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EFFICACIES OF FOUR PHEROMONE-BAITED TRAPS IN CAPTURING MALE *HELICOVERPA* (LEPIDOPTERA: NOCTUIDAE) MOTHS IN NORTHERN FLORIDA

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ABSTRACT

Helicoverpa armigera (Hübner) (Lepidoptera: Noctuidae) is a serious pest of grain, row, and vegetable crops throughout much of the world, although it is currently not established in the United States. USDA-APHIS and the Cooperative Agricultural Pest Survey program are charged with the responsibility to monitor for this insect pest. The adult stage is the easiest to monitor using pheromone-baited traps. Traps must be easy to handle, portable and cost effective so that they can provide high quality specimens for identifiers. This study was conducted from spring through the fall in 2010 and 2011 to compare the trapping efficacy and cost-effectiveness of 4 pheromone-baited traps for male *Helicoverpa* moths. Over 11,600 *Helicoverpa* moths were captured, all identified as the corn earworm, *H. zea* (Boddie). The Pherocon® 1C “sticky” trap generally captured the fewest number of males, while equal numbers of moths were captured in a wire cone, Scentry™ Heliothis, and Universal (Unitrap) Moth “bucket” trap when moderate populations were present. Wire cone traps performed statistically better when high populations were present. The sticky traps captured the highest number of non-target insects, most being ants, flies, and beetles. Overall, the average corn earworm per trap vs. cost ratio for bucket traps was higher than the other traps, suggesting that more moths per dollar would be captured using these traps.

Key Words: pheromone trapping, corn earworm, Old World bollworm; *Helicoverpa*

RESUMEN

Helicoverpa armigera (Hübner) (Lepidoptera: Noctuidae) es una plaga importante de granos, cultivos en fila y hortalizas en una gran parte del mundo, aunque hasta ahora no se ha establecido en los Estados Unidos. El USDA-APHIS y el programa de Sondeo de Plagas Agrícolas Cooperativo tienen el cargo de responsabilidad de vigilar este insecto plaga. Las trampas usadas deben ser fáciles de manejar, portátil y rentable para que puedan proporcionar muestras de alta calidad para los identificadores. Se realizó estos estudios desde la primavera hasta el otoño del 2010 y 2011 para comparar la eficacia de la captura y la efectividad de costo de las cuatro trampas cebadas con feromonas para polillas *Helicoverpa* masculinas. Más de 11,600 polillas *Helicoverpa* fueron capturadas, todas identificadas como el gusano elotero del maíz, *H. zea* (Boddie). La trampa “pegajosa” 1C de Pherocon® generalmente capturó el menor número de machos, mientras que el mismo número de polillas fueron capturadas en un cono de alambre, Scentry™ Heliothis, y la trampa de “cubeta” Universal (Unitrap) para polillas. Las trampas pegajosas capturan el mayor número de insectos no objetivo, de estos la mayoría fueron hormigas, moscas y escarabajos. Todas las trampas fueron iguales en su capacidad para capturar por lo menos una polilla cuando las poblaciones fueron bajas. En general, el número promedio de gusanos del elote de maíz por trampa comparado con la tasa del costo para las trampas cubetas fue mayor que las otras trampas, lo que sugiere que más polillas por dólar serían capturados usando estas trampas.

Palabras Clave: captura por feromonas, gusano del maíz, gusano de la cápsula del Viejo Mundo; *Helicoverpa*

The genus *Helicoverpa* (Lepidoptera: Noctuidae) contains several species worldwide that impact a variety of row, vegetable, fiber, and ornamental crops. The Old World bollworm, *H. armigera* (Hübner), has the largest geographic distribution, attacking crops in Africa, the Middle East, southern Europe, central, southern, and southeastern Asia, Australia, and New Zealand (Common 1953; Zalucki et al. 1986; Topper 1987; Fitt 1989). The oriental tobacco budworm, *H. assulta* (Guenée), is found in Africa, the Far East, and Australia where it attacks onion and solanaceous crops such as peppers and tobacco (Hill 1983; Cho & Boo 1988; Cork et al. 1992). An Australian species, *H. punctigera* (Wallengren), infests a similar range of crops as *H. armigera* (Common 1953; Zalucki et al. 1986; Fitt 1989). Native to the Americas, *H. zea* (Boddie), attacks many economically important crops such as corn (as corn earworm, Archer & Bynum 1994; Wiseman & Widstrom 1992), cotton (as cotton bollworm, Ellsworth & Bradley 1992), tomatoes (as tomato fruitworm, Walgenbach et al. 1991), soybeans (Eckel et al. 1992), and tobacco (Neunzig 1969). This species has many wild host plants that can sustain its population between crop plant cycles (Neunzig 1963).

Helicoverpa armigera attacks many of the same crop plants as *H. zea*, having a diverse host plant range of over 180 cultivated and wild plant species (Fitt 1989). Larval feeding injury and plant damage is similar to that of *H. zea*, and worldwide annual control costs and production losses of over \$5 billion have been estimated (Lammers et al. 2007). This species also appears to have many of the same migration behaviors of *H. zea* (Westbrook et al. 1995), moving long distances in Australia (Gregg et al. 1993), northern and northeastern China (Feng et al. 2009, 2010), and India (Riley et al. 1992). Because of its host plant range, feeding behavior, and ability to move long distances, *H. armigera* is considered a serious threat to American agriculture (Venette et al. 2003).

Interceptions of *H. armigera* within the United States have occurred through trade cargo access points such as airport and maritime ports of entry. Pogue (2004) reported that 20 interceptions were made by the U.S. Department of Agriculture –Animal and Plant Health Inspection Services –Plant Protection Quarantine (USDA-APHIS-PPQ) inspectors at multiple U.S. ports-of-entry in 2003. Due to the sheer volume of imported plant cargo entering the United States, only 2% of incoming containers carrying plant material are visually inspected (Magarey et al. 2009). In Florida, the Miami Inspection Station is the port-of-entry for nearly 85% of all non-indigenous plant material entering into the United States (Simberloff et al. 1997).

To proactively ensure that the Old World bollworm is neither present nor established within sweet corn plantings in Florida, the Florida Department of Agriculture and Consumer Services, Division of Plant Industry, Cooperative Agricultural Pest Survey program (CAPS), along with USDA-APHIS-PPQ initiated the Small Grains Survey. By using Universal (Unitraps) moth “bucket” traps baited with pheromone designed to capture male *Helicoverpa* moths, CAPS survey specialists monitored for the presence of *H. armigera* in Escambia, Calhoun, and Jackson counties, Florida (Whilby & Smith 2009). Another trap, the Pherocon® 1C “sticky” trap, has been used successfully for several noctuid species (Tingle & Mitchell 1975). Traditionally, wire cone traps or the vinyl mesh netting version (Scentry™ Heliothis trap) have been used to capture heliothine moths (Hartstack et al. 1979; Gauthier et al. 1991; Lopez et al. 1994). Each moth trap varies greatly in their size, cost, processing time, specimen quality, and trapping efficacy.

Although bucket traps have been successful in capturing moths related to the genus *Helicoverpa* (Kehat et al. 1991), no study has determined which moth trap may ultimately yield larger numbers of quality specimens coupled with a short processing time and low cost. The alternative is also important, the ability of traps to detect moths when population densities are low (trap sensitivity). This study was designed to determine the most efficacious, cost effective trap readily available for *Helicoverpa* spp. male moths. These results are needed by CAPS and APHIS survey specialists, extension personnel, company scientists, and others who need to monitor populations of these moths.

MATERIALS AND METHODS

Field Sites and Moth Trapping

Two field sites in northern Florida were used in the study. The first site was the University of Florida's Dairy Research Unit in Hague. The research dairy and surrounding farm comprise 344 ha and has continuous production of field corn for silage from March through October. Traps were placed April through July 2010 (14 dates) and then May through September 2011 (16 dates). The second site was a 400-ha commercial peanut farm in Williston; traps were placed August through October 2010 (9 dates). Four trap blocks were positioned along the roads and edges of crops. Each trap block included one of each pheromone-baited trap: a sticky trap (Pherocon® 1C, Trécé, Inc., Adair, Oklahoma; trap cost US\$4.12), a wire cone trap constructed in the 1990s by a local metal worker (US\$150); a Heliothis trap (Scentry Biologicals,



Fig. 1. Pherocon 1C (top left), wire cone (top right), Scentry *Heliothis* (bottom left), and Universal moth traps (bottom right).

Inc., Billings, Montana; US\$54.25), and a bucket trap (Great Lakes IPM, Vestaburg, Michigan; US\$11.55) (Fig. 1).

The Pherocon 1C trap top measured 28 cm long by 23 cm wide; the bottom sticky liner was 27 cm by 22.5 cm. Two plastic spacers provided a 4-cm space in between the top and liner. Moths captured in the sticky trap generally died overnight. The wire cone traps used in our study were smaller than the traditional 75-50 Hartstack (Hartstack et al. 1979) or 75-25 traps (Mitchell et al. 1985). Traps had a base cone that measured 50 cm tall with a bottom diameter of 50 cm that narrowed to an opening of 30 cm (50-30 trap). The apex cone was 30 cm tall with a 5-cm funnel opening that allowed insects into the apex cone. This top portion was attached by Bungee cords to the larger bottom cone. Lures were pinned to a cork that was attached to the bottom support rod. Therefore, moths attracted to the pheromone flew upwards through the base cone and into the apex cone, where they were captured. *Heliothis* cone traps were also composed of 2 cones. The base cone measured

80 cm long, with a bottom opening of 34 cm that narrowed to 15 cm at the top; the apex cone measured 27 cm long, with a bottom opening of 15 cm that narrowed to 6 cm at the top. The bottom portion of the apex cone was secured to the top portion of the base cone with Velcro material. The lure was placed along a cord stretching across the bottom of the base cone. Moths captured in the wire cone and *Heliothis* traps died over a couple of days. Standard bucket traps consisted of a white bucket (12.5 cm tall and 16 cm wide), a yellow funnel (bottom opening 3.2 cm) on top of the bucket, and a dark green cover (16 cm diam.) attached to the funnel by 4 posts that allowed a 3-cm circular space in between cover and funnel. The pheromone lure was placed in a green basket (5.3 cm long) that hung in the middle of the green cover. Therefore, moths attracted to the pheromone “fell” downwards (usually after flying inside the posts and bouncing off the top cover) through the yellow funnel into the bucket where they were killed by an insecticide strip (Hercon® Vapor-tape II, Hercon® Environmental, Emigsville,

Pennsylvania). Bucket traps also contained a small cellulose sponge (cat. #7920-00-633-9906; GSA Global Supply, Ft. Worth, Texas) to absorb water from rain or irrigation. The sticky, wire cone, and bucket traps were hung on metal poles so that the lure height was approximately 1.5 m above the ground. Because they had to be tied to the poles rather than placed on top, the *Heliothis* traps were placed on 2-m poles.

Corn earworm and Old World bollworm rubber septa lures with the same components (Suterra LLC, Bend, Oregon and Trécé, Inc., Adair, Oklahoma) were alternated and replaced every 2 weeks. Traps within each trap block were rotated on a monthly basis. Traps within blocks were at least 30 m apart; the separation among blocks was at least 100 m.

Moth Identification and Time to Process Specimens

Weekly moth samples were collected and processed. For the Williston 2010 samples, the total time in minutes for the whole process was compiled for the following components: extraction (removal from trap), target screening (sorting of moths that appeared to be *Helicoverpa*, chemical processing (see below), and proper identification (observing genitalic characters). All *Helicoverpa* species were identified by softening and clearing abdomens in warm 10% KOH for 45 min (Meagher et al. 2008). To remove excess sticky film, moths collected from the sticky traps were treated with Histo-clear II® (National Diagnostics, Atlanta, Georgia) for 5 min, air dried, soaked in alcohol, then placed in 10% KOH for 45 min (Miller et al. 1993). Under a dissecting microscope, genitalia were extracted from the abdomen with fine tweezers. The most important character used for distinction between *H. zea* and *H. armigera* is the number of small lobes, or diverticula, at the base of the vesica (3 lobes for *H. zea*, 1 for *H. armigera*) (Pogue 2004, Figs. 9 and 10). The

number of cornuti, size and shape of the valves, and the shape of the 8th abdominal sternite were also examined.

Trap Sensitivity

Three calculations were made to measure trap sensitivity. First, the number of times (dates × replications for the 3 sites; $n = 36, 56$, and 64 for Williston 2010, Hague 2010, and Hague 2011, respectively) that a particular trap was the only trap of the 4 that captured moths. The second calculation was the number of times that a particular trap was the only trap of the 4 without moths. The final calculation was the number of times that a trap contained zero moths.

Statistical Analysis

All analyses were conducted using SAS (SAS 9.2, SAS Institute 2008). All data were analyzed using Box-Cox (PROC TRANSREG) and PROC UNIVARIATE to find the optimal normalizing transformation, if needed (Osborne 2010). Analysis of variance (PROC MIXED, LSMEANS) was then used to separate means among trap types, with date, the date by trap interaction, and block as random variables. Trap sensitivity was analyzed by PROC GLM.

RESULTS

Over 11,600 *Helicoverpa* moths were collected in 2010 and 2011 and genitalia analysis indicated that no *H. armigera* was detected. Results from the field tests are shown in Table 1. Collections from Hague in 2010 showed that fewer *H. zea* moths were found in sticky traps than in the other traps. In Williston in 2010, there was no significant difference among trap captures, although there was a trend for bucket traps to capture the most moths and for sticky traps to capture the fewest moths. Over 8,600

TABLE 1. NUMBER (MEAN ± SE) OF *HELICOVERPA ZEA* MALES IN EACH TRAP TYPE AT HAGUE 2010 ($N = 56$), WILLISTON 2010 ($N = 36$), AND HAGUE 2011 ($N = 64$).

Trap Type	Hague 2010 ^a	Williston 2010 ^a	Hague 2011 ^b
Wire cone	14.1 ± 3.6 a	9.7 ± 2.8 a	80.5 ± 12.6 a
Heliothis	9.6 ± 2.2 a	7.9 ± 2.6 a	33.7 ± 5.4 b
Bucket	9.4 ± 1.8 a	12.8 ± 3.7 a	18.6 ± 2.4 b
Sticky	1.0 ± 0.2 b	2.4 ± 0.5 a	6.2 ± 0.9 c
	$F = 6.3$; $df = 3, 39$ $P = 0.0014$	$F = 2.0$; $df = 3, 24$ $P = 0.1481$	$F = 27.4$; $df = 3, 45$ $P < 0.0001$

Means in the same column followed by the same letter are not significantly different using LS MEANS ($P > 0.05$).
^alog (x + 0.1) transformed before analysis.
^b4th-root (x + 0.1) transformed before analysis; non-transformed means are presented.

H. zea males were captured in Hague in 2011, with the highest capture in wire cone traps, lowest capture in sticky traps, and intermediate capture in *Heliothis* and bucket traps.

Although the capability of a trap to capture large numbers of moths is important, another factor to consider in selecting a trap is its capability to capture moths when populations are very sparse. The first measure of trap sensitivity happened infrequently as there were few times that one trap was the only one of the group to capture moths and the difference in this variable among traps was not significant (sticky 2/156, bucket 3/156, wire cone 6/156, *Heliothis* 8/156; $P = 0.2452$). The number of times that a particular trap was the only trap of the 4 without moths occurred more frequently, but there was still no significant difference among the various types of traps (bucket 6/156, *Heliothis* 7/156, wire cone 10/156, sticky 17/156; $P = 0.4273$). The number of times that a trap contained zero moths occurred in over ¼ of the samples, but again there was no significant difference among the various types of traps (*Heliothis* 38/156, bucket 43/156, wire cone 45/156, sticky 61/156; $P = 0.4273$). Therefore, all traps were similar in their ability to capture at least one moth when populations were sparse.

Non-target moths captured by the various traps were counted and included the following species: *Leucania* spp., *Chloridea* (= *Heliothis*) *virescens* (F.), *Spodoptera dolichos* (F.), and *S. frugiperda* (J. E. Smith). Other non-target insects captured included ants, small flies, thrips, and beetles. In total, more than 35,500 non-target insects were collected (Table 2). Sticky traps contained the most non-target insects, with the smaller insects dominating. Bucket and wire cone traps contained the fewest non-target insects. All moths that looked like *Helicoverpa* spp. were prepared for genitalia analysis. The time needed to remove moths from traps and prepare genitalia for identification was calculated for samples collected from Williston in

2010. Our hypothesis was that specimens from sticky traps would require a significantly longer time to process because of glue removal and the large number of non-targets. This was proven correct as more than 44 min were required to process a moth from the sticky traps compared to slightly more than 12 min per moth in bucket traps (Table 3).

One of the objectives of this study was to compare the numbers of moths captured with the cost of traps in dollars to determine the moth per trap cost ratio. For each site, the bucket and sticky traps captured more moths per dollar than the *Heliothis* and wire cone traps (Table 4). In fact, in Hague in 2011 when high numbers of moths were captured, the bucket traps caught 1.6 moths per dollar compared to only 0.55 moths per dollar with the wire cone traps. Therefore, more than twice as many moths could be captured in bucket traps per dollar than in wire cone traps in a monitoring program.

DISCUSSION

Even though we didn't collect *H. armigera*, we expect that its response to lures and traps would be the same as that of *H. zea*. *Helicoverpa armigera* has been successfully trapped by wire cone, dry funnel (Kehat & Greenberg 1978), and delta (sticky) traps baited with sex pheromone in Australia (Baker et al. 2011; Fitt et al. 1989; Wilson & Morton 1989), Egypt (Salem et al. 2008), Greece (Mironidis et al. 2010), Hungary (Dömötör et al. 2007; Keszthelyi et al. 2011), and India (Basavaraj et al. 2013; Srivastava 2010). Sex pheromone traps, along with light traps, were successfully used to document new distributions of *H. armigera* in parts of Australia where populations were not thought to exist (Fitt et al. 1995).

Past studies showed that large wire cone traps captured more heliothines than Scentry

TABLE 2. NUMBER (MEAN ± SE) OF NONTARGET INSECTS IN EACH TRAP TYPE AT HAGUE 2010, WILLISTON 2010, AND HAGUE 2011.

Trap Type	Hague 2010 ^a	Williston 2010 ^b	Hague 2011 ^c
Sticky	124.2 ± 7.3 a	178.8 ± 42.7 a	289.4 ± 15.1 a
<i>Heliothis</i>	37.2 ± 3.1 b	33.6 ± 4.8 b	29.1 ± 2.4 b
Bucket	10.2 ± 1.7 c	15.7 ± 2.5 c	10.5 ± 1.4 c
Wire cone	8.7 ± 1.1 c	18.3 ± 3.1 c	5.7 ± 0.6 d
	$F = 97.5$; $df = 3, 24$ $P < 0.0001$	$F = 56.4$; $df = 3, 24$ $P < 0.0001$	$F = 219.5$; $df = 3, 45$ $P < 0.0001$

Means in the same column followed by the same letter are not significantly different using LSMEANS ($P > 0.05$).
^a $\sqrt[4]{x + 0.1}$ transformed before analysis.
^blog transformed before analysis. ^clog ($x + 0.1$) transformed before analysis; non-transformed means are presented.

TABLE 3. NUMBER OF MINUTES TO PROCESS ONE *HELI-COVERPA ZEA* MALE MOTH (MEAN \pm SE) FOR IDENTIFICATION, WILLISTON 2010. ONLY SAM- PLES THAT CONTAINED MOTHS WERE USED IN THE ANALYSIS. A LOG TRANSFORMATION WAS USED BEFORE ANALYSIS ($F = 8.9$; $DF = 3, 22$; $P = 0.0005$).

Trap Type	Minutes	n
Sticky	44.3 \pm 6.3 a	23
Heliothis	25.3 \pm 5.5 b	22
Wire cone	23.5 \pm 5.9 b	21
Bucket	12.2 \pm 3.4 b	24

Heliothis, bucket, or sticky traps (Gauthier et al. 1991; Lopez et al. 1994). In our study, wire cone traps outperformed other traps only in the trial where large populations of *H. zea* moths were present (Hague in 2011). The large surface area of the bottom cone and relatively large area of the collecting top helps to retain more moths, which is probably the reason that under high densities, more moths can be captured in the wire cone traps (Gauthier et al. 1991). In all trials the sticky traps captured the least numbers of moths and the greatest number of non-target insects.

Wire cone traps are expensive to construct, not commercially available, and are difficult to handle. Scentry™ Heliothis traps are cheaper and are easier to handle, but require more maintenance to keep them able to hold moths that are captured. We also had a problem with ants constructing nests in the traps and removing captured moths. Bucket traps are cheaper than Scentry™ Heliothis traps, but do require extra costs for the insecticide strips to kill moths (US\$1.18 – US\$1.60 per strip depending on number purchased) and some type of hanging mechanism. We used metal angle pieces that slid into the poles, but something as simple

as bamboo poles are also a convenient hanging mechanism (Unbehend et al. 2013). Spiders and frogs can also add to trap maintenance of bucket traps as they web the bottom funnel or consume captured moths in the bucket, respectively. Sticky traps are the cheapest and require little maintenance. However, the sticky area can quickly become covered with other in- sects and dust so that only a limited number of moths can be captured. Additional processing steps, which increase processing time to count and identify moths, must be considered when using sticky traps.

Finally, trap efficiency as suggested by the trap cost ratio showed that more moths per dollar could be captured by bucket traps, and this ratio becomes greater as the moth population increased. Therefore, our results suggest that for *Helicoverpa* monitoring programs, where the goal is to collect large numbers of high quality specimens for identification purposes, the efficiency and manageability of bucket traps is the optimal solution.

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TABLE 4. NUMBER (MEAN \pm SE) OF *HELICOVERPA ZEA* MALES PER DOLLAR TRAP COST IN EACH TRAP TYPE AT HAGUE 2010, WILLISTON 2010, AND HAGUE 2011. THE SAME LETTER IN EACH LOCATION INDICATES NO SIGNIFICANT DIFFERENCE.

Trap Type	Hague 2010 ^a	Williston 2010 ^a	Hague 2011 ^b
Bucket	0.81 \pm 0.16 a	1.11 \pm 0.32 a	1.61 \pm 0.30 a
Sticky	0.23 \pm 0.05 a	0.58 \pm 0.12 a	1.49 \pm 0.41 b
Heliothis	0.18 \pm 0.04 b	0.15 \pm 0.05 b	0.64 \pm 0.17 c
Wire cone	0.09 \pm 0.02 c	0.06 \pm 0.02 c	0.55 \pm 0.15 c
	$F = 16.4$; $df = 3, 39$ $P < 0.0001$	$F = 25.3$; $df = 3, 24$ $P < 0.0001$	$F = 20.8$; $df = 3, 45$ $P < 0.0001$

Means in the same column followed by the same letter are not significantly different using LSMEANS ($P > 0.05$).

^a log transformed before analysis.

^b 4th-root transformed before analysis; non-transformed means are presented.

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