Biological control of the codling moth by \textit{Trichogramma embryophagum} based on Degree-Hours forecasting model

Hossein Ranjbar Aghdam\textsuperscript{1}\textsuperscript{*} and Mohammadreza Attaran\textsuperscript{2}

1. Assistant Professor, Department of Biological Control Research, Iranian Research Institute of Plant Protection, Tehran, Iran
2. Assistant Professor, Laboratory of Biological Control, Iranian Research Institute of Plant Protection, Amol, Iran

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ABSTRACT

The codling moth, \textit{Cydia pomonella}, is the most important pest insect in apple orchards. Efficacy of a native strain of the egg parasitoid wasp, \textit{Trichogramma embryophagum}, was statistically compared with chemical control and check treatments in an apple orchard. The best times for releasing the parasitoid wasps were determined by using a forecasting model based on estimation of Growing Degree Hours (GDH). Codling moth biofix was determined by using pheromone traps, and environmental temperature in the apple orchard recorded hourly in order to estimate of GDH. Based on the recorded temperatures and also, by using phenological forecasting model, the best times for releasing egg parasitoid wasps were determined. The wasps were released five times for two generations of the codling moth. The results of the efficacy test showed that there was no statistically difference between chemical and biological control treatments using Tukey range test ($P<0.05$), considering the percentage of damage. However, the damage percentage was statistically more in the mentioned above treatments as compared to check treatment. Additionally, considering the results, it is confirmed that the percentage of fruit damage by the codling moth in biological and chemical treatments were 47.76 and 50.73 lower than the value estimated for check treatment, respectively. Finally, based on the findings of the present study, it can be suggested that acceptable biological control of the codling moth by using \textit{T. embryophagum} is possible. It necessary to consider all the technical points from selection of a suitable biocontrol strain to its application and evaluation of the obtained results.

Keywords: biological control, codling moth, efficacy, phenological model, thrichogramma.

\textsuperscript{*} Corresponding author E-mail: hossein_aghan2003@yahoo.com    Tel: +98 912 2621715
Production and application of *Trichoderma harzianum* Tr6 for controlling of damping-off caused by *Phytophthora drechsleri* and its effect on the growth promotion of cucumber

Zhila Delkhah and Keivan Behboudi*
M.Sc. Student and Associate Professor, Department of Plant Protection, University College of Agriculture and Natural Resources, University of Tehran, Karaj, Iran
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**ABSTRACT**

*Trichoderma* genus is one of the most important biocontrol agents for certain plant disease with different control mechanisms. These fungi, like the other biocontrol agents are under the influence of environmental factors, such as culture medium type. The effect of the medium (Mol.C.B.M.) containing sugar beet molasses, corn steep liquor, baker’s yeast and malt extract as the liquid medium and two solid media including wheat bran (W.b.) and wheat seed (W.s.) were experimented on sporulation, biocontrol efficacy of *Trichoderma harzianum* Tr6 against *Phytophthora drechsleri* and growth promotion of the plant. The media were inoculated with 1ml of suspension containing $10^8$ spores of the antagonist and were maintained in an incubator at light density of 206 Lux and 28°C for 10 days. The Mol.C.B.M. medium had the most effect on sporulation (1.83×$10^8$ spore/gdw), disease control (62.5%), height, fresh and dry weight of shoots and roots of the plant, but the percentage of inhibition was less in solid media at laboratory surveys. Therefore, the Mol.C.B.M. medium is efficient to encompass the objectives of this study.

**Keywords:** antagonist, culture medium, sporulation, Trichoderma, wheat bran.
Biological properties of *Pseudomonas fluorescens* UTPF68 and its biocontrol activity against *Phytophthora drechsleri* on cucumber

Somayeh Sadat Ghafelebashi¹, Fatemeh Jamali² and Masoud Ahmadzadeh¹*  
¹. Former Graduate Student and Professor, Department of Plant Protection, Faculty of Science and Agricultural Engineering, University College of Agriculture and Natural Resources, University of Tehran, Karaj, Iran  
². Assistant Professor, Department of Agronomy and Plant Breeding, Faculty of Agriculture and Natural Resources, Persian Gulf University, Bushehr, Iran  
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**ABSTRACT**

*Pseudomonas fluorescens* UTPF68, a well-known biocontrol bacterium was used in this study. The volatile metabolites released from UTPF68 in NA, NAG, PDA media, could inhibit the growth of fungi *P. drechsleri* at the rate of 84.44, 83.55 and 67.33 percent, respectively. The results showed that antibiotics production on PDA and CMA media prevented the growth of *P. drechsleri*, 100% and 93%, respectively. The results showed that this strain is able to produce all three antibiotics DAPG, MAPG and Plt. Also, the extracellular extracts of UTPF68 reduced the vegetative growth of *P. drechsleri*. In addition, the bacterium effectiveness was studied against the pathogen in greenhouse cucumber. The bacterium was able to control the pathogen under the *in vitro* and *in vivo* conditions. So that the application of bacterial concentration at 10⁸ CFU/ml in to the soil reduced the amount of the disease up to 60%. Also, the use of this bacterium, both in the presence or absence of the pathogen used as control treatments, increased the growth of cucumber plants. The evaluation of bacterial colonization of roots showed that the population density of strain UTPF68 was increased on cucumber root system up to 2.3×10⁹ CFU.

**Keywords:** antibiotic, biocontrol, colonization, extracellular extracts, volatile metabolites.

* Corresponding author E-mail: ahmadz@ut.ac.ir  
Tel: +98 26 32225517
Biological Control of Root-knot Nematode 
(*Meloidogyne javanica*) by *Pseudomonas fluorescens* CHA0 and *Trichoderma harzianum* BI in Tomato

Somayeh Mokhtari*, Navaz-allah Sahebani and Hassan Reza Etebarian

1. Former Graduate Student, Perisan Gulf University, Iran  
2. Associate Professor and Assistant Professor, University College of Abureihan, University of Tehran, Iran  
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ABSTRACT

Root-knot nematodes (*Meloidogyne* spp.) are among the most important groups of plant-parasitic nematodes causing damage and yield losses to many cultivated plants. The bacterium *Pseudomonas fluorescens* CHA0 and the fungus *Trichoderma harzianum* BI were investigated for biological control capability and systemic induction of plant resistance against the nematode *M. javanica* under glasshouse conditions. Furthermore, the changes of peroxidase (POX) and polyphenol oxidase (PPO) activity in tomato were evaluated. In glasshouse tests, the results showed that tomato plants inoculated with both *T. harzianum* BI and *P. fluorescens* CHA0, the disease index of infected plants were significantly decreased as compared to application *P. fluorescens* CHA0 as the sole biological agent, but not statistically significant with respect to *T. harzianum* BI. The results showed that the POX activity was started at the first day after nematode inoculation and reached to a maximum level following the fourth day and after that it continuously decreased. There was no significantly difference in enzymatic chanes of plants between co-incultion of *T. harzianum* BI and *P. fluorescens* CHA0 in comparison to *P. fluorescens* CHA0. The polyphenol oxidase (PPO) activity reached to a maximum level after five day in *T. harzianum* BI and *P. fluorescens* CHA0 inoculated plants. There was a significant difference between all treatments. So, both antagonists can induce plant defense mechanisms such as POX and PPO in tomato plant against nematode infection.

Keywords: ISR, *Pseudomonas fluorescens*, root-knot nematode, *Trichoderma harzianum*. 

* Corresponding author E-mail: mokhtari_so@yahoo.com  
Tel: +98 9171257967
The use of silicon on systemic resistance induction against fusarium head blight disease of wheat

Vahid Ghazimohseni1, Seyed Kazem Sabbagh2*, Sedigheh Esmaeilzadeh Bahabadi3 and Morteza Ghorbani1

1. M.Sc. Student of Plant Pathology and Assistant Professor, Department of Plant Protection, University of Zabol, Iran
2. Assistant Professor, Department of Plant Pathology and Institute of Plant Biotechnology, University of Zabol, Iran
3. Assistant Professor, Department of Biology, Faculty of Basic Sciences, University of Zabol, Iran
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ABSTRACT
In this study the effect of different concentrations of silicon (0, 2, 4 and 6 mM) on expression of some genes involved in induced resistance and also the activity of several antioxidant enzymes were investigated. The plants was treated in flowering stage via soil solution. The treated plants were inoculated with a fungal susppension containing 106 spore per 1 mL by injection into the lower part of the spikelets followed by harvesting at 0, 24, 72 and 120 h after inoculation. The induction of resistance was determined by measuering enzymatic activity of gene expression analysis. The results showed that disease severity was reduced in treated plants compared to the control. Biochemical analysis showed that activity of peroxidase and polyphenol oxidase enzymes at all concentrations of silicon was significantly increased. Gene expression analysis showed that expression level of the genes encoding beta-1, 3-glucanase and oxalate oxidase in plants treated with silicon concentration of 6 mmol/l were increased 120h after inoculation. In all, the results of this research indicate that silicon could increase plant resistance against some pathogenic fungi by activation of induced systemic resistance.

Keywords: Head Blight, silicone, qRT-PCR, gene expression.
Comparison of effect of organic media on population of both wild-type and its mutant of biocontrol bacterium, \textit{Pseudomonas fluorescens} VUPf5

Arezoo Lagzian*, Roohallah Saberi-Riseh and Pejman Khodaygan  
Former Graduate Student and Assistant Professors of Plant Pathology, Faculty of Agriculture,  
Vali-e-Asr University of Rafsanjan, Iran  
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ABSTRACT
In this study, the growth of two strains \textit{Pseudomonas fluorescens} VUPf5 and \textit{P. fluorescens} VUPf5-1 (a mutant bacterium showing phenotypic similarity to mutants of regulatory GacS/GacA system) were evaluated in the organic media. This spontaneous mutation blocked the biosynthesis of some antimicrobial compounds including hydrogen cyanide, phenazine and exoprotease in biocontrol strain \textit{Pseudomonas fluorescens} VUPf5. Using organic media amendments by zinc sulfate reduced the growth of mutants. With the aim of increasing VUPf5 population and decreasing VUPf5-1 population, six carbon sources (sugar beet molasses, starch, potato, date, sugar and apple), three nitrogen sources (soybean, urea, ammonium sulfate) and one nitrogen-carbon source (wheat germ) were assessed. Medium 13B (molasses, wheat germ, soybean, sugar, apple, manganese sulfate) was more significantly effective for increasing the growth of strain VUPf5 (5.56×10^{10} cells per ml) as compared to other treatments. Medium 1D (molasses, wheat germ, soybean, sugar, apple, zinc sulfate) greatly caused a reduction in reproduction of strain VUPf5-1. The results could provide a reliable basis for optimized mass production of biocontrol agent in fermentation process.

Keywords: fermentation, mutant, optimization, pseudomonas.

* Corresponding author E-mail: lagzian.az@gmail.com Tel: +98 9157333415
Biological parameters of *Chrysoperla carnea* (Stephens) feeding on common pistachio psylla treated by three plant extracts and amitraz

Faranak Mirzaii¹, Mohammad Amin Samih¹* and Hossein Allahyari³
1. Former M.Sc. Student, and Associate Professor, Department of Plant Protection, Faculty of Agriculture, Vali-e-Asr University of Rafsanjan, Iran
3. Associate Professor, Department of Plant Protection, University College of Agriculture and Natural Resources, University of Tehran, Karaj, Iran
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**ABSTRACT**

The green lacewing *Chrysoperla carnea* (Stephens) (Neuroptera: Chrysopidae) is one of the most important natural enemies of pest insects. The present study was conducted to evaluate the side effects of extracts of three different plant, *Calotropis procera* (Willd.) R. Br. (Asclepiadaceae), *Fumaria parviflora* Lam. (Fumariaceae), *Eucalyptus* spp. (Myrtaceae) and the pesticide amitraz on biological parameters of the second instar larvae of green lacewing. Experiments were conducted at controlled conditions (26±2°C, 65±5 RH and 16:8 h-L:D) feeding on common pistachio psylla, *Agonoscena pistaciae* Burckhardt and Lauterer using two methods of exposure, ingestion and topical application. The results showed that total developmental period (second instar larva to adult) when 2nd instar larvae was treated with *C. procera* extract and amitraz in ingestion was longer than control (water) and there was no significant difference between treatments with control. The results also showed that maximum survival period for adult occurs in *Eucalyptus* spp. with 127 days and minimum with 110 and 113 days in amitraz, and *C. procera* treatment. In topical application method, there was no significant difference between treatment effects on developmental period in immature stage. The developmental period in immature stage for *F. parviflora* with 22.76 was the most effective treatment, and for *C. procera* treatment with 21.96 was the lowest. The results showed that the effect of predators feeding on infected psylla was more than local exposure to pesticides. Increase effect of treatments on developmental stages of the predator with increase of life, shows the effects of sublethal effect of treatments. In integrated pest management strategy, better understanding of these effects will help us to attend to all aspects of safe combination application on natural enemies.

**Keywords:** *Agonoscena pistaciae*, *Chrysoperla carnea*, developmental period, side effects.

* Corresponding author E-mail: samia_aminir@yahoo.com   Tel: +98 9133139550
Optimizations of *Bacillus thuringiensis* native isolate (6R) production in laboratory fermenter

Fatemeh Saberi¹*, Rasoul Marzban² and Mahdi Arjomand¹

1. Former M. Sc. Student and Associate Professor, Department of Biotechnology Chemical Engineering, Islamic Azad University, Science and Research Branch, Tehran, Iran
2. Assistant Professor, Plant Protection Research Institute of the Research, Education and Agricultural Extension, Velenjak, Tehran, Iran
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ABSTRACT

Biological control plays a very important role in pest management. *Bacillus thuringiensis* Ber. has been widely used as a biological control agent in over 60% of microbial-based pesticides and moreover, insect resistance Bt-transgenic crops has been used in the commercial production of a large number of plants (Bt-crops). The successful performance of some of these Bt-based bio-pesticides depends on the concentration of Delta endotoxin and spores in the final product. The medium composition and the conditions under which the bacteria are grown strongly influence these concentrations. Therefore, this study was set to optimize the conditions of Bt growth, i.e. pH, temperature and aeration rate. To reduce the number of experiments, Taguchi statistical method and Qualitek 4 were used. The results showed that the optimum conditions leading to the highest bacterial spores of $1.50 \times 10^{12}$ were the temperature of 28°C, pH of 5.7, and oxygen saturation of 80%. Based on the results, the highest and the lowest interactions among the process parameters were at 39.92% and 0.25% for temperature/aeration rate and temperature/pH, respectively.

Keywords: aeration rate, *Bacillus thuringiensis* pH, optimization, temperature.