Deltamethrin-Incorporated Nets as an Integrated Pest Management Tool for the Invasive *Halyomorpha halys* (Hemiptera: Pentatomidae)

T. P. Kuhar, 1,2 B. D. Short, 3 G. Krawczyk, 4 and T. C. Leskey 3

¹Department of Entomology, Virginia Tech, Blacksburg, VA 24061-0319 (tkuhar@vt.edu), ²Corresponding author, e-mail: tkuhar@vt.edu, ³USDA-ARS, Appalachian Fruit Research Station, Kearneysville, WV 25430-2771 (brent.short@ars.usda.gov; tracy.leskey@ars.usda.gov), and ⁴Fruit Research and Extension Center, Pennsylvania State University, Biglerville, PA 17307-0330 (gxk13@psu.edu)

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Abstract

Long-lasting insecticide nets (LLINs), which have insecticide incorporated within the fibers, have been widely used for control of malaria and other insect-vectored diseases. Only recently have researchers begun exploring their use for control of agricultural pests. In this study, we evaluated the toxicity of a deltamethrin-incorporated LLIN, ZeroFly (Vestergaard–Frandsen, Washington, DC) for control of the brown marmorated stink bug, *Halyomorpha halys* (Stål). In the lab, exposure to the ZeroFly net for 10 s resulted in >90% mortality of *H. halys* nymphs and >40% mortality of *H. halys* adults. Longer exposure to the net resulted in higher mortality. In another experiment, a 15-cm² sheet of ZeroFly net placed inside of the stink bug trap provided long-lasting kill of *H. halys* adults equal to or better than standard dichlorvos kill strip. Potential for the use of ZeroFly nets for *H. halys* IPM is discussed.

Key words: : Halyomorpha halys, long-lasting insecticide net, trap, attract and kill

Insecticide-treated nets have been widely used as a malaria vector control tool around the world since the mid-1990s (Hill et al. 2006). These nets are typically treated with a pyrethroid insecticide, such as permethrin or deltamethrin, which repel, incapacitate, and kill mosquitoes (Diptera: Culicidae) that land on the nets. As an advancement over nets that are dipped in insecticides, and which require retreating after a few washings or a year of use, long-lasting insecticidal nets (LLINs) have been developed with the netting material (usually polyethylene, or polyester) that has insecticide incorporated within or bound around the fibers (Martin et al. 2007, Reimer et al. 2016). With LLINs, insecticide from within the fibers continues to move to the surface over time, replacing the residue that may have been removed by washing; this enables long (multiyear) residual efficacy of the nets (Martin et al. 2007).

Researchers have recently begun exploring the use of LLINs for management of agricultural pests. In particular, pyrethroid-incorporated nets applied as floating row covers, or 2-m high fences, have been shown to be efficacious for control of various lepidopteran and aphid pests of vegetables (Martin et al. 2006, Licciardi et al. 2008, Dáder et al. 2015), and are considered to be cost-effective because they control multiple key pests, last multiple years, and because larger mesh sizes can be used compared with untreated row covers (Dáder et al. 2015). To our knowledge, the efficacy of LLINs for control of pestiferous stink bugs (Hemiptera: Pentatomidae) has not been investigated.

Herein, we report on the toxicity of a commercially available deltamethrin-incorporated LLIN, ZeroFly screen (Vestergaard–Frandsen, Washington, DC), on an important invasive pest in the United States and Europe, the brown marmorated stink bug, *Halyomorpha halys* (Stål) (Hemiptera: Pentatomidae) (Leskey et al. 2012b, Rice et al. 2014), and explore strategies for the use of the ZeroFly nets in stink bug IPM.

Materials and Methods

Netting

All experiments were conducted with black ZeroFly netting obtained from Vestergaard–Frandsen (Lausanne, Switzerland) in spring 2015. The netting was supplied as a 15- by 1.8-m roll, mesh size of 32–33 holes per cm², and was incorporated with deltamethin at ~3.85 mg a.i./g fiber. For untreated controls, we used a roll of 7.6- by 1.2-m charcoal fiberglass window screen FCS8480-M (Saint-Gobain ADFORS Clear Advantage); the untreated screen had the same mesh size and color as the ZeroFly screen.

Insects

Adult *H. halys* were collected from buildings (fall), trees, and agricultural plants during the summer. Nymphs for bioassays were obtained from a colony at Virginia Tech, Blacksburg, VA, maintained

in a growth chamber at 60–80% RH, $27 \pm 2^{\circ}$ C, with a photoperiod of 16:8 (L:D) h, and provisioned with green beans, *Phaseolus vulga-ris* L., raw shelled peanuts, *Arachis hypogaea* L., and a water wick.

Residual Efficacy Bioassay

In summer 2015 and 2016 in Blacksburg, VA, bioassays were conducted on H. halys nymphs (third and fourth instars) and in fall 2016, similar bioassays were conducted on adults. For each bioassay, ~10 insects were placed simultaneously in a 1-liter plastic container lined with the netting, so that the insects could forage freely, but remained in contact with the netting. Various intervals of exposure were evaluated including 10 s, 1 min, and 10 min on the ZeroFly netting and 1 min on the untreated screen as a control. Following exposure, the insects were immediately transferred to untreated containers provisioned with a green bean pod and a moistened water wick. Mortality of the bugs was assessed after 24 h. The bioassay was replicated four times each on different dates and with different bugs. Following arc-sine square root transformation, mortality data were analyzed using one-way ANOVA and Fisher's Protected LSD to separate means at the P < 0.05. All statistical analyses were carried out using JMP Pro 11.0 (SAS 2013). Probit analysis (LeOra Software 2002) was used to calculate LT₉₀ values (lethal exposure time) and 95% fiducial limits (FL) for both nymphs and adults.

Comparison of Killing Agents in Pheromone Trap Jar Tops

Experiments were conducted at the Appalachian Fruit Research Station in Kearneysville, WV, in 2016 using Dead-Inn Stink Bug traps (40.6 cm height, AgBio, Inc., Westminster, CO). Each trap top was composed of a plastic jar that was 10.2 by 10.2 by 15.2 cm (W by L by H), with a funnel that had a 7.6-cm opening. Inside each jar top, one of the four treatments was evaluated for efficacy and speed of kill of adult *H. halys*: 1) ZeroFly netting, 2) lambdacyhalothrin-treated netting, 3) dichlorvos kill strip, current standard for insect traps (Leskey et al. 2012a), and 4) no killing agent

(control). The ZeroFly net was cut into 15-cm² sheets and wrapped tightly around the funnel cone on the inside of the jar and fastened with a paper clip. The lambda-cyhalothrin treated netting was produced by soaking 15-cm² pieces of polyester no-see-um mesh in a 2.3% a.i. solution of Warrior II with Zeon Technology (Syngenta Crop Protection, Greensboro, NC) for 1h. The netting was fully dried before being attached to the funnel as the ZeroFly netting. For the dichlorvos kill strip, a half piece of Hercon Vaportape II (Hercon Environmental, Emigsville, PA) was suspended inside the jar by a paper clip. Adults H. halys were released individually into one of the aforementioned treatments. Four response variables were assessed: 1) escape from jar, 2) time until escape, 3) mortality in jar, and 4) time until death. Evaluations of H. halys presence and condition were taken at 10-15-min intervals for a period of 3 h. Treatment effects on the binomial variables (escape and mortality) were analyzed using logistic regression and then chi-square pairwise comparisons to distinguish among treatments. Treatment effects on the continuous variables (time until escape and death) were analyzed using a t-test and ANOVA with Tukey's HSD test, respectively (JMP 11.0, SAS 2013).

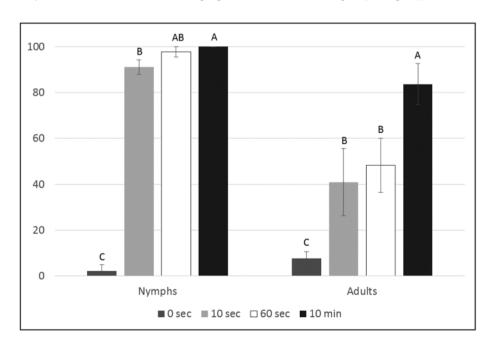
Results and Discussion

Residual Efficacy Bioassay

The ZeroFly net was highly toxic to *H. halys* nymphs and adults (Fig. 1). There was an effect of exposure time on the net on mortality of *H. halys* nymphs (F=534.4; df=3; P<0.001), with an LT₉₀=8.27 s (95% FL=0.65–121.11; slope=0.783). There was also an effect of exposure time on mortality of *H. halys* adults (F=8.46; df=3; P<0.003). Adult mortality averaged 40.8, 48.3, and 83.6% for 10-s, 1-min, and 10-min exposures, respectively, with an LT₉₀=39.8 min (95% FL=9.30–141.66; slope=0.692).

Comparison of Killing Agents in Pheromone Jar Tops

There was an effect of treatment on the percentage of *H. halys* adults that escaped jar tops ($\chi^2 = 17.75$; df = 3; P = 0.0005).



 $\textbf{Fig. 1.} \ Percentage \ mortality \ (mean \pm SE) \ of \ \textit{H. halys} \ nymphs \ and \ adults \ 24 \ hafter \ brief \ exposure \ periods \ to \ ZeroFly \ net \ in \ the \ lab.$

Table 1. Percentage and time until escape or death (mean ± SEM) of H. halys adults placed in commercial Dead-Inn stink bug trap jars

| Treatment | Mean ± SEM | | | |
|--------------------------------|------------------------|--------------------------------------|---------------------|------------------------------------|
| | % Escaped ^a | Time until escape (min) ^b | % Dead ^c | Time until dead (min) ^d |
| Dichlorvos kill strip | 16.67 ± 7.78a | 42.75 ± 10.13a | 83.33 ± 7.78b | 54.75 ± 4.65b |
| Lambda-cyhalothrin-treated net | $0.00 \pm 0.00b$ | N/A | $100.00 \pm 0.00a$ | $61.17 \pm 4.27b$ |
| ZeroFly net | $0.00 \pm 0.00b$ | N/A | 88.00 ± 6.63 ab | $76.59 \pm 4.83a$ |
| Control | $29.17 \pm 9.48a$ | 44.14 ± 12.60a | $4.17 \pm 4.17c$ | 45.00 ± 0.00 |

[&]quot;Hercon Vaportape II (N = 24); Lambda-cyhalothrin Netting (N = 23); ZeroFly Netting (N = 25); Control (N = 24).

Halyomorpha halys adults escaped only from untreated control jar tops and those containing dichlorvos, but there was no difference in the percentage of ($\chi^2 = 1.07$; df = 1; P = 0.3005) or time until escape (t=0.09; df=8.86; P=0.9332) between these two treatments. There was an effect of treatment on the percentage of dead H. halys adults ($\chi^2 = 70.96$; df=3; P < 0.0001) and the time until death (F=5.90; df=2, 62; P=0.0045). All jars provisioned with killing agents resulted in significant mortality of adults compared with the untreated control, with the highest rates, 100% and 88%, recorded from the lambda-cyhalothrin-treated and ZeroFly netting, respectively (Table 1).

Our experiments showed ZeroFly nets are quite toxic to *H. halys*, delivering a lethal dose of deltamethrin to bugs within several seconds or minutes of exposure depending on stage of the bug. We also showed a small square of the net can be used effectively as a killing agent in traps replacing dichlorvos. Additional research on other uses of the ZeroFly net for *H. halys* IPM is ongoing. In particular, the use of nets in combination with aggregation pheromone lures (Weber et al. 2014) has shown promise as an attract-and-kill device (TPK and GK, unpublished data). ZeroFly nets and other LLINs show tremendous potential for use in pest management of *H. halys* and many other insects as well.

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^bHercon Vaportape II (N=4); Control (N=7).

^{&#}x27;Hercon Vaportape II (N = 20); Lambda-cyhalothrin Netting (N = 23); ZeroFly Netting (N = 25); Control (N = 24).

^dHercon Vaportape II (N = 20); Lambda-cyhalothrin Netting (N = 23); ZeroFly Netting (N = 22). Control excluded from analysis due to a rep of 1.