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Source: Florida Entomologist, 89(2) : 245-250

Published By: Florida Entomological Society

URL: [https://doi.org/10.1653/0015-4040\(2006\)89\[245:MDAAR\]2.0.CO;2](https://doi.org/10.1653/0015-4040(2006)89[245:MDAAR]2.0.CO;2)

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MATING DISRUPTION AND ATTRACT-AND-KILL AS REDUCED-RISK STRATEGIES FOR CONTROL OF GRAPE ROOT BORER *VITACEA POLISTIFORMIS* (LEPIDOPTERA: SESIIDAE) IN FLORIDA VINEYARDS

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ABSTRACT

Mating disruption and attract-and-kill (A&K) gels were evaluated for control of grape root borer (GRB), *Vitacea polistiformis* Harris (Lepidoptera: Sesiidae), in Florida grape (*Vitis* sp.) vineyards. For mating disruption, pheromone twist-ties with leopard moth, *Zeuzera pyrina* L. (Lepidoptera: Cossidae), pheromone were placed in vines at a rate of 635 per ha. Attract-and-kill (A&K) gels containing the GRB pheromone and a pyrethrin (botanical insecticide) were applied to vine trunks at a rate of 112.5 g per hectare. These treatments were compared with chlorpyrifos (Lorsban), an organophosphate insecticide, and an untreated control in a randomized complete block design. Two wing-style sticky traps with GRB pheromone were placed in each treatment to monitor male moth activity and determine levels of trap shutdown. Complete trap shutdown occurred in the twist-tie sections for both 2003 and 2004 suggesting disruption of mating. Traps placed in the A&K and the pheromone twist-tie sections caught significantly fewer GRB than the Lorsban treatments in 2003. In 2004, significantly fewer moths were caught in the pheromone twist-tie and Lorsban treatments than the A&K and untreated controls. The differences between 2003 and 2004 for A&K treatments were due to the use of the incorrect pheromone blend in the 2004 batch of A&K. Whereas the differences in Lorsban treatments between both years may be related to residual activity of the pesticide. The economics of adopting A&K and mating disruption with twist-ties containing leopard moth pheromone is discussed. Both A&K and mating disruption with the leopard moth pheromone show promise as reduced-risk control tactics to be used in a GRB integrated pest management program and warrant further study.

Key Words: grape root borer, *Vitacea polistiformis*, mating disruption, attracticides, off-blend pheromones, IPM

RESUMEN

Sistemas de confusión de atracción sexual y geles para atraer y eliminar, fueron evaluados para el control de *Vitacea polistiformis* Harris (Lepidoptera: Sesiidae) en viñas de Florida. En el sistema de confusión de la atracción sexual, se utilizaron "twist-ties" con feromonas de *Zeuzera pyrina* L. (Lepidoptera: Cossidae). Seiscientos treinta y cinco unidades de este atrayente fueron usados por hectárea. El gel para atraer y eliminar usando una mezcla de feromonas de *V. polistiformis* y pyrethrin (insecticida botánico) se aplicó en una dosis de 112.5 g/ha. Estos tratamientos fueron comparados con chlorpyrifos (Lorsban), un insecticida organofosforado, y con un control sin ningún tratamiento químico, usando un diseño de bloques completos al azar. Dos trampas adhesivas con feromona de *V. polistiformis* se colocaron en cada tratamiento para monitorear la actividad de los machos. Los tratamientos con "twist-ties" consiguieron confundir los machos al punto que no se capturó ninguno en las trampas en 2003 ó 2004 sugiriendo que no hubo cópula. Las trampas colocadas en el tratamiento con "twist-ties" y con gel para atraer y eliminar, capturaron significativamente menos machos que las trampas colocadas en los tratamientos con Lorsban en 2003. En 2004, significativamente menos machos fueron capturados en los tratamientos con Lorsban y con "twist-ties" que los tratamientos que usaron el gel o que en el control sin tratamiento. Las diferencias entre 2003 y 2004 para el tratamiento de atraer y eliminar se debió a la el uso de una mezcla incorrecta de feromonas en 2004. La diferencia de las capturas en los tratamientos con Lorsban puede estar relacionada con la actividad en el residuo del pesticida. Las ventajas y desventajas económicas de el uso de estas dos tecnologías son discutido en el artículo Ambos tratamientos, el gel y los "twist-ties" con feromonas de *Z. pyrina*, son promisorios como candidatos para incluir en un programa de manejo integrado de *V. polistiformis* y merece más estudios en el futuro.

Translation provided by the authors.

The grape root borer (GRB), *Vitacea polistiformis* (Harris) (Lepidoptera: Sesiidae), is the key pest of grapes (*Vitis* spp.) in Florida (Liburd et al.

2004). Larvae feed on the roots, resulting in reduced yields and eventually, death of the vine. In severe cases, entire vineyards are lost (Dutcher &

All 1979). Traditionally, chlorpyrifos (Lorsban 4E) has been used as a soil drench to control primarily first instars as they move to the soil surface and enter the soil seeking roots, and newly emerged adults. Lorsban, an organophosphate, is suspected of being carcinogenic (Food Quality Protection Act 1996) and its future use is not certain.

The potential for modifying an insect pest's behavior through the use of pheromones in order to control its impact on a crop has been investigated widely in the last 30 years. Mating disruption first proved to be successful in controlling cabbage looper moths, *Trichoplusia ni* (Hubner) (Shorey et al. 1967), and since then has been used successfully on a number of insect pests (Cardé & Minks 1995). Among the sesiids, some success has been obtained with the currant clearwing moth *Synanthedon tipuliformis* (Clerck) (Cardé & Minks 1995), the peachtree borer *Synanthedon exitosa* (Say), and the lesser peachtree borer *S. pictipes* (Grote and Robinson) (Yonce 1981).

Pheromone twist-ties with GRB pheromone, 99% (*E,Z*)-2,13-octadecadien-1-ol: 1% (*Z,Z*)-3-13-octadecadien-1-ol [99% (*E,Z*)-2,13-ODDA: 1% (*Z,Z*)-3,13-ODDA] (Snow et al. 1987) have shown great potential as a viable control tactic for GRB management. Pearson & Meyer (1996) used 254 dispensers per ha in vineyards and examined females in the treatment plots. They found a significant reduction in the number of mated GRB females compared with females taken from the untreated controls. Also, Webb (1991) recorded a significant reduction in trap-catches, mated females, and pupal case counts in vineyards treated with GRB pheromones compared with the untreated vineyard, potentially indicating a high degree of mating disruption.

Some studies suggest that "off-blends" (incomplete pheromones) may work better at mating disruption than the synthetic pure pheromone blend that is most similar to the natural pheromone (Minks & Cardé 1988). The mechanisms by which off-blend pheromones function to inhibit mating are not well understood. However, one theory is that the off-blend camouflages the true female pheromone, rendering it indistinguishable from the background (Minks & Cardé 1988). Another theory is that by creating a sensory imbalance, the male becomes attuned to the more predominant off-blend, and the ratio in the true blend is interpreted as unnatural (Bartell 1982).

Attract-and-kill (A&K) is a promising new technology that involves an attractant such as a pheromone and a toxicant. Unlike mating disruption, which functions by "confusing" the insect, attract-and-kill technology attracts the insect to a pesticide laden gel matrix, which, upon contact, kills the insect. Attract-and-kill has been successfully used on several lepidopteran species including codling moth (Ebbinghaus et al. 2001), and Oriental fruit moth (Evenden & McLaughlin

2004). Recently, IPM Tech (Portland, OR) developed an attracticide for grape root borer, called Last Call™ GRB, which has not previously been tested under field conditions.

The overall goal was to evaluate the use of an off-blend pheromone for mating disruption as well as an attract-and-kill technique for the control of grape root borer in Florida vineyards.

MATERIALS AND METHODS

Four muscadine grape (*Vitis* sp.) vineyards with similar management practices were chosen for this experiment. All vineyards were pruned annually in early winter, had similar fertilization schedules, were treated with glyphosate 3-4 times a year in a 1.2-m band under the trellis, and were mowed between rows every 2-3 weeks. No other vineyards occurred within 16 kilometers, but areas of naturally occurring wild grapes were nearby.

Each vineyard consisted of four treatments and was divided into four, 0.4-ha plots. Treatments included: 1) Mating disruption with pheromone twist-ties (Shin-Etsu Chemical Co. Ltd. Tokyo, Japan), 2) Attract-and-kill with Last Call™ GRB (IPM Tech Portland, OR), 3) Chemical control with chlorpyrifos (Lorsban 4E) (Dow Agro-Sciences LLC, Indianapolis IN), and an untreated control. A 15-m buffer zone was left between treatments. Experimental design was a randomized complete block (blocked by vineyard) with four treatments and four replicates. Two wing-style sticky traps baited with the GRB pheromone [99% (*E,Z*)-2,13-ODDA: 1% (*Z,Z*)-3,13 ODDA] (Great Lakes IPM, Vestaburg, MI) were hung on the trellis wire at 1.0 to 1.5 m above the ground in each treatment at least 20 m apart to monitor populations of male GRB moths. This study was initiated in the 2003 grape-growing season and repeated in 2004.

Pheromone Twist-Ties

Pheromone twist-ties emitting the leopard moth, *Zeuzera pyrina* L. (Lepidoptera: Cossidae) pheromone (95% (*E,Z*)-2,13-ODDA: 5% (*E,Z*)-3,13-ODDA) (70 mg of active ingredients per unit) were applied to 0.4-ha treatment plots at a rate of 635 per ha (254 per 0.4 ha), approximately one twist-tie per vine. The dispensers were evenly distributed throughout the plot, and hung from the vine near the trellis wire at roughly 1 to 1.5 meters above the ground.

The leopard moth pheromone has not been previously tested in GRB mating disruption experiments. We chose to use this off-blend pheromone because it contains the same major component as the GRB pheromone and it is commercially available and significantly cheaper. Also, the findings of Johnson & Mayes (1980); Johnson et al. (1981, 1986); and Pearson & Meyer (1996) showed that it

is possible to cause mating disruption with attractants other than the complete blend.

Attract-and-Kill with Last Call-GRB

The Last Call™ GRB used in the 2003 and 2004 field trials contained 0.16% GRB pheromone, 6.0% pyrethrins (CAS 8003-34-7), and 93.984% inert ingredients. We applied 2,250 drops per ha (900 drops for 0.4 ha). Each drop contained 50 µl Last Call-GRB matrix. To achieve a uniform distribution of drops throughout the vineyard, the 900 drops were divided by the number of vines per 0.4 ha plot, approximating 2-4 drops per vine. Last Call™ GRB drops were applied to the trunks of vines ~0.5 to 1.5 meters from the ground through a calibrated hand-pump, manufactured by IPM Tech specifically for attract-and-kill gels. The pump fits in the palm of the hand and can be operated with one hand. Attract-and-kill was reapplied every 6 weeks for the duration of the season, according to the protocol determined by IPM Tech.

Chemical Control with Lorsban

Lorsban 4E® (chlorpyrifos) (Dow Chemical U.S.A., Midland, MI) (44.9% a.i.) was applied once per season at the labeled rate of 1.06 L to 378 L of water to treat 200 vines. It was applied closest to the period of greatest GRB emergence, based on earlier findings (Webb et al. 1992).

Statistical Analysis

Trap catches were counted weekly and recorded from the beginning of the season until the end of the GRB flight. Total mean number of GRB captures was analyzed by repeated measures Analysis of Variance (ANOVA), and differences among means were determined by Tukey's multiple comparison test ($P < 0.05$) (SAS Institute 2004).

RESULTS

During 2003, the number of male moths captured in traps in the areas treated with leopard moth pheromone twist-ties was significantly

fewer than the male moths captured in the untreated control and the Lorsban treatments ($F = 9.81$; $df = 3, 264$; $P < 0.0001$) (Table 1). The number of male moths captured in traps in the twist-tie sections was not significantly different from those captured in traps in the areas treated with A&K. Traps deployed in the A&K plots caught significantly fewer male moths than traps deployed in the Lorsban sections. There were no significant differences in trap catches between the Lorsban and the untreated control sections (Table 1).

During 2004, significantly fewer GRB were captured in areas treated with leopard moth pheromone twist-ties compared with the A&K and untreated control sections (Table 1) ($F = 11.42$; $df = 3, 234$; $P < 0.0001$). There were no differences between the twist-tie and Lorsban treatments or between the attract-and-kill and the untreated control sections (Table 1).

Figure 1 shows the mean number of GRB males caught per week per treatment for 2003 and 2004. The GRB captures in the untreated control sections were similar during each year. For the A&K treatments, larger numbers of GRB were captured later in the season than earlier for 2003 (Fig. 1A). Also, a relatively high number of GRB were caught in the traps in the Lorsban section in 2003. During 2004, the periods of low captures coincided with periods of extreme weather such as hurricane activity (early September). Also, trap catches in the A&K treatments were more erratic with periods of high and low captures occurring throughout the season (Fig. 1B). Grape root borer trap catches in the Lorsban treatment of 2004 was much lower than the previous year, especially later in the season during the 4-week period when Lorsban was active (Fig. 1B).

DISCUSSION

Mating Disruption

Wing traps were used to measure male moth activity throughout the vineyards and consequently, mating disruption success by trap shutdown. If GRB males were not able to locate the trap pheromone source, it is assumed that it

TABLE 1. WEEKLY MEAN $\pm$ SEM NUMBER OF GRAPE ROOT BORERS PER TRAP FOR MATING DISRUPTION, ATTRACT-AND-KILL, LORSBAN, AND UNTREATED CONTROL TREATMENTS IN FOUR FLORIDA VINEYARDS FOR 2003 AND 2004.

Treatment	Weekly mean trap capture $\pm$ SEM	
	2003	2004
Mating disruption	0.00 $\pm$ 0 c	0.00 $\pm$ 0 b
Attract-and-kill	1.07 $\pm$ 0.44 bc	3.50 $\pm$ 0.76 a
Lorsban	3.07 $\pm$ 0.73 a	0.84 $\pm$ 0.32 b
Untreated control	2.49 $\pm$ 0.62 ba	3.00 $\pm$ 0.75 a

Means in columns followed by the same letter are not significantly different ($P = 0.05$, Tukey's test)

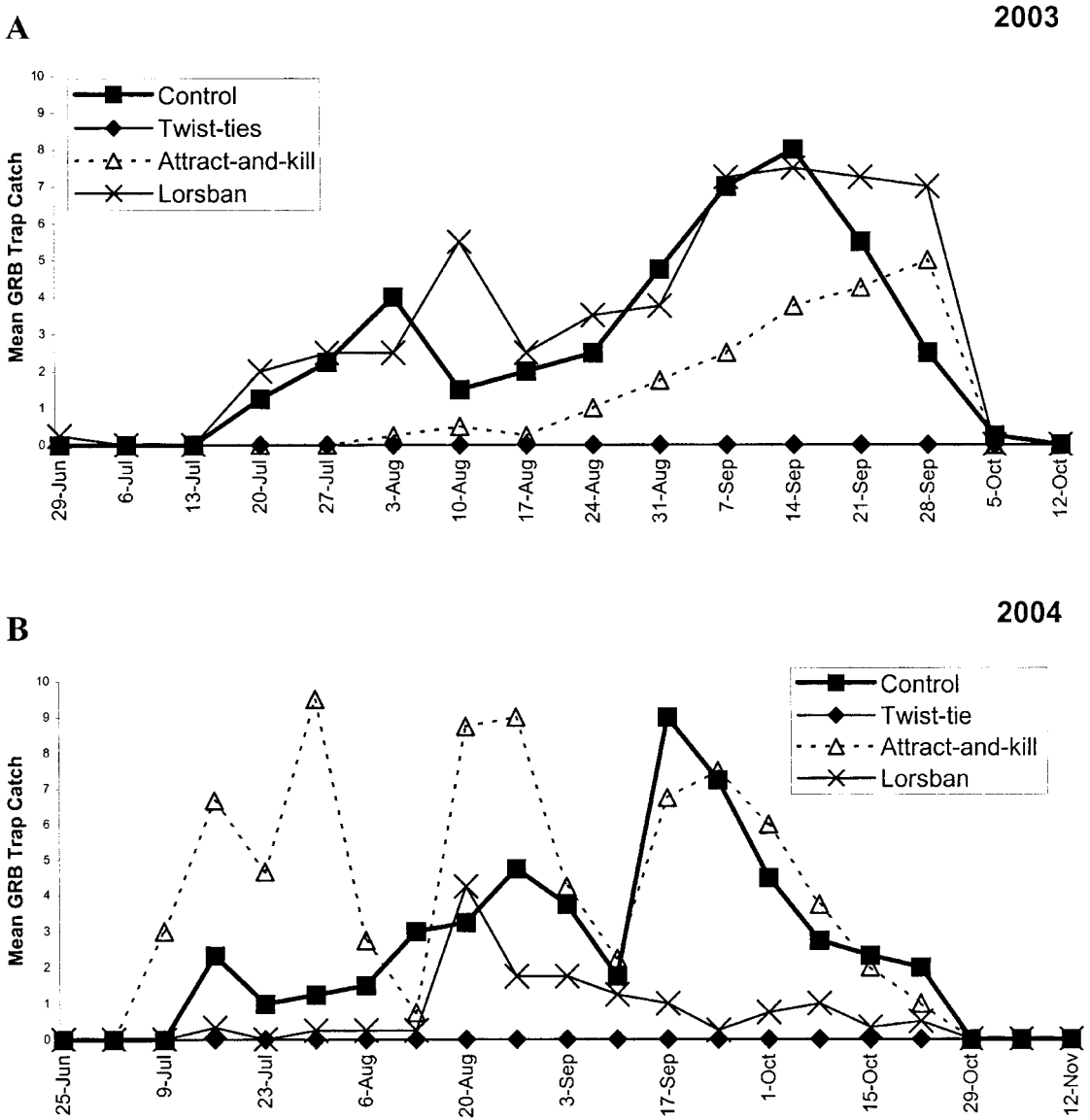


Fig. 1 A&B. Mean number of grape root borer males caught in monitoring traps per week for four treatments: untreated control, pheromone twist-ties, attract-and-kill gels, and Lorsban 4E for 2003 and 2004.

would be unlikely for them to find a calling female. In an experiment by Webb (1990), males were unable to locate caged calling females in traps in a vineyard saturated with synthetic pheromone. In our study, complete trap shutdown was achieved in all of the pheromone twist-tie treatments for both years, indicating that males were unable to orient to the female pheromone source. Therefore, it is reasonable to assume that disruption of mating occurred.

It is possible that some mating may still occur despite complete trap shutdown. In addition, gravid females have been observed immigrating

into pheromone-saturated vineyards and laying eggs (Johnson et al. 1986). This behavior is unexplained since Pearson & Schal (1999) demonstrated a weak attraction to the GRB pheromone by mated females. It is not known how often or to what extent gravid females enter pheromone-saturated vineyards, but it can have serious consequences on a mating disruption program.

Trap shutdown alone does not prove that mating disruption has occurred. Previous studies have confirmed mating disruption success in pheromone-saturated vineyards by other methods in addition to trap shutdown including deter-

mining if males could locate caged calling females, counting pupal skins, and production of fertile or infertile eggs by females caught within the vineyards. Webb (1990) showed a significant degree of mating disruption by trap shutdown as well as the reduction of pupal skin counts indicating a correlation between the two. Johnson et al. (1991) and Yonce (1981) also showed a correlation between reductions of pupal skin counts and trap shutdown. Attempts were made to recover pupal skins but a shortage of labor and inclement weather coupled with low recovery of pupal skins forced us to cancel this activity.

We used twist-ties with the pheromone of the leopard moth *Zeuzera pyrina* L. (Lepidoptera: Cossidae) 95% (*E,Z*)-2,13-ODDA: 5% (*E,Z*)-3,13-ODDA. It shares the major component with the GRB, although in a smaller percentage (95% rather than 99%), and has a different minor component. Due to the fact that it was possible to cause mating disruption with off-blends (Hodges et al. 1984; Minks and Cardé 1988), and the fact that the leopard moth pheromone was commercially available and significantly cheaper, it was chosen as our disruption tool.

Attract-and-Kill

Trap catches in the A&K treatment plots during 2003 were not significantly different from the twist-tie sections, indicating low activity by male moths in the area treated with A&K. It is unclear whether the success of the A&K was due to insecticide poisoning of the males because of contact with A&K or mating disruption. The A&K treatment did not work as well in 2004 when traps showed no difference from the untreated control. Traps in the A&K treatments caught 48% of the total GRB captured in 2004, compared with only 13% in 2003. Initially, the reason for this was unclear. Further investigation revealed that the producers of A&K failed to include the correct pheromone blend in the 2004 batch. Analysis of the gel revealed the ratio to be 95:5 (*E,Z*)-2,13-ODDA: 4.5%(*Z,Z*)-3,13-ODDA instead of 99:1. Previous studies demonstrated that a 95:5 ratio was mostly unattractive to male GRB (Snow et al. 1987).

We observed that the A&K drops often deteriorated quite rapidly under Florida weather conditions. During the summer growing season, Florida vineyards usually experience powerful storms with pelting rain, intense heat, and solar radiation. These conditions may affect the stability and longevity of the A&K. The protocol for A&K indicates that it must be reapplied every 6 weeks. However, we noticed that many drops were almost dry after 3 weeks and totally missing during the 4th through 6th week. Only in a few instances did we observe drops that lasted the entire 6-week period. Attract-and-kill warrants further investigation to determine the frequency of appli-

cation under Florida conditions and its overall effectiveness.

The Lorsban treatment was included in the study as a standard chemical treatment for GRB control. Lorsban primarily controls first instars as they emerge from eggs and burrow to the roots. It can also reduce the number of adults as they emerge from their cocoons. In 2003, there were no significant differences in the number of male moths caught in traps between the untreated (control) and the Lorsban-treated sections. However, during 2004, traps in the vineyards treated with Lorsban caught significantly fewer GRB than the untreated controls. The reason for the differences between 2003 and 2004 is not clear.

Costs

At our current application rates, LastCall-GRB costs \$250 per hectare and it requires roughly 2.5 h/ha to apply and 3-4 applications per season (under Florida conditions). This could be labor-intensive, depending on the size of the vineyard. Future studies should evaluate how many drops per hectare would provide effective control. For instance, instead of 900 drops per 0.4 ha, perhaps the drops (1 drop = 0.05 g) could be consolidated into larger amounts and applied on fewer vines.

Our pheromone twist-tie application rate of 635 units per ha is fairly high. Practically, 635 twist-ties per ha may be expensive for farmers (\$287/ha compared with \$32-\$65/ha for Lorsban). It takes an average worker 3 h/ha to deploy the twist-ties. However, the pheromone lasts the entire season under normal conditions.

Future studies should focus on different deployment tactics as well as rates of application (number of twist-ties per ha). It may be important to compare the leopard moth pheromone to the true GRB blend in further mating disruption studies. This was beyond the scope of this study, but future studies should also incorporate the counting of pupal skins as a means to determine GRB reductions from the treatments.

Mating disruption with the use of the leopard moth (*Zeuzera pyrina*) pheromone may be an effective, reduced-risk strategy for controlling GRB, and a good alternative to conventional chemical control. Attract-and-kill technology may also be a potentially effective strategy for GRB control, but more research is needed.

ACKNOWLEDGMENTS

We thank the grape growers who participated in this research and contributed their farms and time, especially Roger and Marcia Price (Harmony Vineyard), Bob, RuthAnn, and Kellie Thropp (Log Cabin Vineyard), Jim and Lois Hangar (Orange Blossom Vineyard), and Robert and Bonniejean Paulish (Blue Heron Vineyard). We also thank the Florida Grape Grower's Association for funding this research for three consecutive years. We ap-

preciate the assistance of the staff from the Small Fruit and Vegetable IPM Lab at the Department of Entomology and Nematology at the University of Florida, especially Gisette Seferina and Alejandro Arevalo. We thank Dr. Robert McSorley and Dr. Susan Webb for reviewing earlier drafts of this manuscript. We acknowledge Dr. John R. McLaughlin and the staff of IPM Tech, Inc. for supply of LastCall™ GRB. We are especially indebted to Jack W. Jenkins with Pacific Biocontrol Corp. for his assistance in acquiring the leopard moth twist-ties. We thank Dr. Peter Teal with the USDA in Gainesville Florida for his assistance in analyzing pheromone samples.

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