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EVALUATION OF INSECTICIDE CHEMISTRIES AGAINST THE LEEK MOTH (LEPIDOPTERA: ACROLEPIIDAE), A NEW PEST IN NORTH AMERICA

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Abstract

The leek moth, *Acrolepiopsis assectella* (Zeller), is a newly introduced micro-lepidopteran pest in North America that attacks *Allium* crops, including onion, leek, and garlic. Eggs are laid on leaves and emerging larvae may cause extensive damage by mining leaves, feeding on leaf surfaces and feeding directly on bulbs. Little is known about existing natural enemies for this pest in North America, but classical biological control introductions are underway in Canada. However, other management options are needed because the threat to the onion production industry in New York State and the Great Lakes Region is imminent. Laboratory studies showed that lambda cyhalothrin (Warrior® II), spinetoram (Radiant® SC), methomyl (Lannate® LV), chlorantraniliprole (Coragen®), and spinosad (Entrust®) significantly increased larval mortality, compared to the control, at 2, 4, and 8 days after treatment, while *Bacillus thuringiensis* and azadirachtin insecticides did not. These results are explained in part by the behavior of the insect.

Key Words: *Acrolepiopsis assectella*; leek moth; onion leaf miner; teigne du poireau; onion; leek; allium

RESUMEN

La polilla del puerro, *Acrolepiopsis assectella* (Zeller) es un micro-lepidóptero plaga recientemente introducido en América del Norte, que ataca los cultivos de *Allium* incluyendo cebolla, puerro y ajo. Los huevos son puestos en las hojas y las larvas recién salidas pueden causar daño severo por las minas que hacen en las hojas, la alimentación sobre la superficie de las hojas y la alimentación directa a los bulbos. Se sabe poco sobre los enemigos naturales existentes para esta plaga en América del Norte, pero están realizando introducciones del control biológico clásico en Canadá. Sin embargo, otras opciones de manejo son necesarias debido a su amenaza inminente a la industria de producción de cebolla en Nueva York y la región de los Grandes Lagos. Ensayos de laboratorio mostraron que el cialotrina lambda (Warrior II), spinetoram (Radiante SC), metomilo (Lannate LV), clorantraniliprol (Coragen) y spinosad (Entrust) aumentó significativamente la mortalidad de larvas, en comparación con el control, a los 2, 4 y 8 días después del tratamiento, mientras que insecticidas *Bacillus thuringiensis* y azadiractina no lo hizo. Estos resultados se explican en parte por el comportamiento del insecto.

Palabras Clave: *Acrolepiopsis assectella*, la polilla de puerro, cebolla, minador de la hoja; teigne du poireau, cebolla, puerro, Allium

The Leek moth, *Acrolepiopsis assectella* Zeller (Lepidoptera: Acrolepiidae), also called the onion leaf miner or teigne du poireau (Garland 2002), is a micro-lepidopteran pest of *Allium* crops, including onion, leek, and garlic (Allison et al. 2007). Eggs are laid on leaves and emerging larvae can cause damage by mining leaves or feeding on leaf surfaces and bulbs (Allen et al. 2008; Garland 2002).

Acrolepiopsis assectella has been a pest of *Allium* crops in Western Europe, especially Belgium, France, and the Netherlands, for hundreds of years. The range of *A. assectella* extends across the Europe from Sweden to Russia and North Africa (Asman 2001; Garland 2002; Landry 2007;

Jenner et al. 2010b). In North America, *A. assectella* was first discovered in Ottawa, Canada in 1993 (Garland 2002; Allen et al. 2008; Mason et al. 2010). Range expansion has continued and *A. assectella* is presently established throughout southeastern Ontario and western Quebec (Landry 2007; Jenner et al. 2010b). In 2009, *A. assectella* was first found in the United States near Plattsburg, northern New York State along the St. Lawrence Seaway less than 230 km from Ottawa. In 2012, *A. assectella* was present in Jefferson, St. Lawrence, and Clinton Counties of northern New York (Ivy 2011), but found only in small-scale production areas and home gardens containing onions, leeks, and garlic. However,

large commercial onion production occurs in Oswego County, NY, located 300 km to the south. *A. assectella* represents a threat to this area and other onion production areas in New York that report annual harvest values of greater than \$54 million (USDA-ERS 2011). If *A. assectella* becomes established in New York, it may also spread to other states in the northeast where *Allium* vegetable production is important.

Acrolepiopsis assectella has few known natural enemies in North America and pest pressure is often more severe than in Europe (Jenner et al. 2010b; Jenner et al. 2010c). Biological control with a parasitoid has worked well in Europe, and introduction of *Diadromus pulchella* (Wesmael) is underway in Canada (Jenner et al. 2010a; Jenner et al. 2010c). These efforts are important but additional options are needed because of the immediate threat to onion production in New York State and the Great Lakes Region (Mason et al. 2011). Cultural control options have been evaluated for *A. assectella*. Intercropping has not proven effective, but trap cropping shows promise (Asman 2002; Asman & Ekbohm 2006).

Insecticides will play an important role in a control strategy for this pest of high value vegetable crops, and this has been demonstrated in the case of swede midge, *Contarinia nasturtii* Kieffer (Diptera: Cecidomyiidae) (Chen 2011), another newly introduced pest of vegetables from Canada. Cornell University petitioned the New York State Department of Environmental Conservation (NYSDEC), and was granted 2(ee) emergency exemptions, for 5 insecticides for control of *A. assectella* in 2010 based on the immediate threat of *A. assectella*. Requests for Entrust® (spinosad) and DiPel® DF (*Bacillus thuringiensis* var. *kurstaki*) were approved for homeowners and organic growers in affected regions. Approvals for Warrior® II (lambda-cyhalothrin), Lannate® LV (methomyl), and Radiant® SC (spinetoram) were granted for large-scale conventional growers. Continued registration of these products is important to prevent crop damage, and proven efficacy against *A. assectella* is key to this process.

Other insecticides might also be effective but a literature review indicated a lack of research on efficacy against *A. assectella* for many chemistries, including those with the 2(ee) exempted products mentioned above. This laboratory study was conducted to provide preliminary efficacy data for a variety of insecticides in different chemical classes that could be suitable against *A. assectella*.

MATERIALS AND METHODS

Mature leek plants, *Allium ampeloprasum* var. 'Lincoln' (Bejo Seeds Inc., Geneva, New York), were grown in a greenhouse at a constant 15 °C and 16:8 h L:D. Plants were grown in 15 cm diam,

15 cm deep plastic pots from seeds. Slow release fertilizer (Scotts Osmocote, 15-9-12) was applied twice at 3-mo intervals initiated at planting.

Acrolepiopsis assectella were reared in a laboratory colony that originated from field-collected pupae and late instar larvae found in a home garden in Essex County, New York in August 2011. Pupae of this population were placed in clear acrylic cylinders (11 cm diam × 15 cm high) fitted with screened caps (i.e. oviposition chambers) and a flask of 10% sucrose solution for adult nutrition. When adults emerged, a 10 cm × 3 cm piece of fresh leek leaf, rubbed with masticated leek pulp (Garland 2002), was hung from the oviposition chamber lid for a 24-h period during which adults were given the opportunity to lay eggs on the leaf. Egg-laden leaves were collected daily. To facilitate handling and collection of larvae, freshly collected leaves were cut into 0.6 cm squares and allowed to dry to prevent leaf mining by emerging neonates.

Dried egg-laden leaves were checked daily for neonate larvae. *Acrolepiopsis assectella* larvae were collected using a size 0 round tip paintbrush dipped in water to prevent static charge. Individual larvae were transferred to single flat-bottom 6mm diam wells of a 96-well plate (Falcon 3912 MicroTest III Flexible Assay Plate, Becton Dickinson Labware, Oxnard, California) containing a small amount of leek pulp. Leek pulp was prepared by adding 100 mL of distilled water to 25 g of fresh, washed leek leaves in an Osterizer Model 471 blender set to high for 2 min, and then drained of liquid with a small strainer. A sheet of Parafilm M was firmly pressed over the entire plate to form a seal over all individual wells containing insects to retain moisture and humidity, but allow gas exchange. Plates were held at 20 °C and 16:8 h L:D. Larvae were transferred to clean trays with fresh leek pulp every 2 to 3 d. Insect stage was calculated using head capsule size. When larvae were 2nd or 3rd instars, they were individually placed in designated 96-well plates at 10 °C and 16:8 h L:D.

Insecticide treatments were applied at a standard volume of 285.2 L per ha and 2.8 kg per cm² of pressure through a single Teejet flat fan 8002 nozzle tip with an Allen Track Sprayer (Allen Machine Works, Midland Michigan). A single potted leek plant, as described above, was placed into the spray chamber and treated. Maximum label rates, indicated on New York State registration labels, were applied with a surfactant at 1% v/v (Dyne-Amic®, Helena Chemical Co.) (Table 1). A single leaf of sufficient size was marked on each leek with a small pin and positioned during application to receive an even dose of insecticide. Treatments were replicated 3 times for use in bioassays completed at 2, 4, and 8 d after treatment (DAT).

Bioassays were completed using 7, 8, or 9 discs (3.2 cm diam), cut from a treated leaf, and placed

TABLE 1. MORTALITIES OF *ACROLEPIOPSIS ASSECTELLA* LEEK MOTH LARVAE IN INSECTICIDE BIOASSAYS.

Insecticide	Label rate/h ^a	Rate g (AI)/ha	2 DAT	4 DAT	8 DAT
			% mortality (±SE) ^{bc}	% mortality (±SE) ^{bc}	% mortality (±SE) ^{bc}
Lambda-cyhalothrin (Warrior II)	0.140 L	35.0	96.3 ± 3.7 aA	81.0 ± 9.9 aB	66.7 ± 5.4 bC
spinetoram (Radiant SC)	0.730 L	182.1	92.6 ± 4.9 aA	95.2 ± 4.8 aA	91.7 ± 5.4 aA
methomyl (Lannate LV)	3.505 L	874.3	81.5 ± 8.1 abA	85.7 ± 6.7 aA	83.4 ± 8.9 abA
chlorantraniliprole (Coragen)	0.365 L	127.5	77.8 ± 7.8 abA	90.5 ± 6.1 aA	91.7 ± 5.4 aA
spinosad (Entrust)	0.140 Kg	168.1	66.7 ± 7.8 bA	71.4 ± 15.3 aA	70.9 ± 9.8 abA
Bt aizawai (Agree WG)	2.243 Kg	1120.8	14.8 ± 8.1 cA	9.5 ± 6.2 bA	16.7 ± 6.3 cA
azadirachtin (Neemix 4.5)	0.511 L	127.5	11.1 ± 5.6 cA	14.3 ± 6.7 bA	12.5 ± 8.8 cA
Bt kurstaki (DiPel DF)	1.121 Kg	605.3	11.1 ± 5.6 cA	14.3 ± 9.9 bA	12.5 ± 6.1 cA
untreated check	—	—	14.8 ± 8.1 cA	19.1 ± 9.9 bA	16.7 ± 12.6 cA

^aLabel rates English equivalent: Warrior II, 1.96 fl oz/A; Radiant SC, 10.0 fl oz/A; Lannate LV, 3.0 pints/A; Coragen, 5.0 fl oz/A; Entrust, 2.0 oz/A; Agree WG, 2.0 lb/A; Neemix 4.5, 7.0 fl oz/A; DiPel DF, 1.0 lb/A.

^bMeans within a column followed by the same lower case letters are not significantly different ($\alpha = 0.05$, Fisher LSD).

^cMean within a row followed by the same upper case letter are not significantly different ($\alpha = 0.05$, Fisher LSD).

in individual Comet™ 1 oz. portion/shot glasses (WNA, Covington, Kentucky, www.wna.biz, SKU P10). Three 2nd or 3rd instars were then placed on the surface of a leaf disc, and a clear plastic lid was placed on the cup. To determine the activity of each insecticide over time, mortality rates at 2, 4, and 8 DAT were evaluated at 48 h after exposure using different leaf discs and insects for each DAT treatment.

Mortality was analyzed using one-way ANOVA and means were separated using Fisher LSD tests. Data were arcsine square root transformed before analysis but untransformed means are presented. Analyses were carried out in JMP 9.0 for Macintosh (SAS Institute, Cary, South Carolina).

RESULTS

There were significant differences in mortality between insecticides at each DAT and within an insecticide treatment over time (Table 1). Warrior® II, Radiant® SC, Lannate® LV, Coragen®, and Entrust® caused significant larval mortality, compared to the untreated check, at 2, 4, and 8 DAT. At 2 DAT, Warrior® II caused the highest level of mortality at $96.3 \pm 3.7\%$. Radiant® SC ($92.6 \pm 4.9\%$), Lannate® LV ($81.5 \pm 8.1\%$), and Coragen® ($77.8 \pm 7.8\%$) performed well and were not statistically different from Warrior® II. While Entrust® ($66.7 \pm 7.8\%$) performed significantly better than the untreated check, it was significantly worse than the top 4 treatments. In contrast, several biologically-based insecticides did not provide significantly elevated mortality compared to the untreated check ($14.8 \pm 8.1\%$) at 2 DAT: Agree® WG ($14.8 \pm 8.1\%$), Neemix® 4.5 ($11.1 \pm 5.6\%$) and DiPel® DF ($11.1 \pm 5.6\%$).

At 4 DAT, Radiant® SC ($95.2 \pm 4.8\%$) caused the highest mortality levels, followed by Cora-

gen® ($90.5 \pm 6.1\%$), Lannate® LV ($85.7 \pm 6.7\%$), Warrior® II ($81.0 \pm 9.9\%$), and Entrust® ($71.4 \pm 15.3\%$). No significant differences among these treatments were detected, but all showed significantly higher levels of mortality compared to the untreated check at 4 DAT. Compared to the untreated check ($19.1 \pm 9.9\%$) at 4 DAT, there were no significant differences for the biologically-based insecticides Agree® WG ($9.5 \pm 6.2\%$), Neemix® 4.5 ($14.3 \pm 6.7\%$), and DiPel® DF ($14.3 \pm 9.9\%$).

At 8 DAT, Radiant® SC ($91.7 \pm 5.4\%$) and Coragen® ($91.7 \pm 5.4\%$) had the highest levels of mortality among all treatments, followed by Lannate® LV ($83.4 \pm 8.9\%$) and Entrust® ($70.9 \pm 9.8\%$), but there were no significant differences among these treatments. Warrior® II ($66.7 \pm 5.4\%$) showed significantly higher mortality compared with the untreated check at 8 DAT, but the level of control was significantly lower than the other four insecticides. The biologically-based insecticides Agree® WG ($16.7 \pm 6.3\%$), Neemix® 4.5 ($12.5 \pm 8.8\%$), and DiPel® DF ($12.5 \pm 6.1\%$) were not significantly different from the untreated check ($16.7 \pm 12.6\%$).

ANOVA and Fishers LSD analysis of mortality at 2 DAT, 4 DAT, and 8 DAT within treatments did not reveal significant effects of treatment timing, with the exception of Warrior® II, where at 2 DAT, mortality was significantly higher than at 4 DAT and 8 DAT. Warrior® II mortality at 4 DAT was significantly higher than at 8 DAT and significantly lower than 2 DAT.

DISCUSSION

Acrolepiopsis assectella's behavior was an important factor in this experiment and likely influenced whether an insecticide was effective, as Mason et al. (2010) suggested. *Acrolepiopsis*

assectella mortality in the pyrethroid-sprayed plants was initially high but decreased as time progressed. Pyrethroids degrade quickly and the active ingredient in Warrior®, lambda cyhalothrin, has a half-life of 5 d on plant surfaces, breaking down readily in sunlight (National Pesticide Information Center 2001). Our results showed that insects exposed to the plant surface shortly after treatment were controlled before entering the plant. As time between application and insect exposure increased, the treatment became significantly less effective and declined by 29.6% (Table 1).

Spinetoram (Radiant® SC) and spinosad (Entrust®) both belong to the spinosyn class of insecticides and had longer residual activities. Spinosad has a unique mode of action, acting quickly on the insect nervous system through contact and ingestion (Thompson et al. 2000). This is suitable for *A. assectella* because of the short time period larvae are exposed to the plant surface. These positive results suggest more work should be done to evaluate spinosyn products.

Methomyl (Lannate® LV) and chlorantraniliprole (Coragen®) were also effective at controlling *A. assectella*. Unlike lambda-cyhalothrin (Warrior®), they were effective through 8 DAT. The exact exposure pathways of these chemicals are not known. Both insecticides have contact and ingestion activity against arthropods (DuPont 2007). Methomyl does not persist long on plant surfaces, having a half-life of 3 to 7 d (Extension Toxicology Network 1993).

The bacteria *Bacillus thuringiensis* (Bt) must be ingested to reach target sites in the digestive tract and have an effect on the target organism. Neither of the Bt treatments (Agree® WG and DiPel® DF) effectively controlled *A. assectella*. This contradicts the literature in which Bt was reported to control *A. assectella* before harvest (Garland 2002). Based on *A. assectella*'s leaf mining behavior, it is likely that our larvae did not ingest sufficient Bt proteins. The spray chamber used to treat leek plants provided very good coverage prior to mining and likely was much better than what would occur in the field. Bt is also broken down quickly by sunlight in the environment, so good control wouldn't be expected.

Results show that insecticides can be effective but will be influenced by chemical composition, mode of action and/or behavior of *A. assectella*. Lambda-cyhalothrin may be an effective option in situations where *A. assectella* have not hatched or penetrated the leaf tissue. It should be applied no more than 2 d intervals to compensate for degradation of the active ingredient, and should not be used as a one-time treatment if adults oviposit for a prolonged period. Spinetoram, chlorantraniliprole, and spinosad also provided effective consistent control for up to 8 d, so time between reapplication could be lengthened. Products containing

Bt (Agree® WG and DiPel® DF) and azadirachtin (Neemix® 4.5) did not provide high levels of mortality of *A. assectella* and are not likely to be suitable control agents.

More work is needed because some of our findings contradict current recommendations about the effectiveness of Bt products (e.g., Allen et al. 2008). Neonate larvae burrowed into the host within min to h of hatching and 2nd instars placed on fresh leaf material also burrowed into mesophyll quickly (personal observation). It is unlikely they ingested sufficient amounts of Bt proteins to be harmed, despite the use of good foliar spray technology. Of the other options we tested for organic producers, only spinosad (Entrust®) provided decent control. Conventional growers have more effective options. Based on these tests and our knowledge of spinetoram (Radiant® SC) and chlorantraniliprole (Coragen®) for other Lepidoptera, field tests should be conducted with these newer insecticide chemistries, as well as pyrethroid and carbamate insecticides.

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