Susceptibility of Microtheca ochroloma (Coleoptera:Chrysomelidae) to Botanical and Microbial Insecticide Formulations

Authors: Balusu, Rammohan, and Fadamiro, Henry Y.

Source: Florida Entomologist, 96(3) : 914-921

Published By: Florida Entomological Society

URL: https://doi.org/10.1653/024.096.0327
SUSCEPTIBILITY OF *MICROTHECA OCHROLOMA* (COLEOPTERA: CHrysomelidae) TO BOTANICAL AND MICROBIAL INSECTICIDE FORMULATIONS

RAMMOHAN BALUSU* AND HENRY Y. FADAMIRO

Department of Entomology and Plant Pathology, 301 Funchess Hall, Auburn University, Auburn, AL 38649 U.S.A

*Corresponding author; E-mail: balusrr@auburn.edu

ABSTRACT

The yellowmargined leaf beetle, *Microtheca ochroloma* Stål (Coleoptera: Chrysomelidae), is a key pest of organic crucifer production in the southern United States. The susceptibility of larvae and adults of *M. ochroloma* to some botanical and microbial insecticide formulations was evaluated using laboratory leaf-dip bioassays. Insecticides evaluated included OMRI (Organic Material Review Institute) approved formulations of PyGanic® (pyrethrum), Entrust® (spinosad), Mycotrol® (*Beauveria bassiana* strain GHA), and NOFLY® (*Isaria fumosoroseus* strain FE 9901). Others were MBI-203 (experimental organic formulation of *Chromobacterium subtsugae*) and BotaniGard® 22WP (conventional formulation of *Beauveria bassiana* strain GHA). The insecticides were first evaluated at the field recommended rate against *M. ochroloma* larvae and adults, followed by multiple-concentration assays to determine the LC$_{50}$ and LT$_{50}$ for promising formulations. At the field recommended rates, all tested formulations were toxic to the larvae compared to the untreated control, whereas only Entrust® and PyGanic® were effective against the adults. Entrust® and PyGanic® caused 100% mortality to the larvae and adults after just 24 h of exposure. The LC$_{50}$ values of Entrust® and PyGanic® were 200 X and 15 X less than the field recommended rates, respectively. MBI-203 was effective against the larvae (100% mortality after 5 days) but not against the adults. The entomopathogenic fungal formulations, Mycotrol®, NOFLY®, and BotaniGard®, were much less toxic with LT$_{50}$ values of 10, 12, and 9 days, respectively. Although all 3 fungal formulations caused significantly higher larval mortality than the untreated control after 5 days of exposure, none resulted in more than 50% larval or 14% adult mortalities over the 9-day exposure period.

Key Words: Yellowmargined leaf beetle, botanicals, biopesticides, LC$_{50}$, pyrethrum, spinosad

RESUMEN

El escarabajo de las hojas de margen amarillo, *Microtheca ochroloma* Stål (Coleoptera: Chrysomelidae), es una plaga clave de la producción de crucíferas orgánicas en el sur de los Estados Unidos. Se evaluó la susceptibilidad de las larvas y adultos de *M. ochroloma* a algunas formulaciones de insecticidas botánicos y microbianos mediante bioensayos de hojas sumergidas en el laboratorio. Los insecticidas evaluados incluyeron formulaciones aprobadas por el OMRI (Instituto de Revision de Materia Organica) como PyGanic® (piretro), Entrust® (spinosad), Mycotrol O® (*Beauveria bassiana* cepa GHA) y NOFLY® (*Isaria fumosoroseus* cepa FE 9901). Los otras formulaciones fueron MBI-203 (formulación orgánica experimental de *Chromobacterium subtsugae*) y BotaniGard® 22WP (formulación convencional de *Beauveria bassiana* cepa GHA). Los insecticidas fueron evaluados con dosis de campo recomendadas contra larvas y adultos de *M. ochroloma*, seguido por los ensayos de concentración múltiple para determinar la CL$_{50}$ y LT$_{50}$ de las formulaciones prometedoras. En las proporciones recomendadas de campo, todas las formulaciones probadas fueron tóxicas para las larvas en comparación con el control sin tratar, mientras que sólo Entrust® y PyGanic® fueron eficaces contra los adultos. Entrust® y PyGanic® causó una mortalidad del 100% de las larvas y los adultos después de sólo 24 horas de exposición. Los valores de LC$_{50}$ de Entrust® y PyGanic® fueron 200x y 15x menores que las proporciones recomendadas de campo, respectivamente. La MBI-203 fue eficaz contra las larvas (100% de mortalidad después de 5 días), pero no contra los adultos. Las formulaciones de hongos entomopatógenos, Mycotrol®, NOFLY® y BotaniGard®, fueron menos tóxicos, con LT$_{50}$ valores de 10, 12 y 9 días, respectivamente. Aunque todas las 3 formulaciones de hongos causaron una mortalidad de larvas significativamente más alta que el control sin tratar después de 5 días de
The yellowmargined leaf beetle, *Microthea ochroloma* Stål (Coleoptera: Chrysomelidae), is a major pest of cruciferous vegetables (Brassicaceae: Brassicaceae) in the southern United States (USA) (Chamberlin & Tippins 1948; Ameen & Story 1997a). This beetle, which was accidentally introduced into the USA from South America in the 1940s (Chamberlin & Tippins 1948), is now widely distributed in the southern USA with major field infestations reported in Alabama, Florida, Louisiana, Mississippi, South Carolina, North Carolina, and Texas (Ameen & Story 1997b). Both larvae and adults of *M. ochroloma* feed in groups on foliage and may cause complete defoliation of crucifers. Although *M. ochroloma* is often not a major problem in conventional crucifer production due to its susceptibility to synthetic foliar insecticides (Bowers 2003), the beetle poses a major threat to organic vegetable production since organic farmers cannot use synthetic insecticides. Organic production of crucifers is presently an emerging industry in Alabama and other parts of the southern USA, and *M. ochroloma* is often the predominant pest and a major factor limiting the growth and expansion of the industry (Balusu & Fadamiro 2012).

Several years of field studies by our group and others have identified only a few effective OMRI (Organic Material Review Institute) approved formulations, specifically Entrust® (spinosad), against *M. ochroloma* (Overall 2008; Balusu & Fadamiro 2012). PyGanic® (pyrethrum) was moderately effective, while other tested insecticides, including some entomopathogenic fungal formulations showed very little or no efficacy against larvae or adults (Balusu & Fadamiro 2012). Therefore, spinosad (Entrust®) and pyrethrum (PyGanic®) are currently recommended for management of this pest in organic crucifer vegetable production in Alabama. However, repeated application of these insecticides could lead to development of insecticide resistance. Identification of additional effective products could provide growers more control options and help to reduce the risk of insecticide resistance. Also, knowledge of baseline susceptibility response of *M. ochroloma* to spinosad (Entrust®) and pyrethrum (PyGanic®) is essential for keeping track of changes in toxicological response of the pest over time. Baseline susceptibility data can be used as reference values to diagnose shifts in susceptibility or to monitor insecticide resistance (Prabhaker et al. 2006).

Many of the microbial insecticides, including entomopathogenic fungal formulations, which showed poor field efficacy against *M. ochroloma* in our field study (Balusu & Fadamiro, 2012), have been reported as effective against other Chrysomelidae (Poprawski et al. 1997; Butt et al. 1992). Thus, an understanding of the factors responsible for the limited field efficacy of entomopathogenic fungal formulations against *M. ochroloma* is important to allow us to overcome constraints and thereby achieve efficacious control of *M. ochroloma*. Poor field performance of entomopathogenic fungal formulations is typically related to suboptimal ambient conditions such as relative humidity, temperature, and UV-radiation (Inglis et al. 1996). However, studying these factors under field conditions is extremely complex. Evaluation of the microbial formulations against *M. ochroloma* under controlled laboratory conditions will likely provide an insight into the factors responsible for their poor field efficacy.

Thus, the present study was conducted to evaluate the susceptibility of *M. ochroloma* larvae and adults to various botanical and microbial formulations under laboratory conditions. Many of the insecticides tested in the current paper are known to be effective against other Coleopteran insect pests (Andersen et al. 2006; Igrc Barcic et al. 2006; Isman 2006) and thus are expected to show activity against *M. ochroloma*.

Ultimately, it is hoped that the results of this laboratory study will help to understand the basis for the poor performance of the microbial insecticides tested in our field trials (Balusu & Fadamiro, 2012), and allow identification of additional effective organically acceptable insecticides which can be applied as stand-alone treatments or in rotation with Entrust® for effective management of *M. ochroloma* in organic crucifer production. Furthermore, knowledge of baseline susceptibility of *M. ochroloma* to various insecticides can be used to determine changes in susceptibility over time and the onset of resistance development.

**MATERIALS AND METHODS**

Plants

Turnip (Brassica rapa L.var. rapa cv ‘purple top white globe’) seedlings were raised from seeds purchased from Johnny’s Selected Seeds (Winslow, Maine) in 60-well seed trays at one seed per well under controlled greenhouse conditions (26 ± 2 °C and 55 ± 5% RH). Seedlings (3 wk old) were

Palabras clave: El escarabajo de las hojas del margen amarillo, botánicos, bioplágicidas, ensayo de hojas sumergidas, LC₅₀ , piretro, spinosad, *Isaria fumosoroseus*
transplanted into 0.5-L pots in Sunshine potting mixture #8 consisting of 70-80% Canadian sphagnum grower grade peat moss, coarse grade perlite, coarse grade vermiculite, dolomitic limestone for pH adjustment, gypsum, and wetting agent (SunGro Horticulture, Washington). Plants were irrigated daily and fertigated twice a wk with Scotts® peat lite special fertilizer (Scotts-Sierra Horticultural Product Company, Marysville, Ohio), a 20-10-20 water soluble NPK fertilizer mixture with micronutrients. Plants were grown using organic practices and no pesticides were applied. Plants used for the bioassays and insect rearing were about 4-5 wk post transplanting.

Insects

Adults of *M. ochroloma* collected from a commercial organic farm in central Alabama in October 2006 were used to start laboratory colonies, which were supplemented annually with field-collected adults. *Microtheca ochroloma* was reared on foliage of turnip plants (grown in the greenhouse as described above) in clear plastic Petri dishes (150 mm diam × 30 mm height) lined with paper towels (Bounty®, Procter & Gamble, Cincinnati, Ohio). The colony was maintained at 25 ± 2 °C, 50 ± 10% RH, and a photoperiod of 14:10 h L: D.

Treatments

The materials evaluated (Table 1) included OMRI (Organic Material Review Institute) approved formulations such as PyGanic® (pyrethrum), Entrust® (spinosad), Mycotrol O® (*Beauveria bassiana* strain GHA), and NOFLY® (*Isaria fumosoroseus* strain FE 9901). Others were MBI-203 (experimental organic formulation of *Chromobacterium subtsugae*) and BotaniGard® 22WP (conventional formulation of *Beauveria bassiana* strain GHA). Entrust® WP (spinosad), the most effective treatment in our field trials, was evaluated as positive control, whereas BotaniGard® 22WP was evaluated as a positive control for organic standard.

Bioassay Method

Toxicity of the insecticides (Table 1) against *M. ochroloma* larvae and adults was determined in 2 experiments. Single (field recommended rate) concentration screening assays were first carried out, and the most promising treatments were further evaluated in multi-concentration assays. Solutions of each insecticide were made in distilled water (= control) and no adjuvants were used. All bioassays were performed using the leaf-dip method at ambient conditions, 26 °C, 14:10 h L: D, and 50% RH. Briefly, intact leaves on a turnip plant were

### Table 1. Insecticides Tested Against Microtheca ochroloma.

| Registered name | Active ingredient | Type | Field recommended rate | Company |
|-----------------|-------------------|------|------------------------|---------|
| PyGanic® 1.4 EC | Pyrethrum         | OMRI approved | 2 qt/ac (4.68 L/ha) | McLaughlin Gormley King Company, Minneapolis, MN |
| Entrust® WP     | Spinosad          | OMRI approved | 2 oz/ac (140 g/ha)  | Dow AgroSciences LLC, Indianapolis, IN |
| Mycotrol O® ES  | *Beauveria bassiana* strain GHA | OMRI approved | 1 qt/ac (2.34 l/ha) | Laverlam International Corporation, Butte, MT |
| NOFLY® WP       | *Isaria fumosoroseus* strain FE 9901 | OMRI approved | 2 lb/ac (2.25 kg/ha) | Natural Industries, Inc. Houston, Houston, TX |
| MBI-203 SC      | *Chromobacterium subtsugae* | Experimental | 2 qt/ac (4.68 L/ha) | Marrone Bio Innovations, Davis, CA |
| BotaniGard® 22WP| *Beauveria bassiana* strain GHA | Conventional | 2 lb/ac (2.25 kg/ha) | Laverlam International Corporation, Butte, MT |
directly immersed in each insecticide solution for 5 s. The leaves were then removed and air dried for ~4 h to remove excess water on the leaf surface. A leaf disc (~90 mm) cut out of an excised treated leaf was placed in a 100 mm diam Petri dish lined with moist filter paper. To maintain higher humidity levels than the ambient RH, the Petri dishes were covered with damp paper towel and placed inside a large plastic container (25 × 45 cm). Mortality of insects was recorded daily for 14 days (or until pupation of larvae). Insects that failed to move when probed with a dissecting needle were recorded as dead and removed from the Petri dish. For the entomopathogenic fungal formulations, dead insects were removed and incubated separately in Petri dishes lined with damp filter paper. The Petri dishes were placed in a humidifier and inspected for the presence of mycelia on cadavers (mycosis) starting on day 7.

Toxicity at the Field Recommended Rates

In the first experiment, all insecticide formulations were evaluated at field recommended rates (i.e., single concentration assays) for efficacy against M. ochroloma larvae and adults. Test solutions of field recommended rates of the insecticides (Table 1) were prepared with distilled water. A group of 20 1st instar larvae (larvae that emerged within 24 h) from the same batch was placed in a Petri dish containing a treated leaf disc. Similarly, a group of 20 adults (4-5 days old) from the same batch was placed in a Petri dish containing a treated leaf disc by using a fine camel’s hair brush (#00). The experiment was replicated 5 times per insecticide over time and insect mortality was determined as described above.

Multi-Concentration Assays

Promising treatments in the first experiments were selected for further evaluation in multi-concentration assays to determine the LC50 (lethal concentration that kills 50% of test insects or median lethal concentration) and LT50 (lethal time to kill 50% of test insects). Based on the results of the first experiment, all 6 insecticides were evaluated against the larvae, but only 2 (Entrust® and PyGanic®) were evaluated against the adults. Each insecticide was tested at 5 concentrations plus distilled water control for a total of 6 rates. The concentration range for each insecticide was determined based on the results of preliminary bioassays, which gave a mortality range of 10 to 90%. For each concentration, a group of 20 adults (4-5 days old) or 20 larvae (1 day old) from the same batch were placed in a Petri dish containing a treated leaf disc. The experiment was replicated 5 times per concentration over time and insect mortality was determined as described above.

Data Analysis

Mortality data did not meet the assumptions of independence. Thus, the data were analyzed using the Kruskal-Wallis non-parametric test (P < 0.05; JMP® 7.0.1, SAS Institute 2007). The data were further analyzed using the ordinary F-test after which the results were compared with the non-parametric test. When both procedures gave similar results, the ANOVA assumptions were assumed to be satisfied (http://faculty.evansville.edu/ch81/bio415f02/BIO415Topic7.pdf). Means were then separated using the Tukey Kramer honestly significant difference (HSD) test. The LC50 values expressed in ppm, LT50 values (in days), 95% fiducial limits (FL), and regression slopes were estimated by probit analysis (Finney 1971) using POLO PLUS software for Windows (LeOra software 2007). Tests of parallelism of probit regression lines for all treatments were conducted using chi-square goodness-of-fit tests (POLO PLUS, LeOra software 2007).

RESULTS

Toxicity at Field Recommended Rates

There was a significant effect of insecticide treatment at the field recommended rate on mortality of M. ochroloma larvae (F = 1251.2, df = 6, 28, P < 0.0001) as early as 24 h of exposure to treated leaf discs (Fig. 1). Entrust® and PyGanic® resulted in 100% larval mortality after 24 h. MBI-203 also caused significantly greater mortality than the untreated control starting at 24 h; however 100% mortality was not attained until day 5. In contrast, no significant differences were recorded between the entomopathogenic fungal formulations (i.e. Botanigard®, Mycotrol O®, and

Fig. 1. Percent mortality of Microtheca ochroloma larvae exposed to field recommended rates of various insecticide formulations in leaf-dip bioassays. Means within a date having no letter in common are significantly different (ANOVA, Tukey-Kramer HSD, P < 0.05).
The results of this laboratory study demonstrated varying levels of susceptibility of *M. ochroloma* to the tested insecticides. Among the formulations, Entrust® was the most effective causing 100% larval and adult mortality within 24 h, as well as having the lowest LC₅₀ values and survival time. PyGanic®, the second best treatment, also caused complete mortality of both life stages within 24 h, but was ~100-fold and 10-fold less toxic than Entrust® to the larvae and adults, respectively. The results further showed that the LC₅₀ values of Entrust® and PyGanic® were only a fraction, 200X and 15X less than the actual field recommended rate, respectively. MBI-203 (experimental organic formulation of *Chromobacterium subtsugae*) was effective against the larvae but not against the adults. At the field recommended rate, the entomopathogenic fungal formulations (Mycotrol®, NOFLY®, and BotaniGard®) were comparatively less efficacious against the larvae and showed no efficacy against the adults.

**DISCUSSION**

terms-use
The efficacy of Entrust® or its active ingredient (spinosad) has also been documented against other beetles in the same family (Chrysomelidae) as *M. ochroloma*, including flea beetles, *Phyllotreta* spp., in crucifer crops (Andersen et al. 2006), *Epitrix tuberis* Gentner in potato (Chu et al. 2006), and Colorado potato beetle, *Leptinotarsa decemlineata* (Say) in potato (Igrc Barcic et al. 2006). Entrust® was also effective against lepidopteran pests of crucifer crops in Alabama (Maxwell & Fadamiro 2006). The efficacy of Entrust® recorded in the present study may be attributed to its broad spectrum activity (Cisneros et al. 2002), multiple modes of entry (Eger & Lindenberry 1998, Liu et al. 1999). The active ingredient in Entrust® is both a contact and stomach poison (Eger & Lindenberry 1998; Liu et al. 1999). The efficacy of PyGanic®, a botanical insecticide with pyrethrum as the active ingredient, against *M. ochroloma* was not surprising, since pyrethrum is known for its rapid knockdown effect on insects. PyGanic® has also been reported as effective against other insect pests including Colorado potato beetle (Igrc Barcic 2006), and harlequin bug, *Murgantia histrionica* (Hahn) (Overall 2008). In general, the results of this laboratory study are in agreement with our field data which identified Entrust® as the only effective treatment and PyGanic® as moderately effective (Balusu & Fadamiro 2012). The high activity of the experimental formulation MBI-203 against *M. ochroloma* larvae is very encouraging and suggests that (if registered) this insecticide may play a role in the management of *M. ochroloma*. Chromobacterium subtugae, the active ingredient in MBI-203, has been reported to be toxic to several insects including larvae of Colorado potato beetle (Martin et al. 2004), as well as adults of southern corn rootworms, *Diabrotica undecimpunctata howardi* Barber and *Diabrotica virgifera virgifera* LeConte, and southern green stink bug, *Nezara viridula* (L.) (Martin et al. 2007).

The entomopathogenic fungal formulations were only slightly effective against *M. ochroloma* larvae and did not work against the adults. Although some studies have reported the efficacy of entomopathogenic fungal formulations against Colorado potato beetle (Poprawski et al. 1997) and other chrysomelid beetle species (Butt et al. 1992), our laboratory results, which also agree with our field data (Balusu & Fadamiro 2012), suggest that these fungal formulations are relatively non-toxic to *M. ochroloma*. The poor efficacy of the fungal formulations against *M. ochroloma* in this laboratory study is unlikely due to suboptimal environmental conditions, but may be attributed to some plant or insect-related factors as follows: First, low pathogenicity of fungal formulations may be due to potential negative interactions of surface chemicals of crucifer plants. Crucifer crops contain specialized secondary plant metabolites called glucosinolates that are transformed into isothiocyanates during insect damage. Isothiocyanates are known for their fungistatic properties. In fact, many studies have reported the inhibitory activity of isothiocyanates, specifically phenylethyl-, 2-chlorophenyl-

### Table 2. Concentration-Mortality Response of *Microtheca ochroloma* Larvae Exposed to Various Insecticide Formulations in Leaf-Dip Assays.

| Treatment      | No. insects | Slope ± SE | LC₅₀ (ppm of a.i) | 95% Fiducial limits (ppm) | χ²  |
|----------------|-------------|------------|------------------|---------------------------|-----|
| PyGanic®       | 121         | 2.86 ± 0.23| 10.9 b           | 8.48 - 14.03              | 8.85|
| Mycotel O®     | 118         | 1.99 ± 0.25| 6.67 × 10⁴a      | 534.9 - 922.2             | 2.46|
| NOFLY®         | 114         | 1.28 ± 0.29| 4.08 × 10⁴a      | 2018.5 - 25038            | 0.78|
| MBI-203        | 118         | 6.21 ± 0.66| 5.21 × 10⁴a      | 4064.2 - 6417.2           | 8.55|
| Entrust®       | 118         | 1.15 ± 0.08| 0.1 c            | 0.061 - 0.18              | 10.81|
| BotaniGard®    | 117         | 1.96 ± 0.27| 1.3 × 10⁴a       | 829.4 - 5938.6            | 6.1 |

### Table 3. Probit Analysis of Time-Mortality Response of Bioassay with Test Formulation Against *Microtheca ochroloma* Larvae.

| Treatment      | No. insects | Slope ± Se  | LT₅₀ (days) | 95% Fiducial limits (days) | χ²  |
|----------------|-------------|-------------|-------------|---------------------------|-----|
| Mycotel O®     | 108         | 2.54 ± 0.25 | 10.44 a     | 7.82 - 21.52              | 34.88|
| NOFLY®         | 114         | 4.77 ± 0.68 | 12.26 a     | 10.77 - 15.27             | 3.09 |
| MBI-203        | 112         | 5.34 ± 0.31 | 2.21 b      | 1.94 - 2.58               | 23.9 |
| BotaniGard®    | 111         | 5.88 ± 0.58 | 9.03 a      | 8.52 - 9.76               | 0.67 |
and allyl-isothiocyanates, on entomopathogenic fungal conidia germination and its ability to infect the insects (Inyang et al. 1999; Klingen et al. 2002). Second, the physio-chemical properties of host insect cuticle could also account for its lower susceptibility to fungal pathogens. *Microtheca ochroloma* may sequester plant-derived fungistatic compounds (isothiocyanates) from turnip plant as defense against potential microbial pathogens. The presence of fungistatic compounds in the insect cuticle is well demonstrated (Smith & Grula 1982; Butt et al. 1995). Butt (1990) showed that insect cuticle influences all stages of the infection process: adhesion, germination, and appresorium differentiation. Lastly, insect nutrition is another factor that plays an important role in the success of entomopathogens (Vago 1963). Inadequate nutrition often results in increased susceptibility to pathogens. Balusu & Fadamiro (2011) demonstrated that turnip is a preferred host of *M. ochroloma*. Therefore, it is possible that *M. ochroloma* raised on turnip leaves are more vigorous and less susceptible to fungal infection. Despite their poor efficacy in reducing adult mortality, we observed in this study some sub-lethal effects of some microbial formulations (i.e., MBI-203, Mycotrol O®, and BotaniGard® 22WP) on *M. ochroloma* in terms of reduced feeding and oviposition. If confirmed, these sub-lethal effects can be as valuable as direct insect mortality (Liu & Story 1999: 803-810). In summary, this study has identified some promising OMRI – acceptable biopesticides and effective rates against *M. ochroloma* larvae and adults. Entrust® and PyGanic® were the most effective insecticides followed by MBI-203. The data also showed that the actual lethal concentrations of Entrust® and PyGanic® were only fractions of the field recommended rates. Additional studies are necessary to further evaluate the field activity of the promising treatments identified in this study, in particular in rotation with Entrust® for effective management of *M. ochroloma* in organic crucifer production.

**ACKNOWLEDGMENTS**

The authors thank the following companies for providing insecticide formulations for this study: McLaughlin Gormley King Company (Minneapolis, Minnesota), Dow AgroSciences LLC (Indianapolis, Indiana), Laverlam International Corporation (Butte, Montana), Natural Industries, Inc. (Houston, Texas), and Marrone Bio Innovations (Davis, California). We also thank our undergraduate student workers, Shelia Boyt, David Appel, and Allison Tyler, for helping with insect rearing. Funding for this study was provided in part through grants by the Alabama Agricultural Experiment Station (Auburn University) and USDA-IR-4 program, as well as limited funding support from Marrone Bio Innovations and McLaughlin Gormley King Company.

**REFERENCES CITED**

AMEEN, A. O., AND STORY, R. N. 1997a. Biology of the yellowmargined leaf beetle (Coleoptera: Chrysomelidae) on crucifers. J. Entomol. Sci. 32: 478-486.

AMEEN, A. O., AND STORY, R. N. 1997b. Feeding preferences of larval and adult *Microtheca ochroloma* (Coleoptera: Chrysomelidae) for crucifer foliage. J. Agric. Entomol. 14: 363-368.

ANDERSEN, C. L., HAZZARD, R., VAN DRIESCHE, R., AND MANGAN, F. X. 2006. Alternative management tactics for control of *Phyllotreta cruciferae* and *Phyllotreta striolata* (Coleoptera: Chrysomelidae) on *Brassica rapa* in Massachusetts. J. Econ. Entomol. 99: 803-810.

BALUSU, R. R., AND FADAMIRO, H. Y. 2011. Host finding and acceptance preference of the yellowmargined leaf beetle, *Microtheca ochroloma* (Coleoptera: Chrysomelidae), on cruciferous crops. Environ. Entomol. 40: 1471-1477.

BALUSU, R. R., AND FADAMIRO, H. Y. 2012. Evaluation of organically acceptable insecticides as stand-alone treatments and in rotation for managing yellowmar-
ginned leaf beetle, Microtheca ochroloma (Coleoptera: Chrysomelidae), in organic crucifer production. Pest Mgt. Sci. 68: 573-579.

Bowers, K. 2003. Effects of within-field location of host plants and intercropping on the distribution of Microtheca ochroloma (Stål) in mizuna. M.S. thesis. Univ. Florida, Gainesville.

Butt, T. M. 1990. Fungal infection processes—a mini-review. Vth Intl. Colloq. Invertebr. Pathol., Adelaide, Australia, pp. 121-124.

Butt, T. M., Barrisever, M., Drummond, J., Schuler, T. H., Tillemans, F. T., and Wilding, N. 1992. Pathogenicity of the entomogenous, hyphomycete fungus, Metarhizium anisopliae, against the chrysomelid beetles Pseudolides chrysopoecophala and Phaedo cochleariae. Biocontrol Sci. Technol. 2: 325-332.

Butt, T. M., Ibrahim, L., Clark, S. J., and Beckett, A. 1995. The germination behaviour of Metarhizium anisopliae on the surface of aphid and flea beetle cuticles. Mycol. Res. 99: 945-950.

Chamberlin, F. S., and Tippins, H. H. 1948. Microtheca ochroloma, an introduced pest of crucifers, found in Alabama. J. Econ. Entomol. 41: 979.

Christie, P. T., and Wright, D. J. 1980. Activity of abamectin against larval stages of Spodoptera litoralis Boisduval and Heliothis armigera Hubner (Lepidoptera: Noctuidae) and possible mechanisms determining differential toxicity. Pesticide Sci. 29: 29-38.

Chu, J. W., SyrovY, L., and Meberg, H. 2006. The Effects of NOVODOR® (Bacillus thuringiensis subsp. tenebrionis) and ENTRUST® (Spinosad) on reproduction and feeding activity of Epitrix tuberis (Coleoptera: Chrysomelidae) in potato. Report from Certified Organic Assoc. British Columbia. 8 pp.

Cisneros, J. D., Goulson, D., Derwent, L. C., Pena-Barcic, J., Bazok, R., Bezjak, S., Gotlin-Culjak, B., Butt, T. M., Bariusever, M., Drummond, J., Schuler, T. H., Tillemans, F. T., and Wilding, N. 1992. Activity of abamectin against larval stages of Spodoptera litoralis Boisduval and Heliothis armigera Hubner (Lepidoptera: Noctuidae) and possible mechanisms determining differential toxicity. Pesticide Sci. 29: 29-38.

Eger, J. E., and Linderberry, J. E. 1998. Utility of spinosad for insect control in Florida vegetables, pp. 55-57 In Proc. Florida State Hort. Soc. Volume. 111.

Finney, D. J. 1971. Probit analysis, 3rd ed. Cambridge University press, Cambridge, United Kingdom.

Igrić Baricć, I., Bazok, R., Bezjak, S., Gotlin Culjak, T., and Baricj, J. 2006. Combinations of several insecticides used for integrated control of Colorado potato beetle (Leptinotarsa decemlineata Say., Coleoptera: Chrysomelidae). J. Pest Sci. 79: 223-232.

Inglis, G. D., Johnson, D. L., and Goettel, M. S. 1996. Effects of temperature and thermoregulation on mycosis by Beauveria bassiana. Biol. Control 7: 131-139.

Inyang, C. N., Butt, T. M., Dougherty, K. J., Todd, A. D., and Archer, S. 1999. The effect of isothiocyanates on the growth of the entomopathogenic fungus Metarhizium anisopliae and its infection of the mustard beetle. Mycol. Res. 103: 974-980.

Isman, M. B. 2006. Botanical insecticides, deterrents, and repellents in modern agriculture and an increasingly regulated world. Annu. Rev. Entomol. 51: 45-66.

Klingle, L., Hajek, A., Meadow, R., and Renwick, J. A. A. 2002. Effect of brassicaceous plants on the survival and infectivity of insect pathogenic fungi. BioControl 47: 411-425

Liu, H. P., and Bauer, L. S. 2008. Microbial control of emerald ash borer, Agrilus planipennis (Coleoptera: Buprestidae) with Beauveria bassiana strain GHA: greenhouse and field trials. Biol. Control 45: 124-132.

Liu, T. X., Sparks, A. N., Hendrix, W. H., and Yue, B. 1999. Effects of SpinTor on cabbage looper (Lepidoptera: Noctuidae): toxicity and persistence of leaf residue on cabbage under field and laboratory conditions. J. Econ. Entomol. 92: 1266-1273.

Martin, P. A. W., Blackburn, M., and Shropshire, A. S. 2004. Two new bacterial pathogens of Colorado potato beetle (Coleoptera: Chrysomelidae). J. Econ. Entomol. 97: 774-780.

Martin, P. A. W., Hirose, E., and Aldrich, J. R. 2007. Toxicity of Chromobacterium subsugae to Southern Green Stink Bug (Heteroptera: Pentatomidae) and Corn Rootworm (Coleoptera: Chrysomelidae). J. Econ. Entomol. 100: 680-684.

Maxwell, E., and Fadamiro, H. Y. 2006. Evaluation of several reduced-risk insecticides in combination with an action threshold for managing lepidopteran pests of cole crops in Alabama. Florida Entomol. 89: 117-126.

Overall, L. M. 2008. Evaluation of organic insecticides to control harlequin bug, Murgantia histrionica (Hahn), and yellowmargined leaf beetle, Microtheca ochroloma (Stål), on leafy greens. M.S. thesis, Oklahoma State University, Stillwater, Oklahoma.

Poprawski, T. J., Carruthers, R. I., Speese III, J., Vacek, D. C., and Wendel, L. E. 1997. Early-season applications of the fungus Beauveria bassiana and introduction of the hemipteran predator Perillus bioculatus for control of the Colorado potato beetle. Biol. Control 10: 48-57.

Prabhaiker, N., Castle, S., Byrne, F., Henneberry, T. J., and Toscano, N. C. 2006. Establishment of baseline susceptibility data to various insecticides for Homalodisca coagulata (Homoptera: Cicadellidae) by comparative bioassay techniques. J. Econ. Entomol. 99: 141-154.

Smith, R. J., and Grula, A. E. 1982. Toxic components on the larval surface of the corn earworm (Heliothis zea) and their effects on germination and growth of Beauveria bassiana. J. Invertebr. Pathol. 39: 15-22.

Vago, C. 1963. Predispositions and interrelations in insect diseases, pp. 339-379 In E. A. Steinhaus [ed.], Insect Pathology: An Advanced Treatise. Academic Press, New York.