of *B. bassiana* and *M. anisopliae* that are virulent for this pest. Isolates were obtained from soil and insect samples as well as from the mycology collection of the Entomopathogenic Fungus Laboratory (IMYZA, INTA). The pathogenicity and virulence of 28 strains of *M. anisopliae* and 66 strains of *B. bassiana* were evaluated on adult boll weevils, and the sublethal effects on feeding and oviposition produced by these fungi were studied. The Ma 50 and Ma 20 strains were the most virulent; there was decreased feeding of females infected with Ma 20 and Bb 23 and decreased oviposition with Ma 20. The evaluated *M. anisopliae* strains were compatible with pyrethroid insecticides and more tolerant to high temperatures; these results will allow the incorporation of EPF strains to the integrated management programs of this pest (Nussenbaum 2014).

Another important pest in Argentina is the fruit fly *Anastrepha fraterculus* (Wiedemann) (Tephritidae) (native) and *Ceratitis capitata* (Wiedemann) (Tephritidae) (exotic), which is a quarantine pest that limits fresh fruit and vegetable exports. Studies conducted by Albornoz (2014) allowed the selection of EPF strains to control these species in walnut, peach, and guava trees. A search of different EPFs resulted in four strains of *B. bassiana* whose pathogenicity was later evaluated on different fly stages. Larva and pupa mortality was greater than 55% for *C. capitata* and less than 27% for *A. fraterculus* in sprinkling applications. Adult mortality was 22% for both species. Results were promising to control this species, but field evaluations are necessary. Other studies conducted under laboratory conditions in the Institute of Agricultural Microbiology and Zoology INTA-Castelar evaluated the lethal concentration (90), mean survival, and conidial production of *B. bassiana* strains to control *C. capitata.* Furthermore, the combativeness of the fungus was studied with the Mercaptothion 100% CE and Dimetoato 40% CE insecticides. The study results concluded that the Bb 238, Bb 259, and Bb 132 strains were promising for the development of a commercial product to control the insect. These strains were also compatible with Mercaptothion, but exhibited compatibility limitations with Dimetoato (Porras and Lecuona 2008).

Argentina has a pilot plant within INTA called MICOPLAR that mass-produces biopesticides. This plant was initially intended to manufacture mycoinsecticide bait against leaf-cutter ants and to develop more efficient methodologies to produce EPF. It is hoped that the research developed in the plant will result in technology transfer to different productive sectors of the country in such a way as to promote local (public and/or private) micro-ventures and/or generate a network of mycoinsecticide pilot plants in strategic locations to benefit small- and medium-sized producers.

11.5.2 Experience in Brazil

Among the reasons to develop biological control in Brazil, the ample use of agrochemicals is highlighted; this has caused serious biological imbalances, such as water and soil contamination, as well as the appearance of secondary pests. Brazil is one of the Latin American countries that has achieved great progress in the use of EPF to control pests in pasturelands, sugarcane, coffee, citrus fruits, and vegetables. Just as in other countries of the region, *M. anisopliae* and *B. bassiana* are the most used fungi. The Brazilian Agricultural Research Corporation (Embrapa) has made significant contributions to the development of biological control technologies using EPF. It has an important fungus collection currently conserved in the National Genetic Resources and Biotechnology Research Center (Cenargen). It has also implemented technology transfer programs that include consultancy, training, and publications.

Various biological control strategies have been developed for the pest *Bemisia tabaci* (Gennadius) (Aleyrodidae); Brazil has been seriously affected by this insect since 1995 and accumulated losses exceed 5 billion dollars (Maranhão and Maranhão 2009). Among the most affected crops are tomato, cucumber, melon, watermelon, cotton, and other ornamental plants. The insect was initially detected in the southeast region (São Paulo) and was rapidly disseminated to almost all the regions of the country (Villas Bôas et al. 1997; Lima et al. 2000). Vicentini et al. (2001) evaluated 50 strains of *B. bassiana* to control nymphs of *B. tabaci* biotype B in melon. Results of strains CG 136 and CG 149 for nymph mortality were approximately 90%. These strains were isolated from insects of the same taxon as *B. tabaci* and performed better than the strains contained in commercial products recommended to control this insect.

11.5.3 Experience in Chile

There has been increasing interest in Chile since the 1990s to develop this technology, and several institutions have conducted research in this field. Important collections were established, especially those provided by the Institute of Agricultural Research (INIA) through its Technology Center for Biological Control, which consists of more than 1000 fungal strains that were collected throughout the country and currently conserved in the Microbial Genetic Resources Bank. Currently, INIA is the institution that has conducted the most studies with EPF and is developing many initiatives to control economically important pests.

Fruit growing for exportation faces a significant group of pests that are economically important that consist of Curculionidae (weevils), especially the species *Aegorhinus superciliosus* (Guérin-Méneville), *Aegorhinus nodipennis* Hope, and *Naupactus xanthographus* (Germar). One of the main limiting factors in grapevine production and other fruit trees in Chile is related to the control of the vine weevil (*N. xanthographus*) (Artigas 1994). It is a native quarantine species conventionally controlled by repeated applications of organophosphate pesticides in the aerial part and in the past by the establishment of sticky bands with azinphos-methyl on the trunk of each plant (González 1983). It is critical to attack this pest in vineyards under organic management where the use of synthetic chemical insecticides is prohibited. In 2002, the vine weevil attack was massive in organic vineyards and provoked serious economic damage, exhibiting a mean of 12 adults per plant. Consequently, biological control alternatives in the soil began to be explored.

After large laboratory screening tests using different native strains of *B. bassiana* and *M. anisopliae* against fruit tree weevil larvae, the strains 1 CET and B323 of *B. bassiana* and M82 and M430 of *M. anisopliae* (Fig. 11.2) were selected. Field evaluations were then performed by applying lyophilized conidia, which controlled larvae by more than 90% as compared to the control; strains M430 and B323 were the most effective (Mejías 2004). The Agroecology R&D Center made large-scale applications of these strains through a drip irrigation system, which directly arrived at the area where the larvae were attacking the vine roots. Applications took place

Fig. 11.2 Biological control experience using entomopathogenic fungi (EPFs) in organic grapevines: (**a**) vine weevil adult female, (**b**) vine weevil larvae in the soil, (**c**) larvae parasitized with EPF. (Photo: Carlos Pino, Agroecological R&D Center)

in autumn and spring 2003 and control levels were approximately 90% of larvae; in the aerial part, the level decreased to less than 1 adult per plant. These evaluations became common management practice for this Chilean native pest and stopped being one of the main limiting factors of ecological management. This is one of the milestones of successful large-scale biological control, which facilitated the conversion of more than 3000 ha to organic management and proved that ecological management requires science, proactivity, and collective ingenuity for its development (Pino 2013).

Another pest that is currently provoking serious damage in Chilean fruit growing is *Lobesia botrana* (Denis & Schiffermüller) (Tortricidae), which affects *Vitis vinifera* and other fruit trees in urban and productive sectors of the country. Formulations are being evaluated to control this quarantine pest, which are based on different strains of the genera *Beauveria* and *Metarhizium* management at field level.

For horticulture, evaluations with native strains of *B. bassiana* and *M. anisopliae* are in progress to control the tomato moth [*Tuta absoluta* (Meyrick) (Gelechiidae)], which is an important pest in the tomato crop (*Solanun lycopersicon*) because it can cause yield losses of approximately 90% of the fruit (Apablaza 1988). Strains of *Metarhizium* exhibit high control levels, 80% mortality in eggs and 90% in larvae, higher than *B. bassiana* strains which had 60% mortality in eggs and 50% in larvae (Rodríguez et al. 2006a, b).

11.5.4 Experience in Colombia

The history of insect biological control in Colombia using EPF arise from the need of producers to have access to more sustainable and economical products to control important agricultural pests. Together with research on the use of EPF on different pests of economic interest, several small artisanal companies were established to mass-produce these agents. Colombia currently has both large national and international companies that produce, commercialize, and export several EPF strains. The development of EPFs has been associated with the implementation of integrated pest management programs for different crops, such as cotton, maize, soy, sorghum, yucca, tomato, bean, banana, and fruit trees.

As demonstrated in different research studies, Colombia is working hard to evaluate the potential of EPF to control extensive crop pests, such as the coffee berry borer (*Hypothenemus hampei* (Ferrari) (Curculionidae) and the white potato worm (*Premnotrypes vorax* Hustache) (Curculionidae). For example, efforts have been made in recent years to reduce the losses caused by the white worm in potato crops, which can reach 100% depending on the infestation level and crop management. Studies conducted by Villamil et al. (2016) demonstrated that commercial strains combined with Metaril® W.P + *B. brongniartii*® W.P (T6) and the native isolate of *Beauveria* sp. Bv01 (T1) reduced damage caused by the insect with significant control levels (77.0% and 77.6%), which represents a promising alternative to control *P. vorax* when it is incorporated in an integrated pest management scheme (Delgado 2015). In the case of coffee, the most important pest is the berry borer, which was introduced from Africa and arrived without any natural enemies. The National Coffee Research, Innovation, and Technological Development Center (Cenicafé) has established an EPF collection (native and exotic strains) with important biocontrol activity on the insect. Numerous studies have been conducted over time using EPF with excellent control results. The EPF introduction program sponsored by the National Coffee Growers Federation is highlighted; the total berry borer population in 1995 decreased by 49% on the average because of *Beauveria* (Ruíz 1996).

In the last 20 years, the Colombian Agricultural Institute (ICA) has concentrated on research to find biological alternatives for the integrated pest management programs. This is coupled with the effort by the Colombian Agricultural Research Corporation (Corpoica, now Agrosavia) to conduct studies of biocontrol applications in which EPFs are emphasized in different economically important crops for the country.

11.6 Conclusion

Entomopathogenic fungi have evolved from the “calcinaccio” described from the *Beauveria bassiana*-covered spore caterpillars, by the “Father of Insect Pathology” Agostino Bassi (Davidson 2012), to a complex science with a tremendous potential for insect control. Thus, EPFs can serve as effective biological control agents for pest control and also as plant-beneficial microbes by protecting plants against insects or microbial plant pathogens. EPFs can affect insects by changing its behavior, ability to feed, reproduce, and survive. Moreover, some EPFs can colonize various tissues and persist for months, even pass to the next generation such as endophytic microorganisms, protecting plants through direct interactions with insect herbivores by producing insecticidal metabolites or by inducing systemic resistance.

It is clear that EPFs play an important role as effective biocontrol agents and pest management worldwide as well as in Latin American countries, but our understanding of their effectiveness, mass production, effective deliveries, and impacts on plant and insect communities is still at the beginning stages. Research is needed in order to understand their additional roles in nature, as endophytes, antagonists to plant pathogens, in association with the rhizosphere, and even plant growth-promoting agents. Thus, these multiple roles deserve to the EPF an auspicious future in the development of agriculture.

References

Albornoz P (2014) Hongos patógenos asociados a moscas de la fruta (Diptera: Tephritidae) en el sector austral de Las Yungas del noroeste argentino. Dissertation, Universidad Nacional de La Plata

Apablaza J (1988) Avances en el control de la polilla del tomate. Evaluación del insecticida cartap. Cienc Tecnol 35(9):27–30

Arnold A, Mejía L, Kyllo D et al (2003) Fungal endophytes limit pathogen damage in a tropical tree. Proc Natl Acad Sci U S A 100(26):15649–15654

Artigas J (1994) Entomología económica: insectos de interés agrícola, forestal, médico y veterinario, vol 2. Ediciones Universidad de Concepción, Concepción

Cerdá-Olmedo E, Martín-Rojas V, Cubero B (1996) Causes of cell death following ultraviolet B and C exposures and the role of carotenes. Photochem Photobiol 64(3):547–551

Davidson EW (2012) History of insect pathology. In: Vega F, Kaya H (eds) Insect pathology, 4th edn. Academic Press, New York, pp 13–28

Delgado XLM (2015) Control biológico con hongos entomopatógenos: una estrategia de aula para promover el cuidado del medio ambiente. Dissertation, Universidad Nacional de Colombia

Diffey BL (1991) Solar ultraviolet radiation effects on biological systems. Phys Med Biol 36(3):299–328

Faria MR, Wraight SP (2007) Mycoinsecticides and mycoacaricides: a comprehensive list with worldwide coverage and international classification of formulation types. Biol Control 43(3):237–256

Fernandes EK, Rangel DE, Moraes AM et al (2007) Variability in tolerance to UV-B radiation among *Beauveria* spp. isolates. J Invertebr Pathol 96(3):237–243

Furlong MJ, Pell JK (2005) Interactions between entomopathogenic fungi and arthropod natural enemies. In: Vega FE, Blackwell M (eds) Insect-fungal associations: ecology and evolution. Oxford University Press, New York, pp 51–73

Glare T, Caradus J, Gelernter W et al (2012) Have biopesticides come of age? Trends Biotechnol 30(5):250–258

Goettel MS, Eilenberg J, Glare T (2005) Entomopathogenic fungi and their role in regulation of insect populations. In: Gilbert L, Iatrou K (eds) Comprehensive molecular insect science. Elsevier, Amsterdam, pp 361–406

González R (1983) Manejo de plagas de la vid. Universidad de Chile, Santiago

Gutiérrez CG, Maldonado MBG (2010) Uso de bioinsecticidas para el control de plagas de hortalizas en comunidades rurales. Ra Ximhai 6(1):17–22

Hamil R, Higgens C, Boaz H et al (1969) The structure of beauvericin, a new depsipeptide antibiotic toxic to *Artemia salina*. Tetrahedron Lett 49(49):4255–4258

Hoffmann M, Frodsham A (1993) In: Cooperativa de la Universidad de Cornell (ed) Los enemigos naturales de las plagas de insectos vegetales, Ithaca

Jaronski ST, Mascarin GM (2017) Mass production of fungal entomopathogens. In: Lacey L (ed) Microbial control of insect and mite pests: from theory to practice. Academic Press, New York, pp 141–155

Lacey LA, Grzywacz D, Shapiro-Ilan DI et al (2015) Insect pathogens as biological control agents: back to the future. J Invertebr Pathol 132:1–41

Lima LHC, Návia D, Inglis PW et al (2000) Survey of *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) biotypes in Brazil using RAPD markers. Genet Mol Biol 23(4):781–785

Machado L (2016) Efectos de plaguicidas químicos empleados en *Phaseolus vulgaris* L. sobre el crecimiento, colonización y patogenicidad de *Beauveria bassiana* (Bals.) Vuill y *Metarhizium anisopliae* (Mest.) Sorok. Dissertation, Universidad Central Marta Abreu de Las Villas

Maranhão EAA, Maranhão EHA (2009) Hongos entomopatógenos: importante herramienta para el control de “moscas blancas” (Homoptera: Aleyrodidae). An Acad Pernamb Ciênc Agron 5(6):209–242

Mejías P (2004) Efectividad de *Beauveria bassiana*, *Metarhizium anisoliae* y *Steinernema* sp. en el control de *Naupactus xanthographus*. Tesis, Universidad Católica del Maule

Meyling NV, Eilenberg J (2007) Ecology of the entomopathogenic fungi *Beauveria bassiana* and *Metarhizium anisopliae* in temperate agroecosystems: potential for conservation biological control. Biol Control 43(2):145–155

Nourrisson C, Dupont D, Lavergne RA et al (2017) Species of *Metarhizium anisopliae* complex implicated in human infections: retrospective sequencing study. Clin Microbiol Infect 23(12):994–999

Nussenbaum AL (2014) Aislamientos de *Beauveria bassiana* y *Metarhizium anisopliae* virulentos para el control del picudo del algodonero, *Anthonomus grandis* (Coleoptera: Curculionidae). Tesis, Universidad de Buenos Aires

Ownley B, Gwinn K, Vega F (2010) Endophytic fungal entomopathogens with activity against plant pathogens: ecology and evolution. BioControl 55(1):113–128

Parine NR, Devinder K, Khan PAA et al (2010) Antifungal efficacy of secondary metabolites from entomopathogenic fungi *Beauveria bassiana*. J Pharm Res 3(4):855–856

Pino C (2013) Manual de vitivinicultura orgánica. Cespedes, Curicó

Porras L, Lecuona R (2008) Estudios de laboratorio para el control de *Ceratitis capitata* (Wiedmann) (Diptera: Tephritidae) (Mosca del Mediterráneo) con *Beauveria bassiana*. Agron Costarricense 32(2):119–128

Quesada-Moraga E, Maranhao E, Valverde-García P et al (2006) Selection of *Beauveria bassiana* isolates for control of the whiteflies *Bemisia tabaci* and *Trialeurodes vaporariorum* on the basis of their virulence, thermal requirements and toxicogenic activity. Biol Control 36(3):274–287

Roberts DW, Humber RA (1981) Entomogenous fungi. In: Cole GT, Kendrick B (eds) Biology of conidial fungi. Academic Press, New York, pp 201–236

Rodríguez M, Gerding M, France A (2006a) Efectividad de aislamientos de hongos entomopatógenos sobre larvas de polilla del tomate *Tuta absoluta* Meyrick (Lepidoptera: Gelechiidae). Agric Téc 66(2):159–165

Rodríguez M, Gerding M, France A (2006b) Entomopathogenic fungi isolates selection for egg control of tomato moth Tuta absoluta Meyrick (Lepidoptera: Gelechiidae) eggs. Agric Téc 66(2):151–158

Roy HE, Steinkraus DC, Eilenberg J et al (2006) Bizarre interactions and endgames: entomopathogenic fungi and their arthropod hosts. Annu Rev Entomol 51:331–357

Ruíz R (1996) Efecto de la fenología del fruto del café sobre los parámetros de la tabla de vida de la broca del café, *Hypothenemus hampei* (Ferrari). Tesis, Universidad de Calda

Schumacher V, Poehling HM (2012) In vitro effect of pesticides on the germination, vegetative growth, and conidial production of two strains of *Metarhizium anisopliae*. Fungal Biol 116(1):121–132

Serebrov VV, Gerber ON, Malyarchuk AA et al (2006) Effect of entomopathogenic fungi on detoxification enzyme activity in greater wax moth *Galleria mellonella* L. (Lepidoptera, Pyralidae) and role of detoxification enzymes in development of insect resistance to entomopathogenic fungi. Biol Bull 33:581

Shah PA, Pell JK (2003) Entomopathogenic fungi as biological control agents. Appl Microbiol Biotechnol 61(5/6):413–423

Shah FA, Ansari MA, Watkins J et al (2009) Influence of commercial fungicides on the germination, growth and virulence of four species of entomopathogenic fungi. Biocontrol Sci Tech 19(7):743–753

Susuki A, Kanaoka M, Isogai A et al (1977) Bassianolide, a new insecticidal cyclodepsipeptide from *Beauveria bassiana* and *Verticillium lecanni*. Tetrahedrom Lett 18(25):2167–2170

Vega F, Posada F, Aime M et al (2008) Entomopathogenic fungal endophytes. Biol Control 46(1):72–82

Vega FE, Meyling NV, Luangsa-Ard JJ et al (2012) Fungal entomopathogens. In: Vega FE, Kaya HK (eds) Insect pathology, 2th edn. Academic Press, New York, pp 171–122

Vicentini S, Faria M, Oliveira MR (2001) Screening of *Beauveria bassiana* (Deuteromycotina: Hyphomycetes) isolates against nymphs of *Bemisia tabaci* (Genn.) biotype B (Hemiptera: Aleyrodidae) with description of a new bioassay method. Neotrop Entomol 30(1):97–103

Villamil JE, Martínez JW, Pinzón EH (2016) Actividad biológica de hongos entomopatógenos sobre *Premnotrypes vorax* Hustache (Coleoptera: Curculionidae). Rev Ciênc Agr 33(1):34–42

Villas Bôas GL, França FH, Ávila AC et al (1997) Manejo integrado da mosca-branca *Bemisia argentifolii*. Embrapa, Brasília. (Circular Técnica da Embrapa Hortaliças, 9)

Wagner BL, Lewis LC (2000) Colonization of corn, *Zea mays*, by the entomopathogenic fungus *Beauveria bassiana*. Appl Environ Microbiol 66(8):3468–3473

Wang C, Wang S (2017) Insect pathogenic fungi: genomics, molecular interactions, and genetic improvements. Annu Rev Entomol 62:73–90

Yáñez M, France A (2010) Effects of fungicides on the development of the entomopathogenic fungus *Metarhizium anisopliae* var. anisopliae. Chil J Agric Res 70(3):390–398