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SUSCEPTIBILITY OF MICROTHECA OCHROLOMA (COLEOPTERA: CHRYSOMELIDAE) TO BOTANICAL AND MICROBIAL INSECTICIDE FORMULATIONS

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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 LC50 and LT50 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 LC50 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 LT50 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, LC50, 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 CL50 o LT50 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 LC50 de Entrust® y PyGanic® fueron 200 X y 15 X 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 LT50 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 exposición, ninguna resultó en más de 50% larval or 14% adult mortalities over the 9-day exposure period.

Key Words: Escarabajo de las hojas de margen amarillo, botánicos, biopesticidas, LC50, piretro, spinosad
The yellowmargined leaf beetle, Microtheca ochroloma Stål (Coleoptera: Chrysomelidae), is a major pest of cruciferous vegetables (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
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 x 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
直接浸入每种杀虫剂的溶液中5秒。叶子被移除并风干约4小时，移除叶子表面的水分。从叶子中切出一个90毫米的叶子圆片，放入一个100毫米直径的培养皿，用湿的滤纸覆盖。培养皿放置在一个湿度发生器中，并在培养皿内放置湿的滤纸。培养皿被取出并单独放置在培养皿中。对叶片表面进行消毒处理，杀死昆虫。

在试验中，所有杀虫剂均在田间推荐的速率下进行评估。试验溶液在5个浓度下，每种杀虫剂重复5次。昆虫的死亡率用上述方法确定。

数据处理

数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；JMP® 7.0.1，SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；SAS Institute 2007）。数据未达到参数化独立性的假设。因此，数据采用Kruskal-Wallis非参数测试（P < 0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；SAS Institute 2007）。数据进一步分析了普通的ANOVA假设，即P值在0.05；SAS Institute 2007）。
NOFLY® and the untreated control on days 1-4. On day 5 the fungal formulations caused significantly higher mortality than the untreated control ($F = 492.9$, df = 6, 28, $P < 0.0001$). However no fungal formulation resulted in more than 50% larval mortality throughout the exposure period (Fig. 1). The average survival time for larvae treated with Entrust®, PyGanic®, MBI-203, Botanigard® Mycotrol O®, and NOFLY® at the field recommended rate was 1, 5, 8, 8, and 7 days after treatment, respectively (Fig. 1).

For adults, Entrust® and PyGanic® were the most effective treatments with 100% mortality after 24 h, which was significantly greater than the other treatments or the untreated control ($F = 1974$, df = 6, 28, $P < 0.0001$) (Fig. 2). No significant differences were recorded between the other treatments and the untreated control on days 1-7. On day 8, Botanigard® (14% mortality) and Mycotrol O® (12% mortality) were significantly better than the untreated control ($F = 979.1$, df = 6, 28, $P < 0.0001$), whereas mortalities caused by MBI-203 (2%) and NOFLY® (2%) were not significantly different from the untreated control (Fig. 2). The average survival time for adults treated with Entrust®, PyGanic®, Botanigard®, and Mycotrol O® at the field recommended rate was 1, 1, 9, and 9 days, respectively (Fig. 2).

In general, the fungal formulations were slow-acting and relatively less efficacious against *Microtheca ochroloma*. None resulted in more than 50% larval or 14% adult mortality over the 9-day exposure period. Comparing the 2 life stages, the larvae were more susceptible to the insecticides than adults.

**Multi-Concentration Assays**

The LC$_{50}$ and LT$_{50}$ values, 95% fiducial limits, slope, and chi-square values for the insecticides tested against larvae are presented in Table 2. All chi-square values were not significant ($\alpha = 0.05$) in Pearson’s goodness-of-fit test on the probit model, indicating a good fit of regression line. Entrust® and PyGanic® had the lowest LC$_{50}$ values against the larvae (Table 2), indicating higher toxicity. However, Entrust® (LC$_{50} = 0.1$ ppm) was ~100 times more toxic to the larvae than PyGanic® (LC$_{50} = 10.9$ ppm). All other treatments (Mycotrol O®, NOFLY®, MBI-203, and BotaniGard®) were not significantly different after 24 h of exposure. Significant concentration-mortality responses of the larvae were observed for all insecticides tested, as indicated by the positive slope values (Table 2). MBI-203 had the highest slope (6.21 ± 0.66), followed by PyGanic® (2.86 ± 0.23), Mycotrol O® (1.99 ± 0.25), BotaniGard® (1.96 ± 0.27), NOFLY® (1.28 ± 0.29), and Entrust® (1.15 ± 0.08); higher slopes indicate greater concentration-mortality response. The second measure of efficacy was the LT$_{50}$ values that were calculated for the field recommended rate (Table 3). LT$_{50}$ values were not estimated for Entrust® and PyGanic® since both treatments caused complete mortality of larvae within 24 h at the field recommended rate. Among the remaining treatments, MBI-203 had significantly lower LT$_{50}$ (2 days) than BotaniGard® (9 days), Mycotrol O® (10 days), or NOFLY® (12 days) (Table 3).

Since Entrust® and PyGanic® were the only effective treatments against the adults at the field recommended rate (experiment 1), LC$_{50}$ values were estimated only for these two formulations against the adults (Table 4). Entrust® (LC$_{50} = 2.4$ ppm) was ~10 times more toxic to the adults than PyGanic® (LC$_{50} = 24.1$ ppm). LT$_{50}$ values were not estimated since both insecticides caused complete mortality within 24 h of exposure.

**DISCUSSION**

The results of this laboratory study demonstrated varying levels of susceptibility of *Microtheca 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 LC$_{50}$ 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 LC$_{50}$ values of Entrust® and PyGanic® were only a fraction, 200 X and 15 X less than the actual field recommended rate, respectively. MBI-203 (experimental organic formulation of *Chromobacterium subtsgae*) 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.
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 subtsgae*, 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 undecimtacta 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-,
and allyl-isothiocyanates, on entomopathogenic fungal conidia germination and its ability to infect the insects (Inyang et al. 1999; Klingien 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 appressorium 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 & Bauer 2008) in decreasing the pest status of M. ochroloma, and are worthy of further investigation.

The results showed that M. ochroloma larvae were more susceptible than the adults to the tested insecticides. For instance, the LC50 value of Entrust® against the larvae (0.1 ppm) was 20X lower than that of the adults (2.36 ppm). This difference in susceptibility may be attributed to the differential feeding rate of both life stages; larvae are more voracious feeders than adults (unpublished data). Another possible explanation for the differential susceptibility observed between the 2 life stages may be a difference in the ability of the insecticides to penetrate through the cuticle or a difference in the composition of the cuticle. Christie & Wright (1990) attributed marked differences in relative toxicity of the insecticide abamectin between larval instars of Spodoptera littoralis (Boisdruval) to differences in the insecticide’s penetration rate.

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.

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<table>
<thead>
<tr>
<th>Treatment</th>
<th>No. insects</th>
<th>Slope ± SE</th>
<th>LC50 (ppm of a.i)</th>
<th>Lower - Upper</th>
<th>χ²</th>
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<tbody>
<tr>
<td>PyGanic®</td>
<td>117</td>
<td>3.24 ± 0.27</td>
<td>24.14 a</td>
<td>19.71 - 31.22</td>
<td>8.85</td>
</tr>
<tr>
<td>Entrust®</td>
<td>118</td>
<td>1.15 ± 0.08</td>
<td>2.36 b</td>
<td>1.30 - 4.29</td>
<td>10.81</td>
</tr>
</tbody>
</table>

TABLE 4. CONCENTRATION-MORTALITY RESPONSE OF Microtheca ochroloma ADULTS EXPOSED TO VARIOUS INSECTICIDE FORMULATIONS IN LEAF-DIP ASSAYS


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