

**BIOLOGICAL CONTROL OF EUROPEAN
COCKCHAFFER LARVAE (*Melolontha melolontha* L.)
– PRELIMINARY RESULTS**

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A B S T R A C T

The most serious insect pest in Hungary is the European or common cockchafer (*Melolontha melolontha* L.). The larvae cause extensive and lethal damage to the roots of young trees. No procedure for chemically controlling soil-dwelling cockchafer larvae has been approved for use in integrated fruit production (IFP), the most widespread environmentally sound production program in Hungary. Growers therefore have no option to control cockchafer larvae except for biological control. Entomopathogenic nematodes (EPNs) are the most promising control agents. The aim of this project is to develop a EPN biocontrol product effective against European cockchafer larvae. The main steps of this new project are:

- Isolating new nematode strains from natural *Melolontha* habitats in Hungary;
- Screening the nematode collection for EPN strains effective against *Melolontha* larvae;
- Establishing a pilot scale bioreactor and elaborating the technology for industrial scale fermentation of selected EPN strains; and
- Developing techniques to apply EPNs in horticultural practice.

Key words: biological control, entomopathogenic nematodes, european cockchafer, *Melolontha melolontha*

INTRODUCTION

The most serious insect pest in Hungary is the European or common cockchafer (*Melolontha melolontha* L.). From time to time, localized outbreaks also occur in other central European countries. The adults feed on the blossoms and young leaves of fruit trees and other deciduous forest and ornamental trees, while the larvae cause extensive and lethal damage to the roots of young trees.

No procedure for chemically controlling soil-dwelling cockchafer larvae has been approved for use in integrated fruit production (IFP), the most widespread environmentally sound production program in Hungary. Mechanical control is not possible with perennial crops. Growers therefore have no option to control cockchafer larvae except for biological control.

The only product available which is effective against cockchafer larvae is based on the entomopathogenic fungus, *Beauveria brongniartii*. It has been approved for use and is marketed under several product names in several countries, including Austria, Italy and Switzerland. Unfortunately, it has not yet been approved for use in Hungary.

The usefulness of *Beauveria* products is also limited by the fact that soil temperatures in Hungary are often higher than 27°C, which is the upper threshold of the growth range for *Beauveria*. Higher temperatures kill spores, which prevents the development of the hyphal network necessary for effective control (Kessler et al., 2003).

Other potential biological control agents include entomopathogenic nematodes (EPNs), which are efficient pathogens in over 200 insect hosts (Shapiro-Ilan et al., 2002). The main advantage of EPNs is that they quickly spread infection. Infective juveniles (IJs) generally enter the insect host one or two days after inoculation and kill the insect larva in 24 to 36 hours. Only a few days of favorable weather conditions might be enough for effective pest control.

In Europe, there are five companies which produce EPNs (Kaya et al., 2005). Four of them produce strains of *Heterorhabditis bacteriophora* and *H. megidis* which are effective against some important scarab larvae such as *Phyllopertha horticola*. However, none of the strains currently available is effective against larvae of *Melolontha melolontha* and *M. hippocastani*.

The aim of this project is to develop a EPN biocontrol product effective against European cockchafer larvae. Preliminary results of this ongoing project are presented in the present paper.

MATERIAL AND METHODS

EPN strains

The EPN strains used in this study came from the EPN collection at our institute (Tóth, in press). All of the strains which had been isolated in 2003 were evaluated. Tests on the remaining EPNs are currently underway.

Pathogenicity tests

Pathogenicity of the EPN strains to cockchafer larvae was tested by two different methods.

All of the strains were first screened to select virulent EPNs. Five third instar cockchafer larvae which had been collected from the field were washed in tap water and placed on filter paper disks in separate petri dishes. The filter paper disks were wetted with 1 ml of a suspension containing 1,000 IJs per milliliter of sterile tap water. Mortality was recorded after seven and fourteen days. Five strains killed four or five of the larvae and were selected for further study.

Ten washed larvae were put separately into plastic vials containing 8 grams of sterile sandy soil wetted to 50% of the water holding capacity. The soil was then inoculated with 1000 IJs per gram of soil with each of the five selected strains. The vials were kept at 25°C. The most effective strains were tested at different temperatures and at different concentrations.

RESULTS

Five strains were selected in the original screening procedure, but only one of them, *Heterorhabditis downesi* Strain 267, was highly effective in the soil test (data not shown). The dose effect of this strain is presented in Figure 1. Strain 267 caused about 90% mortality at a dose of 1,000 IJs per gram of soil, and about 50% mortality at a dose of 100 IJs per gram of soil. The optimum temperature was 20°C, although mortality was also relatively high at 15°C (Fig. 2). IJs were inactive at 10°C.

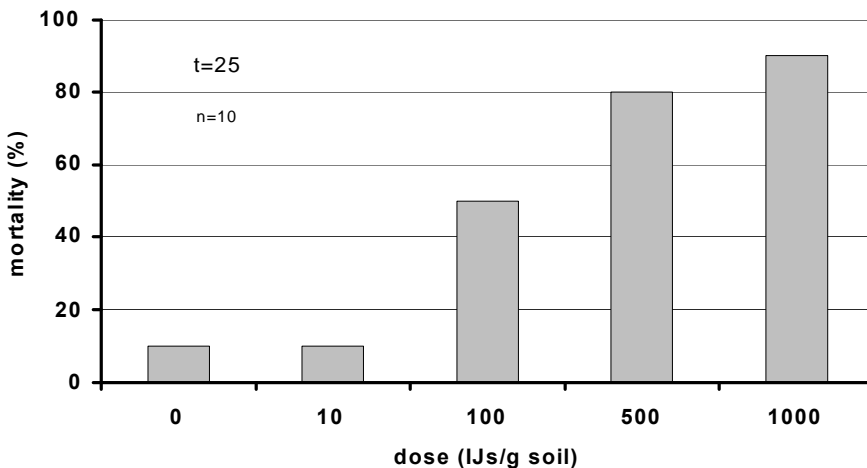


Figure 1. Effectiveness of *Heterorhabditis downesi* Strain 267 against larvae of *Melolontha melolontha* at different doses

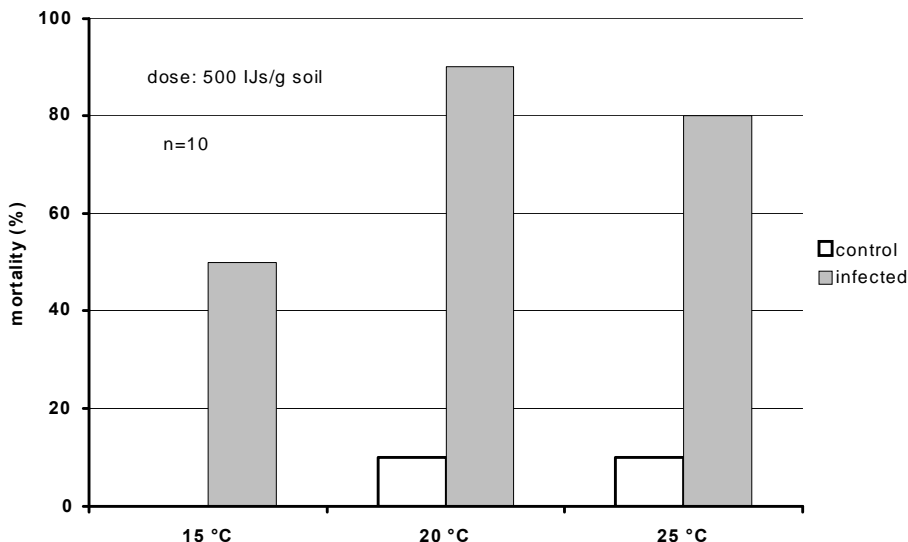


Figure 2. Effectiveness of *Heterorhabditis downesi* Strain 267 against larvae of *Melolontha melolontha* at different temperatures

DISCUSSION

EPNs have been used as effective biological control agents against the larvae of some scarabs, including *Popillia japonica* and *Phyllopertha horticola* (Kaya et al., 2005). However, none of the EPNs which are commercially available are effective against larvae of the European cockchafer (Peters and Galarza, 2002).

In laboratory studies, the nematode species which promised to be the most effective in controlling the European cockchafer was *Steinernema glaseri* (Peters, 2000). However, because *S. glaseri* comes from the USA, it cannot be widely used in Europe.

Different strains of two European *Heterorhabditis* species, *H. bacteriophora* and *H. megidis*, were both ineffective against second and third instar cockchafer larvae (Berner and Schnetter, 2001).

Of the strains collected during the survey of the Hungarian EPN fauna in 2003, one strain, *Heterorhabditis downesi* Strain 267, proved to be an effective biocontrol agent against the European cockchafer.

The temperature optimum of Strain 267 is relatively low, so that it can be applied in the spring or autumn. However, Strain 267 may not be effective if applied during hot summers. Fortunately, the infection process is fast and most infected larvae are killed in four to six days. Therefore, only a few days of favorable weather conditions might be enough for effective pest control.

Large amounts of nematodes are needed to confirm our laboratory data in the field. An *in vitro* liquid technique has been developed to mass produce Strain 267. Field tests are planned for next year.

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BIOLOGICZNE ZWALCZANIE CHRABĄSZCZA MAJOWEGO (*Melolontha melolontha* L.): BADANIA WSTĘPNE

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S T R E S Z C Z E N I E

Na Węgrzech, najbardziej szkodliwym owadem jest chrabąszcz majowy (*Melolontha melolontha* L.). Jego larwy powodują rozległe, często letalne uszkodzenia korzeni młodych drzew. Dla celów Integrowanej Produkcji (IP), najbardziej rozpowszechnionej na Węgrzech przyjaznej środowisku metody produkcji, nie zostało zaakceptowane żadne zwalczanie chemiczne żerujących w glebie larw chrabąszcza majowego. Dlatego też sadownicy nie mają wyboru i do niszczenia larw chrabąszcza majowego muszą stosować metody biologiczne.

Nicienie – pasożyty owadów (Entomopathogenic nematods – EPNs), są najbardziej obiecującymi organizmami zwalczającymi tego szkodnika. Celem przedstawianego projektu badawczego jest wytworzenie na bazie EPN nowego produktu pochodzenia biologicznego do zwalczania larw chrabąszcza majowego. Głównymi punktami tego projektu jest:

- Znalezienie nowych szczepów nicieni w naturalnych środowiskach chrabąszcza na Węgrzech;
- Skrining kolekcji nicieni, w celu znalezienia EPNs, efektywnych przeciwko larwom chrabąszcza;
- Wykonanie prototypu bioreaktora i opracowanie technologii dla przemysłowej hodowli wyselekcjonowanych szczepów EPN;
- Rozwinięcie technik dla zastosowania EPNs w praktyce ogrodniczej.

Słowa Kluczowe: biologiczne zwalczanie, entomopatogeniczne nicienie, chrabąszcz majowy, *Melolontha melolontha*