

THE USE OF ENTOMOPATHOGENS IN THE CONTROLLING OF INSECT PESTS OF STORED PRODUCT

Ramazan CANHILAL

Erciyes University, Faculty of Agriculture,
Department of Plant Protection, 38039 Kayseri, Turkey

Corresponding author email: r_canhilal@hotmail.com; ramazancanhilal@erciyes.edu.tr

Abstract

*A chemical method called fumigation is mainly applied to control of stored product pests in the world and in Turkey. The alteration of natural balance among living organisms, the resistance of pests for pesticides, and residues on crops are produced by application of pesticides widely. Nowadays, incremental necessity has been occurred to find out alternatives to chemicals. Biological control is a novel method to replace chemicals. Insect pathogens which kill insects causing disease are agents such as bacteria, fungi, nematodes, viruses, protozoa. The mass of entomopathogenic bacteria are in genera *Coccobacillus* and *Bacillus*. *Coccobacillus acridiorum* produced disease in grasshoppers. *Bacillus thuringiensis* and *Bacillus popillia* are important disease causing agents against lepidopteran pests. Entomopathogenic fungi *Beauveria bassiana*, *Metarhizium anisopliae*, and *Verticillium lecanii* cause disease on larger insects than other pathogens. These are rather prevalent on the insects in orders Lepidoptera, Homoptera, Hymenoptera, Coleoptera, and Diptera. Insects, especially living in soil at one of its life cycle such as larva, pupa or adult are highly susceptible against entomopathogenic nematodes. Neosteinerematidae, Steinernematidae and Heterorhabditidae are the families containing these nematodes. They are obligatory insect pathogens in nature. Entomopathogenic nematodes impacts many insect species with a broad host range. Entomopathogenic viruses at least 16 families are very important in biological control to affect insects pests. Baculoviruses are produced commercially and applied as a biological control agent to manage significant agricultural and forestry insects, especially in Orders Lepidoptera and Hymenoptera. Entomopathogenic protozoans such as *Nosema locusta* are a substantial role in the ecologically management of populations of insect pests. In this review, application and potentials of entomopathogens as biological control agents of insect species harmful on stored products has been abstracted.*

Key words: entomopathogens, insect pests, stored products.

INTRODUCTION

Infestation of stored bulk grain and processed commodities by insects causes big economic loss (Hagstrum and Flinn, 1995). These insects damage the product by physical yield and quality loss, inducing mould growth, contamination of products with insect bodies [1]. They can shelter inaccessible places and survive on even little bit food. They reproduce and increase their population quickly. Then move from cracks and crevices, perforated floors, and inside machinery into stored bulk products to infest them (Campbell et al., 2004). The order Coleoptera includes about 250.000 species. Forty families of this order contain insects harmful on stored products world-wide. Bostrichidae, Bruchidae, Cucujidae, Curculionidae, Dermestidae, Silvanidae and

Tenebrionidae are some of these families (Rees, 1996).

Government regulations, environmental and human health concerns, resistances of insects to insecticides, pesticide residues on crops, changing consumer demands limit the presence and use of chemical insecticides against these pests (Durmuşoğlu et al., 2010).

Entomopathogens are biological control agents causing diseases in insect populations. These are organisms such as bacteria, fungi, nematodes, viruses, and protozoa. They can be a safe alternative for stored-product pests in unreachable places, because some biological control agents actively search out pests in these cryptic habitats (Schöller and Flinn, 2000). In this paper, the research and applications of entomopathogens against stored products are summarized.

ENTOMOPATHOGENIC BACTERIA

Entomopathogenic bacteria are most commonly used microorganisms against insect pests present. The most widely used ones, spore-forming facultative bacteria producing crystals. The bacteria enter the insect body through the mouth with food. They form endospore and protein crystals. These crystals contain toxins. The insects are killed because the toxins or bacteria wrap the body of insect.

The majority of entomopathogenic bacteria take place in *Coccobacillus* and *Bacillus* species. *Coccobacillus acridiorum* is a type of grasshopper pathogen. *Bacillus thuringiensis* and *Bacillus popillia* are the other two important species. In our country, bacteria are recommended and used against harmful species in Order Lepidoptera below.



Heliothis armigera



Lobesia botrana



Hyponomeuta sp.



Cydia pomonella



Lymantria dispar



Archips sp.



Thaumetopoea pityocampa

ENTOMOPATHOGENIC FUNGI

Fungal diseases in insects, the lighting in the Italian Agostino Bass silk white musk nature of

fungal diseases in insects were known since the 1834-1835 year.

Entomopathogenic fungi, capable of infecting insects in Order Lepidoptera, Homoptera, Hymenoptera, Coleoptera and Diptera included, are quite common. The fungi encounters host with chance in environmental conditions. Population density depends on the amount of surrounding fungal spores and insect pests. The host entrance into cuticle is realized by both with lytic enzymes and by means of mechanical formation of appressorium (Figure 1). After penetrating the cuticle and epidermis, spores germinate and multiply in the insect body. The metabolites formed by fungi cause physiological and biochemical changes in the host and results in insect death.

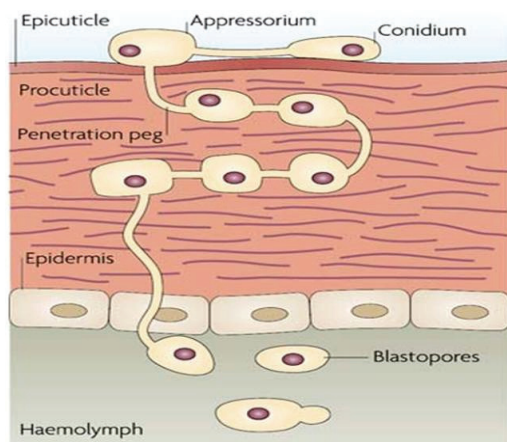


Figure 1. Penetrating of fungus into the cuticle and epidermis, and appressorium formation

Lord (2011) used *Beauveria bassiana* against *Dermestes maculatus* and it was successful with 82% in 75% humidity. Khashaveh (2011) observed 68-92% mortality in the population of *Sitophilus granarius* and *Tribolium castaneum* adults against which *Beauveria bassiana* applied. Ahmad (2010) used *Lecanicili lecanii*, *Isaria fumosarose* and *Metarhizium anisoplia* against *Sitophilus zeamays* adults and determined 100% insect mortality.

Lord (2009) obtained 10% more mature death on *Rhyzopertha dominica*, *Oryzaephilus surinamensis*, *Cryptolestes ferrusineus* using *Beauveria bassiana* with diatomaceous powder formulation.

Batta (2008) used *Beauveria bassiana* and *Metarhizium anisoplia* against *Rhyzopertha*

dominica, *Sitophilus oryzae*, *Tribolium castaneum*, *Sitophilus oryzae* adults and got the highest mortality rate (85-96%) on *Sitophilus oryzae*. Hansen (2007) determined *Beauveria bassiana* to be a successful control agent against *Sitophilus granarius* with rate of 83-98% mortality.

ENTOMOPATHOGENIC NEMATODES

Entomopathogenic nematodes (EPNs) (*Rhabditida*: *Neosteinerternematidae* *Steinernematidae* and *Heterorhabditidae*) do not cause contamination of ground water and are harmless to plants and animals. As biological control agents, EPNs attract attention increasingly in research area recently. Their ideal properties such as; the broad host spectrum, to be able to kill their hosts within 24-48 hours, to be producible commercially easily in vivo or in vitro, having ability to search actively their hosts, settling in application areas and staying effective for a long time, having easy applicability, being in compliance with many chemicals and being safe for the environment are important for their preferability.

They are soil dwelling, aquatic organism. They have motile bacteria (*Xenorhabdus* spp. and *Photorhabdus* spp.) living in their intestine. Nematode and bacterium are mutualistic symbionts and obligate, lethal parasites of insects. EPNs can be found everywhere on earth and infect many different insects (Smart, 1995). Infective juveniles of the nematodes living in the soil enter the host insect's haemocoel through mouth, anus, and respiratory openings or cuticle's thin sections. Once in the insect, infective juveniles of the nematode release the bacteria that are carried in the intestine. Bacteria block the insect's immune system, multiply and kill the insect using many different toxins and causing septicemia in the hemolymph (Figure 2).

Bacterial cells reproduce rapidly. The nematodes eat the bacteria and some of body tissue of the insect, and reproduce for 3 or 4 generations depending on the food source. Over 100,000 nematodes exit the insect (Burnell and Stock, 2000) (Figure 2).

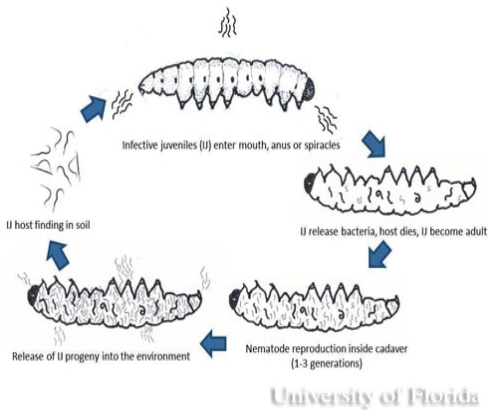


Figure 2. The simple life cycle of entomopathogenic nematodes

Tradan et al., (2006) in their study used four species of entomopathogenic nematodes (*Steinernema feltiae*, *S. carpocapsa*, *Heterorhabditis bacteriophora* and *H. megidis*) against storage pests, *Sitophilus granarius* and *Oryzaephilus surinamensis* in the laboratory to determine the activity of nematodes against adults. LC_{50} value was found as 803-1195 Ijs/adult on *S. granarius* and 921-1335 Ijs/adult on *O. surinamensis*.

Shahina and Salma (2010) studied seven local (Pakistani) entomopathogenic nematodes (*Steinernema pakistanense*, *S. asiaticum*, *S. abbasi*, *S. siamkayai*, *S. feltiae*, *Heterorhabditis bacteriophora* and *H. indica*) on *Sitophilus oryzae*'s adults and pupae in the laboratory. Consequently, the LC_{50} value for pupae of *S. oryzae* was 42-169 Ijs/pupae and for adults, 55-370 Ijs/adult.

Canhilal et al., (2013) determined the biological activity of nine endemic nematodes obtained from a survey conducted in the various districts of Kayseri Province against *Sitophilus oryzae* adults. The lowest LC_{50} value was 57.96 IJs/adult for *S. carpocapsae* 076 isolate, while the highest LC_{50} values was 922.95 Ijs/adults for *S. feltiae* OZV-5-S isolate.

ENTOMOPATHOGENIC VIRUSES

Baculovirus is important as biological control agents especially in the control of pests in agriculture and forestry. It is produced commercially and used against pests belonging to Lepidoptera and Hymenoptera.

In Brazil, *Baculovirus anticarsii* is used successfully against *Anticarsia gemmatilis* causing substantial harm in soybeans.

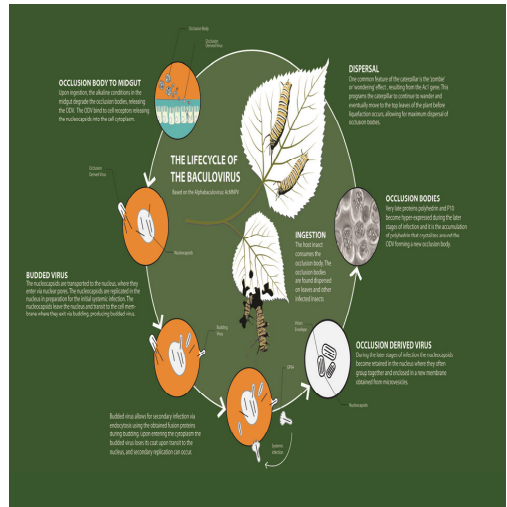


Figure 3. The life cycle of the baculovirus.

Nakai (2013) in Japan used granulovirus (GV) as a biological agent against the tea leaf crumbers (*Adoxophyes honmai* and *Homona magnanima*, Lepidoptera: Tortricidae) and there was no harmful infections during the four growing seasons.

ENTOMOPATHOGENIC PROTOZOA

Entomopathogenic protozoa are usually host-specific and slow effect is caused due to chronic infection. *Nosema locustae* has been developed as a commercial product for grasshoppers control (Henry and Oma, 1981). Entomopathogenic protozoa needs live hosts to be produced and shows quite slow effect. Therefore, it has limited application for biological control.

CONCLUSIONS

Chemical fumigation is the most commonly used method against of stored product pests in the world and in our country. Widely used insecticides against harmful organisms damage the natural balance existing among organisms, cause harmful organisms to acquire resistance to pesticides and residues in the crops. In recent years, biological control is emphasized as an

alternative to chemical control. Entomopathogens have an important place in the biological control because they have a wide host range, are harmless to the environment and human, and could be applied with conventional sprayers. They can be used more against stored product pests with the development of new biotechnical methods such as collecting pests in some stations to meet them with entomopathogens.

ACKNOWLEDGEMENTS

I would like to thank my graduate student, Sevim Dogan of Yozgat Bozok University, Faculty of Agriculture and Natural Sciences, Department of Plant Protection for literature collection.

REFERENCES

- Ahmed B.I., 2010. Potentials of Entomopathogenic Fungi in Controlling the Menace of Maize Weevil *Sitophilus zeamais* on Stored Maize Grain. Archives of Phytopathology and Plant Protection, 43(2): 107-115.
- Akhurst R., Smith K., 2002. Regulation and Safety. Entomopathogenic Nematology. Gaugler, R. (ed), CABI, Wallingford UK, p. 311-332.
- Batta Y.A., 2008. Control of Main Stored-Grain Insects with New Formulations of Entomopathogenic Fungi in Diatomaceous Earth Dusts. International Journal of Food Engineering, 4(1): 1556-3758.
- Bornstein F.S., Kiger H., Rector A., 2005. Impacts of fluctuating temperature on the development and infectivity of entomopathogenic nematode *Steinernema carpocapsae*-A10. Journal of Invertebrate Pathology, 88: 147-153.
- Campbell J.F., Arthur, F.H., Mullen, M.A., 2004. Insect management in food processing facilities. Advances in Food and Nutrition Research 48, 240-295.
- Canhilal R., Borazan F., Doğan S., Özdemir Y.E., Eşgin G., Aksoy H., 2013. Determination of efficacy of entomopathogenic nematodes (Rhabditida: Heterorhabditidae and Steinernematidae) against a stored crop pest, *Sitophilus oryzae* (Coleoptera: Curculionidae). 4th International Participated Entomopathogens and Microbial Control Symposium, Artvin, Turkey, p. 28.
- Charmley A.K., Leger R.J., 1989. The role of cuticle degrading enzymes in fungal pathogenesis in insects. The Fungal Spore and Disease Initiation in Plants and Animals, Eds: Cole, G.T. and Kock, H.C., Plenum Press, New York, p. 267-286.
- Durmuşoğlu E., Tiryaki O., Canhilal R., 2010. Türkiye’de Pestisit Kullanımı, Kalıntı ve Dayanıklılık Sorunları. Türkiye Ziraat Mühendisliği 7. Teknik Kongresi. Ankara, 11-15 Ocak, Bildiriler Kitabı, 2:589-607.[In Turkish].
- Ehlers R.U., 2005. Forum on Safety and Regulation. Nematodes as Biocontrol Agents. Grewal, P.S., Ehlers, R.-U., and Shapiro-Ilan, D.I. (eds.), CABI, Cambridge, p. 107-114.
- Hagstrum D.W., Flinn, P.W., 1995. Integrated pest management. In: Subramanyam, B., Hagstrum, D.W. (Eds.), Integrated Management of Insects in Stored Products. Marcel Dekker, New York, p. 399-408.
- Hall R.A., Papierok B., 1982. Fungi as biological control agents of arthropods of agricultural and medical importance. Parasitology. Anderson, R.M., and Canning, E.U., (eds.), Cambridge University, Great Britain, 84, p. 205-240.
- Hansen L.S., 2007. Combining larval parasitoids and an entomopathogenic fungus for biological control of *Sitophilus granarius* (Coleoptera: Curculionidae) in stored grain. Biological Control, 40(2): 237-242.
- Hansoylu R.B., 2003. Obtained from Ground Turkey Entomopathogenic fungi *Beauveria bassiana* (Bals.) Using the race as a biological control agent. Master of Science Thesis, Hacettepe University, Institute of Science and Technology, Ankara, 84s.
- Henry J.E., Oma E.A., 1981. Pest Control by Nosema locustae, A pathogen of Grasshoppers and Crickets, pp. 573-586, In Microbial Control of Pests and Plant Diseases, 1970-1980, Ed. Burges, H. D., Academic Press, London.
- Inceoglu A.B., 2001. Recombinant Baculoviruses for Insect Control, Pest Manag. Sci, 57: 981-987.
- Kaya H.K., Gaugler R., 1993. Entomopathogenic nematodes, Annual Review of Entomology, 38: 181-206.
- Khashaveh A., 2011. The Use of Entomopathogenic Fungus, *Beauveria bassiana* (Bals.) Vuill. in Assays with Storage Grain Beetles. Journal of Agricultural Science and Technology, 13(1): 35-43.
- Knaak N., Fiuza M.L., 2005. Histopathology of *Anticarsia gemmatilis* Hübner (Lepidoptera: Noctuidae) Treated with Nucleopolyhedrovirus and *Bacillus thuringiensis* serovar kurstaki, Brazilian J. Microbiol, 36: 196-200.
- Koppenhöfer A.M., 2000. Nematodes. In: Lacey, L.A. Kaya, H.K. Eds. Field Manual of Techniques in Invertebrate Pathology. Dordrecht, The Netherlands, Kluwer, 283-301.
- Leger R.J., Charmley A.K., Cooper R.M., 1988. Production of polyphenol pigments and phenoloxidase by the entomopathogen, *Metarhizium anisopliae*. Journal of Invertebrate Pathology, 53: 211-215.
- Lord J. C., 2009. *Beauveria bassiana* infection of eggs of stored-product beetles. Entomological Research, 39(2): 155-157.
- Lord J.C., 2011. Influence of substrate and relative humidity on the efficacy of three entomopathogenic fungi on *Dermestes maculatus*. Biocontrol Science of Technology, 21(4): 475-483.
- Nakai M., 2013. Potential of slow-killing insect viruses to control leaf-rollers in tea fields, 4th International Participated Entomopathogens and Microbial Control Symposium, Artvin, Turkey, p.36.

- Payne C.C., 1986. Insect Pathogenic Viruses As Pest Control Agents. *Fortschritte der Zoologie*, 32: 183-200.
- Rees D. Coleoptera. In *Integrated Management of Insects in Stored Products*; Subramanyam, B., Hagstrum, D.W., Eds., Marcel Dekker: New York, 1996; 1-40.
- Schöller M., Flinn P.W., 2000. Parasitoids and predators. In: Subramanyam, B., Hagstrum, D.W. (Eds.), *Alternatives to Pesticides in Stored-Product IPM*. Kluwer Academic Publishers, Norwell, Massachusetts, p. 229-271.
- Sezen K., 2008. Nematodlar ve biyolojik mücadele. *Entomopatogenler ve biyolojik mücadele*. Demirbağ, Z. (ed.), Esen Ofset Matbaacılık, Trabzon, p. 245-288.
- Shahina F., Salma J., 2010. Laboratory Evaluation Of Seven Pakistani Strains Of Entomopathogenic Nematode Against Stored Grain Insect Pest *Sitophilus oryzae* L. *Pakistan Journal Nematology*, 28(2): 295-305.
- Smart Jr. G.C., 1995. Entomopathogenic Nematodes for the Biological Control of Insects, Supplement to the *Journal of Nematology*, 27(4S): 529-534.
- Trdan S., Vidrih M., Vali N., 2006. Activity of four entomopathogenic nematode species against young adults of *Sitophilus granarius* (Coleoptera: Curculionidae) and *Oryzaephilus surinamensis* (Coleoptera: Silvanidae) under laboratory conditions, *Journal of Plant Diseases and Protection*, 113(4): S. 168–173, 2006, ISSN 1861-3829.
- Zimmermann G., 2007. Review on safety of the entomopathogenic fungi *Beauveria bassiana* and *Beauveria brongniartii*, *Biocontrol Science and Technology*, 17(5/6): 553-596.